CANINE INTERVERTEBRAL DISC DISEASE: THE CURRENT STATE OF KNOWLEDGE

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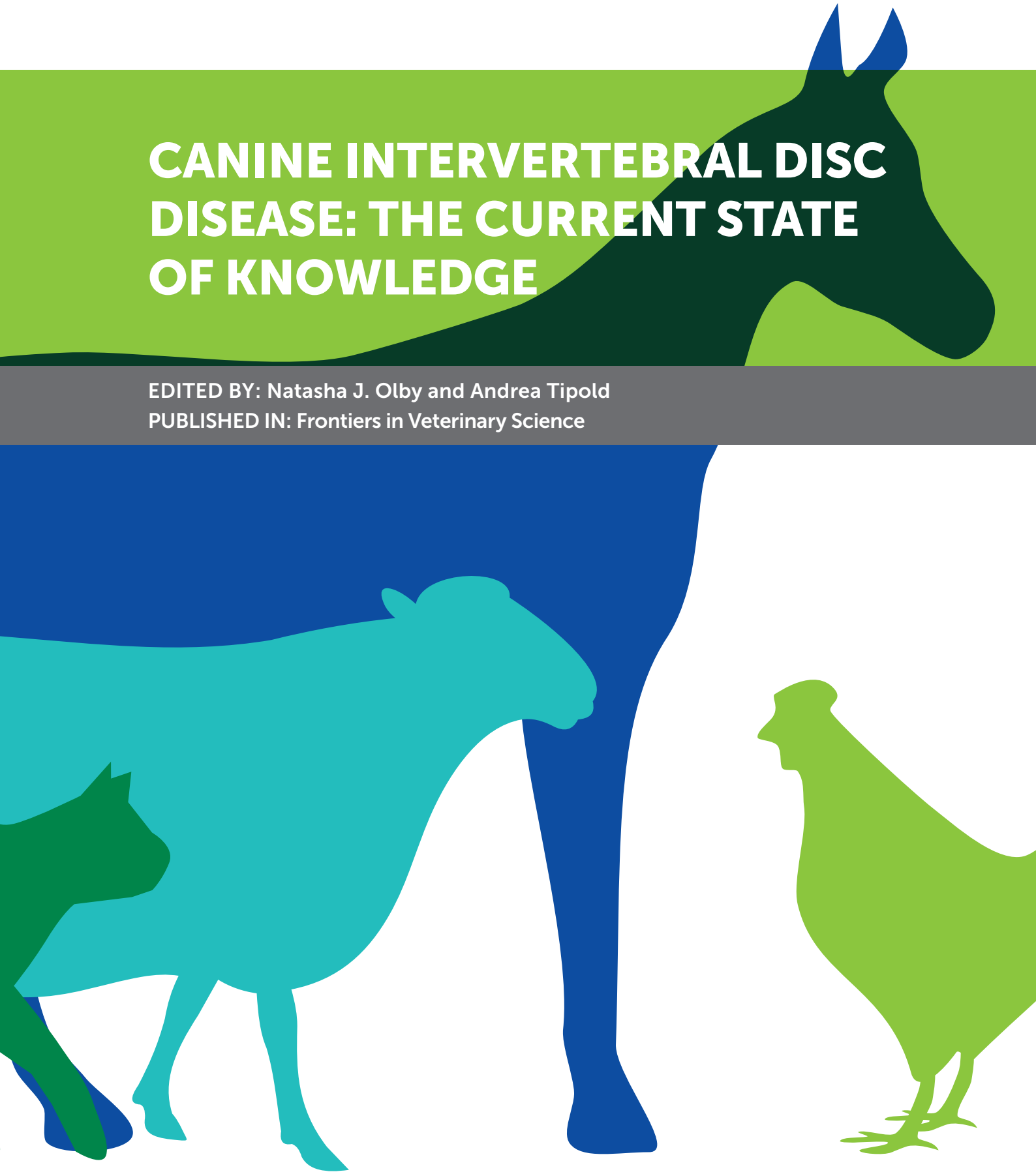

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CANINE INTERVERTEBRAL DISC DISEASE: THE CURRENT STATE OF KNOWLEDGE

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EDITORIAL
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Editorial: Canine Intervertebral Disc Disease: The Current State of Knowledge

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Keywords: intervertebral disc disease, canine, spinal cord injury, disc extrusion, disc degeneration

Editorial on the Research Topic

Canine Intervertebral Disc Disease: The Current State of Knowledge

Intervertebral disc disease in dogs was first described in the late 1800s with a case report in which the author puzzled over whether the cartilaginous mass within the vertebral canal of a dachshund could be related to the underlying intervertebral disc. Degeneration of intervertebral discs was recognized and described in detail in the 1940s and 1950s and since that time the seminal work of Hansen has imprinted types I and II intervertebral disc disease on every veterinarian. Due to the popularity of certain breeds of dog, intervertebral disc disease is by far the most common cause of canine paralysis and is treated on a daily basis by veterinarians. More recently, the advent of accessible magnetic resonance imaging and dramatic advances in molecular tools have led to expansion and refining of our categorization of intervertebral disc disease, understanding of the genetic risk factors, and improved ability to describe, quantify, and treat the associated spinal cord injury.

This collection of 10 articles takes a holistic look at what we now know about intervertebral disc disease. One of the most exciting advances in the field is the identification of a strong genetic risk factor in chondrodystrophic breeds of dog. The finding of an expressed FGF4 retrogene on chromosome 12 in these breeds of dog is described in detail by Dickinson and Bannasch. This paper documents the discovery of the variant in Nova Scotia Duck Tolling Retrievers and then its validation across other breeds. Mechanisms by which overexpression of FGF4 might exert its influence on the developing intervertebral disc are presented and discussed.

The impact of this finding has been incorporated into a discussion of the classification of intervertebral disc disease in a paper by Fenn et al. This paper also reviews the numerous types of acute intervertebral disc extrusion that are now well-recognized thanks to MRI and incorporates them into a classification scheme. This scheme classifies disc herniation according to the degenerative process underlying it, and takes into account genetic, imaging, and pathological findings. Diagnosis of intervertebral disc degeneration and herniation using imaging is presented in careful detail in a review paper by da Costa et al.. This paper describes key findings reported in the literature using plain radiography, myelography, and computed tomography but then updates the reader with a description of the most current magnetic resonance imaging (MRI) research. Specific imaging features of different types of IVDD are described and illustrated in detail and advanced MR imaging techniques are explained and their potential for future clinical utility is discussed.

Establishing prognosis for recovery, and understanding factors that alter prognosis is important for owners and veterinarians, and can play a role in optimizing case allocation in clinical trials. There are abundant data on prognosis for intervertebral disc extrusions and this literature is summarized and presented in tabular form in a paper on prognosis by Olby et al. Signalment,

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clinical presentation, imaging, plasma, and CSF biomarkers are all discussed. These data are taken into consideration in a paper describing Current Approaches to the Management of Acute Thoracolumbar Disc Extrusion by Moore et al. This paper is careful to identify knowledge gaps where high level evidence is lacking and draws on published evidence available to make recommendations on surgical vs. conservative management, timing, type and extent of surgery, the role of fenestration, and adjunctive neuroprotective and post-operative care.

Given the high frequency of intervertebral disc-induced spinal cord injury, there is much interest in performing well-designed clinical trials to identify the best therapy for this condition. The many different considerations required to design a clinical trial are described by Jeffery et al.. This article explains the pros and cons of different styles of trial and explains how the information generated might be used clinically. Readers of the article by Leiws et al. on emerging and adjunctive therapies for disc-induced spinal cord injury will learn of the myriad of different therapies that have been investigated for this condition. The evidence for efficacy of locally applied therapies such as laser and pulsed electromagnetic therapy, different pharmaceuticals, cellular therapies, adjunctive surgical techniques and rehabilitation among others are all presented with a careful consideration of the evidence available.

The consequences of acute disc extrusion on the spinal cord are described by Spitzbarth et al. in a well-illustrated paper on the pathology of disc induced spinal cord injury. This paper gathers together detailed descriptions of histopathological changes and highlights what is known on the molecular processes underlying these changes with a particular emphasis on the complex inflammatory response. In addition, the unique changes that occur with progressive myelomalacia are described. Less frequently discussed, but extremely important outcomes of spinal cord injury are addressed in two articles, by Lewis et al. and Granger et al. The article by Leiws et al. takes a careful look at the recovery of walking in dogs that do not recover pain perception following severe, disc induced spinal cord injuries. This paper explains what is known about the circuitry behind locomotion and discusses the evidence presented for the mechanism of recovery of walking in this population of dogs. The article by Granger et al. focuses on urinary and fecal incontinence, a consequence of spinal cord injury that is particularly difficult to manage. The pathways involved in normal control are explained and the impact of spinal cord injury on those pathways is discussed in detail. The frequency and circumstances in which there are long term issues with incontinence are described and methods of management are discussed.

The purpose of this topic is to provide a detailed summary of what is known about intervertebral disc disease in dogs, harnessing the great strides made in recent years. The majority of these articles were written by members of the Canine Spinal Cord Injury Consortium, CANSORT SCI, a group of veterinarians actively involved in the field of spinal cord injury research. These papers collectively have provided a clear summary of the genetic, imaging, and histopathological characteristics of intervertebral disc disease, allowing the development of a new classification system. The detailed summaries of the body of evidence for a wide range of different therapies, and of the factors that influence recovery of motor and autonomic function enable the reader to weigh the most appropriate therapeutic approach for their cases and to identify unexpected outcomes that might need intervention. Our hope is that this collection will provide an accessible and efficient means of updating knowledge of this extremely broad topic and prove to be a springboard from which new ideas can flow.

AUTHOR CONTRIBUTIONS

NO and AT have overseen the entire Research Topic. NO wrote the original draft of this editorial with editing input and advice from AT. All authors contributed to the article and approved the submitted version.

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REVIEW
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Current Understanding of the Genetics of Intervertebral Disc Degeneration

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Premature degeneration of the intervertebral disc and its association with specific chondrodystrophic dog breeds has been recognized for over a century. Several lines of evidence including disease breed predisposition, studies suggesting heritability of premature intervertebral disc degeneration (IVDD) and association of a dog chromosome 12 (CFA 12) locus with intervertebral disc calcification have strongly supported a genetic component in IVDD in dogs. Recent studies documenting association of IVDD with an overexpressing *FGF4* retrogene on CFA 12 have opened up new areas of investigation to further define the pathophysiology of premature IVDD. While preliminary data from studies investigating FGF4 retrogenes in IVDD implicate FGF4 overexpression as a major disease factor, they have also highlighted knowledge gaps in our understanding of intervertebral disc herniation which is a complex and multifactorial disease process.

Keywords: chondrodystrophy, fibroblast growth factor 4, heritable, intervertebral disc degeneration, retrogene

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INTRODUCTION

The list of inherited neurological disorders in companion and production animals is ever expanding. There are over 120 known genetic variants for neurological disorders in dogs alone (1), and with advances in molecular genetic technology and consistently decreasing costs, the list is continuing to expand at a rapid rate. Many of these disorders are associated with breed specific syndromes and have a relatively "localized" effect on the health of the overall dog population. The vast phenotypic diversity within domesticated dogs is the result of selection for genetic variants that define key traits such as skeletal size, body size, skull shape, snout length, coat color, leg length, and other breed-defining characteristics (2, 3). Beyond the "desirable" morphological traits, undesirable "disease" syndromes may be associated with these genetic loci due to either multiple phenotypic sequelae of specific variants, or associated genetic variants carried within long regions of linkage disequilibrium. This can be particularly problematic when disease causing genes define key characteristics of the breed e.g., leg length or head shape resulting in the variant being essentially fixed (homozygous for the associated allele) in a majority if not all animals within certain breeds; premature degeneration of the intervertebral disc (IVD) in chondrodystrophic dog breeds provides a quintessential example of this dilemma. The high penetrance of intervertebral disc disease (IVDD) associated genes in many popular dog breeds presents a daunting clinical challenge and results in millions, if not billions of dollars of annual veterinary treatment-related expense and suffering. However, as with the profound impact of preventative and screening practices in cancer medicine, the potential for genetic interventions to have dramatic effects on clinical IVDD in dogs far outweighs any likely impacts from advances in specific treatments.

"SHORT LIMBED" DOGS AND IVDD

Extreme differences in limb length define many of the dog breeds around the world, and the association between specific "short-legged" breeds and premature intervertebral disc degeneration has been documented since the early twentieth century [referenced in (4, 5)] Skeletal dysplasia is a general term describing abnormalities of growth and development of cartilage and/or bone and associated alterations in stature (6, 7). The molecular genetic underpinnings of limb length variability in dog breeds are becoming more completely understood although many unexplained types of skeletal dysplasia remain. Several skeletal dysplasias in specific dog breeds have been associated with mutations in members of the collagen gene family or its binding proteins (8–10), fibrilin related protein (11), as well as an altered sulfate transporter protein (12). However, overexpression of *FGF4* associated with insertion of *FGF4* retrogenes on CFA12 and CFA18 appear to have broader influences on limb length across many breeds and are the only genes to have been implicated in body size in across-breed association studies (3, 13–16). While many breed specific mutations are considered undesirable (8–10, 12) some of these genes have been under positive selection in specific breeds due to their effects on height, despite associated pathologies including glaucoma and IVDD (11, 13).

Terminology applied to skeletal dysplasia subgroups can be confusing; the term chondrodysplasia covers a broad group of skeletal dysplasia disorders in humans with abnormal development of the endochondral components of the skeletal system (present at birth) and has been used to describe extreme differences in limb length in several dog breeds, such as the Basset Hound, Dachshund, and Pekingese (14). Historically, the term chondrodystrophy has been applied as a more general terminology to include terms such as chondrodysplasia. In the veterinary literature it has come to be used as a term describing "short limbed" dogs with skeletal dysplasia that additionally have progressive degeneration of the intervertebral disc after birth, with the progressive nature of the IVDD informing the use of the term "dystrophy" (4, 5, 17). While the genetic alterations listed above have all been associated with altered limb length, only the overexpression of *FGF4* secondary to retrogene insertion on CFA12 has also been specifically associated with premature degeneration of the intervertebral disc (13).

THE *FGF4* GENE

FGF4 is one of a family of 18 secreted canonical FGF proteins that interact with 4 signaling tyrosine kinase FGF receptors (FGFR1-4) (18). The FGF4 subfamily (FGF4,5,6) bind to receptors expressed predominantly in mesenchymal tissues (FGFR1c, 2c, 3c, 4), and as with most FGFs they have important roles in early stages of embryonic development and organogenesis (18). *Fibroblast Growth Factor 4* is most highly expressed in the embryonic ectoderm, axial, paraxial, and lateral plate mesoderm, and tail bud (19). Later in development, *Fgf4* is highly expressed in the apical ectodermal ridge of the developing limb bud, as well as somites, which go on to form the vertebral column and non-nuclear components of the intervertebral disc (19–21). FGF signaling is required for proper embryonic axial growth and segmentation and *Fgf4Fgf8* murine hypomorphs are characterized by altered vertebral morphology and smaller limb buds (20, 22). In a mouse model, creation of a gain of function *Fgf4* copy to replace an inactive *Fgf8* gene was able to rescue limb development; however, it also caused abnormal tissue deposition and postaxial polydactyly, highlighting that levels of FGF proteins throughout embryonic development must be properly controlled for normal limb formation (23). FGF signaling is important in development of the ear, and both *Fgf4* and *Fgfr1* are expressed in embryonic structures that give rise to the pinna (19, 24). Hypomorphic alleles of *Fgf4* and *Fgfr1c* (25, 26) both result in reduced pinna size although it is unclear whether FGF4 overexpression specifically results in increased pinna size as is common in many chondrodystrophic dog breeds. Expression of *Fgf4* is not documented specifically in the nucleus pulposus during development (19, 27, 28), however FGF signaling is involved in the differentiation of notochordal cells (29), is present in the developing end plate and annulus fibrosus (28) and mutual interaction between the notochord and vertebral bodies are instrumental in the proper formation of the IVD (21).

FGF4 RETROGENES

Two separate retrogenes, derived from the parental *FGF4* gene on CFA18, have been described in dogs resulting in various degrees of skeletal dysplasia and disproportionate dwarfism (13, 14). An *FGF4* retrogene on CFA18, 25 Mb from the parental gene, associated with marked limb length variation (14), and an *FGF4* retrogene on CFA12 associated with chondrodystrophy characterized by moderate variation in limb length and an odds ratio of 51.23 (95% CI = 46.69, 56.20) for Hansen Type I intervertebral disc disease (13). The CFA12 *FGF4* retrogene was identified concurrently by separate genome wide association studies investigating alteration in limb length in Nova Scotia Duck Tolling Retrievers and with IVDD across breeds (13). The same chromosomal region had previously been identified associated with limb morphology in Portuguese Water Dogs and intervertebral disc calcification in Dachshunds without defining a causative mutation (30, 31). Comparatively, activating mutations of the FGFR3, one of the receptors for FGF4, are responsible for some of the most common causes of disproportionate dwarfism in humans including achondroplasia (6, 32). Intervertebral disc degeneration is also a common finding in human achondroplasia, however histopathological characterization is lacking, and many factors including vertebral malformations, spinal canal stenosis and secondary degenerative changes are a major component of disease pathology (33).

Retroposition is a gene duplication mechanism that utilizes an RNA intermediate to randomly insert intronless retrocopies of genes into the genome following reverse transcription (**Figure 1**) (34, 35). Reverse transcriptase activity can be provided from a variety of sources, however in mammals it is most commonly associated with long interspersed nuclear elements (LINEs) (35).

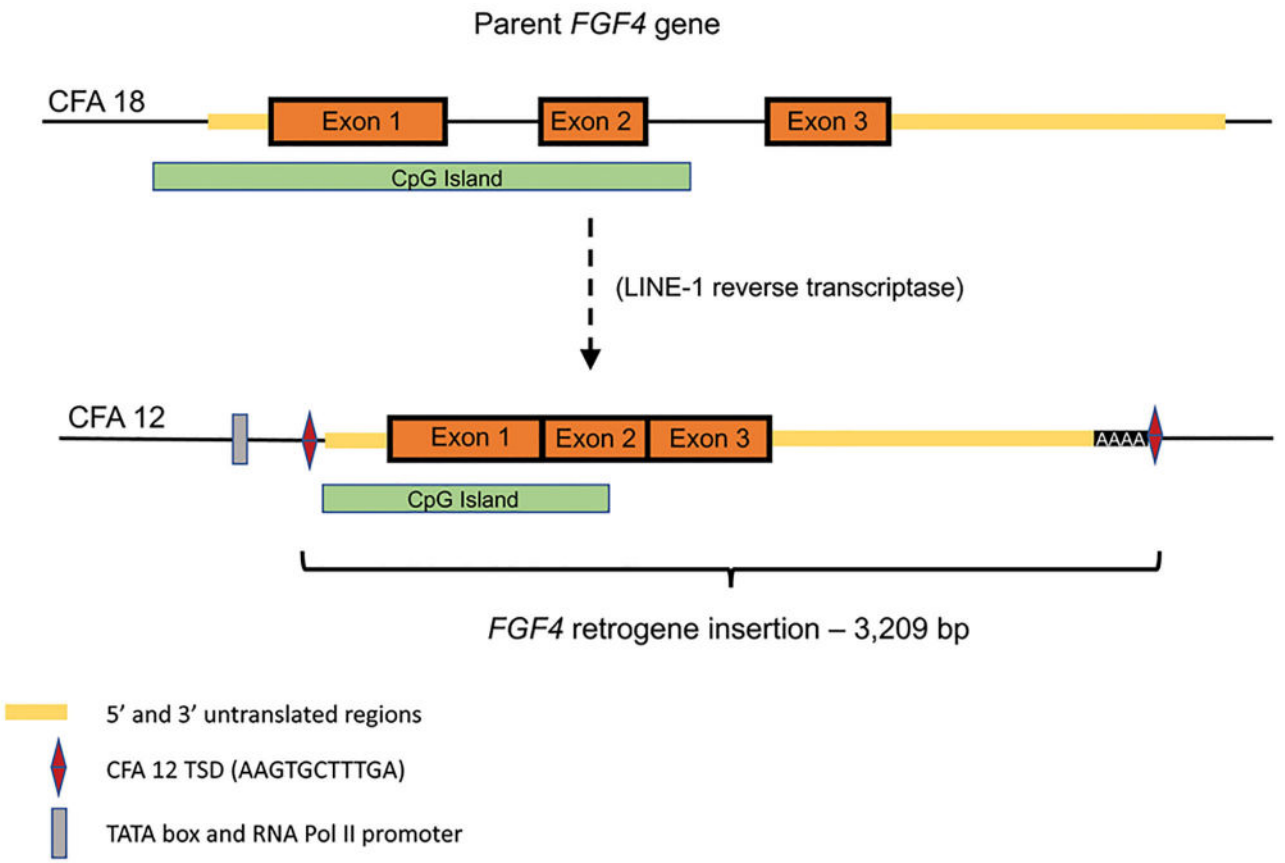

FIGURE 1 | Schematic of endogenous *FGF4* (CFA18) retrotransposition to CFA12:33,710,178 (canFam3): Predicted TATA box at chr12:33,709,940-947 (canFam3) and predicted RNA Pol II promoter at chr12:33,709,964-976 (canFam3) are shown. The retrogene insert includes all of the predicted 3′UTR, followed by 42 adenine residues and the duplicated target site duplication (TSD) sequence (AAG TGC TTT GA). Retrotransposed sequence also includes a large CpG island.

LINEs are one of several types of transposable DNA sequences that have the ability to change their position within a genome. LINEs utilize their own reverse transcriptase activity to copy and paste themselves into new locations, however this activity may also create DNA copies of mRNA from functional genes which, if inserted, may result in retrogene copies. Both of the canine *FGF4* retrogenes appear to have arisen from RNA retrotransposed by LINE-1 integrase and reverse transcriptase, including flanking target site duplications (TSDs) and polyA tracts (class 1 templated sequence insertion polymorphism) (36).

Retrocopies have historically been considered to be inactive in most cases due to lack of appropriate regulatory elements as well as genetic alterations that remove open reading frames, and documented association with disease states is relatively uncommon. However, retrotransposition is believed to play an important role in genome evolution (35) and as the field develops, examples of disease related retrotransposition are likely to increase in frequency. Currently only the CFA12 and CFA18 *FGF4* retrogenes have been associated with clinical phenotypes in dogs.

The CFA12 *FGF4* retrogene is 3,209 bp long (Gen Bank accession no. MF040221) and consists of the parental chromosome 18 *FGF4* exons (**Figure 1**) and a majority of the 5′-untranslated region (UTR) including the transcription start site and cis-regulatory elements including a TATA box, RNA Pol II promoter sequences and many conserved transcription factor binding sites. The insertion is at 33.7 Mb between the *OGFRL1* and *RIMS1* genes. [The CFA18 *FGF4* retrogene is a similar size (2,665 bp) with common 5′ UTR and exons but a shorter 3′ UTR and is inserted within a LINE element on the parental chromosome 18]. The CFA12 *FGF4* retrogene is transcriptionally active resulting in a 20-fold increase in FGF expression in neonatal intervertebral disc from dogs homozygous for the CFA12 *FGF4* retrogene insertion compared to disc from neonatal dogs with just the parental *FGF4* genes (13). Although the CFA12 *FGF4* retrogene was inserted near sequences with promoter properties, it is more likely that the *FGF4*-associated CpG island (**Figure 1**) included in the retrogene and shared with other species, including humans, is driving expression (37). CpG islands are genomic regions with a higher frequency of C-G dinucleotides than the genome average, often co-localizing with gene promoters and having important roles in transcriptional regulation. In fact, retrogene expression has been shown to be dependent on the genomic context of its insertion

and contribution of CpG islands more than the use of nearby promoters (38). Seven of eight genes in the direct neighborhood of the CFA12 *FGF4* insertion are actively expressed in neonatal IVD and vertebral body (13) suggesting that the retrogene was inserted in a gene milieu conducive to expression in IVD. Specific embryonic expression of FGF4 in dogs with 2, 4, or 6 copies of the gene (from parental, or either retrogene) is to be defined, however quantitative and/or tissue specific differences associated with the CFA12 *FGF4* retrogene expression may explain the IVD-associated phenotype seen with CFA12 but not the CFA18 *FGF4* retrogene expression.

IVDD AND *FGF4* RETROGENES

Although we have little ancient historical data documenting IVDD prior to the late nineteenth century (39), we do know that descriptions of short-legged dog breeds go back over 4000 years (40). These include depiction of a short-legged dog on a tomb wall in Egypt (2000 BC), ceramic works of the Colima Dog in Mexico from AD 300-600, the description of the Turnspit in the first English dog book in 1576, and the description of the first Dachshund in Germany in 1735. Although the genetic makeup of these dogs is unknown, it is likely that one or both of the *FGF4* retrogenes have been involved in the generation of these ancient short legged phenotypes. Retrotransposition of the *FGF4* gene has occurred at least twice in recent history, although the lack of accumulation of mutations in either retrocopy suggest these events are still relatively recent in genomic evolutionary terms. Once the *FGF4* retrogene(s) appeared and produced an obvious phenotype, strong selection was likely applied to retain them.

Differential expression of the CFA12 and CFA18 *FGF4* retrogenes generally reflects the morphological phenotype and susceptibility to IVDD we recognize in clinical practice (**Table 1**, **Figure 2**). Breeds carrying one *FGF4* retrogene copy (CFA12 or CFA18) tend to have skeletal dysplasia with moderately short limbs, while those carrying both copies, such as Dachshunds, Corgis and Bassett Hounds, have the most severe form of disproportionate dwarfism. The breeds with a higher frequency of the CFA12 *FGF4* insertion are the same breeds identified in the last 50 years as being predisposed to IVDD (4, 5, 41). Although the CFA18 *FGF4* retrogene is found commonly in breeds that also have the CFA12 *FGF4* retrogene and IVDD (Dachshunds, Corgis, Bassett Hounds), it does not appear to be directly associated with development of IVDD since several of the highly susceptible short legged breeds carrying the CFA12 *FGF4* retrogene such as Beagles, French Bulldogs and Cocker Spaniels do not carry the CFA18 *FGF4* retrogene and interestingly contrast with the short legged breeds such as Cairn Terriers and West Highland White Terriers that are rarely reported to have IVDD and carry only the CFA18 *FGF4* retrogene (5, 13, 42).

FGF4 GENE DOSAGE

While it is clear that the CFA12 *FGF4* retrogene is highly associated with premature chondroid metaplasia and degeneration of the intervertebral discs (13, 42, 43), assessing the effects of the CFA12 *FGF4* retrogene is complicated as many high susceptibility breeds also carry the CFA18 *FGF4* retrogene, and many breeds susceptible to IVDD are homozygous for both of the *FGF4* retrogenes making assessment of relative risk challenging. It is also probable that there are additional modifying or causative genetic loci as well as morphological and environmental factors contributing to the overall clinical presentation of IVDD across breeds (44–50). We do know that the presence of the CFA12 *FGF4* retrogene alone is sufficient to cause loss of the normal physaliferous notochordal cells and replacement of the nucleus pulposus by cartilaginous material in puppies as young as 10 weeks of age (**Figure 3**) (43). Sufficiency was demonstrated in Nova Scotia Duck Tolling Retriever dogs homozygous for the CFA12 *FGF4* retrogene with no CFA18 *FGF4* retrogene and compared to breed matched controls with neither retrogene (43). The effect of the CFA12 *FGF4* retrogene on IVD degeneration also appears to be dominant since IVDD is seen in dogs that are homozygous or heterozygous for the CFA12 insertion (13, 42).

Allele Frequency

Genotyping of over 3,000 dogs from 75 breeds showed that the CFA12 *FGF4* retrogene was present in 40 breeds, the CFA18 *FGF4* retrogene in 32 breeds and both retrogenes in 23 breeds (**Table 1**) (42). The CFA12 *FGF4* retrogene has an extremely high allele frequency (>90%) in the general population of breeds such as Beagles, Dachshunds, French Bulldogs, and most spaniel breeds, and not surprisingly, this frequency increases when looking at the population of dogs that present with clinical IVDD (42). Batcher et al. investigated 569 dogs presenting for decompressive surgical treatment of intervertebral disc disease and described differences in cases typified by calcified/mineralized disc herniations compared to those with "fibrous" type herniation with respect to retrogene frequency. Consistent with typical clinical neurological caseload, 75% of all cases involved throacolumbar discs, and affected dogs frequently had 1 or 2 copies of the CFA12 *FGF4* retrogene (allele frequency 0.636). Consistent with the skeletal dysplasia phenotype common to both retrogenes, presence of 1 or 2 copies of either the CFA12 or CFA18 *FGF4* retrogenes was also associated with "smaller" dogs (based on body weight).

Age Related Factors

Defining age of onset of IVDD is challenging for many reasons, not least because histopathological evidence of premature degeneration is already present in affected dogs before 1 year of age (4, 41, 43). Presence of 1 or 2 copies of the CFA12 *FGF4* retrogene is associated with a younger age of presentation for decompressive surgery (mean 6.1 vs. 8.5 years) (**Figure 2**) (42). Interestingly and as reported previously, French Bulldogs (51, 52) that are essentially fixed for the CFA12 *FGF4* retrogene had a significantly younger age at surgery (median 3.7 years) compared to other breeds homozygous for the CFA12 *FGF4* retrogene such as Dachshunds (median age 6.5 years) (42). Number of CFA 12 *FGF4* retrogene copies does not appear to affect age at presentation for surgery (42). The role of the CFA 18 *FGF4* retrogene is less clear; a single copy was associated with a younger

TABLE 1 | *FGF4* retrogene allele frequencies.

		CFA12 *FGF4* Retrogene				CFA18 *FGF4* Retrogene			
Breed	**Total**	**0**	**1**	**2**	**Frequency**	**0**	**1**	**2**	**Frequency**
Beagle	29	0	0	29	1.00	19	0	0	0
Cavalier King Charles Spaniel	25	0	0	25	1.00	6	0	0	0
Clumber Spaniel	6	0	0	6	1.00	6	0	0	0
Dachshund	509	0	30	479	0.97	3	9	482	0.98
Cocker Spaniel, American	13	0	1	12	0.96	6	0	0	0
Cocker Spaniel, English	14	0	1	13	0.96	14	0	0	0
Bulldog, French	113	0	14	99	0.94	71	2	0	0.01
Dandie Dinmont Terrier	28	1	5	22	0.88	0	1	27	0.98
Welsh Corgi, Pembroke	63	3	15	45	0.83	0	2	61	0.98
Welsh Corgi, Cardigan	7	1	1	5	0.79	0	2	4	0.83
Skye Terrier	13	2	2	9	0.77	0	0	9	1.00
Basset Hound	38	3	20	15	0.66	1	4	27	0.92
Pekingese	32	5	15	12	0.61	1	1	22	0.94
Coton de Tulear	14	3	6	5	0.57	0	1	12	0.89
Poodle, Miniature and Toy	119	28	46	45	0.57	38	10	5	0.19
Springer Spaniel, English	23	10	8	5	0.39	13	0	0	0
Nova Scotia Duck Tolling Retriever	172	69	87	16	0.35	7	0	0	0
Shih Tzu	128	69	42	17	0.30	0	3	16	0.92
Bichon Frise	79	51	23	5	0.21	1	2	5	0.75
Mixed Breed	678	477	148	53	0.19	495	118	65	0.18
Chihuahua	224	170	46	8	0.14	4	15	43	0.81
Jack Russel Terrier	14	11	3	0	0.11	3	1	1	0.3
Danish Swedish Farmdog	29	23	6	0	0.10	12	0	0	0
Chesapeake Bay Retriever	41	34	7	0	0.09	10	0	0	0
Brittany	17	17	0	0	0.07	6	0	0	0
Maltese	95	85	8	2	0.06	0	0	27	1.00
Pinscher, Miniature	9	8	1	0	0.06	9	0	0	0
Schnauzer, Miniature	9	8	1	0	0.06	9	0	0	0
Scottish Terrier	12	11	1	0	0.04	0	1	6	0.93
Australian Shepherd	48	45	3	0	0.03	37	0	0	0
Yorkshire Terrier	15	14	1	0	0.03	0	0	5	1.00
German Shepherd Dog	25	24	1	0	0.02	15	0	0	0
Labrador Retriever	38	37	1	0	0.01	28	0	0	0
Australian Cattle Dog	12	12	0	0	0	5	0	0	0
Bernese Mountain Dog	11	11	0	0	0	5	0	0	0
Border Collie	5	5	0	0	0	5	0	0	0
Border Terrier	6	6	0	0	0	6	0	0	0
Boston Terrier	7	6	1	0	0	7	0	0	0
Bull Terrier	5	5	0	0	0	5	0	0	0
Bulldog, English	13	13	0	0	0	5	0	0	0
Cairn Terrier	10	10	0	0	0	0	1	9	0.95
Doberman Pinscher	23	23	0	0	0	10	0	0	0
Fox Terrier	12	12	0	0	0	7	0	1	0.13
Golden Retriever	14	14	0	0	0	4	0	0	0
Great Dane	13	13	0	0	0	5	0	0	0
Irish Setter	8	8	0	0	0	5	0	0	0

(Continued)

TABLE 1 | Continued

Breed	Total	CFA12 *FGF4* Retrogene				CFA18 *FGF4* Retrogene			
		0	1	2	Frequency	0	1	2	Frequency
Newfoundland	14	14	0	0	0	5	0	0	0
Norwich Terrier	19	19	0	0	0	3	0	16	0.84
Poodle, Standard	55	55	0	0	0	30	0	1	0.03
Pug	9	9	0	0	0	7	0	0	0
Rottweiler	15	15	0	0	0	5	0	0	0
Shetland Sheepdog	13	13	0	0	0	5	0	0	0
Siberian Husky	13	13	0	0	0	5	0	0	0
St. Bernard	12	12	0	0	0	5	0	0	0
Weimaraner	14	14	0	0	0	5	0	0	0
West Highland White Terrier	10	10	0	0	0	0	0	8	1.00
Whippet	6	6	0	0	0	5	0	0	0

age at surgery, however no difference was seen comparing 2 copies vs. zero copies or 1 and 2 copies (42).

Calcification: Type I vs. Type Ii

Calcification/mineralization of intervertebral discs, either surgically or radiographically, is typically used as a surrogate for the presence of Hansen type I vs. Hansen Type II disc disease since it is rarely present in the latter (5, 53). Consistent with previous reports (5, 53–55), studies looking at IVD calcification and CFA12 *FGF4* retrogene frequency also described dogs with calcified (Type I) IVD to be significantly smaller (median 8.1 vs. 25 kg) and to have a significantly younger age at presentation for surgery (5.5 vs. 9 years) compared to non-calcified IVD (Type II) dogs (42). Supportive of the proposed role of the CFA12 *FGF4* retrogene in premature chondroid degeneration and associated mineralization of the IVD, CFA12 *FGF4* retrogenes are more common in surgically treated dogs with evidence of calcification (allele frequency 0.77) compared to surgically treated animals with fibrous/non-calcified disc herniation/protrusion (allele frequency 0.149) (42).

Radiographic Screening

Degree of calcification of the IVD has been shown to be heritable in Dachshunds (56–58) and associated with risk of clinical IVDD in Dachshunds and Pekingese dogs (47, 55, 59, 60). Radiographic screening based on IVD calcification severity scores has been used historically as a potential tool for selective breeding specifically within the Dachshund breed. There are many variables that affect radiographic presence of IVD calcification, and temporal factors can play a major role with appearance and resolution of calcified discs over time (54, 61). Prospective screening with defined time points, that may be breed specific, is important for optimal "scoring" of at-risk dogs. A retrospective analysis of presence or absence of IVD calcification (uncontrolled for age at time of assessment) showed that the observation of calcified discs was significantly more likely in dogs with 2 copies of the CFA12 *FGF4* retrogene (84.8%) compared to 1 copy of the CFA12 *FGF4* retrogene (63.8%) compared to zero copies (18.5%) (42). Multivariable logistic regression identified presence of the CFA12 *FGF4* retrogene as the main contributor to disc calcification with 2 copies of the CFA12 *FGF4* retrogene increasing the odds of disc calcification by a factor of 2.5 compared to 1 copy (42). Presence or absence of the CFA18 *FGF4* retrogene had no significant effect on odds of observing IVD calcification (42).

Decrease in incidence of calcified discs following selective breeding of Dachshunds based on radiographically defined calcification scores has unfortunately been limited (57). Many variables may be influencing progress including precision of scoring (62), limited application and compliance within the breeding population as well as limitations in correlating visual radiographic criteria to underlying genetic status (55, 63). The original mapping of IVD-associated calcification to the CFA12 *FGF4* retrogene location on chromosome 12 was done using high vs. low radiographic calcification scores in Dachshunds (31). Given the extremely high CFA12 *FGF4* retrogene allele frequency in Dachshunds, the low calcification phenotype most likely defined a population of heterozygous (possibly wild type) Dachshunds. This population would be necessary for a successful genome mapping study and is consistent with the CFA12 *FGF4* retrogene dosage effects on radiographically apparent calcification subsequently demonstrated following identification of the CFA12 retrogene (42). Pilot data from a small population of Danish Wire Haired Dachshunds that appear to segregate the CFA12 *FGF4* retrogene showed an OR of 6.1 for high calcification screening scores associated with either 1 or 2 copies of the CFA12 *FGF4* retrogene (64).

The overall dominant role of the CFA12 *FGF4* retrogene with heterozygous and homozygous animals potentially having overlapping degrees of IVDD and calcification together with the variables above are likely reflected in the difficulty obtaining rapid reduction in disease using radiographic screening. Although additional genetic variables still remain to be defined, screening strategies based on the CFA12 *FGF4* retrogene rather than down-stream phenotypes may offer more tractable selection data for breeding.

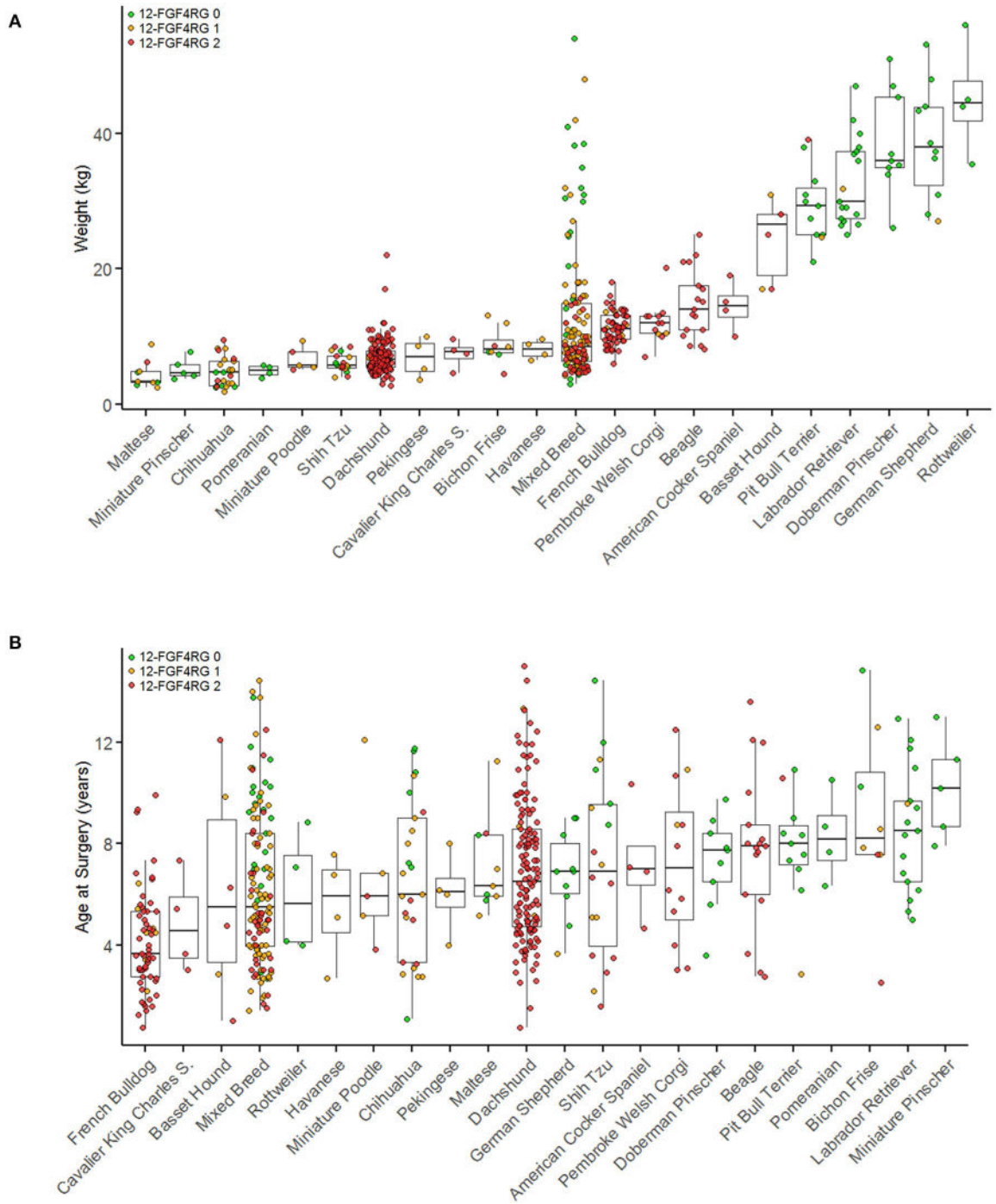

FIGURE 2 | Breed and genotype distribution of surgical IVDD cases by body weight **(A)** and by age at surgery **(B)**. Breeds with fewer than four cases are not included in this figure. Breeds are plotted in order of ascending median weights **(A)** or age **(B)** and colored by CFA12 *FGF4* retrogene genotype. Red indicates two copies of each retrogene, orange indicates one copy, and green indicates zero copies. [Modified from Batcher et al. (42)].

IVDD Relative Risk

Prospective data determining the risk for IVDD associated with presence of the CFA12 *FGF4* retrogene are still to be collected. Assessment in chondrodystrophic breeds such as Dachshunds, Beagles, and French Bulldogs, with very high allele frequencies preludes analysis in retrospective data. However, evaluation of a small group of surgically treated, mixed breed dogs in which segregation of the retrogene occurred and for which historical

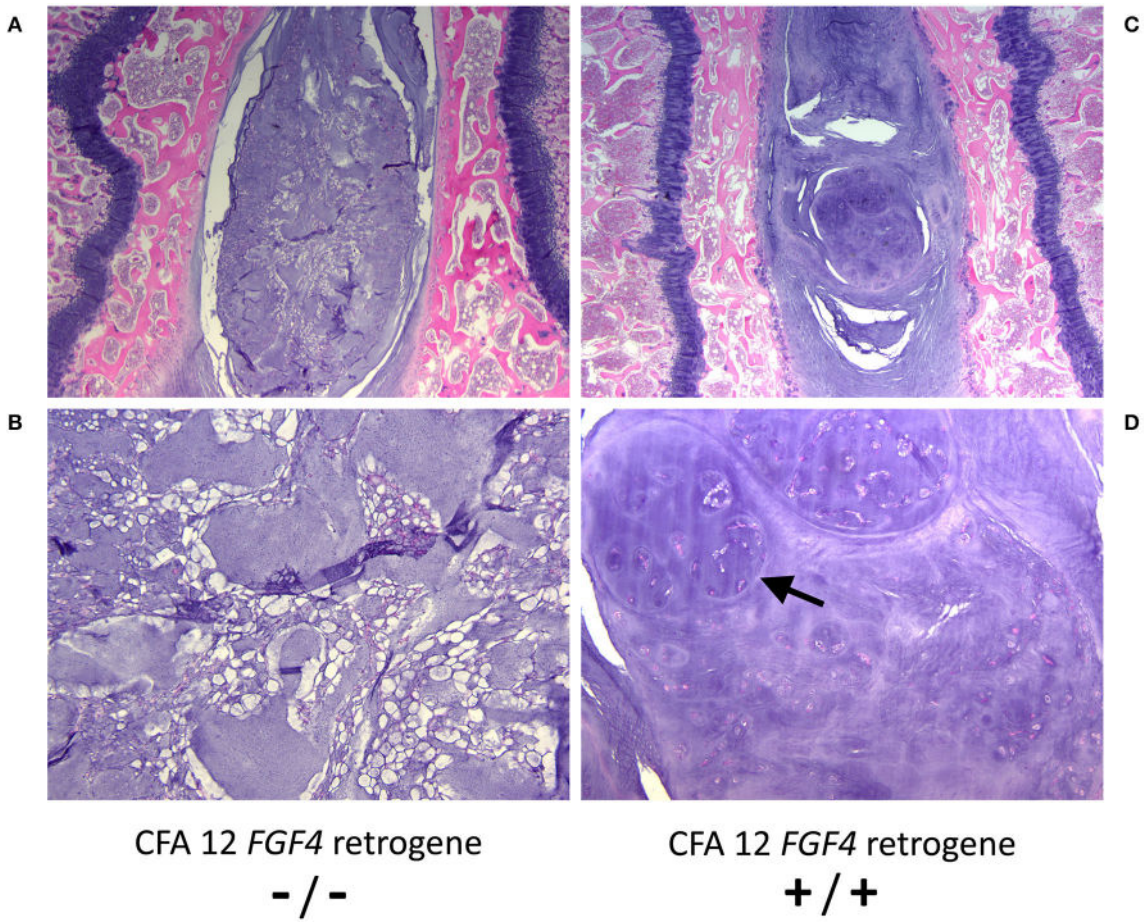

FIGURE 3 | Histopathological images of nucleus pulposus from 10 week-old control (CFA12 *FGF4* retrogene –/–) **(A, B)** and 10 week-old homozygous CFA12 *FGF4* retrogene (CFA12 *FGF4* retrogene +/+) Nova Scotia **(C, D)** Duck Tolling Retriever puppies. The control nucleus pulposus has numerous normal physaliferous notochordal cells with foamy to vacuolated cytoplasm and often stellate cytoplasmic processes. By 10 weeks the CFA12 *FGF4* retrogene-carrying dog's nucleus pulposus **(B)** consists predominantly of round to ovoid chondrocyte-like cells arranged either individually or in nodular clusters (arrow) associated with a dark purple chondroid matrix. Normal notochordal cells are rare. Both dogs have zero copies of the CFA18 *FGF4* retrogene. H&E stain. Magnification **(A, C)** = 20X; **(B, D)** = 100X.

control data for aged, clinically unaffected dogs was available has been performed (42). Looking at number of copies of CFA12 and CFA18 *FGF4* retrogenes, body weight and sex, only presence of the CFA12 *FGF4* retrogene was found to be significantly associated with presentation for decompressive surgery for IVDD, with no difference between 1 and 2 copies. Based on these findings and looking at mixed breeds and other breeds that also segregated the retrogene (allele frequency <0.5 and >0.05), relative risk for IVDD (presenting for surgical treatment) associated with the CFA12 *FGF4* retrogene ranged from 5.5 in Chihuahuas to 15.1 in mixed breed dogs.

WHAT WE DON'T KNOW

Several aspects of the clinical presentation of IVDD in dogs are not easily explained by a simple presence or absence of the CFA12 *FGF4* retrogene. Clarification of these pathophysiological and clinical variables in the presentation of IVDD is important if genotyping is to be used as a tool for reduction in disease incidence. A more comprehensive picture will also increase breeder and owner confidence for eradication strategies that may potentially require major alterations in breed standards for those breeds with high allele frequencies.

CFA12 FGF4 Retrogene Dosage

Risk for clinical disease based on heterozygous or homozygous retrogene status may be powerful data to inform selective breeding strategies. Circumstantial evidence would suggest that there could be an effect of dosage since calcification scores have been shown to be related to risk for IVDD in Dachshunds and Pekingese dogs (47, 55, 59, 60) and disc calcification was shown to be correlated with CFA12 *FGF4* retrogene allele frequency (42). The latter study did not however directly demonstrate a gene dosage effect on age at presentation for surgery or relative risk for surgery (in mixed breed dogs) however scoring was a simple presence or absence and important factors such as age and time of imaging (54, 61) were not controlled. Prospective

studies looking at genotyped dogs within selected breeds as well as selective breeding trials with heterozygous animals from high allele frequency breeds should provide deeper insight into gene dosage effects.

Breed and Environmental-Related Variables

Variables within and across breeds are strongly suggestive for additional genetic, environmental, morphological or metabolic influences on the pathophysiology of IVDD in dogs. The presence of the CFA12 *FGF4* retrogene was associated with a variable risk of presenting for surgery for IVDD from 5.5- to 15.1-fold in different segregating and mixed breeds (42) and even within a relatively non-segregating breed such as the Dachshund, differences in disease prevalence have been noted between different breed types (47, 50). Dachshunds are the most extensively studied breed relating to risk factors for IVDD, however looking retrospectively with current genetic data, historical analyses may have been confounded by variable CFA12 *FGF4* retrogene allele frequencies in different populations of animals. These differences reflect preferences for standard or miniature and variable hair characteristics in different countries. Risk factors including neuter status (45, 50), physical conformation (44, 47–50), axial muscle fascicle length (65), ambient temperature (66), hair characteristics (47, 50), and exercise (46, 50) have been reported with variable consistency in findings across studies.

A shorter T1-S1 vertebral column length and shorter limb (calcaneus-patellar) length were associated with increased risk in one study (48) while a second (49) found increased risk with higher body length to height ratios. Although not documented, the CFA12 *FGF4* retrogene would more likely result in shorter vertebrae, and the later study could have been looking at the dominant effect of the CFA12 *FGF4* retrogene on leg length (rather than vertebral length) as the major determinant of ratio changes. However, several genetic factors are likely to be contributing to conformation including CFA12 and 18 *FGF4* retrogenes. Clinically affected Dachshunds were reported to have longer epaxial muscle fascicle lengths in one study, which could be a secondary effect of clinical IVDD; however, it is interesting to note that FGF4 is also a key signaling molecule in the development of myofibers and tendon of epaxial muscles arising from somitic mesoderm (67, 68).

Data on prevalence of IVDD by Dachshund type varies between studies (47, 50), however in the largest study of over 2,000 Dachshunds, decreased prevalence of IVDD was seen in Standard Wire Haired Dachshunds compared to other standard and miniature types (50). This may be a reflection of the heterozygous CFA12 *FGF4* retrogene status of dogs within the Standard Wire Haired population which can be inferred from the successful GWAS mapping using this Dachshund subtype (31) and further supported by pilot genotyping data from Denmark showing ∼30% (27/91) of Wire Haired (but not Smooth or Short Haired) subtypes were heterozygous for the CFA12 *FGF4* retrogene (64). Long haired Dachshund subtypes (standard and miniature) had the next lowest prevalence of IVDD and the long haired phenotype is known to be caused by mutation within the *FGF5* gene (likely loss of function based on recessive inheritance) (69, 70). Although *FGF5* gene expression has typically been associated with hair follicle development, it is phylogenetically closely related to *FGF4* and similar to *FGF4* has been shown to be expressed in developing limbs (18, 71). Redundancy across the FGF signaling pathways is common (18) and it is interesting to speculate whether there may be a biological relevance in IVDD with over and under expression of these 2 closely related FGF genes. The genetic mutation resulting in the wire haired phenotype is also potentially relevant to IVDD as this involves an activating mutation affecting expression of the *RSPO2* gene (69). RSPO2 synergizes with the WNT-β catenin pathway (72), an important contributor to IVD development (73) and down-regulation of WNT signaling has been shown to be present in early IVD degeneration (74).

Breed Related Calcification

Significant variability in presence of radiographically identified disc calcification (accounting for CFA12 *FGF4* retrogene presence and other factors) based on breed has been reported (42). In dogs presenting for IVDD related surgery, 90.5% Dachshunds, 70.6% French Bulldogs, 60.2% mixed breed dogs, and 40.8% "other pure breed" dogs had at least 1 radiographically defined calcified disc at the time of surgery. The Dachshund data are similar to previous reports (55) and even accounting for potential variability associated with age at assessment, there is a clear breed-associated difference unexplained by the CFA12 *FGF4* retrogene alone.

Breed Related Age Differences

While age at time of surgery for IVDD is significantly lower for CFA12 *FGF4* retrogene dogs as a group (42), differences within the chondrodsytrophic breeds also suggest additional factors in IVDD pathogenesis. Dachshunds and potentially other breeds with high allele frequencies of CFA12 *FGF4* retrogenes presented at an older age for surgery compared to mixed breeds (42). This may reflect additional selection for modifying factors in these high allele frequency breeds resulting in removal of younger-onset dogs from the breeding pool. One can speculate that presence of within-breed "protective/modifying effects could also explain the higher relative risk associated with the CFA12 *FGF4* retrogene in mixed breed dogs. At the opposite end of the spectrum, French Bulldogs have a significantly lower age of disease presentation (42, 51, 52). Unlike potential activation of the WNT pathway by the *ROSP2* gene in Wire Haired dogs, French Bulldogs have down regulation of WNT signaling due to a frameshift mutation in the Disheveled 2 (*DVL2*) gene associated with screw tail and brachycephaly (75). Whether these chondrodystrophic breed associated WNT pathway alterations (in the context of FGF4 overexpression) are clinically relevant to reported IVD-associated down-regulation of WNT signaling (74) remains to be determined, however WNT and FGF signaling pathways cross-talk during a variety of cellular processes (76).

Modifying Effects of Other Retrogenes

Co-expression of the CFA12 and CFA18 *FGF4* retrogenes is common in many chondrodystrophic breeds making exclusion of effects of the CFA18 *FGF4* retrogene from retrospective data challenging. However, effects of the CFA18 *FGF4* retrogene appear to be modest (42), potentially affecting age at presentation but not relative risk or prevalence of radiographic disc calcification based on multivariable analyses (42). The CFA12 *FGF4* retrogene alone appears to be sufficient to cause premature degeneration of the IVD (43) although histopathological studies of IVD from young dogs of breeds homozygous for the CFA18 *FGF4* retrogene such as Cairn Terriers, West Highland White Terriers (typified by low clinical incidence of IVDD) would provide further insight into the relative effects of the two retrogenes.

The location of a large CpG island in the two *FGF4* retrogenes documented to date may facilitate its expression in other chromosomal locations as well. This provokes the question of how many other times the *FGF4* gene has been retrotransposed and then eliminated by selective breeding, or if additional retrocopies remain in some breeds and are responsible for other morphological phenotypes or contribute to IVDD.

IVDD in Dogs Without the CFA12 *FGF4* Retrogene

Previous studies have described Hansen type I IVDD in non-chondrodystrophic breeds (77, 78), and zero copies of the CFA12 *FGF4* retrogene were found in 12% (46/378) of dogs presenting for IVDD related surgery and with documented presence of calcified intervertebral discs (42). Breeds represented included Labrador Retriever, Doberman Pinscher, German Shepherd, Pit Bull Terrier, Rottweiler and Pomeranian and the age at time of surgery was 1.5–2 years older compared to dogs carrying the CFA12 *FGF4* retrogene. Type II degeneration of the IVD is typically seen in older non-chondrodystrophic breeds and rarely is associated with IVD calcification (5, 53), however histopathologically defined disc degeneration in chondrodystrophic and non-chondrodystrophic dogs has many similarities (79). It is possible that some older non-chondrodystrophic dogs could present with IVDD disease similar to chondrodystrophic dogs reflecting the heterogeneity seen within both groups, however the population of CFA12 *FGF4* retrogene negative dogs with calcified IVD could also reflect alternative genetic causes of IVDD characterized by calcification but with a later age of onset.

Human Genetic Correlates

Canine IVDD has been proposed as a potential model for human degenerative disc disease over many years (53, 80). Specific human disease conditions such as achondroplasia, where underlying genetic causes (*FGFR3* gain-of-function mutations) are defined, may have specific molecular mechanistic similarities to *FGF4* overexpression and IVDD (potentially signaling through FGFR3) in dogs. However, less definitive data are available regarding genetics of IVDD and associated low back pain and sciatica in the general human population. Heritability of risk factors for IVDD in humans was initially established using twin studies (81–84) and a variety of candidate gene polymorphisms have been associated with several aspects of IVDD in a variety of ethnic and age-related groups [reviewed in (85)]. Genome-wide association studies similar to those defining the *FGF4* retrogenes in dogs have yielded several potential variants associated with lumbar disc degeneration, sciatica, or "back pain," mostly involving intronic, regulatory or intergenic polymorphisms with no defined causative genetic loci to date (85). It is yet to be determined whether these non-coding polymorphisms may be markers for retrotransposition events similar to findings in dogs with IVDD.

The most critical unanswered question relating to the CFA12 *FGF4* retrogene, is what will be the impact of its discovery on the incidence of IVDD in 10–20 years. While it is clear that there are likely to be additional modifying factors, both genetic and environmental, the evidence that the CFA12 *FGF4* retrogene is a major factor in the development of IVDD in chondrodystrophic dog breeds is compelling. As causative variants for diseases associated with breed morphological traits are defined, the veterinary profession is being forced to face ethical decisions where the primary mission of the profession ("...prevention and relief of animal suffering…") may sometimes conflict with dog breeding phenotypic goals.

Positively, some degree of segregation of the CFA12 *FGF4* retrogene has been seen in almost all IVDD affected breeds, even those with high allele frequencies. In breeds with a high degree of segregation it should be possible to reduce or eliminate the retrogene from the population.

Even in high allele frequency breeds, such as the Dachshund, there are frequency differences between populations [0.98 in USA/UK samples and 0.94 in Swiss samples (42)], indicating that some populations may be less homozygous than others, and segregation may be much higher in specific types such as Wire Haired Dachshunds. Many morphological traits are polygenic in nature (2, 3) and selection for a short-limbed phenotype has likely driven selection of dogs harboring *FGF4* retrogenes. In high IVDD risk breeds such as Dachshunds, Bassets, Corgis and Pekingese that have both the CFA12 and CFA18 *FGF4* retrogene, breeding away from the CFA12 FGF4 retrogene, while still maintaining the aesthetically desirable shortness in stature contributed by the CFA18 *FGF4* retrogene is possible.

The dominant nature of the CFA12 FGF4 retrogene on IVDD and the very high allele frequency in some breeds means that eradication may be challenging. The Wire Haired Dachshund was developed through crossbreeding with Schnauzer and Terrier breeds which may explain the lower frequency of the CFA12 susceptibility locus and lower incidence of IVDD in some wire haired populations. Long term strategies may require a combination of testing and selection of heterozygous dogs, outbreeding, cross breeding, and alteration in breed standards, maybe with inclusion of additional CFA12 FGF4 retrogene-negative phenotypes within the breed standards. Whatever the future holds, from a veterinary perspective, current data suggest that breeding priorities should be for dogs with fewer copies of the CFA12 *FGF4* retrogene, so that the allele frequency can be reduced.

AUTHOR CONTRIBUTIONS

All authors listed have made a substantial, direct and intellectual contribution to the work, and approved it for publication.

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REFERENCES

1. *Online Mendelian Inheritance in Animals* (OMIA) (2020). Availaableonline at: https://omia.org/. (accessed January 5, 2020)
2. Boyko AR, Quignon P, Li L, Schoenebeck JJ, Degenhardt JD, Lohmueller KE, et al. A simple genetic architecture underlies morphological variation in dogs. *PLoS Biol.* (2010) 8:e1000451. doi: 10.1371/journal.pbio.1000451
3. Hayward JJ, Castelhano MG, Oliveira KC, Corey E, Balkman C, Baxter TL, et al. Complex disease and phenotype mapping in the domestic dog. *Nat Commun.* (2016) 7:10460. doi: 10.1038/ncomms10460
4. Hansen HJ. A pathologic-anatomical interpretation of disc degeneration in dogs. *Acta Orthop Scand.* (1951) 20:280–93. doi: 10.3109/17453675108991175
5. Hansen HJ. A pathologic-anatomical study on disc degeneration in dog, with special reference to the so-called enchondrosis intervertebralis. *Acta Orthop Scand Suppl.* (1952) 11:1–117. doi: 10.3109/ort.1952.23.suppl-11.01
6. Mortier GR, Cohn DH, Cormier-Daire V, Hall C, Krakow D, Mundlos S, et al. Nosology and classification of genetic skeletal disorders: 2019 revision. *Am J Med Genet A.* (2019) 179:2393–419. doi: 10.1002/ajmg.a.61366
7. Warman ML, Cormier-Daire V, Hall C, Krakow D, Lachman R, LeMerrer M, et al. Nosology and classification of genetic skeletal disorders: 2010 revision. *Am J Med Genet A.* (2011) 155A:943–68. doi: 10.1002/ajmg.a.33909
8. Frischknecht M, Niehof-Oellers H, Jagannathan V, Owczarek-Lipska M, Drogemuller C, Dietschi E, et al. A COL11A2 mutation in Labrador retrievers with mild disproportionate dwarfism. *PLoS ONE.* (2013) 8:e60149. doi: 10.1371/journal.pone.0060149
9. Goldstein O, Guyon R, Kukekova A, Kuznetsova TN, Pearce-Kelling SE, Johnson J, et al. COL9A2 and COL9A3 mutations in canine autosomal recessive oculoskeletal dysplasia. *Mamm Genome.* (2010) 21:398–408. doi: 10.1007/s00335-010-9276-4
10. Kyostila K, Lappalainen AK, Lohi H. Canine chondrodysplasia caused by a truncating mutation in collagen-binding integrin alpha subunit 10. *PLoS ONE.* (2013) 8:e75621. doi: 10.1371/journal.pone.0075621
11. Jeanes EC, Oliver JAC, Ricketts SL, Gould DJ, Mellersh CS. Glaucoma-causing ADAMTS17 mutations are also reproducibly associated with height in two domestic dog breeds: selection for short stature may have contributed to increased prevalence of glaucoma. *Canine Genet Epidemiol.* (2019) 6:5. doi: 10.1186/s40575-019-0071-6
12. Neff MW, Beck JS, Koeman JM, Boguslawski E, Kefene L, Borgman A, et al. Partial deletion of the sulfate transporter SLC13A1 is associated with an osteochondrodysplasia in the Miniature Poodle breed. *PLoS ONE.* (2012) 7:e51917. doi: 10.1371/journal.pone.0051917
13. Brown EA, Dickinson PJ, Mansour T, Sturges BK, Aguilar M, Young AE, et al. FGF4 retrogene on CFA12 is responsible for chondrodystrophy and intervertebral disc disease in dogs. *Proc Natl Acad Sci USA.* (2017) 114:11476–81. doi: 10.1073/pnas.1709082114
14. Parker HG, VonHoldt BM, Quignon P, Margulies EH, Shao S, Mosher DS, et al. An expressed fgf4 retrogene is associated with breed-defining chondrodysplasia in domestic dogs. *Science.* (2009) 325:995–8. doi: 10.1126/science.1173275
15. Hayward JJ, White ME, Boyle M, Shannon LM, Casal ML, Castelhano MG, et al. Imputation of canine genotype array data using 365 whole-genome sequences improves power of genome-wide association studies. *PLoS Genet.* (2019) 15:e1008003. doi: 10.1371/journal.pgen.1008003
16. Plassais J, Kim J, Davis BW, Karyadi DM, Hogan AN, Harris AC, et al. Whole genome sequencing of canids reveals genomic regions under selection and variants influencing morphology. *Nat Commun.* (2019) 10:1489. doi: 10.1038/s41467-019-09373-w
17. Warburg M, Moller HU. Dystrophy: a revised definition. *J Med Genet.* (1989) 26:769–71. doi: 10.1136/jmg.26.12.769
18. Ornitz DM, Itoh N. The Fibroblast Growth Factor signaling pathway. *Wiley Interdiscip Rev Dev Biol.* (2015) 4:215–66. doi: 10.1002/wdev.176
19. Niswander L, Martin GR. Fgf-4 expression during gastrulation, myogenesis, limb and tooth development in the mouse. *Development.* (1992) 114:755–68.
20. Sun X, Mariani FV, Martin GR. Functions of FGF signalling from the apical ectodermal ridge in limb development. *Nature.* (2002) 418:501–8. doi: 10.1038/nature00902
21. Sivakamasundari V, Lufkin T. Bridging the gap: understanding embryonic intervertebral disc development. *Cell Dev Biol.* (2012) 1:103. doi: 10.4172/2168-9296.1000103
22. Boulet AM, Capecchi MR. Signaling by FGF4 and FGF8 is required for axial elongation of the mouse embryo. *Dev Biol.* (2012) 371:235–45. doi: 10.1016/j.ydbio.2012.08.017
23. Lu P, Minowada G, Martin GR. Increasing Fgf4 expression in the mouse limb bud causes polysyndactyly and rescues the skeletal defects that result from loss of Fgf8 function. *Development.* (2006) 133:33–42. doi: 10.1242/dev.02172
24. Trokovic N, Trokovic R, Mai P, Partanen J. Fgfr1 regulates patterning of the pharyngeal region. *Genes Dev.* (2003) 17:141–53. doi: 10.1101/gad.250703
25. Partanen J, Schwartz L, Rossant J. Opposite phenotypes of hypomorphic and Y766 phosphorylation site mutations reveal a function for Fgfr1 in anteroposterior patterning of mouse embryos. *Genes Dev.* (1998) 12:2332–44.
26. Wright TJ, Mansour SL. FGF signaling in ear development and innervation. *Curr Top Dev Biol.* (2003) 57:225–59. doi: 10.1016/s0070-2153(03)57008-9
27. Peck SH, McKee KK, Tobias JW, Malhotra NR, Harfe BD, Smith LJ. Whole transcriptome analysis of notochord-derived cells during embryonic formation of the nucleus pulposus. *Sci Rep.* (2017) 7:10504. doi: 10.1038/s41598-017-10692-5
28. Dahia CL, Mahoney EJ, Durrani AA, Wylie C. Intercellular signaling pathways active during intervertebral disc growth, differentiation, and aging. *Spine.* (2009) 34:456–62. doi: 10.1097/BRS.0b013e3181913e98
29. Shimauchi Y, Murakami SD, Satoh N. FGF signals are involved in the differentiation of notochord cells and mesenchyme cells of the ascidian *Halocynthia roretzi*. *Development.* (2001) 128:2711–21.
30. Quignon P, Schoenebeck JJ, Chase K, Parker HG, Mosher DS, Johnson GS, et al. Fine mapping a locus controlling leg morphology in the domestic dog. *Cold Spring Harb Symp Quant Biol.* (2009) 74:327–33. doi: 10.1101/sqb.2009.74.009
31. Mogensen MS, Karlskov-Mortensen P, Proschowsky HF, Lingaas F, Lappalainen A, Lohi H, et al. Genome-wide association study in Dachshund: identification of a major locus affecting intervertebral disc calcification. *J Hered.* (2011) 102(Suppl. 1):S81–6. doi: 10.1093/jhered/esr021
32. Webster MK, Donoghue DJ. FGFR activation in skeletal disorders: too much of a good thing. *Trends Genet.* (1997) 13:178–82. doi: 10.1016/S0168-9525(97)01131-1
33. Huet T, Cohen-Solal M, Laredo JD, Collet C, Baujat G, Cormier-Daire V, et al. Lumbar spinal stenosis and disc alterations affect the upper lumbar spine in adults with achondroplasia. *Sci Rep.* (2020) 10:4699. doi: 10.1038/s41598-020-61704-w
34. Mathias SL, Scott AF, Kazazian HH Jr, Boeke JD, Gabriel A. Reverse transcriptase encoded by a human transposable element. *Science.* (1991) 254:1808–10. doi: 10.1126/science.1722352
35. Kubiak MR, Makalowska I. Protein-coding genes' retrocopies and their functions. *Viruses.* (2017) 9:80. doi: 10.3390/v9040080
36. Onozawa M, Goldberg L, Aplan PD. Landscape of insertion polymorphisms in the human genome. *Genome Biol Evol.* (2015) 7:960–8. doi: 10.1093/gbe/evv043
37. Antequera F. Structure, function and evolution of CpG island promoters. *Cell Mol Life Sci.* (2003) 60:1647–58. doi: 10.1007/s00018-003-3088-6

38. Carelli FN, Hayakawa T, Go Y, Imai H, Warnefors M, Kaessmann H. The life history of retrocopies illuminates the evolution of new mammalian genes. *Genome Res.* (2016) 26:301–14. doi: 10.1101/gr.198473.115
39. Dexler H. *Zeitschrift fur Tierheilkunde*. (1896) 7:1.
40. Morris D. *Dogs: A Dictionary of Dog Breeds*. North Pomfret, VT: Trafalgar Square Pub (2002).
41. Braund KG, Ghosh P, Taylor TK, Larsen LH. Morphological studies of the canine intervertebral disc. The assignment of the beagle to the achondroplastic classification. *Res Vet Sci.* (1975) 19:167–72. doi: 10.1016/S0034-5288(18)33527-6
42. Batcher K, Dickinson P, Giuffrida M, Sturges B, Vernau K, Knipe M, et al. Phenotypic effects of FGF4 retrogenes on intervertebral disc disease in dogs. *Genes.* (2019) 10:435. doi: 10.3390/genes10060435
43. Murphy BG, Dickinson P, Marcellin-Little DJ, Batcher K, Raverty S, Bannasch D. Pathologic features of the intervertebral disc in young nova scotia duck tolling retrievers confirms chondrodystrophy degenerative phenotype associated with genotype. *Vet Pathol.* (2019) 56:895–902. doi: 10.1177/0300985819868731
44. Bergknut N, Egenvall A, Hagman R, Gustas P, Hazewinkel HA, Meij BP, et al. Incidence of intervertebral disk degeneration-related diseases and associated mortality rates in dogs. *J Am Vet Med Assoc.* (2012) 240:1300–9. doi: 10.2460/javma.240.11.1300
45. Dorn M, Seath IJ. Neuter status as a risk factor for canine intervertebral disc herniation (IVDH) in dachshunds: a retrospective cohort study. *Canine Genet Epidemiol.* (2018) 5:11. doi: 10.1186/s40575-018-0067-7
46. Jensen VF, Ersboll AK. Mechanical factors affecting the occurrence of intervertebral disc calcification in the dachshund–a population study. *J Vet Med A Physiol Pathol Clin Med.* (2000) 47:283–96. doi: 10.1046/j.1439-0442.2000.00296.x
47. Lappalainen AK, Vaittinen E, Junnila J, Laitinen-Vapaavuori O. Intervertebral disc disease in Dachshunds radiographically screened for intervertebral disc calcifications. *Acta Vet Scand.* (2014) 56:89. doi: 10.1186/s13028-014-0089-4
48. Levine JM, Levine GJ, Kerwin SC, Hettlich BF, Fosgate GT. Association between various physical factors and acute thoracolumbar intervertebral disk extrusion or protrusion in Dachshunds. *J Am Vet Med Assoc.* (2006) 229:370–5. doi: 10.2460/javma.229.3.370
49. Packer RM, Hendricks A, Volk HA, Shihab NK, Burn CC. How long and low can you go? Effect of conformation on the risk of thoracolumbar intervertebral disc extrusion in domestic dogs. *PLoS ONE.* (2013) 8:e69650. doi: 10.1371/journal.pone.0069650
50. Packer RM, Seath IJ, O'Neill DG, De Decker S, Volk HA. DachsLife 2015: an investigation of lifestyle associations with the risk of intervertebral disc disease in Dachshunds. *Canine Genet Epidemiol.* (2016) 3:8. doi: 10.1186/s40575-016-0039-8
51. Aikawa T, Shibata M, Asano M, Hara Y, Tagawa M, Orima H. A comparison of thoracolumbar intervertebral disc extrusion in French Bulldogs and Dachshunds and association with congenital vertebral anomalies. *Vet Surg.* (2014) 43:301–7. doi: 10.1111/j.1532-950X.2014.12102.x
52. Mayousse V, Desquilbet L, Jeandel A, Blot S. Prevalence of neurological disorders in French bulldog: a retrospective study of 343 cases (2002-2016). *BMC Vet Res.* (2017) 13:212. doi: 10.1186/s12917-017-1132-2
53. Hansen HJ. Comparative views of the pathology of disk degeneration in animals. *Lab Invest.* (1959) 8:1242–65.
54. Jensen VF, Arnbjerg J. Development of intervertebral disk calcification in the dachshund: a prospective longitudinal radiographic study. *J Am Anim Hosp Assoc.* (2001) 37:274–82. doi: 10.5326/15473317-37-3-274
55. Rohdin C, Jeserevic J, Viitmaa R, Cizinauskas S. Prevalence of radiographic detectable intervertebral disc calcifications in Dachshunds surgically treated for disc extrusion. *Acta Vet Scand.* (2010) 52:24. doi: 10.1186/1751-0147-52-24
56. Jensen VF, Christensen KA. Inheritance of disc calcification in the dachshund. *J Vet Med A Physiol Pathol Clin Med.* (2000) 47:331–40. doi: 10.1046/j.1439-0442.2000.00297.x
57. Lappalainen AK, Maki K, Laitinen-Vapaavuori O. Estimate of heritability and genetic trend of intervertebral disc calcification in Dachshunds in Finland. *Acta Vet Scand.* (2015) 57:78. doi: 10.1186/s13028-015-0170-7
58. Stigen O, Christensen K. Calcification of intervertebral disks in the dachshund: an estimation of heritability. *Acta Vet Scand.* (1993) 34:357–61.
59. Chai O, Harrosh T, Bdolah-Avram T, Mazaki-Tovi M, Shamir MH. Characteristics of and risk factors for intervertebral disk extrusions in Pekingese. *J Am Vet Med Assoc.* (2018) 252:846–51. doi: 10.2460/javma.252.7.846
60. Jensen VF, Beck S, Christensen KA, Arnbjerg J. Quantification of the association between intervertebral disk calcification and disk herniation in Dachshunds. *J Am Vet Med Assoc.* (2008) 233:1090–5. doi: 10.2460/javma.233.7.1090
61. Stigen O. Calcification of intervertebral disks in the dachshund: a radiographic study of 115 dogs at 1 and 5 years of age. *Acta Vet Scand.* (1996) 37:229–37.
62. Rosenblatt AJ, Hill PB, Davies SE, Webster NS, Lappalainen AK, Bottema CD, et al. Precision of spinal radiographs as a screening test for intervertebral disc calcification in Dachshunds. *Prev Vet Med.* (2015) 122:164–73. doi: 10.1016/j.prevetmed.2015.09.015
63. Stigen O, Kolbjornsen O. Calcification of intervertebral disks in the dachshund: a radiographic and histopathologic study of 20 dogs. *Acta Vet Scand.* (2007) 49:39. doi: 10.1186/1751-0147-49-39
64. Proschowsky F. No useful DNA test for disc prolapse this time. *Dansk Gravhundenklub Magaz Members Danish Dachshund Club.* (2018) 1:12–3.
65. Bostrom A, Channon S, Jokinen T, Junnila J, Hielm-Bjorkman A, Laitinen-Vapaavuori O. Structural characteristics and predicted functional capacities of epaxial muscles in chondrodystrophic and non-chondrodystrophic dogs with and without suspected intervertebral disc herniation- a preliminary study. *Res Vet Sci.* (2019) 123:204–15. doi: 10.1016/j.rvsc.2019.01.008
66. Barandun MA, Bult S, Demierre S, Vidondo B, Forterre F. Colder ambient temperatures influence acute onset canine intervertebral disc extrusion. *Front Vet Sci.* (2020) 7:175. doi: 10.3389/fvets.2020.00175
67. Brent AE, Braun T, Tabin CJ. Genetic analysis of interactions between the somitic muscle, cartilage and tendon cell lineages during mouse development. *Development.* (2005) 132:515–28. doi: 10.1242/dev.01605
68. Kahane N, Cinnamon Y, Bachelet I, Kalcheim C. The third wave of myotome colonization by mitotically competent progenitors: regulating the balance between differentiation and proliferation during muscle development. *Development.* (2001) 128:2187–98.
69. Cadieu E, Neff MW, Quignon P, Walsh K, Chase K, Parker HG, et al. Coat variation in the domestic dog is governed by variants in three genes. *Science.* (2009) 326:150–3. doi: 10.1126/science.1177808
70. Housley DJ, Venta PJ. The long and the short of it: evidence that FGF5 is a major determinant of canine 'hair'-itability. *Anim Genet.* (2006) 37:309–15. doi: 10.1111/j.1365-2052.2006.01448.x
71. Clase KL, Mitchell PJ, Ward PJ, Dorman CM, Johnson SE, Hannon K. FGF5 stimulates expansion of connective tissue fibroblasts and inhibits skeletal muscle development in the limb. *Dev Dyn.* (2000) 219:368–80. doi: 10.1002/1097-0177(2000)9999:9999<::AID-DVDY1056>3.0.CO;2-8
72. Kazanskaya O, Glinka A, del Barco Barrantes I, Stannek P, Niehrs C, Wu W. R-Spondin2 is a secreted activator of Wnt/beta-catenin signaling and is required for Xenopus myogenesis. *Dev Cell.* (2004) 7:525–34. doi: 10.1016/j.devcel.2004.07.019
73. Kondo N, Yuasa T, Shimono K, Tung W, Okabe T, Yasuhara R, et al. Intervertebral disc development is regulated by Wnt/beta-catenin signaling. *Spine.* (2011) 36:E513–8. doi: 10.1097/BRS.0b013e3181f52cb5
74. Smolders LA, Meij BP, Onis D, Riemers FM, Bergknut N, Wubbolts R, et al. Gene expression profiling of early intervertebral disc degeneration reveals a down-regulation of canonical Wnt signaling and caveolin-1 expression: implications for development of regenerative strategies. *Arthritis Res Ther.* (2013) 15:R23. doi: 10.1186/ar4157
75. Mansour TA, Lucot K, Konopelski SE, Dickinson PJ, Sturges BK, Vernau KL, et al. Whole genome variant association across 100 dogs identifies a frame shift mutation in DISHEVELLED 2 which contributes to Robinow-like syndrome in Bulldogs and related screw tail dog breeds. *PLoS Genet.* (2018) 14:e1007850. doi: 10.1371/journal.pgen.1007850
76. Katoh M, Katoh M. Cross-talk of WNT and FGF signaling pathways at GSK3beta to regulate beta-catenin and SNAIL signaling cascades. *Cancer Biol Ther.* (2006) 5:1059–64. doi: 10.4161/cbt.5.9.3151
77. Cherrone KL, Dewey CW, Coates JR, Bergman RL. A retrospective comparison of cervical intervertebral disk disease in nonchondrodystrophic large dogs versus small dogs. *J Am Anim Hosp Assoc.* (2004) 40:316–20. doi: 10.5326/0400316

78. Cudia SP, Duval JM. Thoracolumbar intervertebral disk disease in large, nonchondrodystrophic dogs: a retrospective study. *J Am Anim Hosp Assoc.* (1997) 33:456–60. doi: 10.5326/15473317-33-5-456
79. Hansen T, Smolders LA, Tryfonidou MA, Meij BP, Vernooij JCM, Bergknut N, et al. The myth of fibroid degeneration in the canine intervertebral disc: a histopathological comparison of intervertebral disc degeneration in chondrodystrophic and nonchondrodystrophic dogs. *Vet Pathol.* (2017) 54:945–952. doi: 10.1177/0300985817726834
80. Thompson K, Moore S, Tang S, Wiet M, Purmessur D. The chondrodystrophic dog: A clinically relevant intermediate-sized animal model for the study of intervertebral disc-associated spinal pain. *JOR Spine.* (2018) 1:e1011. doi: 10.1002/jsp2.1011
81. Battie MC, Videman T, Gibbons LE, Fisher LD, Manninen H, Gill K. 1995 Volvo Award in clinical sciences. Determinants of lumbar disc degeneration. A study relating lifetime exposures and magnetic resonance imaging findings in identical twins. *Spine.* (1995) 20:2601–12. doi: 10.1097/00007632-199512150-00001
82. Battie MC, Videman T, Levalahti E, Gill K, Kaprio J. Heritability of low back pain and the role of disc degeneration. *Pain.* (2007) 131:272–80. doi: 10.1016/j.pain.2007.01.010
83. Sambrook PN, MacGregor AJ, Spector TD. Genetic influences on cervical and lumbar disc degeneration: a magnetic resonance imaging study in twins. *Arthritis Rheum.* (1999) 42:366–72.3.0.CO;2-6 "target=" doi: 10.1002/1529-0131(199902)42:2<366::AID-ANR20>3.0.CO;2-6
84. Livshits G, Popham M, Malkin I, Sambrook PN, Macgregor AJ, Spector T, et al. Lumbar disc degeneration and genetic factors are the main risk factors for low back pain in women: the UK Twin Spine Study. *Ann Rheum Dis.* (2011) 70:1740–5. doi: 10.1136/ard.2010.137836
85. Munir S, Rade M, Maatta JH, Freidin MB, Williams FMK. Intervertebral disc biology: genetic basis of disc degeneration. *Curr Mol Biol Rep.* (2018) 4:143–50. doi: 10.1007/s40610-018-0101-2

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REVIEW
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Ambulation in Dogs With Absent Pain Perception After Acute Thoracolumbar Spinal Cord Injury

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Acute thoracolumbar spinal cord injury (SCI) is common in dogs frequently secondary to intervertebral disc herniation. Following severe injury, some dogs never regain sensory function to the pelvic limbs or tail and are designated chronically "deep pain negative." Despite this, a subset of these dogs develop spontaneous motor recovery over time including some that recover sufficient function in their pelvic limbs to walk independently without assistance or weight support. This type of ambulation is commonly known as "spinal walking" and can take up to a year or more to develop. This review provides a comparative overview of locomotion and explores the physiology of locomotor recovery after severe SCI in dogs. We discuss the mechanisms by which post-injury plasticity and coordination between circuitry contained within the spinal cord, peripheral sensory feedback, and residual or recovered supraspinal connections might combine to underpin spinal walking. The clinical characteristics of spinal walking are outlined including what is known about the role of patient or injury features such as lesion location, timeframe post-injury, body size, and spasticity. The relationship between the emergence of spinal walking and electrodiagnostic and magnetic resonance imaging findings are also discussed. Finally, we review possible ways to predict or facilitate recovery of walking in chronically deep pain negative dogs. Improved understanding of the mechanisms of gait generation and plasticity of the surviving tissue after injury might pave the way for further treatment options and enhanced outcomes in severely injured dogs.

Keywords: spinal walking, deep pain negative, intervertebral disc herniation, canine, locomotion, gait generation

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TERMINOLOGY

- "Deep pain negative": term synonymous with "absent pain perception" and defined as an absent behavioral response to noxious stimulation caudal to the injury level. For thoracolumbar SCI, this refers to absent pain perception to a mechanical stimulus in the medial and lateral toes of both pelvic limbs and base of the tail; in dogs, this term is applied in the acute setting with concurrent paraplegia to imply a functionally complete injury though sensory and motor status should be considered separately in the chronic setting.

- Sensorimotor complete injury: term used to describe functionally and/or physically complete injury in people where there is absent voluntary movement or pain perception below the injury level; synonymous with AIS-A designation using human SCI grading parameters.
- Ambulatory: the ability to rise and take at least 10 consecutive weight bearing steps unassisted without falling.
- Chronically paralyzed: broad, non-specific term used to capture the population of dogs with permanent neurologic impairment (motor, sensory, and/or deficits in continence) following severe SCI. Dogs in this group can exhibit paraplegia (i.e., no pelvic limb movement at all) or display varying degrees of pelvic limb movements that fall short of being useful (i.e., they remain non-ambulatory).
- Spinal walking: independent ambulation in a "deep pain negative" dog typically characterized by lack of coordination between thoracic and pelvic limbs, difficulty turning, or going backward, intermittent falling (especially when changing directions), frequently intact toe knuckling response but absent hopping, and increased spasticity.

INTRODUCTION

The majority of dogs suffering from acute spinal cord injury (SCI) will recover adequate or even normal function (1). However, a subset of dogs with severe injury fail to regain pain perception caudal to the injury level ("deep pain negative"), remain incontinent and are classified as having an unsuccessful outcome (1–3). The permanent lack of pain perception has been commonly, and frequently incorrectly, interpreted as an indication of spinal cord transection, complete disconnection from all supraspinal influence and minimal to absent chance of meaningful recovery of function. However, a proportion of permanently deep pain negative dogs demonstrate notable spontaneous motor recovery over time (2, 4, 5). This can range from non-purposeful kicking movements of the limbs, especially following tactile stimulation below the injury level, to overground walking with minimal apparent paresis or ataxia. Ambulation exhibited by this population, typically known as "spinal walking," is commonly considered exclusively reflexive stepping generated by the spinal cord caudal to the level of injury as described by experimental studies of SCI in dogs and other species (6–14). While relatively autonomous circuits within the spinal cord are integral, gait generation is a complex process with extensive coordination between various components of the central and peripheral nervous systems. Understanding the ways in which this circuitry is altered and also how it can recover after injury has broad therapeutic and translational implications.

This review will provide a comparative overview of locomotion and explore the physiological underpinnings of "spinal walking" after severe SCI in dogs. Additionally, the clinical characteristics of motor recovery with absent pain perception as well as proposed means to predict and facilitate its development in this population will be described.

COMPARATIVE REVIEW OF LOCOMOTION/GAIT GENERATION

Normal locomotion is a complex action that involves coordination of multiple brain regions, circuitry within the spinal cord and peripheral nerves and muscles. The basic components of locomotion are evolutionarily conserved with broad overlap even between invertebrate and vertebrate animals. While motor systems within the brain and spinal cord are essential to producing locomotion, integration of sensory input at all levels is also integral to proper functioning and modulation of locomotion in response to environmental surroundings.

Within the brain there are several motor regions from which upper motor neurons arise to produce the descending motor tracts, with some variability in their relative importance across species. These include the primary motor cortex located in the parietal lobe of each cerebral hemisphere, the red nucleus of the midbrain and the reticular formation of the pons and medulla oblongata. Additionally, the mesencephalic locomotor region located just ventral to the caudal colliculi is involved in initiating stepping movements. Axons of neurons from this area do not directly project to the spinal cord but rather interact with other brainstem motor regions, especially within the reticular formation, to produce locomotion. All of these components are also influenced and modulated by the cerebellum and basal nuclei. Input from these areas allow for complex movements and adjustment of locomotor activity. The overall output of the brain activates spinal cord motor circuitry and produces voluntary motor activity (15–18).

Axons of the upper motor neurons (UMN) in the various motor regions of the brain form the descending motor tracts to the spinal cord. These descending motor tracts produce both inhibitory and excitatory influence on spinal cord interneurons and lower motor neurons (LMN) to initiate and regulate voluntary movement. These include the lateral and ventral (the latter being more developed in primates) corticospinal tracts, rubrospinal tract, and pontine and medullary reticulospinal tracts. While the vestibulospinal tracts play a crucial role in posture and influence locomotion, they will not be discussed in detail. The corticospinal tract originates in the primary motor cortex, follows the major descending white matter pathway (internal capsule, crus cerebri, pyramids) to the medulla where the majority of fibers cross at the pyramidal decussation to descend in the lateral funiculus of the contralateral spinal cord. It is primarily involved with complex and precise movements although is reported to retain a role in overall gait generation (17, 19, 20). The rubrospinal tract originates in the red nucleus, immediately crossing midline to travel in the contralateral lateral funiculus of the spinal cord. The pontine and medullary reticulospinal tracts start in the ill-defined reticular formation of the brainstem before descending in the ipsilateral ventral and lateral funiculi, respectively. The rubrospinal and medullary reticulospinal tracts facilitate flexor muscles and inhibit extensors while the pontine reticulospinal tract does the opposite, providing a tonic balance between facilitation and inhibition of spinal cord lower motor neurons (15–18). Direct

evidence in dogs is sparse, but it has been demonstrated in cats that the reticulospinal pathways play an important role in postural control and basic gait generation on a flat surface while the rubrospinal tract is involved in both normal control of locomotion and in producing adaptive movements to changes in the environment (19, 21). The corticospinal tract is less well-developed in domestic species (compared to people and non-human primates) and is not considered essential to generate basic locomotor rhythms; however, it functions in parallel with the other motor pathways to primarily regulate and fine tune movements (19–21).

Within the spinal cord, circuitry involved in gait generation has been identified in multiple species and is known as the central pattern generator (CPG) (13, 16, 18, 22–24). The CPG organizes the basic pattern for stepping, independent of supraspinal or sensory input. This basic rhythmic pattern of the CPG is produced by interconnected, alternating, and mutually inhibitory flexor and extensor interneurons (25). This network is thought to extend the length of the spinal cord but has been most extensively studied in the lumbar region in relation to control of the pelvic limbs (in quadrupeds, or legs in people). In this context, it is located in the intermediate zone of spinal cord gray matter although the precise cranial to caudal location of integral components of the circuitry within the lumbar spinal cord might vary between species (e.g., cranial lumbar in dogs, rats, people; mid-caudal lumbar in cats) (24, 26). These interneurons, in turn, activate lower motor neurons *via* additional intermediary interneurons, the output from which serves as the final common pathway to produce locomotion *via* direct innervation of appendicular muscles (25). The CPG also provides coordination between left and right limbs via integration of commissural interneurons and thoracic and pelvic limbs, important in normal quadrupedal locomotion (15, 18, 27, 28). While autonomously capable of relatively complex patterns of activity, under normal (non-injured) conditions, supraspinal input is necessary for activation (29). Additionally, modifying input to the CPG is necessary to allow adaptation of the basic alternating stepping pattern. Sensory input derived from visual information, vestibular input, and both exteroceptor and proprioceptors located on the body and limbs is also an important component of locomotion, specifically providing information needed to adapt locomotion to an animal's surroundings (16).

Gait generation itself consists of two major phases, the postural stance phase and the protraction or swing phase. However, based on the activation pattern of specific pelvic limb muscles, the step cycle should really be considered as having four phases: flexion and first extension occur during swing while second and third extension occur during stance (30, 31). Second extension happens during the early part of the stance phase when the knee and tarsus joints actually flex despite contracting extensor muscles as the animal prepares to bear weight (30, 31). Third extension is characterized by hip, knee and tarsus extension as the weight of the body is pushed forward (30, 31).

PLASTICITY OF LOCOMOTOR SYSTEMS AFTER SCI

The central nervous system is largely considered to have poor regenerative capacity; however, remarkable plasticity is possible. In fact, much of what is known about the organization and function of locomotor systems has been elucidated *via* various experimental spinal cord transection and decerebrate animal models (6, 7, 9–14, 32). Reorganization and adaptations that occur at all levels might influence recovery of motor function below the level of severe injury. These include regrowth of axons across the epicenter, recovery/reactivation of conduction of residually intact UMN axons traversing the lesion epicenter, a more autonomous role for the CPG, alterations in excitability of interneurons and LMNs below injury, activation of silent synapses, changes in synaptic weight, and alterations in sensory input or how afferent input is integrated at the level of the spinal cord below injury (29, 33–38).

Axonal regeneration of UMN axons has been demonstrated via experimental transection models although the capacity for regeneration varies between axon types and is limited compared to axons in the peripheral nervous system (37, 39–41). While serotonergic axons have demonstrated robust sprouting ability after injury, there are substantial deterrents to meaningful regrowth of most other disrupted axon systems (41). These include the size of the defect, astroglial scar formation, growth inhibitory molecules (e.g., chondroitin sulfate proteoglycans) and myelin-based growth inhibition (37, 39, 40). Additionally, there is no guarantee that regenerating axons will reconnect with the appropriate below-injury targets. These factors lead to minimal functional recovery in most complete transection models. There is active research regarding how to facilitate more effective regrowth through the use of various grafts, scaffolds, inhibitors of scar formation and other modulators of axonal growth (37, 39, 40, 42–47).

Fortunately, even with severe injury, physical spinal cord transection is uncommon. Residually intact, small diameter, subpial UMN axons traversing the lesion epicenter have been shown in various animals and people with functionally complete injury (33, 35, 48, 49). While the degree of loss of large diameter axons and abnormal myelination of residual fibers contribute to persistent neurologic deficits in chronic SCI, there is evidence of reactivation of surviving long tract axons within rubrospinal and other descending motor tracts (50, 51). This might serve to reestablish supraspinal influence on spinal cord circuitry and LMNs and contribute to recovery of voluntary motor control (35, 50). Prior work in rats and cats has shown that as little as 5–10% of the original population of axons can allow voluntary ambulation after severe injury (33, 50, 51).

Additionally, collateral sprouting of spared UMNs and regrowth of local propriospinal fibers traversing the site of injury have each been shown in experimental injury in rodents and lampreys (52–55). These mechanisms serve to produce novel, multisynaptic pathways, and reestablish the connections between UMNs and LMNs with associated improvements in motor function (39, 52). Interestingly, propriospinal neurons have

also been shown to activate CPGs, highlighting their potential importance in achieving useful locomotor recovery after severe injury (39, 55–57).

Below the level of injury, notable changes also occur. There is increased importance of the integration between sensory input and CPG activity to coordinate motor output due to limited or lack of supraspinal control (29). Alterations in both motor neuron pool excitability and sensory input to the dorsal horn occur and likely contribute to functional status after injury (38, 58–62). For example, pharmacologic inhibition of post-synaptic inhibition with strychnine has been used to facilitate spinal walking in experimentally transected dogs (63). However, maladaptive plasticity and development of aberrant neuronal circuits commonly manifested as neuropathic pain or spasticity can also occur and impair functional recovery (58, 60).

SPINAL WALKING DEFINITION AND BRIEF OVERVIEW

Dogs with chronic, permanent (i.e., more than 3 months after injury) loss of pain perception following acute severe, naturally-occurring thoracolumbar SCI are generally considered to have a limited capacity for locomotor recovery. Despite this presumption, a proportion of these dogs regain the ability to walk independently (2, 4). Unassisted ambulation in dogs chronically lacking pain perception ("deep pain negative") has commonly been referred to as "spinal walking."

Dogs without pain perception that exhibit such walking tend to show a spastic pelvic limb gait in which the stepping pattern of the pelvic limbs is not apparently coordinated with the thoracic limbs (the step cycles are out of phase) (**Supplementary Materials 1–3**). There is a tendency to fall to one side, especially when turning. Some dogs will exhibit "attempts" to correct the falling due to excessive spasticity in the limb ipsilateral to the fall. However, other dogs demonstrate the ability to walk much longer distances without falling. Limb movements during ambulation are variable; dragging of the toes is observed in some animals, but many also show excessively high stepping associated with dramatic flexor spasticity, especially when changing directions. It is also common for dogs to lean forward to facilitate standing up when initiating ambulation. This population commonly demonstrates intact toe knuckling response but very delayed to absent hopping, absent extensor postural thrust and inability to step backwards. Spinal reflexes are typically hyper-reflexive, flexor, and extensor spasticity are common. Chronic reflex perturbations are also common including an abnormal crossed extensor reflex between pelvic limbs (stimulating flexion in one pelvic limb that elicits reflex extension of opposite pelvic limb in a non-weight bearing position) and the presence of a "mass reflex" (simultaneous, below-injury movements including flexor spasms of the limbs, tail flagging, and evacuation of the bladder or colon elicited by tactile or other sensory stimulation such as manual bladder expression).

Spinal walking has been proposed to reflect reflexive stepping generated autonomously at the level of the spinal cord CPG in the absence of any supraspinal input (8, 16). This is supported by experimental transection models in dogs showing recovery of treadmill and over ground ambulation in the months after injury with similar electromyographic patterns to normal walking dogs (11, 13, 63–65). While Liu et al. found that no transected dogs without additional therapeutic intervention (polyethylene glycol at the site of transection) regained any pelvic limb motor function, they were only followed for two months which is likely premature to its typical development (65). Other work has showed spontaneous recovery of ambulation in a majority of dogs by an average of four months after transection without any specific therapy (11).

While preservation and/or effective reorganization of the CPG circuitry is integral to motor output after SCI, there are distinct differences between experimental and naturally-occurring SCI as well as between treadmill walking and over ground walking (2, 66). Importantly, simple activation of exclusively CPG-induced reflexive stepping post-injury might not adequately explain the broad variability in when and in which dogs develop independent, over ground ambulation despite persistently absent pain perception. In the normal, uninjured state, supraspinal input is considered necessary for initiation and control of voluntary over-ground locomotion mammals (16). Whether this requirement for supraspinal input to produce functional ambulation is maintained following severe SCI remains uncertain. In one study evaluating electrophysiologic evidence of long tract function in a group of dogs lacking pain perception, results suggested that recovery of supraspinal connections and walking were independent of each other (5). In another study of dogs with permanently absent pain perception, all dogs that recovered ambulation were noted to have a voluntary tail wag within one-month post-injury (2). This finding demonstrated intact brain to tail connections traversing the site of some so-called complete injuries and implied a potential association between such translesional connections and recovery of walking (2). While there is conflicting evidence regarding the role of supraspinal influence in severely injured dogs, there are also other factors to consider such as maintenance of certain sensory input that are likely crucial to guide the appropriate CPG-directed motor output in the post-injury setting (16, 29, 67).

Overall, development of ambulation in pain perception negative dogs likely reflects a reorganized CPG in complex coordination with multiple other factors that might include some degree of spared supraspinal influence, a certain threshold of motor neuron pool excitability, appropriate peripheral sensory input, activity specific locomotor training and yet to be determined combinatorial therapeutic interventions (4, 5, 68, 69).

CLINICAL DESCRIPTION IN DOGS WITH NATURALLY OCCURRING INJURY

Ambulation in pain perception negative dogs secondary to naturally occurring injury is reported to range from 10 to 59%, with the large discrepancy likely due, in part, to differences in the patient population, the injury itself, and variable definitions

of walking and pain perception (2, 4, 5, 69). Although it can be seen with a number of causes of acute SCI, the majority of what we know about this population comes from dogs that suffered intervertebral disc herniation (IVDH), the most common cause of acute SCI in dogs. While an association between injury type and development of spinal walking has not been identified, it appears to be less common in dogs who suffered vertebral column trauma (2, 4, 69). This might reflect that a large percentage of dogs are euthanized at the time of traumatic injuries due to poor prognosis relative to IVDH, but differences in the impact of injury type (e.g., higher rate of more extensive or multiple injuries and physical spinal cord transection in traumatic injuries) on locomotor systems is also possible.

The timeframe during which ambulation develops is also variable. In Olby et al. 2003, 7/18 (38%) dogs with absent pain perception secondary to IVDH regained ambulation on average over 9 months with a range of four to 18 months (2). Among a cohort of 94 dogs examined in the chronic setting in which nine were ambulatory with absent pain perception, the median time since injury at examination was 12 months (range of 3–89 months) (5). While time to develop ambulation was not specifically reported for the nine dogs, the overall timeframe is similar to Olby et al. In contrast, Gallucci et al. found median time to regain ambulation was just 75 days and ranged from 16 to 350 days for the 48/81 (59%) dogs with functionally complete injuries who walked again (4). Differences in study design likely contributed to this discrepancy. Most notably, dogs with shorter average time to ambulation underwent early post-injury, intensive rehabilitation which might have positively impacted recovery (4).

Development of ambulation in pain perception negative dogs is typically considered to require intact local reflex arcs to the pelvic limbs (i.e., an injury level cranial to the fourth lumbar vertebrae) to provide appropriate muscle tone and necessary weight bearing ability (8, 14). However, the importance of lesion location within the T3-L3 spinal cord region remains unclear. It has been suggested that lesions cranial to the thoracolumbar junction might impair supraspinal postural control of epaxial muscles and therefore prevent functional manifestation of the reflexive stepping, even if such spinal circuitry is intact (8). On the contrary, it has also been proposed that more cranial lesions (cranial to L2) might facilitate its development due to sparing of the intrinsic circuitry of the CPG integral to pelvic limb locomotor function (70, 71). The most common site among deep pain negative dogs who walked in one study was T12-T13 and ranged from T4-5 to L2-3 (4). No association between lesion location and ambulation has yet been identified (4, 5, 72).

Body weight but not body condition score has also been reported to influence development ambulation in pain perception negative dogs, with smaller dogs being more likely to become spinal walkers (4). The role of body weight distribution is unclear but compensatory forward loading on to the thoracic limbs has been demonstrated in dogs with SCI (73–75). It is possible that smaller dogs more effectively shift weight off of their pelvic limbs making it easier to "stand" and for stepping movements to become functional walking compared to larger, heavier dogs. The impact of limb length has not been specifically investigated. Anecdotally, taller dogs with a higher center of gravity are less likely to regain ambulation, perhaps due to greater demands on supraspinal postural control to maintain balance which might be lacking after severe injury. Deficiencies in lateral stability have been demonstrated in dogs with both complete and incomplete SCI and might support postural control as an additional factor contributing to return of functional ambulation beyond just regaining pelvic limb stepping movements (76). Clinically, lateral instability can be noted in this population as a tendency to ambulate reasonably well in straight lines but falling when attempting to turn or change directions. Younger age has also been suggested to promote its development (4). Other patient factors that logically might negatively influence motor recovery include lack of behavioral motivation, limb contractures, and severe limb muscle atrophy.

Among chronically deep pain negative dogs, clinical examination of spasticity has also been described in relationship to motor function (77). A canine spasticity scale was developed that specifically quantifies duration of patellar clonus and degree and duration of pelvic limb flexor spasms induced by pin prick to the bottom of the paw. The overall spasticity scale score and duration of flexor spasms were each positively associated with gait scores (77). While spasticity is typically considered a maladaptive response to severe injury in people resulting in pain, reduced quality of life and inconsistent impacts on daily functioning, its potential role in recovery of motor function is poorly understood (77–82). However, the data in dogs suggests that development of flexor spasms might indicate increased excitability of the intraspinal circuitry and improved recovery of stepping (77). Cutaneous sensory stimulation of the hind quarters after injury (especially of the perineum, tail, and paw) has also been suggested to produce stepping movements in dogs (11). The importance of afferent input has been demonstrated in cat and rodent models where sural nerve stimulation, tail electrical stimulation or manual tail or perineum manipulation enhanced pelvic limb stepping (6, 12, 26, 83, 84). In humans with incomplete SCI, cutaneous plantar sensory stimulation during motor training increased spinal cord excitability and has been suggested as a means to enhance recovery of motor function (85). Additionally, it has been advocated to incorporate a variety of walking surfaces for incomplete injury patients supporting an integral role for sensory input in promoting locomotor recovery (86). Although the role of targeted sensory input on the development of spinal walking has not been prospectively evaluated in dogs with naturally occurring injury, providing different sensory environments (e.g., grass versus hard surface flooring) and targeted afferent stimulation might be useful to facilitate walking in this population.

Electrodiagnostic testing has also been utilized to try to shed light on the long tract and local spinal pathways involved in the development of ambulation in dogs with absent pain perception (5, 68, 87). Evaluation of spinal cord long tracts utilizing transcranial magnetic stimulation (TMS) and cortical and spinal cord somatosensory evoked potentials (SSEPs) have produced conflicting results (5, 68, 87). In Lewis et al., no SSEPs traversing the injury site were identified but pelvic limb motor evoked potentials (MEPs) following TMS were noted in

4/20 dogs (including 3/5 ambulatory dogs) (68). Trans-lesional motor conduction was associated with higher open field gait scores and ambulation. One of the four dogs included in this group had present but blunted pain perception which supports a less severe injury and might have explained the MEP and recovery of ambulation (68). In contrast in Hu et al. 2018, cortical SSEPs and MEPs were noted in 12/34 (0/9 spinal walkers) and 19/85 (1/9 spinal walkers) chronically injured dogs, respectively, but no relationship was identified between the presence of either SSEPs or MEPs and ambulation (5). It is possible that trans-lesional conduction in chronic SCI provides insufficient influence in some injuries or is unrelated to the reorganization of spinal cord circuitry that produces walking. Clarification of these electrodiagnostic results and the role of residual or reestablished supraspinal input on long-term recovery of function below clinically complete injuries requires further study.

Local spinal reflex circuitry aimed at evaluating motor neuron pool excitability has also been evaluated in chronically injured dogs using the H-reflex (68). The H-reflex was present recording from the plantar interosseus muscles following tibial nerve stimulation in 19/19 of chronically injured dogs compared to 3/6 controls, and the H-reflex threshold (stimulus intensity at which the waveform first appeared) was lower in SCI dogs than in controls (68). This lowered threshold supports increased motor neuron pool excitability below injury compared to healthy animals without SCI. Notably, the H-reflex threshold was also inversely associated with open field gait scores among the dogs with chronic SCI (68). This suggests that increased motor neuron pool excitability might also play an important role in motor recovery following severe injury.

Magnetic resonance imaging (MRI) features of dogs with chronic SCI have also been described in relationship to below-injury functional status (72, 88, 89). On conventional MRI performed in the chronic setting, a longer length of apparent complete parenchymal compromise (i.e., no normal tissue discernible on consecutive transverse images at the lesion epicenter) was inversely associated with open field gait scores (72). Similarly, more extensive chronic intramedullary lesions or cavitations have been associated with failure to regain ambulation by 7 months after presentation (88). Diffusion tensor imaging (DTI), an MRI application in which images are derived from the cellular motion of water, and associated tractography, which provides a visual representation of spinal cord white matter tracts, have also been evaluated in this population (89). Decreased anisotropy at the lesion epicenter (i.e., loss of directional dependence of water diffusion which is high in the normal spinal cord) as measured by the DTI parameter, fractional anisotropy, and complete loss of fiber tracts traversing the site of injury on tractography were each inversely associated with gait scores (89). Interestingly, of the four deep pain negative dogs that were reported to have no trans-lesional fibers on tractography (two secondary to IVDH and two following vertebral column trauma), none was independently ambulatory. These findings suggest a role for supraspinal input in motor recovery after severe injury in at least some animals but the numbers were small and results require validation in a larger population of dogs.

PREDICTION AND FACILITATION OF SPINAL WALKING

While a variety of factors have been associated with the development of ambulation in dogs with absent pain perception, no predictors in the acute or subacute stage of its subsequent development have yet been established. Considerations worthy of further investigation include clinical parameters such as the onset of spasticity, imaging biomarkers such as DTI indices and tractography, electrodiagnostic evaluation of descending motor tract function or motor neuron pool excitability and serum and cerebrospinal fluid biomarkers of inflammation or structural spinal cord proteins (5, 68, 77, 89–98). While specific markers in serum and cerebrospinal fluid have not been evaluated to predict spinal walking, serum glial fibrillary acidic protein (GFAP) and phosphorylated neurofilament heavy chain (pNFH) have been reported to be useful among deep pain negative dogs in predicting outcome and the development of progressive myelomalacia (96–98). Biomarkers as potential prognostic indicators have been described in detail in the companion article in this issue, "Prognostic Factors in Acute Intervertebral Disc Disease," and it is possible some of these will be useful in this population.

There is also currently limited evidence for specific treatments to facilitate the recovery of ambulation in dogs lacking pain perception. However, a variety of therapeutic interventions have been investigated in experimental models and human SCI to optimize recovery that might prove useful in this population. These include task-specific physical rehabilitation, functional electrical stimulation and epidural stimulation, targeted somatosensory stimulation, treating neuropathic pain, and other pharmacologic interventions (12, 26, 42, 70, 71, 99–108). Importantly, there is growing evidence that multimodal approaches to facilitate motor recovery might prove most useful in improving outcomes in conjunction with traditional approaches directed at the lesion epicenter (36, 70, 109, 110). This is supported by work in chronically sensorimotor complete people and rodent models in which epidural stimulation aimed at motor networks below the level of injury produced some voluntary control of limb function perhaps by unmasking limited residual supraspinal connections (71, 101, 106, 111). Thus, epidural stimulation with locomotor training efforts might be enhanced by combining them with strategies that also promote tissue level recovery at the site of injury. Additionally, combination therapy with task-specific training and chondroitinase ABC in experimental SCI models has been shown to promote regeneration and synergistic plasticity with a greater degree of effective synaptic connections reestablished below injury in an activity dependent manner (112–114). Chondroitinase therapy alone has been shown to be effective in dogs with chronic SCI including recovery of ambulation in 10%, the effect of which might be enhanced by combining it with other treatment modalities (115).

Among dogs lacking pain perception, early in-patient rehabilitation has been suggested as one factor that positively impacted the recovery of ambulation (4). Further evaluation of

specific rehabilitation protocols, focusing on specific components of gait re-training, is warranted. Potassium channel antagonist, 4-aminopyridine, has also been demonstrated to improve ambulation in a subset of chronically paralyzed dogs (69, 116). While promising, the intrinsic value of such therapies as chondroitinase, rehabilitation, or 4-aminopyridine cannot be determined without widespread clinical use in this population. Epidural stimulation or functional electrical stimulation have not been evaluated in dogs with spontaneous SCI, but these techniques might prove useful and preliminary work to develop such devices are underway (117). Overall, exploring multimodal therapeutic approaches will likely prove most useful in enhancing motor recovery after severe, spontaneous SCI in dogs.

AUTHOR CONTRIBUTIONS

ML, NJ, and NO substantially contributed to manuscript concept, preparation, and editing. The additional members of the CANSORT-SCI* contributed to manuscript concept, editing, and review. All authors contributed to the article and approved the submitted version.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fvets.2020.00560/full#supplementary-material

REFERENCES

1. Granger N, Carwardine D. Acute spinal cord injury tetraplegia and paraplegia in small animals. *Vet Clin Small Anim.* (2014) 44:1131–56. doi: 10.1016/j.cvsm.2014.07.013
2. Olby NJ, Levine J, Harris T, Munana K, Skeen T, Sharp N. Long-term functional outcome of dogs with severe injuries of the thoracolumbar spinal cord: 87 cases (1996–2001). *J Am Vet Med Assoc.* (2003) 222:762–9. doi: 10.2460/javma.2003.222.762
3. Scott HW, McKee WM. Laminectomy for 34 dogs with thoracolumbar intervertebral disc disease and loss of deep pain perception. *J Sm Anim Pract.* (1999) 40:417–22. doi: 10.1111/j.1748-5827.1999.tb03114.x
4. Gallucci A, Dragone L, Menchetti M, Gagliardo T, Pietra M, Cardinali M, et al. Acquisition of involuntary spinal locomotion (spinal walking) in dogs with irreversible thoracolumbar spinal cord lesion: 81 dogs. *J Vet Intern Med.* (2017) 31:492–7. doi: 10.1111/jvim.14651
5. Hu HZ, Jeffery ND, Granger N. Somatosensory and motor evoked potentials in dogs with chronic severe thoracolumbar spinal cord injury. *Vet J.* (2018) 237:49–54. doi: 10.1016/j.tvjl.2018.05.007
6. Barbeau H, Rossignol S. Recovery of locomotion after chronic spinalization in the adult cat. *Brain Res.* (1987) 412:84–95. doi: 10.1016/0006-8993(87)91442-9
7. Belanger M, Drew T, Provencher J, Rossignol S. A comparison of treadmill locomotion in adult cats before and after spinal transection. *J Neurophysiol.* (1996) 76:471–91. doi: 10.1152/jn.1996.76.1.471
8. Blauch B. Spinal reflex walking in the dog. *Vet Med Sm Anim Clin.* (1977) 169:173.
9. Brown TG. The intrinsic factors in the act of progression in the mammal. *Proc Royal Soc.* (1911) 84:308–19. doi: 10.1098/rspb.1911.0077

10. Eidelberg E. Consequences of spinal cord lesions upon motor function, with special reference to locomotor activity. *Prog Neurobiol.* (1981) 17:185–202. doi: 10.1016/0301-0082(81)90013-7
11. Handa Y, Naito A, Watanabe S, Komatsu S, Shimizu Y. Functional recovery of locomotive behavior in the adult spinal dog. *Tohoku J Exp Med.* (1986) 148:373–84. doi: 10.1620/tjem.148.373
12. Lovely RG, Gregor RJ, Roy RR, Edgerton VR. Effects of training on the recovery of full-weight-bearing stepping in the adult spinal cat. *Exper Neurol.* (1986) 92:421–35. doi: 10.1016/0014-4886(86)90094-4
13. Naito A, Shimizu Y, Handa Y. Analyses of treadmill locomotion in adult spinal dogs. *Neurosci Res.* (1990) 8:281–90. doi: 10.1016/0168-0102(90)90034-C
14. Sherrington, CS. Flexion-reflex of the limb, crossed extension-reflex and reflex stepping and standing. *J Physiol.* (1910) 40:28–121. doi: 10.1113/jphysiol.1910.sp001362
15. Hall JE. Ch 54 Motor functions of the spinal cord; the cord reflexes. In: Hall JE editor. *Guyton and Hall Textbook of Medical Physiology*. 12th ed. Philadephia: Saunders Elsevier. (2011) p. 655–66.
16. Pearson K Gordon J. Ch 36 Locomotion. In: Kandel E, Schwartz JH, Jessell T, Siegelbaum SA, Hudspeth AJ, editors. *Principles of Neural Science*. 5th ed. New York, NY: McGraw Hill. (2012). p. 812–34.
17. King AS. Pyramidal system and extrapyramidal system. In: King AS, editor. *Physiological and Clinical Anatomy of Domestic Mammals: Central Nervous System Volume 1*. Oxford: Oxford University Press. (1987). p. 141–57.
18. Uemura EE. Ch 16 Motor System. In: Uemura E, editor. *Fundamentals of Canine Neuroanatomy and Neurophysiology*. Ames: Wiley Blackwell. (2015). p. 257–87.
19. Schieber MH. Ch2 comparative anatomy and physiology of the corticospinal system. In: Eisen AA, Shaw PJ, editors. *Handbook of Clinical Neurology*. Amsterdam: Elsevier. (2007). p. 15–37. doi: 10.1016/S0072-9752(07)80005-4
20. Drew T, Jiang W, Widajewicz W. Contributions of the motor cortex to the control of the hindlimbs during locomotion in the cat. *Br Res Reviews.* (2002) 40:178–91. doi: 10.1016/S0165-0173(02)00200-X
21. Lavoie S, Drew T. Discharge characteristics of neurons in the red nucleus during voluntary gait modifications: a comparison with the motor cortex. *J Neurophysiol.* (2002) 88:1791–814. doi: 10.1152/jn.2002.88.4.1791
22. Bussel B, Roby-Brami A, Neris O, Yakovleff A. Evidence for a spinal stepping generator in man. *Paraplegia.* (1996) 34:91–2. doi: 10.1038/sc.1996.15
23. Gerasimenko YP, Makarovskii AN, Nikitin OA. Control of locomotor activity in humans and animals in the absence of supraspinal influences. *Neurosci Behav Physiol.* (2002) 32:417–23. doi: 10.1023/A:1015836428932
24. Guertin PA. The mammalian central pattern generator for locomotion. *Br Res Reviews.* (2009) 62:45–56. doi: 10.1016/j.brainresrev.2009.08.002
25. Rybak, IA, Stecina K, Shevtsova NA, McCrea DA. Modelling spinal circuitry involved in locomotor pattern generation: insights from the effects of afferent stimulation. *J Physiol.* (2006) 577:641–58. doi: 10.1113/jphysiol.2006.118711
26. Zhang SX, Huang F, Gates M, Shen X, Holmberg EG. Early application of tail nerve electrical stimulation-induced walking training promotes locomotor recovery in rats with spinal cord injury. *Spinal Cord.* (2016) 54:942–6. doi: 10.1038/sc.2016.30
27. Kiehn O. Development and functional organization of spinal locomotor circuits. *Curr Opin Neurobiol.* (2011) 21:100–9. doi: 10.1016/j.conb.2010.09.004
28. Shik ML Orlovsky GN. Neurophysiology of locomotor automatism. *Physiol Rev.* (1976) 56:465–501. doi: 10.1152/physrev.1976.56.3.465
29. Edgerton VR, Courtine G, Gerasimenko YP, Lavrov I, Ichiyama RM, Fong AJ, et al. Training locomotor networks. *Brain Res Rev.* (2008) 57:241–54. doi: 10.1016/j.brainresrev.2007.09.002
30. Engberg I, Lundberg A. An electromyographic analysis of muscular activity in the hindlimb of the cat during unrestrained locomotion. *Acta Physiol Scand.* (1969) 75:614–30. doi: 10.1111/j.1748-1716.1969.tb04415.x
31. Rasmussen S, Chan AK, Goslow GE. The cat step cycle: electromyographic patterns for hindlimb muscles during posture and unrestrained locomotion. *J Morp.* (1978) 155:253–70. doi: 10.1002/jmor.1051550302
32. Whelan PJ. Control of locomotion in the decerebrate cat. *Prog Neurobiol.* (1996) 49:481–515. doi: 10.1016/0301-0082(96)00028-7
33. Blight AR, Decrescito V. Morphometric analysis of experimental spinal cord injury in the cat: the relation of injury intensity to survival of myelinated axons. *Neuroscience.* (1986) 19:321–41. doi: 10.1016/0306-4522(86)90025-4
34. Cai LL, Courtine G, Fong AJ, Burdick JW, Roy RR, Edgerton VR. Plasticity of functional connectivity in the adult spinal cord. *Phil Trans R Soc B.* (2006) 361:1635–46. doi: 10.1098/rstb.2006.1884
35. Dimitrijevic MR. Neurophysiology in spinal cord injury. *Paraplegia.* (1987) 25:205–8. doi: 10.1038/sc.1987.35
36. Edgerton VR, Tillakaratne N, Bigbee AJ, de Leon RD, Roy RR. Plasticity of the spinal neural circuitry after injury. *Ann Rev Neurosci.* (2004) 27:145–67. doi: 10.1146/annurev.neuro.27.070203.144308
37. Steward O Willenberg R. Rodent spinal cord injury models for studies of axon regeneration. *Exper Neurol.* (2017) 287:374–83. doi: 10.1016/j.expneurol.2016.06.029
38. Smith AC, Knikou M. A review on locomotor training after spinal cord injury: reorganization of spinal neuronal circuits and recovery of motor function. *Neural Plast.* (2016) 2016:1216258. doi: 10.1155/2016/1216258
39. Bradbury EJ McMahon SB. Spinal cord repair strategies: why do they work? *Nat Rev.* (2006) 7:644–53. doi: 10.1038/nrn1964
40. Filous AR, Schwab JM. Determinants of axon growth, plasticity, and regeneration in the context of spinal cord injury. *Am J Pathol.* (2018) 188:53–62. doi: 10.1016/j.ajpath.2017.09.005
41. Hawthorne AL, Hu H, Kundu B, Steinmetz MP, Wylie CJ, Deneris ES, et al. The unusual response of serotonergic neurons after CNS injury: lack of axonal dieback and enhanced sprouting within the inhibitory environment of the glial scar. *J Neurosci.* (2011) 31:5605–16. doi: 10.1523/JNEUROSCI.6663-10.2011
42. Bradbury EJ, Moon L, Popat RJ, King VR, Bennett GS, Patel PN, et al. Chondroitinase ABC promotes functional recovery after spinal cord injury. *Nature.* (2002) 416:636–40. doi: 10.1038/416636a
43. Brazda N, Estrada V, Voss C, Seide K, Trieu HK, Muller HW, et al. Experimental strategies to bridge large tissue gaps in the injured spinal cord after acute and chronic lesion. *J Vis Exper.* (2016) 110:e53331. doi: 10.3791/53331
44. Hwang DH, Kim HM, Kang YM, Joo In, Cho CS, Yoon BW, et al. Combination of multifaceted strategies to maximize the therapeutic benefits of neural stem cell transplantation for spinal cord repair. *Cell Transplant.* (2011) 20:1361–79. doi: 10.3727/096368910X557155
45. Kang KN, Kim DY, Yoon SM, Lee JY, Lee BN, Kwon JS, et al. Tissue engineered regeneration of completely transected spinal cord using human mesenchymal stem cells. *Biomaterials.* (2012) 33:4828–35. doi: 10.1016/j.biomaterials.2012.03.043
46. Kim BG, Kang YM, Phi JH, Kim YH, Hwang DH, Choi JY, et al. Implantation of polymer scaffolds seeded with neural stem cells in a canine spinal cord injury model. *Cytotherapy.* (2010) 12:841–5. doi: 10.3109/14653249.2010.501784
47. Seo DK, Kim JH, Min J, Yoon HH, Shin ES, Kim SW, et al. Enhanced axonal regeneration by transplanted Wnt3a-secreting human mesenchymal stem cells in a rat model of spinal cord injury. *Acta Neurochir.* (2017) 159:947–57. doi: 10.1007/s00701-017-3097-0
48. Kakulas BA, Kaelan C. The neuropathological foundations for the restorative neurology of spinal cord injury. *Clin Neurol Neurosurg.* (2015) 129:S1–7. doi: 10.1016/j.clineuro.2015.01.012
49. Smith PM, Jeffery ND. Histological and ultrastructural analysis of white matter damage after naturally-occurring spinal cord injury. *Brain Pathol.* (2006) 16:99–109. doi: 10.1111/j.1750-3639.2006.00001.x
50. Blight AR. Cellular morphology of chronic spinal cord injury in the cat: analysis of myelinated axons by line-sampling. *Neuroscience.* (1983) 10:521–43. doi: 10.1016/0306-4522(83)90150-1
51. Fehlings MG, Tator CH. The relationships among the severity of spinal cord injury, residual neurological function, axon counts, and counts of retrogradely labeled neurons after experimental spinal cord injury. *Exper Neurol.* (1995) 132:220–8. doi: 10.1016/0014-4886(95)90027-6
52. Bareyre FM, Kerschensteiner M, Raineteau O, Mettenleiter TC, Weinmann O, Schwab ME. The injured spinal cord spontaneously forms a new intraspinal circuit in adult rats. *Nat Neurosci.* (2004) 7:269–77. doi: 10.1038/nn1195

53. Benthall KN, Hough RA, McClellan AD. Descending propriospinal neurons mediate restoration of locomotor function following spinal cord injury. *J Neurophysiol.* (2017) 117:215–29. doi: 10.1152/jn.00544.2016
54. Courtine G, Song B, Roy RR, Zhong H, Herrmann JE, Ao Y, et al. Recovery of supraspinal control of stepping via indirect propriospinal relay connections after spinal cord injury. *Nature Med.* (2008) 14:69–74. doi: 10.1038/nm1682
55. Flynn JR, Graham BA, Galea MP, Callister RJ. The role of propriospinal interneurons in recovery from spinal cord injury. *Neuropharmacol.* (2011) 60:809–22. doi: 10.1016/j.neuropharm.2011.01.016
56. Ballion B, Morin D, Viala D. Forelimb locomotor generators and quadrupedal locomotion in the neonatal rat. *Eur J Neurosci.* (2001) 14:1727–38. doi: 10.1046/j.0953-816x.2001.01794.x
57. Juvin L, Simmers J, Morin D. Propriospinal circuitry underlying interlimb coordination in mammalian quadrupedal locomotion. *J Neurosci.* (2005) 25:6025–35. doi: 10.1523/JNEUROSCI.0696-05.2005
58. Beauparlant J, van den Brand R, Barraud Q, Friedli L, Musienko P, Dietz V, et al. Undirected compensatory plasticity contributes to neuronal dysfunction after severe spinal cord injury. *Brain.* (2013) 136:3347–61. doi: 10.1093/brain/awt204
59. Gorney AM, Blau SR, Dohse CS, Griffith EH, Williams KD, Lim JH, et al. Mechanical and thermal sensory testing in normal chondrodystrophoid dogs and dogs with spinal cord injury caused by thoracolumbar intervertebral disc herniations. *J Vet Intern Med.* (2016) 30:627–35. doi: 10.1111/jvim.13913
60. Huie JR, Morioka K, Haefeli J, Ferguson AR. What is being trained? How divergent forms of plasticity compete to shape locomotor recovery after spinal cord injury. *J Neurotrauma.* (2017) 34:1831–40. doi: 10.1089/neu.2016.4562
61. Lavrov I, Courtine G, Dy CJ, van den Brand R, Fong AJ, Gerasimenko Y, et al. Facilitations of stepping with epidural stimulation in spinal rats: role of sensory input. *J Neurosci.* (2008) 28:7774–80. doi: 10.1523/JNEUROSCI.1069-08.2008
62. Takeoka A, Vollenweider I, Courtine G, Arber S. Muscle spindle feedback directs locomotor recovery and circuit reorganization after spinal cord injury. *Cell.* (2014) 159:1626–39. doi: 10.1016/j.cell.2014.11.019
63. Hart BL. Facilitation by strychnine of reflex walking in spinal dogs. *Physiol Behav.* (1971) 6:627–8. doi: 10.1016/0031-9384(71)90221-6
64. Naito A, Shimizu Y, Handa Y. Analyses of airstepping movement in adult spinal dogs. *Tohoku J Exp Med.* (1990) 162:41–8. doi: 10.1620/tjem.162.41
65. Liu Z, Ren S, Fu K, Wu Q, Wu J, Hou L, et al. Restoration of motor function after operative reconstruction of the acutely transected spinal cord in the canine model. *Surgery.* (2018) 163:976–83. doi: 10.1016/j.surg.2017.10.015
66. Roeder L, Boonstra TW, Smith SS, Kerr GK. Dynamics of corticospinal motor control during overground walking and treadmill walking in humans. *J Neurophysiol.* (2018) 120:1017–31. doi: 10.1152/jn.00613.2017
67. Gerasimenko Y, Roy RR, Edgerton VR. Epidural stimulation: comparison of the spinal circuits that generate and control locomotion in rats, cats and humans. *Exp Neurol.* (2008) 209:417–25. doi: 10.1016/j.expneurol.2007.07.015
68. Lewis MJ, Howard JF, Olby NJ. The relationship between trans-lesional conduction, motor neuron pool excitability, and motor function in dogs with incomplete recovery from severe spinal cord injury. *J Neurotrauma.* (2017) 34:2994–3002. doi: 10.1089/neu.2017.5012
69. Lewis MJ, Laber E, Olby NJ. Predictors of response to 4-aminopyridine in chronic canine spinal cord injury. *J Neurotrauma.* (2019) 36:1428–34. doi: 10.1089/neu.2018.5975
70. AuYong N, Lu DC. Neuromodulation of the lumbar spinal locomotor circuit. *Neurosurg Clin N Am.* (2014) 25:15–23. doi: 10.1016/j.nec.2013.08.007
71. Van den Brand R, Heutschi J, Barraud Q, DiGiovanna J, Bartholdi K, Huerlimann M, et al. Restoring voluntary control of locomotion after paralyzing spinal cord injury. *Science.* (2012) 336:1182–5. doi: 10.1126/science.1217416
72. Lewis MJ, Cohen EB, Olby NJ. Magnetic resonance imaging features of dogs with incomplete recovery after acute, severe spinal cord injury. *Spinal Cord.* (2018) 56:133–41. doi: 10.1038/s41393-017-0004-8
73. Gordon-Evans WJ, Evans RB, Knap KE, Hildreth JM, Pinel CB, Imhoff DJ, et al. Characterization of spatiotemporal gait characteristics in clinically normal dogs and dogs with spinal cord disease. *Am J Vet Res.* (2009) 70:1444–9. doi: 10.2460/ajvr.70.12.1444
74. Lewis MJ, Williams KD, Langley T, Jarvis LM, Sawicki GS, Olby NJ. Development of a novel gait analysis tool measuring center of pressure for evaluation of canine chronic thoracolumbar spinal cord injury. *J Neurotrauma.* (2019) 36:3018–25. doi: 10.1089/neu.2019.6479
75. Song RB, Oldach MS, Basso DM, da Costa RC, Fisher LC, Mo X, et al. A simplified method of walking track analysis to assess short-term locomotor recovery after acute spinal cord injury caused by thoracolumbar intervertebral disc extrusions in dogs. *Vet J.* (2016) 210:61–7. doi: 10.1016/j.tvjl.2016.01.013
76. Hamilton L, Franklin RJ, Jeffery ND. Quantification of deficits in lateral paw positioning after spinal cord injury in dogs. *BMC Vet Res.* (2008) 4:47. doi: 10.1186/1746-6148-4-47
77. Lewis MJ, Olby NJ. Development of a clinical spasticity scale for evaluation of dogs with chronic thoracolumbar spinal cord injury. *Am J Vet Res.* (2017) 78:854–61. doi: 10.2460/ajvr.78.7.854
78. D' Amico JM, Condliffe EG, Martins KJ, Bennett DJ, Gorassini MA. Recovery of neuronal and network excitability after spinal cord injury and implications for spasticity. *Front Integr Neurosci.* (2014) 8:36. doi: 10.3389/fnint.2014.00049
79. Johnson RL, Gerhart KA, McCray J, Menconi JC, Whiteneck GG. Secondary conditions following spinal cord injury in a population-based sample. *Spinal Cord.* (1998) 36:45–50. doi: 10.1038/sj.sc.3100494
80. Milinis K, Young CA and on behalf of the Trajectories of Outcome in Neurological Conditions (TONiC) study (2105). Systematic review of the influence of spasticity on quality of life in adults with chronic neurological conditions. *Disabil Rehabil.* (2015) 29:1–11. doi: 10.3109/09638288.2015.1106592
81. Noonan VK, Kopec JA, Zhang H, Dvorak MF. Impact of associated conditions resulting from spinal cord injury on health status and quality of life in people with traumatic central cord syndrome. *Arch Phys Med Rehabil.* (2008) 89:1074–82. doi: 10.1016/j.apmr.2007.10.041
82. Westerkam D, Saunders LL, Krause JS. Associations of spasticity and life satisfaction after spinal cord injury. *Spinal Cord.* (2011) 49:990–4. doi: 10.1038/sc.2011.49
83. Edgerton VR, Roy RR, Hodgson JA, Prober RJ, de Guzman CP, de Leon R. Potential of adult mammalian lumbosacral spinal cord to execute and acquire improved locomotion in the absence of supraspinal input. *J Neurotrauma.* (1992) 9:S119–28.
84. Zhang SX, Huang F, Gates M, White J, Holmberg EG. Tail nerve electrical stimulation induces body weight-supported stepping in rats with spinal cord injury. *J Neurosci Methods.* (2010) 187:183–9. doi: 10.1016/j.jneumeth.2010.01.008
85. Piazza S, Serrano-Munoz D, Gomez-Soriano J, Torricelli D, Segura-Fragosa A, Pons JL, et al. Afferent electrical stimulation during cycling improves spinal processing of sensorimotor function after incomplete spinal cord injury. *Neuro Rehabil.* (2017) 40:429–37. doi: 10.3233/NRE-161430
86. Promkeaw D, Arrayawichanon P, Thaweewannakij T, Mato L, Amatachaya P, Amatachaya S. Various surfaces challenge gait characteristics of ambulatory patients with spinal cord injury. *Spinal Cord.* (2019) 57:805–13. doi: 10.1038/s41393-019-0282-4
87. Granger N, Blamires H, Franklin RJ, Jeffery ND. Autologous olfactory mucosal cell transplants in clinical spinal cord injury: a randomized double-blinded trial in a canine translational model. *Brain.* (2012) 135:3227–37. doi: 10.1093/brain/aws268
88. Alisauskaite N, Spitzbarth I, Baumgärtner W, Dziallas P, Kramer S, Dening R, et al. Chronic post-traumatic intramedullary lesions in dogs, a translational model. *PLoS ONE.* (2017) 12:e0187746. doi: 10.1371/journal.pone.0187746
89. Lewis MJ, Yap PT, McCullough S, Olby NJ. The relationship between lesion severity characterized by diffusion tensor imaging and motor function in chronic canine spinal cord injury. *J Neurotrauma.* (2018) 35:500–507. doi: 10.1089/neu.2017.5255
90. Amendt HL, Siedenburg JS, Steffensen N, Kordass U, Rohn K, Tipold A, et al. Correlation between severity of clinical signs and transcranial magnetic motor evoked potentials in dogs with intervertebral

disc herniation. *Vet J.* (2017) 221:48–53. doi: 10.1016/j.tvjl.2017.01.015
91. Kwon BK, Stammers AM, Belanger LM, Bernardo A, Chan D, Bishop CM, et al. Cerebrospinal fluid inflammatory cytokines and biomarkers of injury severity in acute human spinal cord injury. *J Neurotrauma.* (2010) 27:669–82. doi: 10.1089/neu.2009.1080
92. Kwon BK, Streijger F, Fallah N, Noonan VK, Bélanger LM, Ritchie L, et al. Cerebrospinal fluid biomarkers to stratify injury severity and predict outcome in human traumatic spinal cord injury. *J Neurotrauma.* (2017) 34:567–80. doi: 10.1089/neu.2016.4435
93. Lubieniecka JM, Streijger F, Lee JH, Stoynov N, Liu J, Mottus R, et al. Biomarkers for severity of spinal cord injury in the cerebrospinal fluid of rats. *PLoS ONE.* (2011) 6:e19247. doi: 10.1371/journal.pone.0019247
94. Rodrigues LF, Moura-Neto V, Spohr ETCLS. Biomarkers in spinal cord injury: from prognosis to treatment. *Mol Neurobiol.* (2018) 55:6436–48. doi: 10.1007/s12035-017-0858-y
95. Wang-Leandro A, Siedenburg JS, Hobert MK, Dziallas P, Rohn K, Stein VM, et al. Comparison of preoperative quantitative magnetic resonance imaging and clinical assessment of deep pain perception as prognostic tools for early recovery of motor function in paraplegic dogs with intervertebral disk herniations. *J Vet Interm Med.* (2017) 31:842–8. doi: 10.1111/jvim.14715
96. Nishida H, Nakayama M, Tanaka H, Kamishina H, Izawa T, Hatoya S, et al. Evaluation of serum phosphorylated neurofilament subunit NF-H as a prognostic biomarker in dogs with thoracolumbar intervertebral disc herniation. *Vet Surgery.* (2014) 43:289–93. doi: 10.1111/j.1532-950X.2014.12144.x
97. Olby NJ, Lim JH, Wagner N, Zidan N, Early PJ, Mariani CL, et al. Time course and prognostic value of serum GFAP, pNFH, and S100B concentrations in dogs with complete spinal cord injury because of intervertebral disc extrusion. *J Vet Intern Med.* (2019) 33:726–34. doi: 10.1111/jvim.15439
98. Sato Y, Shimamura S, Mashita T, Kobayashi S, Okamura Y, Katayama M, et al. Serum glial fibrillary acidic protein as a diagnostic biomarker in dogs with progressive myelomalacia. *J Vet Med Sci.* (2013). 75:949–53. doi: 10.1292/jvms.12-0483
99. Backus D, Cordo P, Gillott A, Kandilakis C, Mori M, Raslan AM. Assisted movement with proprioceptive stimulation reduces impairment and restores function in incomplete spinal cord injury. *Arch Phys Med Rehabil.* (2014) 95:1447–53. doi: 10.1016/j.apmr.2014.03.011
100. Beekhuizen KS, Field-Fote EC. Sensory stimulation augments the effects of massed practice training in persons with tetraplegia. *Arch Phys Med Rehabil.* (2008) 89:602–8. doi: 10.1016/j.apmr.2007.11.021
101. Calvert JS, Grahn PJ, Zhao KD, Lee KH. Emergence of epidural electrical stimulation to facilitate sensorimotor network functionality after spinal cord injury. *Neuromodulation.* (2019) 22:244–52. doi: 10.1111/ner.12938
102. Cragg JJ, Haefeli J, Jutzeler CR, Röhrich F, Weidner N, Saur M, et al. Effects of pain and pain management on motor recovery of spinal cord-injured patients: a longitudinal study. *Neurorehabil Repair.* (2016) 30:753–61. doi: 10.1177/1545968315624777
103. Harkema SJ, Hilyer J, Schmidt-Read M, Ardolino E, Sisto SA, Behrman AL. Locomotor training: as a treatment of spinal cord injury and in the progression of neurologic rehabilitation. *Arch Phys Med Rehabil.* (2012) 93:1588–97. doi: 10.1016/j.apmr.2012.04.032
104. Ichiyama RM, Gerasimenko YP, Zhong H, Roy RR, Edgerton VR. Hindlimb stepping movements in complete spinal rats induced by epidural spinal cord stimulation. *Neurosci Lett.* (2005) 383:339–44. doi: 10.1016/j.neulet.2005.04.049
105. James ND, McMahon SB, Field-Fote EC, Bradbury EJ. Neuromodulation in the restoration of function after spinal cord injury. *Lancet.* (2018) 17:905–17. doi: 10.1016/S1474-4422(18)30287-4
106. Minassian K Hofstoetter US. Spinal cord stimulation and augmentative control strategies for leg movement after spinal paralysis in humans. *CNS Neurosci Ther.* (2016) 22:262–70. doi: 10.1111/cns.12530
107. Rejc E, Angelia C, Harkema S. Effects of lumbosacral spinal cord epidural stimulation for standing after chronic complete paralysis in humans. *PLoS ONE.* (2015) 10:e0133998. doi: 10.1371/journal.pone.0133998
108. Tillakaratne NJ, de Leon RD, Hoang TX, Roy RR, Edgerton VR, Tobin AJ. Use-dependent modulation of inhibitory capacity in the feline lumbar spinal cord. *J Neurosci.* (2002) 22:3130–43. doi: 10.1523/JNEUROSCI.22-08-03130.2002
109. Branco É, Alves JGR, Pinheiro LL, Coutinho LN, Gomes CRM, Galvão GR, et al. Can paraplegia by disruption of the spinal cord tissue be reversed? The signs of a new perspective. *Anat Rec.* (2019) 303:1812–20. doi: 10.1002/ar.24262
110. Courtine G, Sofroniew MV. Spinal cord repair: advances in biology and technology. *Nat Med.* (2019) 25:898–908. doi: 10.1038/s41591-019-0475-6
111. Angeli CA, Edgerton VR, Gerasimenko YP, Harkema SJ. Altering spinal cord excitability enables voluntary movements after chronic complete paralysis in humans. *Brain.* (2014) 137:1394–409. doi: 10.1093/brain/awu038
112. Alluin O, Delivet-Mongrain H, Gauthier MK, Fehlings MG, Rossignol S, Karimi-Abdolrezaee S. Examination of the combined effects of chondroitinase ABC, growth factors and locomotor training following compressive spinal cord injury on neuroanatomical plasticity and kinematics. *PLoS ONE.* (2015) 9:e111072. doi: 10.1371/journal.pone.0111072
113. García-Alías G, Fawcett JW. Training and anti-CSPG combination therapy for spinal cord injury. *Exper Neurol.* (2012) 235:26–32. doi: 10.1016/j.expneurol.2011.09.009
114. Shinozaki M, Iwanami A, Fujiyoshi K, Tashiro S, Kitamura K, Shibata S, et al. Combined treatment with chondroitinase ABC and treadmill rehabilitation for chronic severe spinal cord injury in adult rats. *Neurosci Res.* (2016) 113:37–47. doi: 10.1016/j.neures.2016.07.005
115. Hu HZ, Granger N, Pai SB, Bellamkonda RV, Jeffery ND. Therapeutic efficacy of microtube-embedded chondroitinase ABC in a canine clinical model of spinal cord injury. *Brain.* (2018) 141:1017–27. doi: 10.1093/brain/awy007
116. Lim JH, Muguet-Chanoit AC, Smith DT, Laber E, Olby NJ. Potassium channel antagonists 4-aminopyridine and the t-butyl carbamate derivative of 4-aminopyridine improve hind limb function in chronically non-ambulatory dogs: a blinded, placebo-controlled trial. *PLoS ONE.* (2014) 9:e0116139. doi: 10.1371/journal.pone.0116139
117. Taghavi N, Luecke GR, Jeffery ND. A wearable body controlling device for application of functional electrical stimulation. *Sensors.* (2018) 18:1251. doi: 10.3390/s18041251

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REVIEW
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Clinical Trial Design—A Review—With Emphasis on Acute Intervertebral Disc Herniation

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There is a clear need for new methods of treatment of acute disc herniation in dogs, most obviously to address the permanent loss of function that can arise because of the associated spinal cord injury. Clinical trials form the optimal method to introduce new therapies into everyday clinical practice because they are a reliable source of unbiased evidence of effectiveness. Although many designs are available, parallel cohort trials are most widely applicable to acute disc herniation in dogs. In this review another key trial design decision—that between pragmatic and explanatory approaches—is highlighted and used as a theme to illustrate the close relationship between trial objective and design. Acute disc herniation, and acute spinal cord injury, is common in dogs and there is a multitude of candidate interventions that could be trialed. Most current obstacles to large-scale clinical trials in dogs can be overcome by collaboration and cooperation amongst interested veterinarians.

Keywords: explanatory, pragmatic, durotomy, glyburide, spinal cord injury

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INTRODUCTION

Introduction of new medical interventions into everyday practice requires assessment of safety, effectiveness and, preferably, comparison with currently available therapies. These assessments are undertaken in the form of clinical trials. The typical process for clinical trial development follows a series of phases. Phase 0 trials involve a small number of subjects and often represent the first exposure of a drug or intervention to the target species, with the goal of learning how a drug interacts with the target. Phase I trials are typically "dose finding" studies that aim to define the optimum dose or regimen for therapy as well as delineating safety and defining toxicities associated with a new treatment in a healthy and, sometimes, a diseased population. Phase II and III trials have the goal of further establishing safety in the target population, assessing effectiveness or efficacy, and then validating those findings in larger populations of patients with the disease of interest.

Phase II and III randomized controlled trials (RCTs) are the "gold standard" for assessment of impact and are rigorously planned clinical experiments designed to minimize all sources of bias (1). There are many trial designs within this category, but the most common is the parallel group trial in which a population of affected participants is randomly allocated to receive either the novel intervention or the standard-of-care therapy (or placebo). The outcome of interest is then measured at a specified time after the intervention and the numbers or proportion of responders, or mean response, compared between groups. Although all clinical trial phases are important for therapeutic development in the context of spinal cord injury for dogs with intervertebral disc herniation, the

focus of this review will be on Phase II and Phase III studies, with some mention of the Phase I studies in terms of the preparatory data they provide.

EXPLANATORY VS. PRAGMATIC

Clinical trials can also be categorized according to whether they aim to be *explanatory* or *pragmatic*, and this distinction is important because it provides critical underpinning to the way that trials are designed. This dichotomy in aim is often overlooked in veterinary medicine but will provide a theme throughout this review (2). There is a range of criteria by which to decide whether a trial is pragmatic or explanatory (3) and many trials do contain aspects of both aims. However, fundamentally an explanatory trial aims to determine whether an intervention will work under ideal circumstances (i.e., "can this intervention work?"), whereas a pragmatic trial aims to determine if there is a benefit when it is applied in everyday practice (4). This difference has a multitude of secondary implications, most notably the tendency to strictly limit participant entry in explanatory trials and to restrict analysis to only those participants that completed every component of intervention and outcome measurement. On the whole, *pragmatic* trials are likely to have greater impact in everyday clinical medicine, because they have broader application.

There is often tension between these two approaches for veterinarians carrying out trials of interventions for spinal cord injury in dogs, because pragmatic trials would tend to be more useful for veterinary clinicians, whereas explanatory trials tend to be more useful in answering questions that arise from the "basic science" of spinal cord injury. In this context, an explanatory trial might tightly limit lesion location, injury severity, time since onset of injury and have detailed and complex outcome measures. All of these aspects might be matched with the tightly-controlled circumstances in which the intervention was previously applied in experimental animals, thereby providing a "proof of principle" that the intervention can translate from laboratory to clinic. Such a trial is more likely to find favor with a "basic science" spinal cord injury researcher. Nevertheless, researchers interested in developing interventions that can be translated from laboratory to clinic would also need to be cognizant that an intervention that works in a limited patient population under ideal circumstances and with a complex outcome measure used to detect benefit might not necessarily have useful clinical impact in humans—or pet dogs—with spinal cord injury. In contrast, a pragmatic trial might include all cases of thoracolumbar spinal cord injury and might focus its outcome measures on owner judgement of each animal's level of function and perceived quality of life. This type of trial will more likely find favor with veterinarians treating such cases, and also with researchers who have a strong interest in translational research: if an intervention can still show effectiveness even when used in sub-optimal circumstances it is likely to be sufficiently robust to translate into human spinal cord injury. The downside to this type of trial is that it may be difficult to understand why a treatment fails in a pragmatic trial; the loose inclusion criteria and outcome assessments may obscure a real effect that is lost in the noise of other competing effects.

Both explanatory and pragmatic trials have value and, in practice, many trials will incorporate aspects of both study aims; nevertheless, it is critical to consider these two, sometimes conflicting, aims during the design process.

KEY ELEMENTS OF A RANDOMIZED CONTROLLED TRIAL

The classic design of Phase II/III RCT may be explanatory or pragmatic and has many prerequisites, notably *random* allocation of individuals to experimental and control groups, *concealment* of allocation before enrolment, assessment of follow-up by *blinded observers*, *pre-specified* definition of outcome assessment methods and comparisons and, usually, enrolment of *large groups* of participants (so as to be able to apply effective randomization). Best practices in the design of RCT have been formalized and published in human medicine under the recommendations of the CONSORT statement (http://www.consort-statement.org/), and this set of guidelines can also readily be applied to veterinary clinical trials.

An important ethical consideration before undertaking an RCT, is whether there is clinical "equipoise." A state of equipoise exists if there is a balance of expert opinion between the two interventions that are being assessed in terms of their effectiveness, or if there is a degree of uncertainty across the field with respect to the efficacy of a particular intervention (5). For instance, although there is evidence that fenestration alone provides similar functional outcomes as decompressive surgery for "deep pain positive" pelvic limb paralysis or paresis following acute thoracolumbar intervertebral disc herniation (6), the expert consensus is that there IS a difference in outcome between these interventions. Therefore, a trial comparing these two options would not currently be considered ethical because expert opinion does not consider them equal in value. Although decompressive surgery has not been proven as the standard of care through a RCT, it has become so by default through synthesis of other types of evidence and through expert opinion.

For a variety of reasons, many aspects of RCTs can be difficult to achieve in veterinary medicine and, in general, there are few reported large-scale RCTs in animals [although note (7)]. However, a small number of clinical trials have already been carried out in dogs with acute and chronic spinal cord injury, most of which take an explanatory approach to trial design. Because one of the ultimate goals of CANSORT-SCI is to provide data on dogs with spinal cord injury that can lead to new therapies effective at the population level, the emphasis in this overview will be on construction of large-scale *pragmatic* trials, a design so far used less commonly in veterinary medicine but most likely to change how spinal cord-injured dogs are handled in future.

When considering an RCT there are several key questions to answer:

- *Does the trial aim to be explanatory or pragmatic?*
- *What population will be examined?*

- *What intervention will be applied?*
- *What will the comparator be?*
- *What outcome measure will be used?*
- *What degree of improvement will be detected? (Including its clinical impact).*

SELECTION OF CASES

Although there is much to be learned about how best to treat dogs with spinal cord injuries of all types, those that have incurred acute thoracolumbar disc herniation are most in need of new therapies. This is partly because it is the most common type of injury (8), and partly because there is a recognized poor prognosis for dogs in some sub-categories of this cohort (9, 10). The main impetus that drives the perceived need for a new therapy for spinal cord injury in dogs is the lower proportion of dogs that recover locomotion (and other functions) after presenting with loss of "deep pain sensation" following acute thoracolumbar intervertebral disc herniation. In this sub-group, the proportion that recover independent quadrupedal locomotion is usually estimated to be around 55%, in contrast to the estimated 90–95% recovery for dogs that present with "deep pain sensation" intact (6, 10, 11). Furthermore, most of these deep pain negative dogs do not recover appropriate autonomic function either. It is currently a major source of frustration for owners and veterinarians alike that we cannot offer anything better for these patients and so this review will focus on this specific sub-set.

Refining Inclusion Criteria

As well as having a clear clinical need for new therapy, dogs that present as "deep pain negative" can almost immediately be identified as potential trial candidates. However, although these cases can be rapidly recognized, it is important to note that this group is not homogenous. Such "deep pain negative" cases have variable duration and rate of onset, delay before presentation, severity of compression and inter-animal variation in body weight or conformation and so they can be further sub-divided if necessary, and this choice might be guided by deciding whether the trial has pragmatic or explanatory aims. For instance, it could be considered that a trial to investigate a putative therapy should be restricted to dogs that present within a specific time window and are of a specific age (i.e., leaning toward a more explanatory design). The advantage of investigating treatment effects in a sub-group of the whole population is that if they are more homogenous then the signal-to-noise ratio of any treatment effect can be more readily discerned. The drawback is that the proportion of cases within each sub-group will of course be smaller than the total population, so causing more difficulties with case recruitment adequate to achieve the prerequisite sample size.

Questions can also be asked about whether to restrict entry to a trial to specific types or sizes of dog. An important corollary is that, strictly, trial results only guide treatment of similar types of patient in the future. For instance if a drug for diabetes (in people) was successful in trials in obese males over 50 years old, there might be doubt about whether the results might apply to underweight 15 year old female patients. This aspect of clinical trial interpretation is known as the *generalizability* (1) and must be used to inform design. For dogs with spinal cord injury after intervertebral disc herniation the majority of cases will be middle-aged chondrodystrophic dogs and so there might be merit in restricting trial entry to these cases. The results could then be used to apply to the most commonly affected patients in future. On the other hand, if a 4 year-old German shepherd were to present with an acute herniation in future then we might not necessarily expect the same results as were obtained in the trial.

The alternative to restricting trial entry is to set up a more *pragmatic* trial, in which all-comers can be included. A possible drawback to more eclectic enrolment, especially when considering spinal cord injury in dogs, is that the trial arms easily become unbalanced through inclusion of relatively unusual cases (because they may randomize to one or other arm only), unless large numbers of cases are included. Another aspect of specific case selection that might apply regarding dog size is the widespread perception that recovery is different between large and small dogs (12). Again, if the treatment groups are sufficiently large this does not cause a problem—large dogs can be assumed to randomize equally to the two arms of the trial.

Although there is reason to think that deep pain negative dogs constitute the group most in need of new therapy, it could also be questioned whether there might be a need to investigate whether the recovery rate for dogs that are deep pain positive might also be enhanced. This enhanced recovery might take the form of a greater proportion recovering to walk or that the recovery could be made more rapid or more complete. Because of the inherent need for more complex outcome measures for these patients, it is likely that designs for studies on these dogs will be explanatory rather than pragmatic.

INTERVENTION

Selection of test interventions in clinical trials is usually based on pre-clinical data, which have generally been derived in experimental animals. When applied to human clinical trials, the steps toward a Phase II/III RCT would usually include a Phase 0 or Phase I "first-in-human" trial to assess toxicity and, depending on the nature of the intervention, often also include pharmacokinetic and pharmacodynamic studies to determine optimal dosing regimens. Appropriate surgical or physical therapy interventions are often less formally assessed at the pre-clinical stage because the relevant procedures may not be feasible or appropriate in experimental animal subjects. Traditionally, new therapeutic interventions in veterinary medicine are often derived from human medicine, but in spinal cord injury there are no therapies available for treatment of humans that are not available for dogs. In both species, treatment consists of care to maintain blood pressure, spinal cord decompression, and vertebral stabilization if appropriate, physical therapy and allowing plenty of time for nervous system recovery and plasticity (10, 13, 14).

There is a huge number of interventions that could potentially be applied to dogs with spinal cord injury, many of which have

been extensively tested in laboratory animals over many years [e.g., (15–17)]. The decision as to which to select for further evaluation through RCTs in clinical cases might be determined by many factors, most notably knowledge about toxicity, the feasibility of appropriate dosing and the feasibility of application within a time period in which the agent has been shown to be effective. For instance, although tetrodotoxin can reduce the loss of spinal cord tissue and function after injury (18) it has very serious potential toxicity and was ineffective when applied 4 h after injury (19). Unfortunately, only about 15% of canine cases of spinal cord injury are presented to a specialty care facility within 8 h, with a much small proportion likely presented within 4 h; therefore, most cases cannot be treated at an appropriate specialist center within such a short period after injury (20). Alternatively, if we were to consider that suitable cases for a clinical trial were dogs that had chronic spinal cord injury—i.e., that they had an acute spinal cord injury from which they made an incomplete recovery—then the time period for the intervention becomes much less critical and a different series of intervention options is available.

Of the multitude of available medication interventions that might be useful based on reported success in laboratory animal models, many could plausibly be converted into clinical therapies in dogs. It is reasonable to consider that a credible subject for a RCT in dogs would be one that has shown benefits in experiments in more than one laboratory and more than one model of injury. Prominent examples of medications that meet these criteria and could be used acutely as neuroprotective strategies for spinal cord injury include riluzole (21), glyburide (22), and minocycline (17), all of which have been the focus of (Phase I or Phase II) clinical trials in people (see ClinicalTrials.gov). Again, it must be asked whether any of these agents can be applied rapidly enough after the injury in pet dogs, for which the median time to presentation is 24 h, to be beneficial (20).

In terms of surgical interventions, there is accumulating evidence that durotomy/duroplasty may be of value in reducing the intraparenchymal spinal cord pressure (thereby improving blood flow) in humans (23), experimental animals (24) and, recently, clinical canine patients (25, 26). This intervention has the benefit of being applicable for many hours, or even days, following an acute spinal cord injury and so could readily be translated into clinical veterinary practice. Currently there is clinical equipoise regarding this intervention, with closely balanced evidence for and against.

None of the interventions mentioned above are complicated to apply and so could all be used within pragmatic and explanatory trial frameworks.

COMPARATOR

The comparison therapy for dogs entered into a RCT to test an intervention for acute spinal cord injury would be "standard care," which would consist of cross-sectional imaging and decompressive surgery (9, 10). Placebo therapy would not be a credible (nor ethical, see below) option in view of current clinical thinking and in some jurisdictions (the UK) is not permitted. Even so, "standard care" is not well-defined, especially in terms of anesthesia protocols, fluid therapy before and during surgery and, especially, post-operative care and physical therapy.

Therefore, other peri-operative therapies might require recording, or might require controlling through defined inclusion/exclusion criteria, when investigating a new intervention, although, again, the need to limit would be determined by how much the balance lay toward an explanatory approach. It is possible that some routine interventions might impact the results or interact with the trial therapy. There is some limited evidence that physical therapy for spinal cord-injured humans can have an impact on outcome (27, 28). Some data suggest benefits in dogs too (29), although a previous RCT on this subject did not support this conclusion (30). Nevertheless, at present there is no single protocol defined for physical therapy after decompressive spinal surgery in dogs, or which sub-population (if any) is most likely to derive benefit, and so this might need attention during a clinical trial. In large pragmatic trials it would not be necessary to define specific physical therapy protocols because the effects of any specific regimen would be expected to randomize equally between treatment groups amongst the large patient numbers, but this might not apply in smaller trials. Similarly, use of other drugs before and after surgery might also require controlling, or recording for inclusion as an analytical covariable. For instance, glucocorticoids have been examined for their effects in numerous studies (31), albeit without strong evidence of effect. More recently, opiates, specifically kappa opioids, have been implicated in worsening outcomes in experimental animals with spinal cord injury (32).

OUTCOME ASSESSMENT

In every clinical trial it is imperative to define an outcome measure that will be used to define whether the intervention has achieved its goal of improving patient outcome. It is also necessary that the outcome be defined BEFORE the trial so as to prevent selective reporting after the data are collected, implying that it requires careful consideration. Useful outcome measures vary a great deal—between those that directly measure a key outcome that is definitely important to a participant, such as death—to those that are termed "*surrogate outcomes*" and measure something related to a key outcome but is not that key outcome—for instance, the area of MRI abnormalities in the brain of patients that have multiple sclerosis [e.g., (33)]. A pragmatic approach will use easily measured outcomes, which might be loosely defined, whereas explanatory trials will aim to quantify outcomes more precisely, often using relatively complex schemes or equipment.

Specific Definitions Are Required

For spinal cord injury trials in dogs the most obvious outcome measure would be the ability to walk, especially since that is usually the lost function that encourages owners to seek veterinary advice. There is much merit in using this as an outcome measure, but there are some details that require attention when designing a trial. For instance, what does "ability

to walk" mean: how many consecutive steps defines a dog as being "able to walk?" Does this apply to every walking surface? The most difficult aspect when recording the ability to walk after severe spinal cord injury in dogs is their ability to (sometimes) develop the ability to "spinal walk"—which is usually defined as the ability to walk without any clinical evidence of neural communication between the head and the limbs (i.e., clinically, dogs with persistent loss of deep pain sensation) [(29, 34) and see Lewis et al., companion article in this issue]. In terms of analysis of the effectiveness of a putative therapy, explanatory trials would wish to exclude recovery that is mediated by "spinal walking" because it does not reflect true regulation of locomotion from the brain and cervical spinal cord (i.e., the trial question is: "can the intervention restore the brain's ability to control the pelvic limbs?"). However, the pragmatic approach would be that if the dog ***can*** walk it does not matter whether the dog is able to voluntarily regulate this motion or not (i.e., the trial question is: "can the dog get around the yard?"). Both are acceptable outcomes but it is essential that these possibilities are considered before the trial and that the appropriate measure is used to address the question that is posed.

Recently the ability to "walk 10 (consecutive) steps" has increasingly been taken as a relatively pragmatic indication of recovery of locomotion [e.g., (35)]. Originally, this measure was used because many dogs that spinal walk are not able to take as many as 10 consecutive steps, although this does not encompass ALL spinal walking dogs (see Lewis et al., this issue) and there is also a question as to whether the flooring surface should be considered too. One advantage of using the 10 step "convention" is that by becoming a recognized standard the results of trials carried out on different interventions can be (broadly) compared. There is of course nothing special about 10 steps as opposed to 20 steps etc., but usually animals that can recover to walk 10 steps can then also go on to improve and walk further.

Outcome Observer

The other question that has to be asked about a simple outcome measure such as walking, is who will make the final decision on the outcome? Will it be the owner or will it be a veterinarian, or a specialist veterinarian? The answers that are given by each may well be different. A specialist neurologist is much more likely to identify spinal walking compared to an owner, and they are much more likely to feel that the distinction is important. Nowadays it might also be asked about whether the outcome can be determined remotely: can the specialist running the trial make a determination of whether a dog can walk (or urinate voluntarily) by observing a video recording? Fortunately, recent work has suggested that it is relatively straightforward to train observers to a common outcome (36). A related question is the *blinding* of outcome observers. It is critical that those running the trial—and ideally owners too—are unaware of which treatment arm their dog has been allocated to. The people running the trial can reasonably be expected to be biased and owners might also interpret outcome in light of wishing to find that the new therapy is beneficial. Therefore, it is imperative that recording of outcome is done by someone who is unaware of the treatment allocation. This requirement can be problematic in veterinary medicine because of the limited personnel available in many clinics, including those trained to ask penetrating questions about voluntary function observed in the dogs, or to carry out more complex evaluations of function.

Although the ability to walk is an obvious and relevant outcome measure there are many other approaches to outcome assessment after spinal cord injury in dogs. In spinal cord injury research on experimental animals, locomotor scoring schemes have been applied for decades, most recently in the open-field "BBB scale" that grades the use of each pelvic limb and the coordination between thoracic and pelvic limb girdles (37). Similar schemes have been devised for use in dogs (38, 39) and all carry the advantage of allowing the ***quality*** of locomotion to be assessed, so implying that grades of recovery can be measured. However, there are also drawbacks, most notably that these scores are not truly numerical (and so are ordinal rather than continuous scales), which complicates interpretation (40), and there is also a great deal of inter-animal variability in outcome, even in rats that have incurred highly-regulated identical injuries (41). In addition, although easily applied in practice, these scales are designed to detect a surrogate outcome—one that is collected for the purposes of a trial rather than to detect a useful clinical benefit. The relationship between (small) improvement on these scales and clinical function is uncertain.

Kinetics and kinematics provide even more finely graded outcomes and kinematic analysis can be especially valuable because it can imply conduction across a lesion in the thoracolumbar area through detection of coordination of phase patterns of thoracic and pelvic limb stepping (42). These outcomes have been used to assess outcome in canine spinal cord injury trials because they are able to detect subtle changes in function (43, 44) that might realistically be expected to occur following an intervention in severely and chronically affected individuals. On the other hand, kinematic measures are clearly *surrogate* outcomes, especially when applied to animals walking on a treadmill, and it can also be argued that the detection of small differences in function might not have much clinical relevance. Again the choice here is outlined as a distinction between pragmatic ("can the dog walk?") and explanatory ("is there a change in kinematics?") outcomes and reiterate the need to select the outcome that is most appropriate for each individual trial.

Alternative outcomes, most notably bladder control, may also be usefully examined. Many owners nowadays are not all that concerned if their dog cannot walk, since they can be adequately mobile in carts, but they may be much more concerned about urinary control. There are many methods to define urinary control, ranging from the pragmatic (e.g., "does the dog urinate in the house?") to more precise, but clinically remote, outcomes such as bladder compliance (the ability of the bladder to accommodate increasing urine volume). The use of bladder compliance as an outcome for a canine spinal cord injury trial has previously been described (44), although there are currently gaps in knowledge regarding normal bladder function in dogs (see companion article in this issue).

Finally, electrodiagnostic tests, particularly sensory and motor evoked potentials that measure long tract function, can be used as

outcome measures (44). These are clearly *explanatory* outcomes and primarily used as an aid to interpreting mechanisms of change in function associated with an intervention.

HOW MANY CASES WILL BE NEEDED?

The number of participants needed to be enrolled in a clinical trial is determined by sample size calculations, which are determined by the desired power of the study and the false positive rate that is acceptable. There is also a difference in numbers needed depending on whether the sample size will be calculated based on a change in proportion reaching a specific criterion, or whether a specified difference in mean values is used. On the whole, power of clinical trials is set at 0.8 or 0.85 (*i.e.* $\beta = 0.2$ or 0.15) and the false positive value is usually set at 0.05 (*i.e.* $\alpha = 0.05$). The methods for calculating sample size are widely available online and contained in various software packages as well as in numerous publications [MedCalc.net (45)].

Sample size calculations also depend on the size of the difference in outcome between groups that is being sought and the variability in the measure between and within individuals. Smaller differences in outcome and greater variability demand larger sample sizes. Variability in outcome can occur because of variability within and between individuals and can also result from imprecision in measurement.

If determining sample size based on proportions reaching a specified criterion (e.g., ability to walk 10 steps) it is necessary to have a reasonable estimate of the outcome after standard therapy (usually derived from previous publications) and to then estimate the proportion that might recover following the test intervention. This estimation is best derived from preliminary data, but can also be based on what might realistically be useful in the clinic, which might, in turn, depend on the invasiveness or toxicity of that intervention. On the whole, if the proportion of the sample that reach the criterion is close to 1 or 0 then sample sizes are much smaller than they are for proportions close to 0.5. For instance if we are to look at the proportion of deep pain dogs recovering to walk after standard therapy ($\sim$0.55) and consider that improving this to 0.65 would be clinically worthwhile, then the necessary sample size is $\sim$375 per group (assuming two-sided testing with $\alpha = 0.05$, $\beta = 0.2$). On the other hand, if we were interested in reducing the proportion of deep pain negative dogs that develop myelomalacia (current therapy is associated with $\sim$0.15) to 0.05, then the sample size needed would be $\sim$138 per group. This statistical efficiency provides a reason to be attracted to trials on very severely, and possibly chronically, injured individuals that are unlikely to recover spontaneously [see (46)].

Using the change in mean values between comparator groups is more statistically efficient, but requires a numeric outcome measure. It is also necessary to know (or estimate reasonably) the mean and standard deviation of the intervention and control groups. A problem that frequently arises is that it is difficult to know how much change in the outcome measure is meaningful. For instance, if we were to examine stride length as an outcome (as is commonly used in experimental rodents), how much change would be clinically meaningful for a paralyzed dog? The other aspect is that this method is highly dependent on the precision of measurement of the outcome. If there is a great deal of variability the ability to be sure that there is a real difference between groups is blunted. Similarly, if the outcome measure turns out not to be normally distributed many of the assumptions in analysis will be breached (although data transformation can often overcome this problem).

Whatever method is used the smaller the difference that is sought, the larger the sample size needed. When calculating sample size it is important that the sample size be realistic. Canine spinal cord injury is very common and so many hundreds of dogs can be accumulated, although that may require multicenter collaboration to achieve. On the other hand, it is also important that the difference between control and intervention group that is sought should be realistic. Most treatments in medicine have a moderate effect size and so, for instance, it is not realistic to power a study to detect a 50% difference in recovery between groups of deep pain negative dogs—no treatment is realistically going to be that effective.

Pragmatic vs. explanatory approaches can differ in terms of numbers needed in two main ways. First, an explanatory trial might be expected to refine the entry criteria with the aim of being able to discern even small differences between intervention and control and the sample size might then be reduced (although the selected cases will be a sub-set of the whole pragmatic population). Second, pragmatic trials tend to rely on "intention-to-treat" analysis in which all cases that enter the trial are included, irrespective of whether they received their allocated treatment or not. On the other hand, explanatory trials tend to rely on "per protocol" analysis, in which only participants that completely complied with the trial protocol are analyzed. Reliance on per protocol analysis tends to increase the numbers needed to achieve appropriate power because there will be many participants lost between enrolment and analysis.

ANALYSIS

Well-designed simple parallel group clinical trials usually do not require complicated statistical testing. Calculation of relative risk for a specific outcome [e.g., see CRASH trial (47)], chi-squared test or *t*-test (or non-parametric equivalent) is often sufficient to answer the basic trial question. Sometimes baseline measurement should be included as a covariate, necessitating use of analysis of covariance methods (usually implying regression techniques). Great care has to be taken with any sub-group analysis, and sub-groups should not be analyzed unless they are *pre-specified* and had relevant power calculations applied before trial commencement. Much harm has occurred in humans through misinterpretation of sub-group analysis (48). Similar considerations apply to analysis of whole study groups for which pre-trial sample size calculations are not available and for which study power is unknown (49).

Exceptions that might require more complicated analysis include the more complicated study designs such as crossover or factorial trials. However, crossover trials will rarely be

appropriate for analyzing effects of interventions for spinal cord injury (especially acute injury, because time will be assumed to have a strong effect) and factorial trials require identification of interventions that might interact with each other (otherwise they have no advantage over parallel group trials) and there are few such combinations that have been identified in laboratory science.

ETHICAL CONSIDERATIONS

The ethics of clinical trials in humans are complicated and rigorously regulated by international treaties and numerous guidelines (Helsinki Declaration: https://www.wma.net/policies-post/wma-declaration-of-helsinki-ethical-principles-for-medical-research-involving-human-subjects/). Those for veterinary clinical trials are less tightly regulated but follow the same basic principles: a trial needs to have a favorable risk/benefit analysis such that animals are carefully protected from detrimental consequences and have a reasonable chance of benefit from an intervention. Both of these aspects are usually addressed through investigations that precede the Phase II trial and may include data collection from experimental animals and Phase I studies in dogs. The question that inevitably follows is how much information is required before a Phase II/III trial can be considered reasonable and ethical. Similar questions have been asked regarding trials in human spinal cord injury (50) and the answer turns out to be different depending upon who is asked (51). On the whole, trials participants are more eager to press ahead with trials and researchers tend to be more cautious.

Beyond the requirement for preliminary data suggesting safety and efficacy, design of the trial itself most be undertaken ethically so as to ensure societal value of the information obtained. It is essential that all outcome measures can be assessed appropriately and that results of the study will be useful, which implies that the results will be disseminated to the scientific community by publication. Most clinical trial work in pet dogs requires ethical oversight from the Institutional Animal Care and Use Committee (IACUC) or a hospital review board at the center that is coordinating the trial, plus informed caregiver consent. Study review and approval generally includes assessment of design and proposed outcomes, component analysis of protocol activities that extend beyond the standard of care and how risks associated with those are mitigated, examination of sample size calculations, and investigator and personnel credentials and training in animal care.

PRACTICAL OBSTACLES TO CLINICAL TRIALS IN SPINAL CORD INJURY IN DOGS

There is a strong rationale for clinical trials on spinal cord injury treatments in dogs and many have been carried out and are readly accessible via PubMed (or other) searches using the keywords "dog" "spinal cord injury" "trial." It is readily apparent that few fulfill all the appropriate requirements for an effective clinical trial and many have design problems, such as lack of a control group (or unclear controls), that limit their value. This current review article is not intended to be a systematic review of previously published trials for but instead provides a checklist of items that can be applied by readers to evaluate each of the previously published trials themselves. On the whole, the most major problem has been the inability to recruit a sufficiently large number of participants, mainly because trials have been located at single centers. Challenges in multicenter trial design include: logistical challenges of coordinating fee structures, study review and approval across different sites; additional costs incurred by incorporation of subawards into funding applications; lack of availability and consistent infrastructure and equipment across sites; need for reasonably consistent training and application of trial methods at each center; barriers assumed to exist with incorporation of trial methodologies into routine private referral practice and the assumed traditional independent-mindedness of veterinarians in general. Retention of recruited clients can also be problematical, especially for spinal cord injury, for which prolonged follow-up is essential. Many previously published clinical trials in canine spinal cord injury are discussed in more detail in the companion article in this issue "*Ambulation in dogs with absent pain perception after acute thoracolumbar spinal cord injury*."

The main question is, how can obstacles to multicenter trials be overcome? Answers might include simplifying trial procedures so that many individuals and centers can be involved, establishing groups with an appropriate democratic structure to allow all collaborators to feel valued and involved in decision-making and, importantly, securing funding that bonds groups together and enables employment of trial-specific personnel.

POSSIBLE CLINICAL TRIAL DESIGNS IN ACUTE SPINAL CORD INJURY IN DOGS: COMPARING AND CONTRASTING PRAGMATIC AND EXPLANATORY EXAMPLES (TABLE 1)

The focus of this review article is on clinical trials for treatment of dogs with thoracolumbar spinal cord injury resulting from acute disc herniation. Whilst there are strong arguments for carrying out trials in dogs that have reached an unacceptable plateau of recovery at a later date—because they definitely require new therapies and the effect of new therapies might be more easily detected (because the baseline recovery rate is so poor)—here we will focus on two possible candidates for trials for dogs that present with loss of "deep pain sensation" in the acute phase. We have selected two candidate interventions that are "ready to go" but have constructed contrasting trial designs to highlight the many choices that need to be made. Although we describe here an explanatory trial focused on glyburide and a pragmatic trial focused on durotomy these specific approaches do not need to be specifically linked in this way to these interventions.

(a) Glyburide, aka glibenclamide, is a hypoglycemic agent that was used as an aid to controlling diabetes mellitus in people. It also has effects on the Sur1/TrpM4 channel that is involved in the progression of spinal cord tissue damage after acute contusion

TABLE 1 | Outline of two contrasting example approaches to spinal cord injury trials in dogs.

	Explanatory	Pragmatic
Population	Chondrodystrophic dogs weighing <15 kg with 7 vertebral-length gap on myelo MRI and serum GFAP >7 ng/mL, presenting within 24 h of when last seen walking	Any deep pain negative dog
Intervention	Glyburide 150 μg/kg, then 75 μg/kg TID for 3 days	4-vertebral length durotomy
Comparator	Sham tablets prepared by pharmacy	Standard care
Outcome	Open-field score repeated at weekly intervals for 3 months	Death or euthanasia by 3 weeks
Other restrictions	Physical therapy applied to animals in both groups for 30 min each day up till 3 months	No rules on physical therapy and adjunctive treatment
Personnel requirements	Training in open-field score analysis. Monitoring of dogs 24/7 for signs of hypoglycemia	No specific requirements
Analysis	Regression/ANOVA to analyze time x treatment interaction effects on open-field score over 3 month period	Chi-squared test to compare proportion of all-cause deaths between groups
Animals included in analysis	Only those that received glyburide at appropriate dose and times for 3 days, received physical therapy according to protocol and have follow-up for all time points until 3 months	All cases; analyzed according to intention-to-treat
Sample size calculation (approximate) $\alpha = 0.05$ $\beta = 0.8$	Expecting a score of 10.7 in control group and 12 in the treated group, with SD of 1.5, would require 22 per group	Expecting 15% deaths at 3 weeks in controls and 5% in intervention group would require 138 per group
Likelihood of a positive outcome changing clinical practice	Minimal	Very high

and its beneficial effects after spinal cord injury have been well-documented in several neuroscience laboratories throughout the world (52). Recently, pharmacokinetic studies have been carried out in dogs, showing that it has a good safety margin (hypoglycemia was not a problem at the doses needed to attain appropriate serum concentrations) and allowing construction of an appropriate dosing regimen for treatment of spinal cord injury (53). This drug therefore appears to have many advantages: it is widely available as a standard commercial preparation, it is cheap, the pharmacokinetics and safety are acceptable for use in dogs and it has shown benefit in many pre-clinical studies of spinal cord injury.

However, one question regarding glyburide is whether we would need to have a time limit on when trial dogs become paraplegic before presentation. In experimental work with glyburide, it would appear that it is most beneficial if it can be given before about 8 h after injury (54). It can be problematical to know for how long dogs have been paraplegic when the owners find them and it can be problematical to get a dog into a specialist clinic for treatment with a trial drug within the 8-h period. Also, if we were to limit inclusion to dogs that presented within 8 h of injury it would greatly reduce our expected recruitment numbers. So the best plan may be to design the trial to accept any dog that has become paraplegic and deep pain negative within 24 h, with a pre-specified analysis of the sub-groups that present <12 and 12–24 h after paralysis.

(b) Durotomy has been posited as a treatment for acute spinal cord injury since it was first modeled in animals in the early twentieth century (55). Since then there have been several studies suggesting that it is, or is not, helpful in dogs with acute spinal cord injury (56–58). The great advantage that durotomy appears to have is that, according to experimental data, the effect of incising the dura persists over at least 3 days after the injury (24), therefore perhaps making it more appropriate than glyburide for translation into dogs. There is also the advantage that anyone who is doing the decompressive surgery can carry out this procedure without having to have additional study materials or equipment.

Although there has been debate over the value of durotomy as an additional decompressive technique that might aid in restoring blood supply (and therefore retaining tissue integrity) after spinal cord injury, there are now data supporting its efficacy in clinically paralyzed dogs (25, 26).

How Might Dogs Be Randomized?

In large human clinical trials it is routine to use a central telephone service that designates treatment to each patient as they are enrolled. This method facilitates multicenter participation but does demand high level staffing and funding. For many veterinary trials for which funding at high levels (if at all) is unavailable, simple randomization that is blocked by center (i.e., each center has their own randomization) is still possible. The most straightforward way to randomize is to prepare a set of opaque envelopes, each containing the treatment allocation on a slip of paper and made up in variable-sized batches (so that the allocating clinician cannot predict what treatment the next patient will receive when they get near the end of a batch). The same procedure can be duplicated at each participating center; it is important that each center should randomly assign cases independently, to ensure that one center does not allocate unevenly compared to another. It is essential that clinicians who will allocate animals to treatment cannot

know which intervention each patient will receive until they open the envelope (i.e., there is appropriate *allocation concealment*). Numbering of envelopes also prevents allocations being selected.

How Might We Measure Outcome?

In these contrasting trials we have selected contrasting outcomes for the two interventions. For the explanatory trial on glyburide we have suggested using an open-field outcome assessment of walking (38) that is applied at weekly intervals for 3 months. These scores can then be assessed using repeated measures ANOVA or equivalent regression analysis, preferably accounting for the non-numerical nature of the outcome scores; animals that die or are euthanased before 3 months will be excluded from this analysis. For the durotomy trial we are interested in possible effect of averting progressive myelomalacia and so the simplest outcome to measure is whether dogs survive for longer then 3 weeks (although this will inevitably include some dogs euthanased for reasons apart from myelomalacia). The proportion surving for longer than 3 weeks will be compared between treatment groups using Fisher's exact test.

Ethical Considerations

A question might also be asked about whether it can be regarded as ethical to carry out a trial if preliminary studies show support for efficacy of an intervention—most notably here regarding durotomy. On the other hand, there may also be long (or even short-term) adverse effects of the new intervention that have not yet been detected and, furthermore, a principle of evidence-based medicine is that new therapies should be rigorously tested before widespread clinical adoption. It is also essential that all participating centers and personnel are adequately trained and equipped to carry out the trial procedures; training videos can often be used to facilitate such preparation.

Another way of looking at the assumed outcomes of a successful trial on durotomy is that if there really is an improvement in outcome from 15 to 5% that are dead by 3 weeks then this would suggest a number-needed-to-treat of $100/10 = 10$, i.e., for every 10 animals that are treated by durotomy only one additional animal survives for more than 3 weeks. While this can be justified if the therapy is beneficial it does also mean that there is little reason to be too concerned about allocating an animal to standard care alone, especially considering that there may also be adverse effects of the novel intervention (see above).

Another aspect is that an independent data and safety monitoring committee should be established to oversee data and, sometimes, to carry out interim analysis as it accumulates. In the trials outlined in **Table 1** this would only be realistic for the pragmatic trial on durotomy (because the other trial will enroll such a small number of cases). Such committees often use statistical stopping rules to aid decisions on interim analyses, to prevent a trial from continuing for too long if there is an unexpected but obvious imbalance in outcomes—an excess of benefit or of harm—before full trial recruitment has occurred (59). However, stopping rules can be controversial because stopping too early can lead to erroneous conclusions—especially with a bias toward larger effect size—or fuel continuing dispute regarding efficacy. One solution is to accept only extreme differences between groups at early stages, with progressive relaxation during the trial (60); the risks of multiplicity must also be incorporated. A more nuanced, and modern, option is to interpret interim results as a whole, taking into account statistics on both primary and secondary outcomes, the relative risk benefit and the problems that might arise in association with stopping too early, in order to provide evidence that is "beyond reasonable doubt" (61).

CONCLUSIONS

Large *pragmatic* clinical trials to determine the optimal methods for treating dogs with spinal cord injury are undoubtedly required. The relatively poor outcome associated with severe (i.e., "deep pain negative") thoracolumbar spinal cord injury following acute intervertebral disc herniation is the most obvious target. On the one hand, trials on such cases should mean that any "signal" resulting from an intervention will be easy to detect (because so little is expected of them); on the other, these cases are the hardest nuts to crack and so it is less probable that a detectable effect will be observed. Trials in less severely affected animals produces the opposite problem: many cases will get better anyway and so the signal of the intervention is lost in the noise of spontaneous recovery. Similar considerations apply to use of alternative outcome measures, including those used to examine autonomic function. There are many candidate therapies that could reasonably be tested and, worldwide, there are many affected dogs available for recruitment. Current barriers are largely problems of our (i.e., veterinarians') own making and can feasibly be overcome.

AUTHOR CONTRIBUTIONS

Subject of review was discussed between NJ, NO, and SM. NJ wrote first draft. All authors contributed to the input and approved the final version.

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REFERENCES

1. Umscheid CA, Margolis DJ, Grossman CE. Key concepts of clinical trials: a narrative review. *Postgrad Med.* (2011) 123:194–204. doi: 10.3810/pgm.2011.09.2475
2. Koppenaal T, Linmans J, Knottnerus JA, Spigt M. Pragmatic vs. explanatory: an adaptation of the PRECIS tool helps to judge the applicability of systematic reviews for daily practice. *J Clin Epidemiol.* (2011) 64:1095–101. doi: 10.1016/j.jclinepi.2010.11.020
3. Tosh G, Soares-Weiser K, Adams CE. Pragmatic vs explanatory trials: the pragmascope tool to help measure differences in protocols of mental health randomized controlled trials. *Dialogues Clin Neurosci.* (2011) 13:209–15.
4. Thorpe KE, Zwarenstein M, Oxman AD, Treweek S, Furberg CD, Altman DG, et al. A pragmatic-explanatory continuum indicator summary (PRECIS): a tool to help trial designers. *CMAJ.* (2009) 180:E47–57. doi: 10.1503/cmaj.090523
5. Freedman B. Equipoise and the ethics of clinical research. *N Engl J Med.* (1987) 317:141–5. doi: 10.1056/NEJM198707163170304
6. Freeman P, Jeffery ND. Re-opening the window on fenestration as a treatment for acute thoracolumbar intervertebral disc herniation in dogs. *J Small Anim Pract.* (2017) 58:199–204. doi: 10.1111/jsap.12653
7. Boswood A, Gordon SG, Häggström J, Wess G, Stepien RL, Oyama MA, et al. Longitudinal analysis of quality of life, clinical, radiographic, echocardiographic, and laboratory variables in dogs with preclinical myxomatous mitral valve disease receiving pimobendan or Placebo: the EPIC Study. *J Vet Intern Med.* (2018) 32:72–85. doi: 10.1111/jvim.14885
8. Moore SA, Granger N, Olby NJ, Spitzbarth I, Jeffery ND, Tipold A, et al. Targeting translational successes through CANSORT-SCI: using pet dogs to identify effective treatments for spinal cord injury. *J Neurotrauma.* (2017) 34:2007–18. doi: 10.1089/neu.2016.4745
9. Jeffery ND, Levine JM, Olby NJ, Stein VM. Intervertebral disk degeneration in dogs: consequences, diagnosis, treatment, and future directions. *J Vet Intern Med.* (2013) 27:1318–33. doi: 10.1111/jvim.12183
10. Granger N, Carwardine D. Acute spinal cord injury: tetraplegia and paraplegia in small animals. *Vet Clin North Am Small Anim Pract.* (2014) 44:1131–56. doi: 10.1016/j.cvsm.2014.07.013
11. Langerhuus L, Miles J. Proportion recovery and times to ambulation for non-ambulatory dogs with thoracolumbar disc extrusions treated with hemilaminectomy or conservative treatment: a systematic review and meta-analysis of case-series studies. *Vet J.* (2017) 220:7–16. doi: 10.1016/j.tvjl.2016.12.008
12. Bull C, Fehr M, Tipold A. Canine intervertebral disk disease: a retrospective study of clinical outcome in 238 dogs (2003-2004). *Berliner Munchener Tierarztliche Wochenschrift.* (2008) 121:159–70.
13. Wuermser LA, Ho CH, Chiodo AE, Priebe MM, Kirshblum SC, Scelza WM. Spinal cord injury medicine. 2. Acute care management of traumatic and nontraumatic injury. *Arch Phys Med Rehabil.* (2007) 88 (Suppl. 1):S55–61. doi: 10.1016/j.apmr.2006.12.002
14. Yue JK, Chan AK, Winkler EA, Upadhyayula PS, Readdy WJ, Dhall SS. A review and update on the guidelines for the acute management of cervical spinal cord injury - Part II. *J Neurosurg Sci.* (2016) 60:367–84.
15. Kwon BK, Okon EB, Plunet W, Baptiste D, Fouad K, Hillyer J, et al. A systematic review of directly applied biologic therapies for acute spinal cord injury. *J Neurotrauma.* (2011) 28:1589–610. doi: 10.1089/neu.2009.1150
16. Tetzlaff W, Okon EB, Karimi-Abdolrezaee S, Hill CE, Sparling JS, Plemel JR, et al. A systematic review of cellular transplantation therapies for spinal cord injury. *J Neurotrauma.* (2011) 28:1611–82. doi: 10.1089/neu.2009.1177
17. Ahuja CS, Fehlings M. Concise review: bridging the gap: novel neuroregenerative and neuroprotective strategies in spinal cord injury. *Stem Cells Transl Med.* (2016) 5:914–24. doi: 10.5966/sctm.2015-0381
18. Teng YD, Wrathall JR. Local blockade of sodium channels by tetrodotoxin ameliorates tissue loss and long-term functional deficits resulting from experimental spinal cord injury. *J Neurosci.* (1997) 17:4359–66. doi: 10.1523/JNEUROSCI.17-11-04359.1997
19. Rosenberg LJ, Wrathall JR. Time course studies on the effectiveness of tetrodotoxin in reducing consequences of spinal cord contusion. *J Neurosci Res.* (2001) 66:191–202. doi: 10.1002/jnr.1211
20. Moore SA, Zidan N, Spitzbarth I, Nout-Lomas YS, Granger N, da Costa RC, et al. Development of an international canine spinal cord Injury observational registry: a collaborative data-sharing network to optimize translational studies of SCI. *Spinal Cord.* (2018) 56:656–65. doi: 10.1038/s41393-018-0145-4

21. Srinivas S, Wali AR, Pham MH. Efficacy of riluzole in the treatment of spinal cord injury: a systematic review of the literature. *Neurosurg Focus.* (2019) 46:E6. doi: 10.3171/2019.1.FOCUS18596
22. Kurland DB, Tosun C, Pampori A, Karimy JK, Caffes NM, Gerzanich V, et al. Glibenclamide for the treatment of acute CNS injury. *Pharmaceuticals.* (2013) 6:1287–303. doi: 10.3390/ph6101287
23. Phang I, Werndle MC, Saadoun S, Varsos G, Czosnyka M, Zoumprouli A, et al. Expansion duroplasty improves intraspinal pressure, spinal cord perfusion pressure, and vascular pressure reactivity index in patients with traumatic spinal cord injury: injured spinal cord pressure evaluation study. *J Neurotrauma.* (2015) 32:865–74. doi: 10.1089/neu.2014.3668
24. Khaing ZZ, Cates LN, Fischedick AE, McClintic AM, Mourad PD, Hofstetter CP. Temporal and spatial evolution of raised intraspinal pressure after traumatic spinal cord injury. *J Neurotrauma.* (2017) 34:645–51. doi: 10.1089/neu.2016.4490
25. Takahashi F, Honnami A, Toki M, Dosaka A, Fujita Y, Hara Y, et al. Effect of durotomy in dogs with thoracolumbar disc herniation and without deep pain perception in the hind limbs. *Vet Surg.* (2020) 49:860–9. doi: 10.1111/vsu.13409
26. Jeffery ND, Mankin JM, Ito D, Boudreau CE, Kerwin SC, Levine JM, et al. Extended durotomy to treat severe spinal cord injury after acute thoracolumbar disc herniation in dogs. *Vet Surg.* (2020) 49:884–93. doi: 10.1111/vsu.13423
27. Morawietz C, Moffat F. Effects of locomotor training after incomplete spinal cord injury: a systematic review. *Arch Phys Med Rehabil.* (2013) 94:2297–308. doi: 10.1016/j.apmr.2013.06.023
28. Burns AS, Marino RJ, Kalsi-Ryan S, Middleton JW, Tetreault LA, Dettori JR, et al. Type and timing of rehabilitation following acute and subacute spinal cord injury: a systematic review. *Global Spine J.* (2017) 7(3 Suppl.):175S−94S. doi: 10.1177/2192568217703084
29. Gallucci A, Dragone L, Menchetti M, Gagliardo T, Pietra M, Cardinali M, et al. Acquisition of involuntary spinal locomotion (spinal walking) in dogs with irreversible thoracolumbar spinal cord lesion: 81 dogs. *J Vet Intern Med.* (2017) 31:492–7. doi: 10.1111/jvim.14651
30. Zidan N, Sims C, Fenn J, Williams K, Griffith E, Early PJ, et al. A randomized, blinded, prospective clinical trial of postoperative rehabilitation in dogs after surgical decompression of acute thoracolumbar intervertebral disc herniation. *J Vet Intern Med.* (2018) 32:1133–44. doi: 10.1111/jvim.15086
31. Akhtar AZ, Pippin JJ, Sandusky CB. Animal studies in spinal cord injury: a systematic review of methylprednisolone. *Altern Lab Anim.* (2009) 37:43–62. doi: 10.1177/026119290903700108
32. Aceves M, Mathai BB, Hook MA. Evaluation of the effects of specific opioid receptor agonists in a rodent model of spinal cord injury. *Spinal Cord.* (2016) 54:767–77. doi: 10.1038/sc.2016.28
33. Barkhof F, Kappos L, Wolinsky JS, Li DKB, Bar-Or A, Hartung HP, et al. Onset of clinical and MRI efficacy of ocrelizumab in relapsing multiple sclerosis. *Neurology.* (2019) 93:e1778–86. doi: 10.1212/WNL.0000000000008189
34. Handa Y, Naito A, Watanabe S, Komatsu S, Shimizu Y. Functional recovery of locomotive behavior in the adult spinal dog. *Tohoku J Exp Med.* (1986) 148:373–84. doi: 10.1620/tjem.148.373
35. Jeffery ND, Barker AK, Hu HZ, Alcott CJ, Kraus KH, Scanlin EM, et al. Factors associated with recovery from paraplegia in dogs with loss of pain perception in the pelvic limbs following intervertebral disk herniation. *J Am Vet Med Assoc.* (2016) 248:386–94. doi: 10.2460/javma.248.4.386
36. Lewis MJ, Williams KD, Langley T, Jarvis LM, Sawicki GS, Olby NJ. Development of a novel gait analysis tool measuring center of pressure for evaluation of canine chronic thoracolumbar spinal cord injury. *J Neurotrauma.* (2019) 36:3018–25 doi: 10.1089/neu.2019.6479
37. Basso DM, Beattie MS, Bresnahan JC. A sensitive and reliable locomotor rating scale for open field testing in rats. *J Neurotrauma.* (1995) 12:1–21. doi: 10.1089/neu.1995.12.1
38. OlbyNJ, De Risio L, Muñana KR, Wosar MA, Skeen TM, Sharp NJ, et al. Development of a functional scoring system in dogs with acute spinal cord injuries. *Am J Vet Res.* (2001) 62:1624–8. doi: 10.2460/ajvr.2001.62.1624
39. Song RB, Basso DM, da Costa RC, Fisher LC, Mo X, Moore SA. Adaptation of the Basso-Beattie-Bresnahan locomotor rating scale for use in a clinical model of spinal cord injury in dogs. *J Neurosci Methods.* (2016) 268:117–24. doi: 10.1016/j.jneumeth.2016.04.023
40. Bhimani AD, Kheirkhah P, Arnone GD, Nahhas CR, Kumar P, Wonais M, et al. Functional analysis in a spinal contusion rat model. *Neurosci Biobehav Rev.* (2017) 83:540–6. doi: 10.1016/j.neubiorev.2017.09.007
41. Jeffery ND, Brakel K, Aceves MR, Hook MA, Jeffery UB. Variability in open-field locomotor scoring following force-defined spinal cord injury in rats: quantification and implications. *Front Neurol.* (2020) 11:650. doi: 10.3389/fneur.2020.00650
42. Hamilton L, Franklin RJ, Jeffery ND. Development of a universal measure of quadrupedal forelimb-hindlimb coordination using digital motion capture and computerised analysis. *BMC Neurosci.* (2007) 8:77. doi: 10.1186/1471-2202-8-77
43. Granger N, Blamires H, Franklin RJ, Jeffery ND. Autologous olfactory mucosal cell transplants in clinical spinal cord injury: a randomized double-blinded trial in a canine translational model. *Brain.* (2012) 135:3227–37. doi: 10.1093/brain/aws268
44. Hu HZ, Granger N, Pai SB, Bellamkonda RV, Jeffery ND. Therapeutic efficacy of microtube-embedded chondroitinase ABC in a canine clinical model of spinal cord injury. *Brain.* (2018) 141:1017–27. doi: 10.1093/brain/awy007
45. Schulz KF, Grimes DA. Sample size calculations in randomised trials: mandatory and mystical. *Lancet.* (2005) 365:1348–53. doi: 10.1016/S0140-6736(05)61034-3
46. Fawcett JW, Curt A, Steeves JD, Coleman WP, Tuszynski MH, Lammertse D, et al. Guidelines for the conduct of clinical trials for spinal cord injury as developed by the ICCP panel: spontaneous recovery after spinal cord injury and statistical power needed for therapeutic clinical trials. *Spinal Cord.* (2007) 45:190–205. doi: 10.1038/sj.sc.3102007
47. Roberts I, Yates D, Sandercock P, Farrell B, Wasserberg J, Lomas G, et al. Effect of intravenous corticosteroids on death within 14 days in 10008 adults with clinically significant head injury (MRC CRASH trial): randomised placebo-controlled trial. *Lancet.* (2004) 364:1321–8. doi: 10.1016/S0140-6736(04)17188-2
48. Rothwell PM. Treating individuals 2. Subgroup analysis in randomised controlled trials: importance, indications, and interpretation. *Lancet.* (2005) 365:176–86. doi: 10.1016/S0140-6736(05)17709-5
49. Button KS, Ioannidis JPA, Mokrysz C, Nosek BA, Flint J, Robinson ESJ, et al. Power failure: why small sample size undermines the reliability of neuroscience. *Nat Rev Neurosci.* (2013) 14:365–76. doi: 10.1038/nrn3475
50. Kwon BK, Soril LJ, Bacon M, Beattie MS, Blesch A, Bresnahan JC, et al. Demonstrating efficacy in preclinical studies of cellular therapies for spinal cord injury - how much is enough? *Exp Neurol.* (2013) 248:30–44. doi: 10.1016/j.expneurol.2013.05.012
51. Kwon BK, Ghag A, Reichl L, Dvorak MF, Illes J, Tetzlaff W. Opinions on the preclinical evaluation of novel therapies for spinal cord injury: a comparison between researchers and spinal cord-injured individuals. *J Neurotrauma.* (2012) 29:2367–74 doi: 10.1089/neu.2012.2479
52. Hosier H, Peterson D, Tsymbalyuk O, Keledjian K, Smith BR, Ivanova S, et al. A direct comparison of three clinically relevant treatments in a rat model of cervical spinal cord injury. *J Neurotrauma.* (2015) 32:1633–44. doi: 10.1089/neu.2015.3892
53. Jeffery N, Boudreau CE, Konarik M, Mays T, Fajt V. Pharmacokinetics and safety of oral glyburide in dogs with acute spinal cord injury. *PeerJ.* (2018) 6:e4387. doi: 10.7717/peerj.4387
54. Simard JM, Tsymbalyuk O, Ivanov A, Ivanova S, Bhatta S, Geng Z, et al. Endothelial sulfonylurea receptor 1-regulated NC Ca-ATP channels mediate progressive hemorrhagic necrosis following spinal cord injury. *J Clin Invest.* (2007) 117:2105–13. doi: 10.1172/JCI32041
55. Allen AR. Surgery of experimental lesion of spinal cord equivalent to crush injury of fracture-dislocation of spinal column. *JAMA.* (1911) 57:878–80. doi: 10.1001/jama.1911.04260090100008
56. Parker AJ, Smith CW. Functional recovery from spinal cord trauma following incision of spinal meninges in dogs. *Res Vet Sci.* (1974) 16:276–9. doi: 10.1016/S0034-5288(18)33725-1
57. Parker AJ, Smith CW. Functional recovery following incision of spinal meninges in dogs. *Res Vet Sci.* (1972) 13:418–21. doi: 10.1016/S0034-5288(18)33985-7

58. Loughin CA, Dewey CW, Ringwood PB, Pettigrew RW, Kent M, Budsberg SC. Effect of durotomy on functional outcome of dogs with type I thoracolumbar disc extrusion and absent deep pain perception. *Vet Comp Orthop Traumatol.* (2005) 18:141–6. doi: 10.1055/s-0038-1632953
59. Whitehead J. Stopping clinical trials by design. *Nat Rev Drug Discov.* (2004) 3:973–7. doi: 10.1038/nrd1553
60. O'Brien PC, Fleming TR. A multiple testing procedure for clinical trials. *Biometrics.* (1979) 35:549–56. doi: 10.2307/2530245
61. Mukherjee SD, Goffin JR, Taylor V, Anderson KK, Pond GR. Early stopping rules in oncology: considerations for clinicians. *Eur J Cancer.* (2011) 47:2381–6. doi: 10.1016/j.ejca.2011.05.019

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REVIEW
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Current Approaches to the Management of Acute Thoracolumbar Disc Extrusion in Dogs

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Intervertebral disc extrusion (IVDE) is one of the most common neurologic problems encountered in veterinary clinical practice. The purpose of this manuscript is to provide an overview of the literature related to treatment of acute canine thoracolumbar IVDE to help construct a framework for standard care of acute canine thoracolumbar IVDE where sufficient evidence exists and to highlight opportunities for future prospective veterinary clinical research useful to strengthen care recommendations in areas where evidence is low or non-existent. While there exist a number of gaps in the veterinary literature with respect to standards of care for dogs with acute thoracolumbar IVDE, recommendations for standard care can be made in some areas, particularly with respect to surgical decompression where the currently available evidence supports that surgery should be recommended for dogs with nonambulatory paraparesis or worse. While additional information is needed about the influence on timing of decompression on outcome in dogs that are deep pain negative for longer than 48 h duration, there is no evidence to support treatment of the 48 h time point as a cut off beyond which it becomes impossible for dogs to achieve locomotor recovery. Surgical decompression is best accomplished by either hemilaminectomy or mini-hemilaminectomy and fenestration of, at a minimum, the acutely ruptured disc. Adjacent discs easily accessed by way of the same approach should be considered for fenestration given the evidence that this substantially reduces future herniation at fenestrated sites. Currently available neuroprotective strategies such as high does MPSS and PEG are not recommended due to lack of demonstrated treatment effect in randomized controlled trials, although the role of anti-inflammatory steroids as a protective strategy against progressive myelomalacia and the question of whether anti-inflammatory steroids or NSAIDs provide superior medical therapy require further evaluation.

Keywords: dog, interveterbral disc, intervertebral disc disease, hemilaminectomy, spinal cord injury, hansen type I

INTRODUCTION

Intervertebral disc extrusion (IVDE) is one of the most common neurologic problems encountered in veterinary clinical practice (1). Dogs with acute IVDE present with a spectrum of neurologic abnormalities caused by a combination of compression and contusion to the spinal cord via sudden extrusion of degenerated and calcified nucleus pulposus of the intervertebral disc through the annulus fibrosus and into the vertebral canal (2). Severity of clinical injury spans a continuum from paraspinal hyperesthesia up to paraplegia with loss of deep pain perception, where these patients with loss of deep pain perception are often termed "deep pain negative." Injury grades are typically described as summarized in **Table 1** (3, 4). Treatment recommendations for an individual dog with IVDE are based on a combination of factors, accounting for the aforementioned severity of neurologic signs presented, availability of specialty care in a geographical area, and preferences and financial limitations of the owner. Available treatment options include medical management (often termed "conservative therapy"), consisting of strict activity restriction, physiotherapy, analgesics and anti-inflammatory medications; or surgical decompression of the spinal cord to remove herniated material from the vertebral canal, followed by similar activity restriction, bladder management if needed, and pain management recommendations. The evidence available in the veterinary literature to guide practitioners in recommending one therapeutic approach over another in dogs with IVDE is relatively low as most published veterinary studies are either retrospective in nature or prospective case series. Some randomized clinical trials and two recent systematic review and meta-analyses are available to inform care recommendations; however, this small database of strong clinical evidence has resulted in a lack of uniform, science-based guidelines for the management of acute canine thoracolumbar IVDE.

The purpose of this manuscript is to build on previously published studies by incorporating broad historical and contemporaneous clinical data to construct a framework for standard care of acute canine thoracolumbar IVDE where sufficient evidence exists and to highlight opportunities for future prospective veterinary clinical research useful to strengthen care recommendations in areas where evidence is low or non-existent. While a number of distinct clinical presentations of intervertebral disc disease occur in dogs, this paper focuses specifically on acute thoracolumbar IVDE, from here on referred to as IVDE, for which the largest body of evidence exists to base treatment recommendations.

TABLE 1 | Modified Frankel scale used to describe the degree of neurologic impairment for dogs with intervertebral disc herniation.

Grade	Clinical signs
0	Paraplegia with absent superficial and deep pain sensation (also termed "deep pain negative" throughout the literature); there is an absence of behavioral response (e.g., vocalizing or orienting movement of the head toward stimulus) when clamping of a hemostat or other instrument to the skin of the limb/paw (superficial) and when applying the same stimulus while clamping the bone of the digit (deep). There is no observable voluntary movement of the hind limbs
1	Paraplegia with absent superficial but intact deep pain sensation; behavioral response (e.g., vocalizing or orienting movement of the head toward stimulus) is absent when clamping the skin (superficial) but present when clamping the bone of the digit (deep). There is no observable voluntary movement of the hind limbs. Note that some studies group this subset of dogs with either grade 0, or grade 2 depending on study design
2	Paraplegia with intact superficial and deep pain sensation (also termed "deep pain positive" throughout the literature); there is presence of a behavioral response (e.g., vocalizing or orienting movement of the head toward stimulus) to both superficial and deep noxious stimuli. There is no observable voluntary movement of the hind limbs
3	Non-ambulatory paraparesis; there is movement of one or both hind limbs, but the animal is unable to take 10 consecutive unassisted weight-bearing steps
4	Ambulatory paraparesis; the animal can take 10 consecutive unassisted weight-bearing steps with the hind limbs but displays an ataxic or paretic gait
5	Paraspinal hyperesthesia only; the animal has a normal hind limb gait but has posture or physical examination findings indicative of paraspinal hyperesthesia
6	Normal

A HISTORICAL PERSPECTIVE

The first description of the clinical presentation of IVDE in dogs is credited to Dexler, who in the late 1800's described a condition of paralysis in dogs caused by compression of the spinal cord from abnormalities of the intervertebral disc that he termed "neoformations" (5). He attributed the neoformations to proliferation of the intervertebral disc, a hypothesis supported by subsequent works that used the term *endochondrosis intervertebralis* to describe the condition. It wasn't until the late 1930's and 1940's that authors began to suggest that these neoformations might in fact be herniation of the intervertebral disc similar to what had been observed in people (6, 7). In 1951 and 1952, Olsson and Hansen (respectively), published in-depth investigations of the syndrome; both supported the fact that these neoformations were in fact disc herniations (5, 8). Hansen's report on the condition further highlighted three breeds with an apparently high risk for the condition: the French bulldog, the dachshund, and the Pekingese (8).

Early diagnosis of IVDE was made by radiography, myelography and pathology. Improvement after conservative treatment was described in individual cases (7). Additional in-depth studies were performed by Hoerlein and published in 1956 and were summarized again after more than 30 years experience (9, 10). Diagnosis evolved over time to become based on physical and neurological examination, radiography, myelography and occasionally tomography. At that time, conservative therapy was recommended for dogs with spinal pain as the only clinical sign, for dogs with a first episode of IVDE and paresis, dogs who had IVDE in combination with other medical disorders, those with paralysis and no conscious perception of deep pain, and those with evidence of progressive myelomalacia (9). Conservative therapy consisted of general good nursing care including proper nutrition, cage rest, ensuring a clean environment, prevention of

decubital sores, and bladder and bowel care. Various protocols for glucocorticoid administration were recommended and applied based on coincident clinical research, starting with dexamethasone, and then, in later years, methylprednisolone (11, 12). Physiotherapy, which included limb exercises and swimming, was recommended although a controlled study had not been undertaken at that time. Medications to control pain were recommended, but clinical texts emphasized that pain should not be completely relieved in the outpatient setting due to concern that a completely comfortable patient might undertake excessive movements and resist cage rest.

Hoerlein personally observed 1,184 dogs with IVDE between 1950 and 1975, finding good surgical results in 87% of paraplegic dogs and in 91% of paretic dogs (10). While a true comparative study was never carried out, the rate of functional recovery was described as much lower for cases managed conservatively, where only 22% of paraplegic dogs recovered the ability to ambulate without assistance (13). Additional cases of dogs who ranged from ambulatory paraparetic to paraplegic with deep pain sensation intact managed conservatively and published between 1950 and 1970 suggested that about half of cases recovered the ability to ambulate without surgery. Therefore, surgical treatment was recommended in cases with pain and/or paresis not responding to conservative care and in cases with substantial neurologic deficits where deep pain sensation was preserved with a reported success rate of 75–90% (9, 10, 14).

TABLE 2 | Synthesis of outcome data from dogs managed medically for intervertebral disc herniation and published before 1983.

Reference	Medical management (N)	Recovered (%)
Olsson (5)	21	12 (57)
Hoerlein (9)	62	32 (52)
Funkquist (13)	33	16 (48)
Funkquist (15)	23	14 (61)
Total	139	74 (53)

Neurologic grade of individual dogs according to the modified Frankel scale at presentation for individual animals is difficult to discern from some papers and these cases represent a heterogenous group of dogs ranging from paraparetic to paraplegic. Wherever possible, based on case description, grade 1 and 5 dogs have been excluded from analysis to assist with comparison to more recent literature. Recovery is defined as return of ambulation based on what is described in the manuscript and only cases with reported post-treatment follow up are included.

CONTEMPORARY QUESTIONS

While historical evaluation of outcomes associated with surgical decompression in dogs with IVDE suggests that decompression offers improved recovery over conservative management for dogs with severe injuries, no studies have systematically compared outcomes between dogs managed medically and surgically for the condition. Beyond this, additional questions remain in the veterinary neurosurgical community regarding the importance of urgent decompression in deep pain negative dogs; the need for prophylactic fenestration to lessen recurrence of IVDE in dogs at high risk; and the value of neuroprotective strategies and post-operative interventions such as activity restriction and physiotherapy. The following sections discuss the relevant veterinary literature with respect to each area.

THE EVIDENCE FOR DECOMPRESSION

Overview of Current Practices

While only a few contemporary studies exist reporting medical management in dogs with severe neurologic deficits due to IVDE, and no randomized controlled studies compare these to surgical treatment, there is a substantial amount of historical literature from which to draw some basis for comparison (13, 15, 16). One difficulty in evaluating outcomes from early reports of canine IVDE is that patient assessment, diagnostic approach, terminology and description of neurologic deficits, and injury grading differ from more recent literature and make it challenging to draw strong conclusions about neurologic grade at presentation and its relationship to long-term outcome. A summary of reported outcomes for dogs with severe IVDE managed medically and published before 1983 is presented in **Table 2**, and a recent systematic review and meta-analysis compares results between medical and surgical management for cases published after 1983 (17). Reported outcomes for dogs managed medically after severe (non-ambulatory paraparetic or worse) neurologic injury due to IVDE described in more recent publications range from 50 to 100%, depending on the severity of injury and the study (17–24). Clinicians anecdotally suggest that while recovery of ambulation after surgical vs. medical management in dogs with paraparesis or paraplegia with intact deep pain sensation may ultimately be comparable, recovery is quicker and more complete for dogs that undergo surgical decompression (25). At present, most board-certified neurologists and orthopedic surgeons recommend surgical decompression for dogs who are non-ambulatory paraparetic or worse secondary to IVDE suggesting that this is the current standard of care (26).

The question of whether or not surgical decompression should be considered standard for dogs with substantially compressive IVDE has been raised and revisited intermittently across the veterinary literature (16, 27, 28). Indeed, several notable studies report that a portion of dogs with substantially compressive IVDE can recover with conservative therapy alone, and that in some cases extruded disc material may even dissipate over time (29–31). Clinicians likely make their current recommendations for surgery based on both the historical literature and having absorbed implications from the field of experimental spinal cord injury suggesting that very early surgical decompression leads to enhanced recovery (32). The drive to perform surgical decompression in the most severe cases may also emanate, in part, from "modern" owner's expectations, our perception of animal welfare in recent decades, and the fact the demonstration of compression on cross-sectional imaging drives an impulse to decompress. Additional influence may also include fairly predictable outcomes with surgery, allowing dog owners to opt for surgery based on concrete numbers; however, no prospective randomized trial has evaluated conservative management *vs.* surgical decompression in dogs with severe IVDE. Currently,

the likelihood of this study happening is low, given the lack of clinical equipoise across the veterinary community related to the currently available data indicating the value of decompression.

Outcome for Severe Injuries (Non-ambulatory Paraparesis or Worse)

While no randomized prospective clinical trials have compared these two treatment options, we can draw some inferences from the existing literature to form a framework for discussing the value of decompressive surgery. In addition to the historical literature available for review, Langerhuus and Miles conducted a systematic review and meta-analysis of dogs with IVDE published between 1983 and 2012 and treated either medically or surgically (17). For dogs with severe injuries (non-ambulatory paraparetic or worse), a statistically greater proportion recovered independent ambulation following decompression *via* hemilaminectomy as compared to conservative treatment. Specifically, 93% of dogs who were non-ambulatory paraparetic or paraplegic with intact deep pain perception recovered ambulation after surgery whereas only 79% of dogs with non-ambulatory paraparesis and 62% of dogs who were paraplegic with intact deep pain perception recovered after conservative treatment alone. For dogs with the most severe injuries, those who were paraplegic and deep pain negative, the recovery rate was 61% after surgical decompression and 10% for those with conservative treatment (although of note here, data from only 25 deep pain negative dogs managed conservatively was available for inclusion). A summary of neurologic outcomes, as reported in this meta-analysis by injury grade is presented in **Table 3**. The influence of surgical decompression on time to return of independent ambulation (e.g., speed of recovery) is more difficult to assess *via* synthesis of previous studies. Fewer data points are available for inclusion in meta-analysis and preclude analysis of recovery by neurologic grade for all severities except paraplegia with intact deep pain perception; however, for dogs with paraplegia with intact deep pain perception, mean time to recovery of ambulation is significantly shorter after surgical decompression compared with conservative therapy (15 vs. 84 days, respectively) (17).

TABLE 3 | Overall percent of dogs achieving neurologic recovery (defined as independent ambulation) based on presenting injury severity, as reported in the literature after 1983 by Langerhuus and others for medical and surgical management of canine thoracolumbar interverterbral disc herniation (IVDH).

Injury severity	Number of cases reported	Medical management	Surgical decompression	References
Paraplegia with absent deep pain sensation	513	10%	61%	(17)
Paraplegia with intact deep pain sensation	603	62%	93%	(17)
Non-ambulatory paraparesis	354	79%	93%	(17)
Ambulatory paraparesis	152	48–84%	95%	(18, 23, 25)
Paraspinal hyperesthesia only	143	60–100%	97%	(18, 23, 25)

Limitations of Available Outcome Data for Severe Injuries (Non-ambulatory Paraparesis or Worse)

Taken in its entirety, synthesis of the previously published veterinary literature supports the standard recommendation for surgical decompression in dogs that are non-ambulatory paraparetic or worse, and that surgery speeds recovery of ambulation and overall locomotor outcome in dogs with severe injuries. There are some limitations to the data used to come to this conclusion that must be acknowledged. Since no prospective, randomized studies are available, all synthetized reports comparing outcome incorporate data only from prospective case series and retrospective studies. Additionally, sufficiently detailed outcomes for dogs managed conservatively are available for a relatively small number of published cases, with only 113 cases described in the literature published since 1983 and a similarly small number described before that date. In comparison, >1,500 surgically treated cases and their associated outcomes are described over the same time period. The paucity of published medically managed cases with severe IVDE, while not surprising given current clinical standards, likely confounds comparison of outcomes. Limited publication of medically managed cases probably results from several factors that bias the entirety of the published literature on canine IVDE. First, there is an overall pre-existing clinical inclination toward recommending decompression for dogs that are non-ambulatory paraparetic or worse due to IVDE. In the face of severe clinical signs, clinicians are naturally driven to administer a treatment because they can. Second, most published case series originate from veterinary specialty referral centers where access to advanced care and recommendation for and compliance with decompressive surgery are likely to be higher. Few published cases originate from primary care facilities, where the rate of and approaches toward medical management are likely to be different. Third, data available and used for synthesis of the literature is largely of retrospective nature. Therefore, caution should be used in over interpretation.

Outcome for Mild Injuries (Paraspinal Hyperesthesia or Ambulatory Paraparesis)

As discussed above, while some population-level analysis of clinical data for dogs with severe injury from IVDE exists to guide treatment recommendations, there is a current gap in the veterinary literature with respect to medical and surgical outcomes for dogs with more mild injuries (those who have paraspinal hyperesthesia or ambulatory paraparesis). Because these dogs were not included in subgroup analysis of outcome in a recent meta-analysis, outcomes for those dogs are summarized in **Table 3** from previous literature (18, 23, 25). While some clinicians steer owners toward conservative management in these more mildly affected cases, 12 and 59% of neurologists and orthopedic surgeons (respectively), report routinely recommending surgical decompression for dogs with

a first episode of IVDE causing back pain or ambulatory paraparesis (26). The evidence to guide practitioners in these more mildly affected cases is currently lacking as few studies specifically examine outcome for dogs in this context. To the authors' knowledge, no large-scale published comparison exists for dogs these dogs with less severe injuries treated medically *vs.* surgically, although one study reported a 54.7% overall success rate for medical management of thoracolumbar IVDE in a cohort of dogs for which 83% were ambulatory on presentation, and several other studies report surgical outcome for a small number of dogs with mild injuries (23). Hurdles to designing randomized controlled studies for dogs with mild IVDE include the difficulty in obtaining a definitive diagnosis for dogs that are not managed surgically, and the fact that most mildly affected dogs are treated at primary care facilities without referral to a specialist. Albeit more challenging to quantify in dogs, and while recovery of locomotion is of considerable importance to owners of dogs with IVDE, relevant long-term outcomes for dogs with mild injuries might also focus on pain and quality of life measures. Given the fact that a substantial portion of dogs with IVDE present with only pain or mild neurologic deficits, large scale prospective studies could be ethically conducted in this area and represent an opportunity to improve our understanding of best practices in treating this substantial patient population (33–37). Well-powered and informative prospective studies in this area will likely require partnership between specialty referral hospitals and primary care facilities and may require the development of novel clinical assessment tools. In particular, it would be useful to conduct a longitudinal study—from puppy to end of life—recording the environment, health and behavior in breeds at risks of IVDE. Feasibility and utility of generating powerful epidemiological data to answer health questions spanning the life course has been previously demonstrated in dogs, cats, and people (38, 39).

SURGICAL APPROACH

A variety of descriptions of surgical approaches to address canine thoracolumbar IVDE have been previously published. These include hemilaminectomy, mini-hemilaminectomy/pediculectomy, dorsal laminectomy, partial corpectomy, and fenestration of the intervertebral disc with or without concurrent laminectomy for removal of herniated disc material (34, 40–42).

Early studies reported spinal decompression procedures for canine IVDE *via* either dorsal laminectomy or hemilaminectomy. Hoerlein and colleagues described a procedure for hemilaminectomy in detail, whereas Funkquist explored procedures for dorsal laminectomy (15, 43). A study published by Hoerlein in 1978, and using a questionnaire gathering information from 50 participating veterinary surgeons about the use of surgical approaches for treatment of IVDE, suggested that the best surgical results were obtained after hemilaminectomy and fenestration (44). These results were later confirmed in a second prospective but non-randomized study comparing hemilaminectomy to dorsal laminectomy for treatment of thoracolumbar IVDE, where hemilaminectomy was reported to significantly improved the surgeon's ability to retrieve herniated disc material and where this enhanced removal of herniated disc was associated with improved early locomotor recovery (45).

Today, hemilaminectomy with or without removal of the articular processes is most commonly described and represents the current decompressive procedure of choice for veterinary spinal surgeons. A recent survey indicated that 95% percent of veterinary neurologists and surgeons typically perform a hemilaminectomy or mini-hemilaminectomy in this scenario (26). While each disc herniation is different, and requires unique consideration for what constitutes the best surgical approach, efficiency in disc retrieval, reduced opportunity for laminectomy membrane formation, and reduced chance of postoperative neurologic decline likely form the relatively broad surgical consensus for hemilaminectomy in the surgical treatment of routine thoracolumbar IVDE (45–47). **Table 4** summarizes the published literature relating to surgical approaches for canine IVDE, including benefits and unique challenges of each approach.

Fenestration of the intervertebral disc without spinal cord decompression has also been historically proposed as a viable option for treatment of IVDE, with the idea re-introduced more recently into the veterinary literature by way of a systematic review presenting outcomes of previously published cases (27, 53, 54). While most articles on fenestration address its role in prophylaxis of recurrent disc extrusion (the arguments for and against that approach are presented below), some authors have suggested that fenestration alone may be useful to facilitate recovery of spinal cord function following IVDE. When first described as a therapeutic intervention for IVDE, the aim of fenestration was, in fact, to reduce intradiscal pressure with the goal of reducing a presumed dynamic lesion of the portion of the disc herniated into the epidural space (56). Dogs with IVDE ranging from pain-only to paraplegia with intact deep pain perception, treated only with lateral fenestration of the intervertebral disc were reported to experience a relatively high recovery rate; however, only a 33% recovery rate was reported for paraplegic deep pain negative dogs as compared to the 50–60% recovery rate typically observed for this group after hemilaminectomy (17, 20, 55). Similar to conservative management in deep pain negative dogs, the number of deep pain negative dogs managed with fenestration alone and published in the literature is quite small. Data also originates almost exclusively from retrospective studies, making outcome evaluation challenging. A recent systematic review suggests that outcome for dogs with mild injuries undergoing fenestration alone could be better than previously suggested; however, a retrospective study of 331 dogs undergoing percutaneous disc ablation without decompression (and thus a procedure similar to fenestration alone) demonstrated a recovery rate of only 38% for dogs with deep pain negative injuries, reinforcing the concept that patients with severe injuries benefit from decompression (27, 57). Currently, <10% of veterinary neurologists and orthopedic surgeons report routinely performing fenestration without concurrent decompression (26). This practice pattern

TABLE 4 | A summary of published surgical approaches and reported outcomes for canine acute intervertebral disc herniation (IVDH) affecting the thoracolumbar spine.

Surgical approach	Description	Advantages	Limitations	References
Dorsal laminectomy	Removal of the spinous process and variable portion of the lamina with conservation of the articular processes	• Increased cord exposure compared to hemilaminectomy • Improved access to dorsal compressive lesions	• No access to ventral portion of the vertebral canal for disc removal • Concern for laminectomy scar formation, particularly if more than one consecutive site	(15, 43, 46, 47)
Hemilaminectomy	Removal of half of the vertebral arch, including the lamina, pedicle, and articular process	• Reduced laminectomy scar • Improved access to ventral portion of the spinal canal for disc removal • Improved access for fenestration	• Residual compression is common (clinical significance unclear)	(17, 19, 48–50)
Mini-hemilaminectoy/ pediculectomy	Similar to a hemilaminectomy but articular process is spared	• Less invasive than hemilaminectomy • Improved access to ventral portion of the spinal canal for disc removal • Improved access for fenestration	• Residual compression is common (clinical significance unclear)	(41, 51, 52)
Partial corpectomy	Partial removal of thoracic or lumbar adjacent vertebral bodies that support the extruded/protruded disk material inside the vertebral canal	• Allows ventral decompression with minimal spinal cord manipulation • May offer an advantage for chronic and ventrally located disc herniations	• Hemorrhage from the venous sinus is common • Transient post-operative deterioration common • Residual compression is common (clinical significance unclear)	(40, 42)
Fenestration without laminectomy	Mechanical removal of the nucleus pulposus through a window created in the annulus fibrosus	• Less invasive than laminectomy • Good outcome for grade 1 and 2 injuries	• Does not relieve spinal cord compression • Reduced and prolonged recovery with severe injuries	(53–56)

may be influenced by previous work suggesting that 80% of dogs presented with back pain as their only clinical sign still have significant spinal cord compression, and thus logically might benefit from decompression (58). Of those surgeons who perform fenestration alone, most indicate they recommend this approach only for dogs with a presenting complaint of spinal pain alone or spinal pain with minimal neurologic deficits (26). At present, evidence to support fenestration alone as a viable surgical approach for canine IVDE is limited and historical literature supports that decompression of the spinal cord, with or without concurrent fenestration, provides improved recovery for dogs with severe (non-ambulatory paraparetic or worse) IVDE (**Table 3**). Lacking from the current literature is data on the incidence of postoperative chronic pain when fenestration is used without concurrent decompression.

THE TIMING OF DECOMPRESSION

When considering the evidence related to the timing of decompression, two clinically important questions arise. Those center on whether decompression should be performed urgently for dogs with severe injuries, and whether dogs that are deep pain negative for a prolonged duration have a reasonable potential for recovery after surgical decompression.

Is There Value in Decompressing Dogs Who Have Been "Deep Pain Negative" for an Extended Period?

Early studies, and most veterinary neurosurgery texts, suggest that timing of decompression influences outcome, particularly for dogs who are deep pain negative secondary to IVDE. Early decompression is typically encouraged, with a recommended timeframe ranging from 12 to 48 h, beyond which prognosis is often suggested to worsen significantly. These recommendations originate from several retrospective studies, most of which include very few dogs with an injury duration of 48 h or greater. This sentiment seems to persist in spite of several studies that have shown the contrary, noting good functional recovery in some deep pain negative dogs with extended injury duration (72 h or more). A challenge in drawing firm conclusions on this topic is the small number of published deep pain negative cases with a duration of injury longer than 48 h and a confirmed postoperative outcome. Jeffery et al. recently evaluated the influence of a variety of clinical factors on outcome in 78 deep pain negative dogs using a prospective multicenter cohort study design (59). Similar to previous reports, his group was also unable to find an association between duration of deep pain negative status and outcome in this patient population; however, a relatively small number of dogs were available for inclusion where the duration of onset of locomotor dysfunction and initial evaluation at a referral center was >48 h. Currently, some clinicians treat this 48 h time point prior to referral as a "cliff" or abrupt point beyond which recovery in deep pain negative dogs cannot be achieved. There is certainly no literature to support this interpretation but the influence of prolonged deep pain negative status on recovery rate, and importantly on extent of recovery, is also not clear because the number of published cases in any one study is low. Designing a large-scale randomized controlled trial assessing the influence of duration of deep pain negative status on outcome after surgical decompression would be ethically challenging; however, a systematic review

and meta-analysis of previously published cases could yield additional valuable information and may assist with guiding owners regarding prognosis for locomotor recovery and the overall utility of surgical decompression for more chronic cases. Larger scale, prospective longitudinal cohort studies could also be helpful and could leverage existing resources already in use across veterinary referral networks (60).

How Quickly Should Decompression Occur to Maximize Opportunity for Neurologic Recovery in Dogs With Severe Injuries?

Recommendations for urgent decompression, particularly for dogs who are deep pain negative, likely stem from some of the previously mentioned studies on surgical outcomes as well as from the experimental and human spinal cord injury literature where some studies suggest that early decompression is associated with enhanced locomotor recovery; however, the human clinical literature is mixed with regard to the effect of timing of decompression on outcome. A more recent prospective, multi-institutional cohort study of 888 patients with acute spinal cord injury failed to demonstrate an influence of timing of decompression on outcome across the entire cohort and specifically in the group of ASIA Impairment Scale (AIS)-A individuals, those with a clinical injury severity somewhat analogous to deep pain negative status. Interestingly, this study did show an association between improved locomotor outcome and early decompression (<24 h) in groups of patients with incomplete (paresis or plegia with pain sensation intact) injuries (61). What becomes difficult in terms of comparison between dogs and people is that standard recovery curves differ substantially between the two species. Where the reported outcome for locomotor recovery in deep pain negative dogs ranges from 50 to 60%, the incidence of recovery in people with equivalent injuries is much lower; thus drawing strong direct parallels between recovery curves for the two can be challenging. It is likely that people with sensorimotor complete (deep pain negative) injuries represent a much more "complete" injury in many cases and therefore the ability to influence recovery may be more limited whereas those with less complete injuries may be more amenable to intervention. Additionally, most people presenting with spinal cord injury are polytrauma patients with acute concerns related to hypotension, internal injuries and other co-morbid conditions. Therefore, delay in decompression is often necessary in favor of stabilization of the patient for general anesthesia. In most dogs with IVDE, this is not the case and delay of anesthesia for medical reasons is rarely necessary.

As noted above, a recent study was not able to demonstrate improved neurologic outcome in deep pain negative dogs with early surgical intervention, although most patients included in that study were referred for decompressive surgery within 24 h, therefore potentially confounding the ability to demonstrate associations (59). Interestingly, while another recent retrospective study by Castel et al. also supported the lack of influence of timing of surgery on locomotor recovery, this study did find an association between delay of decompression beyond 12 h and increased risk of progressive myelomalacia, an uncommon but often fatal phenomenon observed almost exclusively in dogs who are deep pain negative secondary to IVDE (62).

Taken in total, the evidence in the veterinary literature supporting the need for emergent/immediate decompression in dogs that are deep pain negative secondary to IVDE is low and there likely exists a subset of dogs with severe spinal cord injury that, due to the severity of their injury, will not improve regardless of speed of intervention; however, the evidence to the contrary is also low and a threshold beyond which outcome may worsen has not been established. While warranting further investigation, an increased risk of myelomalacia with delayed decompression might support the recommendation to undertake decompression in severely injured dogs ideally within 12–24 h. The current literature lacks data specific to dogs that are paraplegic with intact deep pain, as these dogs are often grouped either with non-ambulatory paraparetic dogs, or with paraplegic deep pain negative dogs depending on study design. Specifically, it is not known how many dogs progress from paraplegic or paraplegic and deep pain negative when left briefly untreated, e.g., overnight.

THE NEED FOR FENESTRATION

Perhaps the most historically controversial issue in veterinary neurosurgery centers on the question of prophylactic fenestration of the intervertebral disc both at sites of current extrusion and at distant sites. The concept of intervertebral disc fenestration has been advanced by some veterinary spinal surgeons as a preventative measure which can be taken at the time of decompressive surgery to reduce future extrusion of disc material at sites adjacent to those affected at the time of the original procedure (34). Typically, fenestration of intervertebral discs between T11 and L4 are approached dorsolaterally or laterally at the time of surgery for a extruded disc, and a window is made into the annulus fibrosus with various means employed to evacuate any degenerated nucleus pulposus *in situ* (63–68). Fenestration of the L4-5 and L5-6 spaces is not typically performed due to concern for injury of the nerve roots essential for weight bearing at that location (69). In the context of acute canine IVDE, fenestration is performed "always" or "most of the time" by 69% of board-certified neurologists and 36% of board-certified surgeons (26). Clinicians who do not routinely fenestrate cite concerns including questionable efficacy; prolonged surgical time; complications such as hemorrhage, pneumothorax or nerve root injury; variable success in removal of *in situ* nucleus pulposus; potential for introduction of additional disc material into the vertebral canal; induction or worsening of degenerative changes to non-herniated discs, and the concern for adjacent segment disease (70, 71). Clinicians who do routinely fenestrate cite a recurrence rate as high as 40% for IVDE and the fact that dogs who present for a second bout of surgical IVDE have a rate of euthanasia as high as 44%, often due to financial concerns of the owner (34, 72).

A number of large-scale retrospectives, and two prospective studies have evaluated the effect of fenestration on recurrence

of IVDE (34, 36, 66, 69, 72, 73). It is clear from these and other studies that the likelihood of recurrence increases with the number of calcified discs *in situ* present. All studies support the concept that prophylactic fenestration is generally successful in reducing future extrusion of disc material at fenestrated disc spaces and that second disc extrusions, when they occur in dogs that have undergone prophylactic fenestration in conjunction with a history of previous decompressive surgery, are more likely to happen at non-fenestrated sites (34, 73). Results of large-scale contemporary studies evaluating outcome and recurrence after hemilaminectomy with fenestration are detailed in **Table 5**. Of note, of the >1,100 cases of surgical fenestration and associated outcome reported in the studies, complications from fenestration were noted in only 15 cases (0.01%), suggesting that fenestration is a safe procedure and concern for surgical complication of various types may not be a valid reason for choosing not to fenestrate.

Several challenges exist in interpreting the existing literature on fenestration. First, there is no study that prospectively compares, in a randomized fashion, recurrence rate of IVDE in dogs that undergo hemilaminectomy with and without fenestration. As such, clinical demographic factors, surgeon preference and experience, fenestration technique, and other patient- and clinician- level factors likely bias the current literature. Developing a randomized trial to evaluate this question is difficult because veterinarians who fenestrate do so because they believe the literature supports that it is effective to reduce recurrence, and those who do not may not have experience or enthusiasm to do so if participating in a trial. Additionally, comparing recurrence rates between studies is difficult due to differences in outcomes monitoring and reporting. Some patients with a recurrence of IVDE may not return to a specialty care facility for a second incidence of clinical signs, or those with mild signs may be managed medically without confirmatory imaging. Thus, the true incidence and etiology of signs may be underestimated or unclear. To address this concern, longitudinal studies involving the owner could provide valuable data. Inclusion of only confirmed cases of IVDE recurrence in some studies, but not others, also limits comparison between techniques. Even so, the current literature supports prophylactic fenestration as a safe way to reduce future disc herniation at fenestrated sites. Published studies suggest that fenestration reduces the recurrence rate of IVDE, and surgeons should particularly consider fenestration of calcified discs. However, high quality evidence in support of this is not available and it is not known how many sites should be fenestrated to achieve the best outcome, nor is it known what is the effect of fenestration on development of IVDE in adjacent unfenestrated sites of substantial consequence (ex. L4-5 in the patient fenestrated from T11-L4).

THE USE OF NEUROPROTECTIVE STRATEGIES

Various interventions have been evaluated in the context of acute spinal cord injury, many targeting secondary injury processes such as ischemia and vasospasm, inflammation, free radical production, ion channel disturbances and glutamate excitotoxicity.

Methylprednisolone Sodium Succinate (MPSS)

The use of high dose steroids, particularly methylprednisolone sodium succinate, in acute spinal cord injury takes its roots from the experimental spinal cord injury literature, where the proposed mechanism of therapy was prevention of lipid peroxidation and secondary free radical injury (74–77). This therapy was evaluated in several high-profile human spinal cord injury trials which showed a potential small treatment effect when administered within 8 h after injury, although the results of those studies, and their clinical implications, remain controversial (78–81). Some experimental evidence, including a study in dogs, had also suggested that this therapy might be less useful than originally anticipated (82). In a recent prospective randomized placebo controlled blinded clinical trial evaluating the effect of MPSS on outcome in paraplegic deep pain negative dogs with IVDE, no treatment effect was observed with respect to locomotor recovery (83).

Many veterinary clinicians continue to use corticosteroids such as prednisone or dexamethasone routinely at lower, anti-inflammatory doses for the management of canine IVDE (26). The question of whether treatment with non-steroidal anti-inflammatories (NSAIDs) or steroids is most appropriate represents a somewhat polarizing issue in veterinary medicine and is highly clinician-dependent. One retrospective study demonstrated decreased odds of successful outcome with conservative therapy, and lower owner-reported quality of life scores, with the use of corticosteroids, irrespective of dose, duration, or specific steroid administered (23). While no study provides a prospective comparison of NSAIDs vs. anti-inflammatory doses of steroids in the management of IVDE in dogs, a recent study retrospectively evaluated clinical risk factors for the development of progressive myelomalacia in deep pain negative dogs and suggested that administration of corticosteroids may have a protective effect (62). The use of steroids at anti-inflammatory doses or NSAIDs in management of canine IVDE is an area where clinical equipoise exists and while controlling for pre-treatment of dogs prior to referral would present a challenge, this question could lend itself to prospective randomized trials evaluating outcome, quality of life, and incidence of myelomalacia between the two treatments.

Polyethylene Glycol (PEG)

Polyethylene glycol (PEG) is a surfactant that gained popularity as a possible neuroprotective strategy for acute spinal cord injury. While not entirely understood, the proposed mechanism of PEG is that it acts as a fusogen to repair damaged neuronal cell membranes, prevent ion channel disturbances that lead to cytotoxic edema and secondary injury, and may also stimulate angiogenesis and promote axonal regeneration (84, 85). Studies in experimental models of SCI were encouraging for a positive treatment effect (86–88). Laverty et al. examined

TABLE 5 | Results of recent large-scale contemporaneous studies evaluating outcome and recurrence after hemilaminectomy with fenestration.

Reference	Number of cases	Study design	Surgical approach	Recurrence rate	Fenestration-associated complications	Notes
Mayhew et al. (72)	229	Retrospective case series	Hemilaminectomy without fenestration for all cases.	19.2%	Not applicable	None
Brisson et al. (34)	265	Retrospective case series	Hemilaminectomy for all cases. Prophylactic blade or power-assisted fenestration at various sites	4.4%	Pneumothorax ($n = 1$) Hemothorax ($n = 1$)	Hemothorax secondary to collagenase used for chemonucleolysis
Forterre et al. (69)	19	Prospective cohort	Hemilaminectomy for all cases. Single site power-assisted fenestration of the herniated disc vs. no fenestration	No recurrence in fenestrated group; 60% recurrence rate in non-fenestrated group although only 30% were clinical	None	Recurrence followed by MRI, not just clinical signs
Brisson et al. (66)	207	Prospective, randomized trial	Hemilaminectomy for all cases. Randomized to either single (herniated site only) or multi-site fenestration (T11-L4)	7.5% for multi-site fenestration and 18% for single site fenestration	Hemorrhage during fenestration ($n = 7$) Nerve root trauma ($n = 4$) Broken curette tip within the disc ($n = 1$)	Recurrence rate includes only confirmed recurrences; 92% of recurrences at a non-fenestrated site
Aikawa et al. (73)	662	Retrospective case series	Hemilaminectomy for all cases. Fenestration of all sites T11-12 to L1-2; L2-3 and L3-4 also performed in some dogs	2.3% based on clinical signs and imaging; 10% based on clinical signs alone	Intraoperative iatrogenic disc extrusion into the vertebral column ($n = 1$)	None

Results are compared to recurrence rate noted by Mayhew et al. without fenestration.

the therapy in a prospective open label canine clinical trial for dogs with IVDE (89). The canine study reported a positive treatment effect where a significantly higher number of deep pain negative dogs showed enhanced locomotor improvement but used a group of historical controls for which the recovery rate was less than typically reported for dogs who are deep pain negative secondary to IVDE. Subsequently, a prospective, placebo controlled, randomized, blinded trial evaluating the influence of PEG on outcome in deep pain negative dogs did not demonstrate a positive treatment effect (83).

Other Strategies

A host of other neuroprotective strategies have been evaluated in both the laboratory and human clinical setting for treatment of acute spinal cord injury. Most promising based on positive findings in the experimental setting are minocycline, riluzole, and glybenclamide. At this time, there is no published efficacy data on any of these interventions for use in canine IVDE, although all three medications have known pharmacokinetics in the dog making them amenable to future veterinary clinical trials (90–92).

Several other adjunctive therapeutic strategies are not addressed in the present review but have been suggested throughout the veterinary literature. These interventions have only been evaluated in a small number of published studies, and include acupuncture, pulsed electromagnetic field therapy, chiropractic manipulation, and photobiomodulation (93–95).

THE ROLE OF PHYSICAL ACTIVITY IN THE DEVELOPMENT OF AND RECOVERY FROM IVDE

With respect to the role of physical activity in the development of and recovery from IVDE, several clinical questions exist regarding ideal daily activity levels for prevention of disease in dogs "at risk," what constitutes appropriate restriction of activity after decompressive surgery, and the role of physiotherapy in post-operative recovery.

Daily Activity for Dogs "At Risk" of IVDE

The influence of daily activity level on disc degeneration and the development of IVDE has been previously explored by Packer et al. who evaluated the impact of lifestyle on IVDE risk. It was observed that dogs receiving >1 h of daily exercise were less likely to have IVDE compared to dogs receiving <30 min of daily exercise and not allowed to jump on and off furniture (96). This study suggests that, while activity modification is often recommended by clinicians as a preventative measure against IVDE in chondrodystrophic breeds, this recommendation might be counter-productive.

Post-operative Activity Restriction

Veterinary neurologists and surgeons tend to view strict activity restriction (also termed "cage rest") as a vital component of both conservative and post-operative management of IVDE; however, the impact of this recommendation is poorly studied in dogs (26). Cage rest is usually defined as confinement to a small run or cage

at all times except when the animal needs to eliminate. Cited goals of cage rest include allowing healing of the annulus fibrosus to prevent further extrusion of nucleus pulposus, prevention of further traumatic injury in an ataxic animal, and reducing pain and inflammation associated with affected nerve roots and meninges (25, 97–99). The duration of recommended cage rest is variable; however, some authors recommend as much as 6–8 weeks (26, 97). Whereas, long-term bed rest has not been shown to be beneficial in people with lumbar disc herniations, the argument for cage rest in the management of canine IVDE might be more logical based on anatomy, underlying pathophysiology, and inherent inability to reason with veterinary patients as to why they should consciously self-limit activity (100, 101). The influence of cage rest on outcome has been examined in only one published retrospective veterinary study in which outcome in dogs managed conservatively for IVDE was not influenced by duration of cage rest (23).

Physiotherapy

An additional consideration with respect to activity modification is whether some forms of controlled activity, in the form of physiotherapy, might actually be beneficial to locomotor recovery in the postoperative setting, particularly in more severely affected dogs. This rationale stems from the experimental spinal cord injury literature, where a number of studies have demonstrated that intensive locomotor training can promote anatomic and physiologic changes within the injured spinal cord that might result in improved motor function (102–105). However, the experimental literature relating to physiotherapy is mixed, with persistent questions regarding timing and correct complement of activities to result in improved function vs. maladaptive neuroplasticity (106, 107). To a limited degree, some successful findings have translated to the human clinical setting where very small-scale open label studies have shown mild improvements in weight support, stepping, and spasticity with intensive locomotor training programs often coupled with cell-based therapies, or implantable epidural or nerve stimulation devices (108–112).

Various authors have advocated for the role of physiotherapy in canine IVDE, and physiotherapy is routinely recommended by many veterinary neurologists and orthopedic surgeons (26, 95, 113–115). Several studies have evaluated the role of physiotherapy in dogs with severe spinal cord injury caused by IVDE (93, 115, 116). Bennaim et al. conducted a prospective randomized controlled trial evaluating the influence of both physiotherapy and photobiomodulation on motor recovery in dogs with severe IVDE (non-ambulatory paraparesis or worse) (93). A positive treatment effect was not noted in that study, although the authors suggest that case numbers were not large enough to draw a firm conclusion. Conversely, a large-scale retrospective study of physiotherapy conducted by Jeong et al. evaluated neurologic outcome in dogs with IVDE causing injuries ranging from ambulatory paraparesis to paraplegic with absent nociception (115). The authors noted a significant improvement in locomotor outcome for dogs receiving surgical decompression coupled with physiotherapy when compared to those receiving surgical decompression alone. However, successful locomotor outcome for dogs with paraplegia with or without intact deep pain receiving decompressive surgery alone was only reported to be 17%, which is much lower than the typical outcomes reported in literature which range from 50 to 60%. Zidan et al. also conducted a prospective blinded trial where dogs with incomplete SCI caused by IVDE were randomized after surgical decompression to receive either a basic in-hospital physiotherapy program consisting only of passive range of motion and sling walking activities or a more intensive therapy program (116). No difference in locomotor outcome was observed between groups but this trial was designed to see if the rate of recovery of locomotion could be influenced in a population of non-ambulatory paraparetic and paraplegic deep pain positive dogs. It did not target dogs known to have less chance of recovery and suggests that a randomized controlled trial is indicated to investigate the influence of rehabilitation on recovery in dogs with more severe injuries that fail to show early improvement.

The current state of the veterinary literature does not support a role of routine physiotherapy to improve locomotor outcome in dogs with mild to moderate spinal cord injury caused by IVDE. A challenge in interpreting this literature is the fact that studies include dogs with all injury grades, many of which would assuredly recover with or without other intervention, making it difficult to demonstrate a treatment effect without a very large sample size. Additionally, there is not a standardized physiotherapy program followed across the field, making it is difficult to compare results between studies. Lastly, the type of physiotherapy shown to improve locomotor outcome in experimental injury models, and now employed in people with severe injuries, is a very intensive type of locomotor training. The types of activities described in these locomotor training protocols extend well-beyond the intensity of therapeutic activities implemented in veterinary medicine, which more classically includes under-water treadmill walking, passive range of motion exercises and assisted weight-supported walking. It should also be noted that the primary aim in the acute phase of IVDE is to regain movement, over-ground locomotion and balance but "under-water" treadmill is best suited to improve strength and muscle mass. Activities directed toward restoration of movement, such as the use of "over-ground" treadmill training (as shown in people to improve motor recovery and balance (*e.g.,* using proprioceptive platforms) may actually be more rational early on in the recovery process for IVDE and should be encouraged first (117).

In people with severe spinal cord injury, more intensive protocols are often coupled with external or implantable assistive devices not used in veterinary medicine. For example, robotic assisted gait training clearly reduces spasticity and improves lower limbs motor function in people with severe spinal cord injury (118). Thus, findings from the human clinical setting may not reflect those reasonably expected in veterinary medicine using current approaches or previously evaluated patient populations. A recently published retrospective study by Gallucci et al. showed an increase in the development of "spinal walking" in deep pain negative dogs despite the fact that these dogs did not regain pain perception (119). Further prospective studies are needed to determine what impact

intensive physiotherapy may have in deep pain negative dogs, or on parameters beyond locomotor recovery including important SCI comorbidities such as pressure sores, neuropathic pain, and spasticity. In that respect, consensus about retraining after spinal cord injury lags far behind what is applied in humans where the literature covering that topic is vast. In particular, recent reviews and meta-analysis have clearly shown the benefit of several physiotherapy interventions to improve voluntary muscle strength (120). This includes interventions such as resistance training, functional electrical stimulation or robotic gait training. Some of these are difficult to implement in dogs because of cost and the challenges of eliciting specific voluntary movements on command, but others such as functional electrical stimulation, "over-ground" treadmill training and proprioceptive platform training are more feasible.

CONCLUSIONS AND RECOMMENDATIONS

There exist a number of gaps in the veterinary literature with respect to standards of care for dogs with acute thoracolumbar IVDE. Areas identified for future study include comparison of medical and surgical management for dogs with more mild signs associated with IVDE (those who are only painful or have ambulatory paraparesis), the effect of early decompression in locomotor recovery in dogs with non-ambulatory paraparesis or paraplegic with intact deep pain, and on the incidence of progressive myelomalacia in deep pain negative dogs, the effect of durotomy coupled with spinal decompression in dogs with severe injuries, and the influence of intensive physiotherapy in deep pain negative dogs. Recommendations for standard care can be made in some areas, particularly with respect to surgical decompression where the currently available evidence supports that surgery should be recommended for dogs with non-ambulatory paraparesis or worse. While additional information is needed about the influence on timing of decompression on outcome in dogs that are deep pain negative for longer than 48 h duration, there is no evidence to support treatment of the 48 h time point as a cut off beyond which it becomes impossible for dogs to achieve locomotor recovery. Surgical decompression is best accomplished by either hemilaminectomy or mini-hemilaminectomy and fenestration of, at a minimum, the acutely ruptured disc. Adjacent discs easily accessed by way of the same approach should be considered for fenestration given the evidence that this substantially reduces future herniation at fenestrated sites. Currently available neuroprotective strategies such as high does MPSS and PEG are not recommended due to lack of demonstrated treatment effect in randomized controlled trials. The role of anti-inflammatory steroids as a protective strategy against progressive myelomalacia and the question of whether anti-inflammatory steroids or NSAIDs provide superior medical therapy require further evaluation.

AUTHOR CONTRIBUTIONS

SM, AT, NO, VS, and NG conceived, wrote, and edited the manuscript. CANSORT-SCI edited the manuscript. All authors have approved the final submitted version.

REFERENCES

1. Bergknut N, Egenvall A, Hagman R, Gustås P, Hazewinkel HA, Meij BP, et al. Incidence of intervertebral disk degeneration-related diseases and associated mortality rates in dogs. *J Am Vet Med Assoc*. (2012) 240:1300–9. doi: 10.2460/javma.240.11.1300
2. Hansen HJ. A pathologic-anatomical interpretation of disc degeneration in dogs. *Acta Orthop Scand*. (1951) 20:280–93. doi: 10.3109/17453675108991175
3. Frankel HL, Hancock DO, Hyslop G, Melzak J, Michaelis LS, Ungar GH, et al. The value of postural reduction in the initial management of closed injuries of the spine with paraplegia and tetraplegia I. *Paraplegia*. (1969) 7:179–92. doi: 10.1038/sc.1969.30
4. Levine GJ, Levine JM, Budke CM, Kerwin SC, Au J, Vinayak A, et al. Description and repeatability of a newly developed spinal cord injury scale for dogs. *Prev Vet Med*. (2009) 89:121–7. doi: 10.1016/j.prevetmed.2009.02.016
5. Olsson SE. On disc protrusion in dog (enchondrosis intervertebralis); a study with special reference to roentgen diagnosis and to the value of disc fenestration. *Acta Orthop Scand Suppl*. (1951) 8:1–95. doi: 10.3109/ort.1951.22.suppl-8.01
6. Tillmans S. *Beiträge Zur Enchondrosis Intervertebral. und der Wirbelkanltumoren des Hundes* (Dissertation). University of Giessen, Giessen, Germany (1939).
7. Frauchiger E, Fankhauser R. *Die nervenkrankheiten unserer hunde*. Bern: Verlag Hans Huber (1949).
8. Hansen HJ. A pathologic-anatomical study on disc degeneration in dog, with special reference to the so-called enchondrosis intervertebralis. *Acta Orthop Scand Suppl*. (1952) 11:1–117. doi: 10.3109/ort.1952.23.suppl-11.01
9. Hoerlein BF. Further evaluation of the treatment of disc protrusion paraplegia in the dog. *J Am Vet Med Assoc*. (1956) 129:495–502.
10. Hoerlein BF. Intervertebral disk disease. In: Oliver JE, Hoerlein BF, Mayhew IG, editors. *Veterinary Neurology*. Philadelphia, PA: Saunders (1987). p. 321–42.
11. Hoerlein BF, Redding RW, Hoff EJ, McGuire JA. Evaluation of dexamethasone, DMSO, mannitol, and solcoseryl in acute spinal cord trauma. *J Am Animal Hosp Assoc*. (1983) 19:216–226.
12. Hoerlein BF, Redding RW, Hoff EJ Jr, McGuire JA. Evaluation of naloxone, crocetin, thyrotropin releasing hormone, methylprednisolone, partial myelotomy, and hemilaminectomy in the treatment of acute spinal cord trauma. *J Am Animal Hosp Assoc*. (1985) 21:67.
13. Funkquist B. Thoracolumbar disk protrusion with severe cord compression in the dog II. clinical observations with special reference to the prognosis in conservative treatment. *Acta Vet Scand*. (1962) 3:334.
14. Hoerlein BF. *Canine Neurology: Diagnosis and Treatment. 3rd edn*. Philadelphia, PA: Saunders Company (1978).
15. Funkquist B. Decompressive laminectomy in thoraco-lumbar disc protrusion with paraplegia in the dog. *J Small Anim Pract*. (1970) 11:445–51. doi: 10.1111/j.1748-5827.1970.tb05595.x
16. Knecht CD. The effect of delayed hemilaminectomy in the treatment of intervertebral disc protrusion in dogs. *J Am Animal Hosp Assoc*. (1970) 6:71–7.
17. Langerhuus L, Miles J. Proportion recovery and times to ambulation for non-ambulatory dogs with thoracolumbar disc extrusions treated with hemilaminectomy or conservative treatment: A systematic review and meta-analysis of case-series studies. *Vet J*. (2017) 220:7–16. doi: 10.1016/j.tvjl.2016.12.008

18. Davies JV, Sharp NJH. A comparison of conservative treatment and fenestration for thoracolumbar intervertebral disc disease in the dog. *J Small Animal Pract*. (1983) 24:721–9. doi: 10.1111/j.1748-5827.1983.tb00360.x
19. Scott HW, McKee WM. Laminectomy for 34 dogs with thoracolumbar intervertebral disc disease and loss of deep pain perception. *J Small Anim Pract*. (1999) 40:417–22. doi: 10.1111/j.1748-5827.1999.tb03114.x
20. Olby N, Levine J, Harris T, Muñana K, Skeen T, Sharp N. Long-term functional outcome of dogs with severe injuries of the thoracolumbar spinal cord: 87 cases (1996-2001). *J Am Vet Med Assoc*. (2003) 222:762–9. doi: 10.2460/javma.2003.222.762
21. Laitinen OM, Puerto DA. Surgical decompression in dogs with thoracolumbar intervertebral disc disease and loss of deep pain perception: A retrospective study of 46 cases. *Acta Vet Scand*. (2005). 46:79–85. doi: 10.1186/1751-0147-46-79
22. Ruddle TL, Allen DA, Schertel ER, Barnhart MD, Wilson ER, Lineberger JA, et al. Outcome and prognostic factors in non-ambulatory hansen type I intervertebral disc extrusions: 308 cases. *Vet Comp Orthop Traumatol*. (2006) 19:29–34. doi: 10.1055/s-0038-1632970
23. Levine JM, Levine GJ, Johnson SI, Kerwin SC, Hettlich BF, Fosgate GT. Evaluation of the success of medical management for presumptive thoracolumbar intervertebral disk herniation in dogs. *Vet Surg*. (2007) 36:482–91. doi: 10.1111/j.1532-950X.2007.00295.x
24. Han HJ, Yoon HY, Kim JY, Jang HY, Lee B, Choi SH, et al. Clinical effect of additional electroacupuncture on thoracolumbar intervertebral disc herniation in 80 paraplegic dogs. *Am J Chin Med*. (2010) 38:1015–25. doi: 10.1142/S0192415X10008433
25. Sharp NJH, Wheeler SJ. *Small Animal Spinal Disorders. 2nd edn*. Philadelphia, PA: Elsevier (2005). p. 124–125.
26. Moore SA, Early PJ, Hettlich BF. Practice patterns in the management of acute intervertebral disc herniation in dogs. *J Small Anim Pract*. (2016) 57:409–15. doi: 10.1111/jsap.12496
27. Freeman P, Jeffery ND. Re-opening the window on fenestration as a treatment for acute thoracolumbar intervertebral disc herniation in dogs. *J Small Anim Pract*. (2017) 58:199–204. doi: 10.1111/jsap.12653
28. Jeffery ND, Freeman PM. The role of fenestration in management of Type I thoracolumbar disc degeneration. *Vet Clin North Am Small Anim Pract*. (2018) 48:187–200. doi: 10.1016/j.cvsm.2017.08.012
29. Prata R. Neurosurgical treatment of thoracolumbar disks - the rationale and value. *J Am Anim Hosp Assoc*. (1981) 17:17–26.
30. Steffen F, Kircher PR, Dennler M. Spontaneous regression of lumbar hansen type 1 disk extrusion detected with magnetic resonance imaging in a dog. *J Am Vet Med Assoc*. (2014) 244:715–8. doi: 10.2460/javma.244.6.715
31. Hong J, Ball PA. Images In Clinical Medicine. Resolution of lumbar disk herniation without surgery. *N Engl J Med*. (2016) 374:1564. doi: 10.1056/NEJMicm1511194
32. Dimar JR, Glassman SD, Raque GH, Zhang YP, Shields CB. The influence of spinal canal narrowing and timing of decompression on neurologic recovery after spinal cord contusion in a rat model. *Spine*. (1999) 24:1623–33. doi: 10.1097/00007632-199908150-00002
33. Macias C, McKee WM, May C, Innes JF. Thoracolumbar disc disease in large dogs: A study of 99 cases. *J Small Anim Pract*. (2002) 43:439–46. doi: 10.1111/j.1748-5827.2002.tb00010.x
34. Brisson BA, Moffatt SL, Swayne SL, Parent JM. Recurrence of thoracolumbar intervertebral disk extrusion in chondrodystrophic dogs after surgical decompression with or without prophylactic fenestration: 265 cases (1995-1999). *J Am Vet Med Assoc*. (2004) 224:1808–14. doi: 10.2460/javma.2004.224.1808
35. Itoh H, Hara Y, Yoshimi N, Harada Y, Nezu Y, Yogo T, et al. A retrospective study of intervertebral disc herniation in dogs in japan: 297 cases. *J Vet Med Sci*. (2008) 70:701–6. doi: 10.1292/jvms.70.701
36. Aikawa T, Fujita H, Kanazono S, Shibata M, Yoshigae Y. Long-term neurologic outcome of hemilaminectomy and disk fenestration for treatment of dogs with thoracolumbar intervertebral disk herniation: 831 cases (2000-2007). *J Am Vet Med Assoc*. (2012) 241:1617–26. doi: 10.2460/javma.241.12.1617
37. Granger N, Carwardine D. Acute spinal cord injury: Tetraplegia and paraplegia in small animals. *Vet Clin North Am Small Anim Pract*. (2014) 44:1131–56. doi: 10.1016/j.cvsm.2014.07.013
38. Boyd A, Golding J, Macleod J, Lawlor DA, Fraser A, Henderson J, et al. Cohort profile: the 'children of the 90s'—the index offspring of the avon longitudinal study of parents and children. *Int J Epidemiol*. (2013) 42:111–27. doi: 10.1093/ije/dys064
39. The Bristol Cats Study (2020). Available online at: http://www.bristol.ac.uk/vet-school/research/projects/cats/ (accessed April 21, 2020).
40. Flegel T, Boettcher IC, Ludewig E, Kiefer I, Oechtering G, Böttcher P. Partial lateral corpectomy of the thoracolumbar spine in 51 dogs: Assessment of slot morphometry and spinal cord decompression. *Vet Surg*. (2011) 40:14–21. doi: 10.1111/j.1532-950X.2010.00747.x
41. Huska JL, Gaitero L, Brisson BA, Nykamp S, Thomason J, Sears WC. Comparison of the access window created by hemilaminectomy and mini-hemilaminectomy in the thoracolumbar vertebral canal using computed tomography. *Can Vet J*. (2014) 55:449–55.
42. Salger F, Ziegler L, Böttcher IC, Oechtering G, Böttcher P, Flegel T. Neurologic outcome after thoracolumbar partial lateral corpectomy for intervertebral disc disease in 72 dogs. *Vet Surg*. (2014) 43:581–8. doi: 10.1111/j.1532-950X.2014.12157.x
43. Gage ED, Hoerlein BF. Hemilaminectomy and dorsal laminectomy for relieving compressions of the spinal cord in the dog. *J Am Vet Med Assoc*. (1968) 152:351–9.
44. Hoerlein BF. The status of the various intervertebral disk surgeries for the dog. *J Am Animal Hosp Assoc*. (1978) 14:563.
45. Muir P, Johnson KA, Manley PA, Dueland RT. Comparison of hemilaminectomy and dorsal laminectomy for thoracolumbar intervertebral disc extrusion in dachshunds. *J Small Anim Pract*. (1995) 36:360–7. doi: 10.1111/j.1748-5827.1995.tb02950.x
46. Horne TR, Powers RD, Swaim SF. Dorsal laminectomy techniques in the dog. *J Am Vet Med Assoc*. (1977) 171:742–9.
47. Gambardella PC. Dorsal decompressive laminectomy for treatment of thoracolumbar disc disease in dogs: a retrospective study of 98 cases. *Vet Surg*. (1980) 9:24–6. doi: 10.1111/j.1532-950X.1980.tb01647.x
48. David T. Thoracolumbar hemilaminectomy in the dog using a power skull trephine (a photo essay). *Vet Med Small Anim Clin*. (1976) 71: 477–9.
49. Anderson SM, Lippincott CL, Gill PJ. Hemilaminectomy in dogs without deep pain perception. *Calif Vet*. (1991) 45:24–8.
50. McKee WM. A comparison of hemilaminectomy (with concomitant disc fenestration) and dorsal laminectomy for the treatment of thoracolumbar disc protrusion in dogs. *Vet Rec*. (1992) 130:296–300. doi: 10.1136/vr.130.14.296
51. Jeffery ND. Treatment of acute and chronic thoracolumbar disc disease by "mini hemilaminectomy". *J Small Animal Pract*. (1988) 29:611–6. doi: 10.1111/j.1748-5827.1988.tb02181.x
52. Svensson G, Simonsson US, Danielsson F, Schwarz T. Residual spinal cord compression following hemilaminectomy and mini-hemilaminectomy in dogs: a prospective randomized study. *Front Vet Sci*. (2017) 4:42. doi: 10.3389/fvets.2017.00042
53. Flo GL, Brinker WO. Lateral fenestration of thoracolumbar discs. *J Am Animal Hosp Assoc*. (1975) 11:619–26.
54. Denny HR. The lateral fenestration of canine thoracolumbar disc protrusions: A review of 30 cases. *J Small Anim Pract*. (1978) 19:259–66. doi: 10.1111/j.1748-5827.1978.tb05487.x
55. Butterworth SJ, Denny HR. Follow-up study of 100 cases with thoracolumbar disc protrusions treated by lateral fenestration. *J Small Animal Pract*. (1991) 32:443–7. doi: 10.1111/j.1748-5827.1991.tb00983.x
56. Olsson SE. Observations concerning disc fenestration in dogs. *Acta Orthop Scand*. (1951) 20:349–56. doi: 10.3109/17453675108991182
57. Kinzel S, Wolff M, Buecker A, Krombach GA, Stopinski T, Afify M, et al. Partial percutaneous discectomy for treatment of thoracolumbar disc protrusion: retrospective study of 331 dogs. *J Small Anim Pract*. (2005) 46:479–84. doi: 10.1111/j.1748-5827.2005.tb00276.x
58. Sukhiani HR, Parent JM, Atilola MA, Holmberg DL. Intervertebral disk disease in dogs with signs of back pain alone: 25 cases (1986-1993). *J Am Vet Med Assoc*. (1996) 209:1275–9.
59. Jeffery ND, Barker AK, Hu HZ, Alcott CJ, Kraus KH, Scanlin EM, et al. Factors associated with recovery from paraplegia in dogs with loss of pain perception in the pelvic limbs following intervertebral disk

herniation. *J Am Vet Med Assoc*. (2016) 248:386–94. doi: 10.2460/javma.248.4.386
60. Moore SA, Zidan N, Spitzbarth I, Nout-Lomas YS, Granger N, da Costa RC, et al. Development of an international canine spinal cord injury (CSCI) observational registry: A collaborative data-sharing network to optimize translational studies of SCI. *Spinal Cord*. (2018) 56:656–65. doi: 10.1038/s41393-018-0145-4
61. Dvorak MF, Noonan VK, Fallah N, Fisher CG, Finkelstein J, Kwon BK, et al. The influence of time from injury to surgery on motor recovery and length of hospital stay in acute traumatic spinal cord injury: an observational canadian cohort study. *J Neurotrauma*. (2015) 32:645–54. doi: 10.1089/neu.2014.3632
62. Castel A, Olby NJ, Mariani CL, Muñana KR, Early PJ. Clinical characteristics of dogs with progressive myelomalacia following acute intervertebral disc extrusion. *J Vet Intern Med*. (2017) 31:1782–9. doi: 10.1111/jvim.14829
63. Holmberg DL, Palmer NC, VanPelt D, Willan AR. A comparison of manual and power-assisted thoracolumbar disc fenestration in dogs. *Vet Surg*. (1990) 19:323–7. doi: 10.1111/j.1532-950X.1990.tb01199.x
64. Takahashi T, Nakayama M, Chimura S, Nakahara K, Morozumi M, Horie K, et al. Treatment of canine intervertebral disc displacement with chondroitinase ABC. *Spine*. (1997) 22:1435–47. doi: 10.1097/00007632-199707010-00002
65. Morelius M, Bergadano A, Spreng D, Schawalder P, Doherr M, Forterre F. Influence of surgical approach on the efficacy of the intervertebral disk fenestration: a cadaveric study. *J Small Anim Pract*. (2007) 48:87–92. doi: 10.1111/j.1748-5827.2007.00269.x
66. Brisson BA, Holmberg DL, Parent J, Sears WC, Wick SE. Comparison of the effect of single-site and multiple-site disk fenestration on the rate of recurrence of thoracolumbar intervertebral disk herniation in dogs. *J Am Vet Med Assoc*. (2011) 238:1593–600. doi: 10.2460/javma.238.12.1593
67. Forterre F, Dickomeit M, Senn D, Gorgas D, Spreng D. Microfenestration using the CUSA excel ultrasonic aspiration system in chondrodystrophic dogs with thoracolumbar disk extrusion: a descriptive cadaveric and clinical study. *Vet Surg*. (2011) 40:34–9. doi: 10.1111/j.1532-950X.2010.00780.x
68. Thomovsky SA, Packer RA, Lambrechts NE, Moore GE. Canine intervertebral disc fenestration using a vacuum-assisted tissue resection device. *Vet Surg*. (2012) 41:1011–7. doi: 10.1111/j.1532-950X.2012.01060.x
69. Forterre F, Konar M, Spreng D, Jaggy A, Lang J. Influence of intervertebral disc fenestration at the herniation site in association with hemilaminectomy on recurrence in chondrodystrophic dogs with thoracolumbar disc disease: A prospective MRI study. *Vet Surg*. (2008) 37:399–405. doi: 10.1111/j.1532-950X.2008.00394.x
70. Grunert P, Moriguchi Y, Grossbard BP, Ricart Arbona RJ, Bonassar LJ, Härtl R. Degenerative changes of the canine cervical spine after discectomy procedures, an *in vivo* study. *BMC Vet Res*. (2017) 13:193. doi: 10.1186/s12917-017-1105-5
71. Harris G, Freeman P. Introduction of disc material into the vertebral canal by fenestration of thoracolumbar discs following decompressive surgery. *Vet Comp Orthop Traumatol*. (2020) 33:66–70. doi: 10.1055/s-0039-1700554
72. Mayhew PD, McLear RC, Ziemer LS, Culp WT, Russell KN, Shofer FS, et al. Risk factors for recurrence of clinical signs associated with thoracolumbar intervertebral disk herniation in dogs: 229 cases (1994-2000). *J Am Vet Med Assoc*. (2004) 225:1231–6. doi: 10.2460/javma.2004.225.1231
73. Aikawa T, Fujita H, Shibata M, Takahashi T. Recurrent thoracolumbar intervertebral disc extrusion after hemilaminectomy and concomitant prophylactic fenestration in 662 chondrodystrophic dogs. *Vet Surg*. (2012) 41:381–90. doi: 10.1111/j.1532-950X.2012.00970.x
74. Hall ED. Lipid antioxidants in acute central nervous system injury. *Ann Emerg Med*. (1993) 22:1022–7. doi: 10.1016/S0196-0644(05)82745-3
75. Behrmann DL, Bresnahan JC, Beattie MS. Modeling of acute spinal cord injury in the rat: Neuroprotection and enhanced recovery with methylprednisolone, U-74006F and YM-14673. *Exp Neurol*. (1994) 126:61–75. doi: 10.1006/exnr.1994.1042
76. Ildan F, Oner A, Polat S, Isbir T, Göcer AI, Kaya M, et al. Correlation of alterations on na(+)-K+/Mg+2 ATPase activity, lipid peroxidation and ultrastructural findings following experimental spinal cord injury with and without intravenous methylprednisolone treatment. *Neurosurg Rev*. (1995) 18:35–44. doi: 10.1007/BF00416476
77. Diaz-Ruiz A, Rios C, Duarte I, Correa D, Guizar-Sahagun G, Grijalva I, et al. Lipid peroxidation inhibition in spinal cord injury: cyclosporin-A vs methylprednisolone. *Neuroreport*. (2000) 11:1765–7. doi: 10.1097/00001756-200006050-00033
78. Coleman WP, Benzel D, Cahill DW, Ducker T, Geisler F, Green B, et al. A critical appraisal of the reporting of the national acute spinal cord injury studies (II and III) of methylprednisolone in acute spinal cord injury. *J Spinal Disord*. (2000) 13:185–99. doi: 10.1097/00002517-200006000-00001
79. Hurlbert RJ. Methylprednisolone for acute spinal cord injury: An inappropriate standard of care. *J Neurosurg*. (2000) 93(Suppl 1):1–7. doi: 10.3171/spi.2000.93.1.0001
80. Bracken MB. Steroids for acute spinal cord injury. *Cochrane Database Syst Rev*. (2012) 1:CD001046. doi: 10.1002/14651858.CD001046.pub2
81. Fehlings MG, Wilson JR, Harrop JS, Kwon BK, Tetreault LA, Arnold PM, et al. Efficacy and safety of methylprednisolone sodium succinate in acute spinal cord injury: A systematic review. *Global Spine J*. (2017) 7(Suppl. 3):116S—37. doi: 10.1177/2192568217706366
82. Coates JR, Sorjonen DC, Simpson ST, Cox NR, Wright JC, Hudson JA, et al. Clinicopathologic effects of a 21-aminosteroid compound (U74389G) and high-dose methylprednisolone on spinal cord function after simulated spinal cord trauma. *Vet Surg*. (1995) 24:128–39. doi: 10.1111/j.1532-950X.1995.tb01307.x
83. Olby NJ, Muguet-Chanoit AC, Lim JH, Davidian M, Mariani CL, Freeman AC, et al. A placebo-controlled, prospective, randomized clinical trial of polyethylene glycol and methylprednisolone sodium succinate in dogs with intervertebral disk herniation. *J Vet Intern Med*. (2016) 30:206–14. doi: 10.1111/jvim.13657
84. Shi R. Polyethylene glycol repairs membrane damage and enhances functional recovery: A tissue engineering approach to spinal cord injury. *Neurosci Bull*. (2013) 29:460–6. doi: 10.1007/s12264-013-1364-5
85. Lu X, Perera TH, Aria AB, Callahan LAS. Polyethylene glycol in spinal cord injury repair: A critical review. *J Experi Pharmacol*. (2018) 10:37–49. doi: 10.2147/JEP.S148944
86. Luo J, Borgens R, Shi R. Polyethylene glycol immediately repairs neuronal membranes and inhibits free radical production after acute spinal cord injury. *J Neurochem*. (2002) 83:471–80. doi: 10.1046/j.1471-4159.2002.01160.x
87. Luo J, Borgens R, Shi R. Polyethylene glycol improves function and reduces oxidative stress in synaptosomal preparations following spinal cord injury. *J Neurotrauma*. (2004) 21:994–1007. doi: 10.1089/0897715041651097
88. Luo J, Shi R. Diffusive oxidative stress following acute spinal cord injury in guinea pigs and its inhibition by polyethylene glycol. *Neurosci Lett*. (2004) 359:167–70. doi: 10.1016/j.neulet.2004.02.027
89. Laverty PH, Leskovar A, Breur GJ, Coates JR, Bergman RL, Widmer WR, et al. A preliminary study of intravenous surfactants in paraplegic dogs: polymer therapy in canine clinical SCI. *J Neurotrauma*. (2004) 21:1767–77. doi: 10.1089/neu.2004.21.1767
90. Hnot ML, Cole LK, Lorch G, Rajala-Schultz PJ, Papich MG. Effect of feeding on the pharmacokinetics of oral minocycline in healthy research dogs. *Vet Dermatol*. (2015) 26:399–3. doi: 10.1111/vde.12246
91. Jeffery N, Boudreau CE, Konarik M, Mays T, Fajt V. Fajt Pharmacokinetics V, and safety of oral glyburide in dogs with acute spinal cord injury. *PeerJ*. (2018) 6:e4387. doi: 10.7717/peerj.4387
92. Perdigão APL, Antunes NJ, Juni LT, de Freitas NL, Rojas-Moscoso J, Corrãa SVM, et al. Pharmacokinetics of riluzole in beagle dogs. *Drug Res*. (2019) 69:40–5. doi: 10.1055/a-0645-1248
93. Bennaim M, Porato M, Jarleton A, Hamon M, Carroll JD, Gommeren K, et al. Preliminary evaluation of the effects of photobiomodulation therapy and physical rehabilitation on early postoperative recovery of dogs undergoing hemilaminectomy for treatment of thoracolumbar intervertebral disk disease. *Am J Vet Res*. (2017) 78:195–206. doi: 10.2460/ajvr.78.2.195
94. Zidan N, Fenn J, Griffith E, Early PJ, Mariani CL, Muñana KR, et al. The effect of electromagnetic fields on post-operative pain and locomotor recovery in dogs with acute, severe thoracolumbar intervertebral disc extrusion: A randomized placebo-controlled, prospective clinical trial. *J Neurotrauma*. (2018) 35:1726–36. doi: 10.1089/neu.2017.5485

95. Frank LR, Roynard PFP. Veterinary neurologic rehabilitation: The rationale for a comprehensive approach. *Top Companion Anim Med*. (2018) 33:49–57. doi: 10.1053/j.tcam.2018.04.002
96. Packer RM, Seath IJ, O'Neill DG, De Decker S, Volk HA. DachsLife 2015: An investigation of lifestyle associations with the risk of intervertebral disc disease in dachshunds. *Canine Genet Epidemiol*. (2016) 3:8. doi: 10.1186/s40575-016-0039-8
97. Coates JR. Intervertebral disk disease. *Vet Clin North Am Small Anim Pract*. (2000) 30:77–110. doi: 10.1016/S0195-5616(00)50004-7
98. Bagley R. Basics of treatment of important spinal cord diseases of dogs and cats. *Fund Vet Clin Neurol*. (2005) 14:323–49.
99. Dewey CW, Costa RC. *Practical Guide to Canine and Feline Neurology*. Ames, IA: John Wiley & Sons (2015).
100. Deyo RA, Diehl AK, Rosenthal M. How many days of bed rest for acute low back pain? A randomized clinical trial. *N Engl J Med*. (1986) 315:1064–70. doi: 10.1056/NEJM198610233151705 Available online at: https://www.wiley.com/en-us/Fundamentals+of+Veterinary+Clinical+Neurology-p-9780813828435
101. Vroomen PC, de Krom MC, Wilmink JT, Kester AD, Knottnerus JA. Lack of effectiveness of bed rest for sciatica. *N Engl J Med*. (1999) 340:418–23. doi: 10.1056/NEJM199902113400602
102. Van de Crommert HW, Mulder T, Duysens J. Neural control of locomotion: sensory control of the central pattern generator and its relation to treadmill training. *Gait Posture*. (1998) 7:251–63. doi: 10.1016/S0966-6362(98)00010-1
103. Côté MP, Gossard JP. Step training-dependent plasticity in spinal cutaneous pathways. *J Neurosci*. (2004) 24:11317–27. doi: 10.1523/JNEUROSCI.1486-04.2004
104. Takeoka A, Vollenweider I, Courtine G, Arber S. Muscle spindle feedback directs locomotor recovery and circuit reorganization after spinal cord injury. *Cell*. (2014) 159:1626–39. doi: 10.1016/j.cell.2014.11.019
105. Rossignol S, Martinez M, Escalona M, Kundu A, Delivet-Mongrain H, Alluin O, et al. The "beneficial" effects of locomotor training after various types of spinal lesions in cats and rats. *Prog Brain Res*. (2015) 218:173–98. doi: 10.1016/bs.pbr.2014.12.009
106. Ferguson AR, Huie JR, Crown ED, Baumbauer KM, Hook MA, Garraway SM, et al. Maladaptive spinal plasticity opposes spinal learning and recovery in spinal cord injury. *Front Physiol*. (2012) 3:399. doi: 10.3389/fphys.2012.00399
107. Huie JR, Morioka K, Haefeli J, Ferguson AR. What is being trained? How divergent forms of plasticity compete to shape locomotor recovery after spinal cord injury. *J Neurotrauma*. (2017) 34:1831–40. doi: 10.1089/neu.2016.4562
108. Harkema S, Gerasimenko Y, Hodes J, Burdick J, Angeli C, Chen Y, et al. Effect of epidural stimulation of the lumbosacral spinal cord on voluntary movement, standing, and assisted stepping after motor complete paraplegia: a case study. *Lancet*. (2011) 377:1938–47. doi: 10.1016/S0140-6736(11)60547-3
109. Tabakow P, Jarmundowicz W, Czapiga B, Fortuna W, Miedzybrodzki R, Czyz M, et al. Transplantation of autologous olfactory ensheathing cells in complete human spinal cord injury. *Cell Transplant*. (2013) 22:1591–612. doi: 10.3727/096368912X663532
110. Rejc E, Angeli C, Harkema S. Effects of lumbosacral spinal cord epidural stimulation for standing after chronic complete paralysis in humans. *PLoS ONE*. (2015) 10:e0133998. doi: 10.1371/journal.pone.0133998
111. Zewdie ET, Roy FD, Yang JF, Gorassini MA. Facilitation of descending excitatory and spinal inhibitory networks from training of endurance and precision walking in participants with incomplete spinal cord injury. *Prog Brain Res*. (2015) 218:127–55. doi: 10.1016/bs.pbr.2014.12.005
112. Hofstoetter US, McKay WB, Tansey KE, Mayr W, Kern H, Minassian K. Modification of spasticity by transcutaneous spinal cord stimulation in individuals with incomplete spinal cord injury. *J Spinal Cord Med*. (2014) 37:202–11. doi: 10.1179/2045772313Y.0000000149
113. Drum MG. Physical rehabilitation of the canine neurologic patient. *Vet Clin North Am Small Anim Pract*. (2010) 40:181–93. doi: 10.1016/j.cvsm.2009.09.009
114. Gordon-Evans WJ, Johnson AL, Knap KE, Griffon DJ. The effect of body condition on postoperative recovery of dachshunds with intervertebral disc disease treated with postoperative physical rehabilitation. *Vet Surg*. (2019) 48:159–63. doi: 10.1111/vsu.13142
115. Jeong IS, Piao Z, Rahman MM, Kim S, Kim NS. Canine thoracolumbar intervertebral disk herniation and rehabilitation therapy after surgical decompression: A retrospective study. *J Adv Vet Anim Res*. (2019) 6:394–402. doi: 10.5455/javar.2019.f359
116. Zidan N, Sims C, Fenn J, Williams K, Griffith E, Early PJ, et al. A randomized, blinded, prospective clinical trial of postoperative rehabilitation in dogs after surgical decompression of acute thoracolumbar intervertebral disc herniation. *J Vet Intern Med*. (2018) 32:1133–44. doi: 10.1111/jvim.15086
117. Yu P, Zhang W, Liu Y, Sheng C, So KF, Zhou L, et al. The effects and potential mechanisms of locomotor training on improvements of functional recovery after spinal cord injury. *Int Rev Neurobiol*. (2019) 147:199–217. doi: 10.1016/bs.irn.2019.08.003
118. Fang CY, Tsai JL, Li GS, Lien AS, Chang YJ. Effects of robot-assisted gait training in individuals with spinal cord injury: a meta-analysis. *Biomed Res Int*. (2020) 21:2102785. doi: 10.1155/2020/2102785
119. Gallucci A, Dragone L, Menchetti M, Gagliardo T, Pietra M, Cardinali M, et al. Acquisition of involuntary spinal locomotion (spinal walking) in dogs with irreversible thoracolumbar spinal cord lesion: 81 dogs. *J Vet Intern Med*. (2017) 31:492–7. doi: 10.1111/jvim.14651
120. Aravind N, Harvey LA, Glinsky JV. Physiotherapy interventions for increasing muscle strength in people with spinal cord injuries: A systematic review. *Spinal Cord*. (2019) 57:449–60. doi: 10.1038/s41393-019-0242-z

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REVIEW
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Classification of Intervertebral Disc Disease

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Intervertebral disc disease (IVDD) has been recognized in dogs since the 1800s, when the first descriptions of extruded disc material within the vertebral canal were published. In the intervening time our understanding of intervertebral disc pathology in dogs and cats has increased dramatically, with many variations of IVDD described. Whilst the volume of literature and collective understanding of IVDD has expanded, there has also been scope for confusion as the definition of intervertebral disc disease, with its myriad different manifestations, becomes more complicated. A large volume of literature has aimed to combine the use of histopathology, diagnostic imaging and clinical findings to better understand the various ways in which IVDD can be classified. Much of this research has focused on the classification of mechanisms of intervertebral disc degeneration, centering around the differences between, and overlaps in, IVDD in chondrodystrophic and non-chondrodystrophic dog breeds. However, with the increasing availability of advanced imaging modalities allowing more accurate antemortem diagnosis, the concept of IVDD has expanded to include other clinical presentations that may not fit into traditional models of classification of IVDD. This review aims to provide an up to date overview of both historical and current systems of IVDD classification, highlighting the important findings and controversies underpinning them.

Keywords: IVD, chondrodystrophy, degeneration, Hansen, extrusion, protrusion

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INTRODUCTION

Intervertebral disc disease (IVDD) is a broad term that is widely used in veterinary medicine and encompasses a range of lesions affecting the intervertebral disc (**Table 1**). Since the first descriptions of IVDD in the dog by Dexler in the late 1800s (1, 2), advances in understanding of the underlying etiology have resulted in a gradual evolution in terminology and systems of classification. Initial reports described the presence of cartilaginous material within the epidural space of the vertebral canal (so-called *enchondrosis intervertebralis*), which was subsequently found to be associated with degenerated nucleus pulposus (1, 3). Building on these findings in the 1940s and 50s, Hansen and Olsson made huge advances in understanding the nature of canine IVDD, proposing a system of classification based on histopathological degenerative changes that persists today (2, 4–7). They described two distinct types of IVD degeneration, namely chondroid and fibroid metaplasia, associated with specific breed and signalment signatures. This introduced the classification of dog breeds according to the type of IVDD that was more prevalent, into chondrodystrophic (chondroid metaplasia) and non-chondrodystrophic (fibroid metaplasia) breeds (5, 8). These insights led to the subsequent classification of IVDD into Hansen Type I and Hansen Type II herniations, a system that is still widely used in veterinary medicine (4, 5, 9). At the time of this initial work,

TABLE 1 | Glossary of words commonly used in the description of intervertebral disc disease and suggested definitions.

Term used	Proposed definition
Intervertebral disc (IVD) disease	Very broad, non-specific term suggesting clinical IVD herniation, subclinical IVD herniation, or IVD degeneration without herniation
IVD herniation/ prolapse/displacement	Non-specific descriptors that can be used to describe any form of IVDD that results in loss of structural integrity with part of the IVD displaced beyond its normal boundaries—typically into the vertebral canal
IVD extrusion	Used to describe the herniation/prolapse/displacement of internal contents (predominantly nucleus pulposus) through the annulus fibrosus. Can be associated with varying degrees of degenerative change, hydration, or trauma
IVD protrusion	Used to describe the prolapse/herniation/displacement of the annulus fibrosus beyond its normal boundaries Typically associated with fibroid metaplasia (often referred to as Hansen Type II IVD herniation)

Hansen and others used various terms to describe the subsequent displacement of IVD material into the vertebral canal, with "protrusion," "extrusion," "prolapse" and "herniation" often used interchangeably (2, 5). Indeed, Olsson stated as much in 1951, reporting that "Disc protrusion [is] a synonym for disc herniation and disc prolapse" (2). Since then, the term "IVD extrusion" has become more widely associated with the acute extrusion of nucleus pulposus from an IVD that has features of chondroid metaplasia, whilst "IVD protrusion" has largely been reserved chronic annulus fibrosus thickening originally associated with fibroid metaplasia. In contrast, the term "IVD herniation" continues to be largely used as an umbrella term without specificity to a particular type of degenerative change. For the purposes of this report, in consistency with recent veterinary and human literature (10, 11), we have therefore used IVD herniation as a non-specific term to encompass any type of localized IVD displacement. This basic terminology is outlined in **Table 1**.

In recent years further advances in the understanding of IVDD have been made, building on the seminal early work of Hansen and Olsson. For example, the use of detailed histological grading systems has suggested that both chondrodystrophic and non-chondrodystrophic breeds of dog undergo chondroid metaplasia, albeit at very different rates and times (9, 12–14). Our understanding of the basis of early chondroid metaplasia in chondrodystrophic breeds has also improved dramatically with discovery of expression of a fibroblast growth factor 4 (*FGF4*) retrogene on chromosome 12 as a strong risk factor for IVD extrusion (15–17). In addition to these advances in histological and genetic descriptions of IVDD, advances in diagnostic techniques such as magnetic resonance imaging (MRI) have contributed to the expansion of the group of conditions that might reasonably be referred to as types of IVDD in dogs and cats. The improved diagnostic ability provided by these developments has led to the increasingly frequent diagnosis of IVD conditions that occur with minimal evidence of traditional features of disc degeneration, including the herniation of relatively well-hydrated nucleus pulposus material (18–20). This has created a degree of confusion, as numerous reports of similar clinical presentations have appeared in the literature under a variety of terminology (**Table 2**). This is perhaps best exemplified by the condition variably termed Hansen Type III IVD herniation, traumatic IVD extrusion and high-velocity low-volume IVD extrusion among others, before recently becoming more uniformly accepted as acute non-compressive nucleus pulposus extrusion (ANNPE) (18, 74–76). Whilst these terms are all used to indicate a peracute non-compressive extrusion of nucleus pulposus, traumatic IVD extrusion has also been applied to cases of compressive extrusion of degenerative nucleus pulposus following vertebral column trauma (77). Alongside ANNPE, further conditions that can be considered as types of IVDD have also appeared with increasing frequency in the literature, such as intradural or intramedullary disc extrusions (IIVDE) and acute compressive hydrated nucleus pulposus extrusions (HNPE) (20, 88, 101). In this article we have utilized updated and consistent terminology for these conditions that builds on the traditional two-type IVDD classification model.

As genetic investigations continue and advanced imaging techniques improve, it is likely that this terminology will evolve further. As a result, it is important to periodically review recent and historical literature surrounding IVDD classification with the aim of developing intuitive terminology that reduces confusion and allows a better understanding of the underlying etiology. Specifically, a better understanding of disease classification and terminology is vital in order to facilitate effective research efforts, allowing optimal study design and case selection for clinical, molecular and histological investigations aimed at ultimately improving patient welfare and treatment outcomes.

Whilst there are still many controversies in the categorization of types of disc-associated lesions in veterinary medicine, in this article we aim to review the relevant literature and to outline a consensus where possible. We will provide an up to date overview of reported types of IVDD, highlighting important features, and aiming to explain the theories behind their classification.

ANATOMY OF THE INTERVERTEBRAL DISC

The intervertebral disc plays a critical role in stability of the vertebral column, effectively binding individual vertebrae together to provide support for the entire axial skeleton while allowing multiplanar movement. Additionally, it protects the spinal cord and facilitates the exit/entrance of peripheral nerves. Embryologically it derives from the mesoderm with the exception of the nucleus pulposus, which is a remnant of the notochord (104, 105). The intervertebral disc has 4 regions, all of which contribute to its complex role; the nucleus pulposus, the

TABLE 2 | Terminology used in the veterinary literature to describe different types of intervertebral disc (IVD) disease.

Proposed/current consensus	Examples of reported terms used	References
(Hansen Type I/Acute) IVD extrusion	Hansen Type I IVD herniation	(9, 21–24)
	Hansen Type I IVD extrusion	(25–31)
	Hansen Type I IVD disease	(32–36)
	(Acute-) IVD herniation	(24, 37–46)
	(Acute-) IVD extrusion	(33, 34, 41, 47–53)
	(Acute-) IVD disease	(54–58)
	IVD protrusion	(54, 59, 60)
	Type I protrusion/prolapse	(5)
(Hansen Type II/Chronic) IVD protrusion	Hansen Type II IVD herniation	(9, 22, 23, 61)
	Hansen Type II IVD protrusion	(29, 61, 62)
	Hansen Type II IVD disease	(62)
	IVD protrusion	(23, 48, 51, 61, 63–66)
	Type II protrusion/prolapse	(5)
Acute IVD extrusion (Hansen Type I) with extensive epidural hemorrhage	Acute IVDE with extensive epidural hemorrhage	(37, 67)
	Disc extrusion with extensive epidural hemorrhage (DEEH)	(67)
	Epidural spinal hematoma and IVD extrusion	(68)
Acute non-compressive nucleus pulposus extrusion (ANNPE)	Acute non-compressive nucleus pulposus extrusion (ANNPE)	(18, 35, 52, 69–73)
	Hansen Type III IVDD	(74)
	High-velocity, low-volume IVD extrusion	(75)
	Traumatic IVD extrusion	(76, 77)
	IVD explosion	(78)
	Traumatic IVD prolapse	(5)
	Missile disks	(79, 80)
Hydrated nucleus pulposus extrusion (HNPE)	Hydrated nucleus pulposus extrusion (HNPE)	(19, 20, 81, 82)
	Hydrated nucleus pulposus herniation	(83)
	Acute compressive hydrated nucleus pulposus extrusion	(84)
	Partially degenerated disc extrusion	(85)
	Intraspinal cyst	(86, 87)
	Canine discal cyst	(86)
Intradural/intramedullary IVD extrusion (IIVDE)	Intradural/intramedullary IVD extrusion (IIVDE)	(88)
	Intradural IVD herniation	(72, 89, 90)
	Intramedullary IVD herniation	(91)
	Intramedullary IVD extrusion	(92, 93)
Traumatic IVD extrusion	Traumatic IVD extrusion	(76, 77)
	Traumatic IVD prolapse	(5)
Fibrocartilaginous embolic myelopathy (FCEM)	Fibrocartilaginous embolic myelopathy (FCEM)	(79, 88, 94, 95)
	Fibrocartilaginous embolism (FCE)	(96–99)
	Ischaemic myelopathy	(69, 77, 96, 100–102)
	Spinal cord infarction	(103)

transitional zone, the annulus fibrosus and the cartilaginous endplates (22).

The Nucleus Pulposus

The nucleus pulposus is a gelatinous, bean shaped mass that sits slightly dorsally within the intervertebral disc (**Supplementary Figure 1**). While bounded on all sides by the transitional zone, cranially and caudally it lies close to the cartilaginous endplates. It is derived from the notochord and is characterized by an extremely high-water content, up to 88% in the young animal. Islands of physaliferous notochordal cells can be seen within the ground substance in the young animal and are gradually replaced by chondrocyte like cells with age (5, 22, 106). Notochordal cells produce low levels of type 2 collagen and proteoglycans. The proteoglycans consist of a protein backbone onto which are attached glycosaminoglycans. The most common glycosaminoglycan side chains include chondroitin-6-sulfate and keratan sulfate. These molecules are highly polarized, creating space within the proteoglycan units. The resulting proteoglycans are then aggregated by hyaluronic acid, creating extremely large, highly charged complexes that exert a high osmotic pressure, retaining water within the nucleus (107). Type 2 collagen can

interact with carbohydrates, and thus the network of type 2 collagen found within the nucleus pulposus is associated with the glycosaminoglycan side chains, lending stability in the face of compressive forces.

The Transitional Zone

This zone represents the transition from nucleus pulposus to the annulus fibrosus. Chondrocyte like cells can be seen as well as increasing numbers of fibrocyte like cells moving peripherally away from the nucleus pulposus (22, 108). These cells lie within a fibrous matrix that appears distinct from the more basophilic matrix of the nucleus pulposus. As the transitional zone blends into the annulus fibrosus the fibrous matrix becomes organized into a lamellar orientation.

The Annulus Fibrosus

The annulus fibrosus consists of inner and outer regions, that are both characterized by concentric fibrocartilage lamellae. These lamellae have elongated fibrocytes interspersed between well-organized bundles of collagen with a less well-organized network of elastin fibers found throughout (22, 108). The collagen in turn has a proteoglycan coating and the healthy annulus fibrosus is 60% water as a result. The regions are differentiated by the presence of chondrocytes in the inner annulus fibrosus and the presence of increasing amounts of type 1 collagen in the outer annulus fibrosus (109). The inner annulus fibrosus is anchored to the cartilaginous endplates and the outer annulus fibrosus anchored to the epiphyseal bone of the adjacent vertebrae by Sharpey's fibers. None of the regions discussed thus far have a blood supply but there is light innervation of the outer annulus fibrosus (9, 104).

The Cartilaginous Endplates

The cartilaginous endplates firmly anchor the intervertebral disc to the adjacent vertebra and provide additional shock absorbing functionality. The endplates have ∼5 layers of chondrocytes and contribute ∼6% of the total width of the intervertebral disc (22). They lie immediately adjacent to a rich vascular network from the epiphyseal arterial supply from which nutrients gain access to the IVD. This occurs by osmosis, diffusion and for larger molecules, the central concave regions of the endplates have channels that allow passage of nutrients via bulk flow in response to loading of the disc (22, 104, 110).

INTERVERTEBRAL DISC DEGENERATION

Intervertebral disc degeneration underlies the most common forms of IVD herniation and as such is an extremely important process to understand. Degeneration is effectively an aging process that is heavily influenced by canine genetics and accelerated by biomechanical strain and trauma among other things (9, 110, 111). Overall the process involves replacement of notochordal cells within the nucleus pulposus by chondrocytes with transformation to fibrocartilage (chondroid metaplasia) (5, 14). This is associated with loss of proteoglycans, in particular chondroitin sulfate, and dehydration (12, 107, 111, 112). The resulting biomechanical failure of the intervertebral disc unit is associated with fissuring of the annulus fibrosus and sclerosis of the endplates (5, 12, 110). Throughout this process, the collagen content increases and more type 1 collagen is found toward the center of the disc (**Supplementary Figure 1**) (8, 12, 111). Complete failure of this unit occurs with IVD herniation.

TYPES OF INTERVERTEBRAL DISC HERNIATION

To be consistent with the majority of veterinary and human literature, we are using the term IVD herniation here to describe any form of IVDD that involves the localized displacement of part of the IVD, typically into the vertebral canal (**Table 1**). The following are summaries of the key pathological, clinical and diagnostic features that can be used to discriminate between these types of IVD herniation.

(Hansen Type I/Acute) Intervertebral Disc Extrusion

Historically most often referred to as Hansen Type I IVD disease, herniation or extrusion, this is the most common cause of spinal cord injury in dogs (113, 114). Recently this type of herniation has widely been referred to simply as IVD extrusion, often with the prefix of "acute" applied to indicate the typical clinical presentation and to discriminate from more chronic manifestations of IVD extrusion. The term "extrusion" in this context is defined in **Table 1**. As a result, in this report we have used this term to describe cases with IVD herniation due to chondroid metaplasia and calcification of the disc, with other reported terms listed for reference in **Table 2**.

Pathophysiology

Early studies into canine IVDD described a characteristic chondroid degeneration of the IVD that was particularly prevalent in certain dog breeds, such as French Bulldogs, Dachshunds and Pekingese (5). These dog breeds all have features of altered endochondral ossification with shortened long bones and along with others such as Beagles, Basset Hounds, Cocker Spaniels, and Pembroke Welsh Corgis, have become known as chondrodystrophic breeds. The degenerative change seen in the IVD is characterized by an early onset of progressive dehydration and calcification, with the normally hydrated and notochordal cell-rich gelatinous nucleus pulposus transforming to a dense, dehydrated cartilaginous matrix rich in chondrocyte-like cells by 1 year of age (5, 9). As an extension of this process, the nucleus pulposus becomes calcified and can be identified clearly on spinal radiographs. This is thought to represent dystrophic calcification of necrotic tissue and occurs predominantly but not exclusively in chondrodystrophic breeds of dog. In breeds such as the dachshund, the number of IVDs with calcified nuclei peaks between 24 and 27 months of age and then decreases (115–117). This chondroid metaplasia and calcification ultimately results in a type of IVD herniation whereby the calcified nucleus pulposus acutely extrudes through a ruptured annulus fibrosus into the vertebral canal (**Supplementary Figure 2**) (5). This eventual sudden extrusion of degenerative nucleus pulposus into

the vertebral canal is a consequence of focal degenerative changes within the annulus fibrosus, hypothesized to be the result of altered biomechanics of the intervertebral disc unit, resulting in separation of the lamellae within the (particularly dorsal) annulus (5, 9, 12). The presence of calcified nucleus pulposus on radiographs is now well-established as an indication that chondroid IVD degeneration has occurred and that there is an increased risk of acute IVD extrusion at that site (115). While overwhelmingly recognized in chondrodystrophic breeds, it is important to note that calcified disks can occur in large breeds, frequently at a single site (5, 118). However, it is unclear whether such disks underwent the extremely early chondroid metaplasia seen in chondrodystrophic breeds (62).

Several advances have since been made in the understanding of the pathophysiology of IVD extrusion, explaining the dramatic early chondroid metaplasia and calcification that occurs in chondrodystrophic breeds. The most important discovery in recent years has been the identification first of a locus on chromosome 12 associated with disc calcification in Dachshunds (119), and subsequently the identification of an expressed *FGF4* retrogene at that locus associated with IVD extrusion in chondrodystrophic dogs (16, 17). Breeds with the most extreme short-limbed body conformation, such as Dachshunds, also carry an *FGF4* retrogene insertion on chromosome 18 that is associated with chondrodysplasia, the condition that produces extremely short limbs (15). The previously documented association between IVD extrusion and body conformation in Dachshunds (120), likely reflects both the influence of the *FGF4* retrogene and the importance of biomechanics in these breeds. It has also been suggested that lifestyle factors such as moderate intensity exercise and stair climbing may be associated with a reduced rate of IVD calcification in Dachshunds (121). The genetic investigations and discoveries related to canine IVDD are covered in more detail in the article in this series by Dickinson and Bannasch. These findings explain the extremely early chondroid metaplasia that occurs in chondrodystrophic breeds, but do not explain the single calcified disks that can be identified in large breed dogs (14, 15, 62, 118, 122).

When using the term Hansen Type I IVD extrusion, we are referring to an acute extrusion of degenerative nucleus pulposus, with features of dehydration and cartilaginous calcification, whilst acknowledging that our current understanding of the underlying pathophysiology might be incomplete. Although acute herniations of material from an IVD that has undergone predominantly fibroid degeneration can occur, this is much less common. When the etiology is unclear, the term IVD herniation can be used without specifying the known or presumed underlying pathophysiology (**Table 1**).

Clinical Presentation and Diagnosis

Young to middle-aged chondrodystrophic dog breeds are most commonly affected by IVD extrusion, although as outlined above, they can also occur in non-chondrodystrophic dog breeds (62, 118, 122). Whilst much less frequently reported and less well-described, IVD extrusions with similar features have also been reported in cats (82, 123). The extruded material causes a variable degree of spinal cord contusion and compression, as well as compression of nerve roots and inflammation (124). An extrusion can occur anywhere along the vertebral column, with an increased incidence of IVD extrusion between the T11-12 and L2-3 IVDs (41). Clinical signs reflect the location of the extrusion along the vertebral canal and can range from mild discomfort with no neurological deficits to paralysis of the affected limbs with loss of pain perception. The typical clinical presentation is therefore an acute onset, painful and progressive myelopathy.

Diagnosis of IVD extrusion has evolved markedly over time and is now mostly commonly achieved using either computed tomography (CT) or MRI (**Supplementary Figure 2**). Both imaging modalities have been shown to be superior to techniques such as plain radiographs and myelography (**Supplementary Figure 2**) in diagnosing and localizing IVD extrusions (35, 37, 125). Whilst both MRI and CT can be used to diagnose IVD extrusion, MRI has the benefit of allowing evaluation of soft tissues such as the spinal cord and intervertebral disks (39, 126). The reader is directed to the paper by Da Costa and others in this series for further details on diagnostic imaging in IVDD.

The following is a sub-classification of IVD extrusion, where similar pathological changes of chondroid metaplasia and calcification are likely to underpin the extrusion but result in a different clinical presentation.

Acute Intervertebral Disc Extrusion With Extensive Epidural Hemorrhage

Acute thoracolumbar IVD extrusions can cause multilevel epidural hemorrhage due to laceration of the internal vertebral venous plexus (37, 67, 127, 128). Sometimes this hemorrhage can be dramatic and can cause multilevel spinal cord compression, and indeed appear as a hematoma (67, 68, 127, 128). The term disc extrusion with extensive epidural hemorrhage (DEEH) has been coined to describe this particular phenomenon (67).

Pathophysiology

The affected intervertebral disc undergoes chondroid degenerative changes and calcified nuclear material is extruded into the vertebral canal causing a laceration of the internal vertebral venous plexus and consequent hemorrhage. This phenomenon has been reported in the thoracolumbar spine but not in the cervical spine. The factors that cause ongoing hemorrhage to occur in affected dogs have not been well-defined but likely relate to the relative volume of epidural space. This event is more common in medium to large breed dogs than small chondrodystrophic breeds, leading to speculation that epidural volume is larger in these breeds and thus the vertebral venous plexus is not compressed enough by the extruded material to stop hemorrhage when lacerated. Another possibility is that there is a relatively larger volume of calcified material herniated in chondrodystrophic breeds, effectively compressing the venous plexus to halt hemorrhage.

Clinical Presentation and Diagnosis

Typically, affected dogs develop acute paraparesis that rapidly progresses to paraplegia, often associated with severe spinal pain (67). Medium to large sized breeds of dog such as

the pit bull terrier, American Staffordshire terrier, Labrador retriever, German Shepherd dog and Rottweiler are affected most commonly, but it can occur in small breed and giant breed dogs as well (52, 67, 68, 128). Survey spinal radiographs might reveal calcified nuclear material and narrowing of an intervertebral disc space. Diagnosis is by advanced imaging with classic findings on MRI including evidence of calcified disc material fragments (T2 and T1-weighted hypointense) and multilevel extradural compression by a mass that is hyperintense or has mixed intensity on T2-weighted imaging and is hyper, iso or hypointense on T1-weighted imaging with variable degrees of peripheral contrast enhancement. Gradient echo (T2*) imaging confirms the presence of hemorrhage. The mass often appears like a worm extending along the spinal cord on the sagittal T2-weighted image and gives the appearance of a second spinal cord lying alongside or draped over and compressing the real spinal cord on transverse images. On CT imaging the hemorrhage/hematoma is identified as a moderately hyperattenuating [70–90 Hounsfield units [HU]] extradural mass extending over multiple vertebral levels and lying dorsal, ventral and lateral to the spinal cord. Fragments of calcified disc material identified by higher HU (>100) can be found within the extradural mass, frequently focused over an intervertebral disc space (**Supplementary Figure 3**).

(Hansen Type II/Chronic) Intervertebral Disc Protrusion

Traditionally this type of IVD herniation has been referred to as either Hansen Type II IVD disease, herniation or protrusion (**Table 2**). However, in recent veterinary literature it has been increasingly referred to as simply IVD protrusion, which is the terminology we have used here. The key pathological and clinical features that characterize this type of IVD herniation are outlined below, as well as the areas requiring further investigation.

Pathophysiology

In his investigations into IVDD in dogs in the 1950s, Hansen found that dogs could be grouped according to the type of disc degeneration seen most frequently, into chondrodystrophic and non-chondrodystrophic dog breeds (5). In non-chondrodystrophic dogs, with increasing age, Hansen reported that the disc underwent a slow maturation, whereby the collagen content increased and notochordal cells became more fibrocyte-like, a process termed fibroid metaplasia (5). This fibroid metaplasia typically occurred in non-chondrodystrophic dog breeds over 7 years of age, suggesting that this type of IVD degeneration represents a consequence of later onset age-related changes in comparison to chondrodystrophic dogs (5, 110). At the same time as the nuclear degeneration, it has been postulated that small separations develop in the lamellae of the annulus fibrosus, potentially exacerbated by repeated minor trauma, allowing this degenerative fibroid nuclear material to extend into and between the fibers of the annulus (5, 9, 110). The result was a gradual, discrete thickening and protrusion of the surface of the annulus fibrosus, typically occurring dorsally into the vertebral canal, displacing the dorsal longitudinal ligament and slowly compressing the spinal cord (**Supplementary Figure 4**) (5, 9, 22, 110).

Based on Hansen's description, this type of herniation of the IVD into the vertebral canal became widely referred to as Hansen Type II IVD disease or herniation. However, the distinction between the degenerative processes leading to IVD herniation in the IVD of chondrodystrophic (Hansen Type I) and non-chondrodystrophic (Hansen Type II) dog breeds has been revisited recently. Histopathological comparisons between the IVD of chondrodystrophic and non-chondrodystrophic dog breeds have found features of chondroid metaplasia (chondrification and replacement of notochordal cells by chondrocytes within the nucleus pulposus) in both groups of dogs (13, 14). Investigators have been careful to point out that Hansen's original descriptions had referred to overall degree of fibrosis of the disc, not fibroid metaplasia of the nucleus pulposus specifically, which perhaps had been somewhat misunderstood for many years (14).

IVD protrusion is also often seen in association with other degenerative changes in certain complex disorders of the canine vertebral column, such as disc-associated cervical spondylomyelopathy and degenerative lumbosacral stenosis (71, 129). Whilst a complete discussion of these disorders is beyond the scope of this series, it is likely that nuclear degeneration and annular protrusion occurs as part of a multifactorial etiology in these dogs.

Clinical Presentation and Diagnosis

The clinical presentation of dogs with IVD protrusion is dependent on the location of the affected IVD and the degree of associated compression of relevant structures, such as the spinal cord and nerve roots. Clinical signs tend to reflect the chronic, slowly progressive nature of the IVD degeneration, typically with milder neurological deficits than those seen with acute spinal cord injury secondary to IVD extrusions (63). The characteristic clinical picture is therefore that of a slowly progressive, often non-painful myelopathy in an older, usually non-chondrodystrophic dog (63, 64). Pain can be present depending on the presence of nerve root compression but is less common than in the more acute IVD extrusion.

Changes associated with IVD protrusion on plain radiographs include non-specific signs of IVD degeneration such as vertebral endplate sclerosis, spondylosis deformans and IVD space narrowing (63). Although historically diagnosed using myelography, this has largely been superseded by advanced imaging techniques, particularly MRI in the case of IVD protrusion (**Supplementary Figure 4**) (23, 130). Whilst a specific diagnosis of IVD protrusion can be challenging, MRI criteria have also been reported to assist in differentiating between IVD protrusion and IVD extrusion in dogs (23, 51). Recent studies have also demonstrated the utility of MRI in grading the degree of IVD degeneration using a validated MRI grading scheme, however the grade of degenerative changes seen in IVD protrusions and extrusions were similar (13, 131). Further details regarding diagnostic imaging in IVD protrusions can be found in the paper by Da Costa and others in this series.

Hydrated Nucleus Pulposus Extrusion (HNPE)

A relatively recent addition to the classification of disorders of the IVD in the veterinary literature has been the description of acute compressive HNPE in dogs (20). This term refers to a subtype of acute herniation of a volume of partially or non-degenerate nucleus pulposus that results in a varying degree of extradural spinal cord compression (20). Whilst the condition has been referred to as "acute compressive HNPE" to signify this compression (84), it is typically referred to by the shortened initialism of HNPE (**Table 2**). As with ANNPE, there has been variation in the terminology used to describe this presentation, with initial reports of dogs with similar clinical and diagnostic imaging findings using terms "intraspinal cyst" and "canine discal cyst" (86, 87, 132). There has also been a suggestion to abandon either these terms or HNPE in favor of "partially degenerated disc extrusions" on the basis of histological and cytological findings (85), but most recent literature is consistent in using the terminology of HNPE for this condition (**Table 2**) (81, 84, 133, 134).

Pathophysiology

Reports of acute compressive HNPE in dogs typically describe the presence of well-hydrated extradural material overlying an IVD, suggesting a communication with the annulus fibrosus, as well as associated spinal cord compression (20, 86). The previous use of terms such as intraspinal or discal cysts in dogs has its origin in the observation of MRI similarities with human intraspinal discal cysts (86). People with discal cysts most often present with characteristic clinical signs of a chronic and painful radiculopathy, often affecting the lumbar region (135). Human discal cysts are also typically associated with histological evidence of a well-defined cyst wall and the contents are of a serous or serosanguinous nature (135). In contrast, histological and cytological examination of the extradural material in dogs reveals partially degenerated nucleus pulposus (**Supplementary Figure 5**), whilst a convincing cyst wall has not been consistently identified (81, 83, 133). Furthermore, diagnostic imaging features and microsurgical findings in dogs with HNPE suggest that the extradural material may persist ventral to, or within the dorsal longitudinal ligament (20, 81). Given that dogs with presumed HNPE present with a more acute onset of typically non-painful clinical signs, in combination these findings support the suggestion that the underlying pathophysiology in these dogs represents an acute herniation of hydrated nucleus pulposus (20, 81, 134). However, the extradural material can demonstrate a varying degree of fluidity, detectable on MRI using FLAIR or HASTE sequences, and a good explanation of this range of findings has not yet been forthcoming (**Supplementary Figure 5**).

The exact mechanisms that lead to this herniation have not been established, but there may be similarities with ANNPE whereby a small tear in the annulus fibrosus occurs following acute changes in intradiscal pressure (81, 133). The differences in clinical presentation of HNPE compared to ANNPE, as well as the reported predisposition for the cervical region suggest that this understanding of the underlying pathophysiology is incomplete and worthy of further investigation.

Clinical Presentation and Diagnosis

The vast majority of reported cases of canine HNPE in the veterinary literature have occurred in the cervical vertebral column, suggesting an anatomical predisposition reflected in the typical clinical signs (19, 20, 83, 136). Dogs therefore most often present with an acute onset of tetraparesis or tetraplegia, with symmetrical clinical signs more common in contrast to the lateralisation seen with ANNPE (20). Another characteristic finding reported in dogs with HNPE has been a lack of spinal hyperaesthesia in the majority of cases, as well as more severe neurological deficits in contrast to Hansen Type I IVD extrusion (19, 20, 81, 83, 84). Indeed, tetraplegia with respiratory compromise is not unusual in cervical HNPE (20). No specific breed predisposition has been identified, with both chondrodystrophic or non-chondrodystrophic breeds reported, whilst affected dogs are typically middle-aged or older (19, 20, 136). In most cases the acute onset seen in HNPE appears to occur spontaneously, without inciting causes such as intense exercise or trauma, further differentiating the clinical presentation from that of ANNPE.

Whilst the clinical presentation may provide a high index of suspicion for a diagnosis of HNPE, differentials usually include IVD extrusion, ANNPE and fibrocartilaginous embolic myelopathy (FCEM). Advanced imaging can be used to make a diagnosis of HNPE, with MRI the imaging modality of choice (**Supplementary Figure 5**). Reported characteristic MRI features consistent with HNPE are listed below (20):

- Ventral, midline extradural material (T2-weighted hyperintense, T1-weighted hypointense) overlying an IVD
- Associated spinal cord compression, with or without intramedullary T2-weighted hyperintensity
- Characteristic bi-lobed "seagull" shaped appearance to the extradural material
- Reduced volume of T2-weighted hyperintense nucleus pulposus signal in the affected IVD.

Although most reports of canine HNPE use these MRI features, a recent study has suggested that contrast-enhanced CT can also be used to make a diagnosis of HNPE with a sensitivity of 91% and specificity of 100% (136).

Acute Non-compressive Nucleus Pulposus Extrusion (ANNPE)

There has been considerable variation in the terminology used historically to describe this condition, including: traumatic IVD extrusion, IVD explosion, traumatic IVD prolapse, missile disks, high-velocity low-volume disc extrusion and (Hansen) Type III IVDD (18, 75–78). In most of these reports, clinical and diagnostic imaging descriptions suggest a peracute onset extrusion of non-degenerated nucleus pulposus leading to spinal cord contusion with minimal compression, usually at exercise, with or without evidence of trauma. As a result, the term ANNPE has become widely accepted in the veterinary literature as the most descriptive terminology for this condition

and is therefore used here. Whilst most studies in the veterinary literature involve dogs, ANNPE has also been described in several cats with similar clinical and diagnostic features (75, 80, 137).

Pathophysiology

Within the non-degenerate, normal IVD, the nucleus pulposus draws water down a strong osmotic gradient, creating an innately high intradiscal pressure (22). The surrounding annulus fibrosus on the other hand is composed of dense fibrous tissue with a complex lamellar structure, providing structural integrity as well as mobility (22). As a result, when the IVD is subjected to supra-physiological forces such as those exerted on the vertebral column during brief moments of strenuous exertion or blunt trauma, the annulus fibrosus may tear leading to a sudden extrusion of nuclear material (88). It is hypothesized that the hydrated nuclear material impacts the overlying spinal cord with great force, leading to contusive injury before dissipating or being resorbed due to its hydrated nature and small volume, resulting in minimal or no residual spinal cord compression (18). Reported clinical signs and MRI features reflect this, with several studies describing similar characteristic features (**Supplementary Figure 6**) (18, 69, 70). Although reports of histopathological confirmation of ANNPE are rare, post-mortem findings have revealed small tears in the dorsal annulus fibrosus in affected dogs and non-degenerated nucleus pulposus material within the vertebral canal, supporting this theory (78).

Clinical Presentation and Diagnosis

The characteristic clinical presentation of dogs and cats with ANNPE consists of a peracute onset of signs of myelopathy (ranging from paresis to plegia), usually occurring at strenuous exercise or related to external trauma (18, 69, 138). Clinical signs are lateralised in up to 90% of affected dogs, and are usually non-progressive after the first 24 h (18, 69, 77). Although there are fewer descriptions of feline ANNPE in the literature, more cats have been reported to present with symmetrical clinical signs compared to dogs, and up to 75% of cats with ANNPE present following external trauma (137). Whilst owners may report vocalization at the onset of signs in dogs, clinical examination usually reveals only mild to no spinal hyperaesthesia on palpation (18, 69). In accordance with findings in Hansen Type I IVD extrusion, ANNPE occurs most commonly in the region of the thoracolumbar junction, likely reflecting the increased biomechanical forces at the junction between two stable vertebral segments (18, 69). Whilst clinical signs are usually distinct from the typical presentation seen in compressive IVD extrusions or protrusions described above, an important differential diagnosis for ANNPE in dogs and cats with these clinical signs is an ischaemic myelopathy, most likely reflecting FCEM (69, 88). Spinal MRI is usually required to make a presumptive diagnosis and can be used to differentiate cases of ANNPE from FCEM using specific imaging criteria (88, 139). The imaging findings that can be used to make a diagnosis of ANNPE are listed below (18, 70, 139), with an example shown in **Supplementary Figure 6**:

- Focal intramedullary T2-weighted hyperintensity of the spinal cord
- Spinal cord lesion located overlying an IVD
- Reduced volume of T2-weighted hyperintense nucleus pulposus signal in the affected IVD
- Mild narrowing of the affected IVD in mid-sagittal view
- Small volume extradural material (T2-weighted hyperintense, T1-weighted hypointense) dorsal to the IVD with minimal to no spinal cord compression.

Whilst ANNPE has been reported in association with external trauma in up to 40% of reported cases (18), there are some specific instances of traumatic IVD extrusion that may require separate consideration. As a result, we have proposed an additional classification of traumatic IVD extrusion as a specific subtype of IVD herniation below to reflect these cases that may present with vertebral fractures or luxations, as well as spinal cord compression in some cases.

Traumatic Intervertebral Disc Extrusion

Traumatic IVD extrusion (among other terms, also traumatic IVD "explosion" or "prolapse") has been considered synonymous with ANNPE for some authors, but has also been used to describe IVD extrusion secondary to external trauma (76–78). For the purposes of classification in this article we consider traumatic IVD extrusion as a related subcategory of disorders affecting the IVD. The concept of violent trauma (for example, vehicular trauma) causing a sudden rupture of the annulus fibrosus and subsequent extrusion of disc material into the vertebral canal, regardless of degenerative changes, was highlighted in dogs by Hansen in 1952 (5). At that time Hansen described a case in which "a single extreme violence has ruptured a disc that is not remarkably degenerated," and proposed the term "traumatic disc prolapse" (5).

Although there have been limited descriptions of this clinical presentation since then, one study identified traumatic IVD extrusions in 62% of dogs with spinal trauma, with spinal cord compression seen in 9 (29%) of 31 dogs, without associated fractures or luxations (77). In this study it was suggested that the spinal cord compression, which can further differentiate these cases from the typical characteristics of ANNPE, was related to chondroid degenerative changes of the affected IVD (140). Whilst histopathological findings were not reported to confirm this, histology is typically not available for such cases and MRI features are often supportive. It is important to note that in cases of external trauma, the disc extrusion may or may not be compressive, likely reflecting the underlying pathology in the IVD at the time of trauma in addition to the traumatic event. As a result, consideration of all aspects of the spinal trauma patient, such as signalment (age, chondrodystrophy) and multiple diagnostic imaging modalities may lead to a specific diagnosis of traumatic IVD extrusion (with or without features of IVD degeneration or spinal cord compression) instead of ANNPE in such cases (**Supplementary Figure 7**).

Intradural/Intramedullary Intervertebral Disc Extrusion (IIVDE)

Whilst extruded nucleus pulposus material remains in the extradural space in the case of IVD extrusion, HNPE and ANNPE, there have also been reports of nuclear material penetrating the dura mater (72, 88, 89, 91, 93, 101, 141, 142). The extruded material in this scenario can subsequently remain extramedullary but within the intradural space, or enter the spinal cord parenchyma itself, becoming intramedullary. A recent review article used the term intradural/intramedullary IVD extrusion (IIVDE) to collectively describe these cases, including both cases with and without features of nucleus pulposus degeneration in that classification (88).

IIVDE is an uncommon diagnosis with the largest case series in veterinary literature consisting of 8 dogs, estimated to represent 0.5% of the surgical thoracolumbar IVD extrusion caseload in that study (89). Although most of the reported cases have occurred in the region of the thoracolumbar junction, as with ANNPE and Hansen Type I IVD extrusion, IIVDE has also been described in the cervical vertebral column in dogs and the lumbar region in the cat (93, 141, 143).

Similar to many other reports of IIVDE in dogs (90, 92, 142), that case series describes features of chondroid metaplasia on histological examination of surgically excised nucleus pulposus, suggesting an intradural or intramedullary extrusion of degenerative IVD material and subsequent spinal cord compression (89). However, there are also reports of IIVDE without convincing evidence of advanced degeneration on histological examination, as well as a clinical presentation more fitting with ANNPE, suggestive for IIVDE of non- or partially degenerated nucleus pulposus (72, 88, 101). Some of these cases were originally described as tearing, rupture or laceration of the dura, based on myelography and surgical visualization, with no IVD material identified within the vertebral canal (101). All of these dogs presented with a peracute onset of signs during strenuous activity such as running or jumping, or trauma, typically with an improving clinical course (101, 143–146).

As a result of the similarities in presentation of dogs with IIVDE and either Hansen Type I IVD extrusion, traumatic IVD extusion or ANNPE, antemortem diagnosis is dependent on accurate interpretation of diagnostic imaging, or surgical confirmation of intradural or intramedullary disc material. In most initial reports of IIVDE, myelography was shown to demonstrate either the presence of intramedullary contrast material, a "golf-tee" sign suggestive for intradural-extramedullary material or contrast leakage indicating a dural tear at the site of the presumed IVD extrusion (92, 101). More recent reports suggest that particular imaging characteristics on high-field MRI may be suggestive for a diagnosis of IIVDE, including areas of intramedullary hypointensity on T2-weighted, T1-weighted and gradient echo (T2*) sequences overlying an IVD with reduced nucleus pulposus volume, and a linear tract running from the associated IVD to the spinal cord parenchyma (**Supplementary Figure 8**) (88, 91, 93, 141). A recent case series also suggested that CT-myelography was superior to low-field MRI at diagnosing IIVDE, by allowing the accurate detection of a focal filling defect within the subarachnoid space in cases of intradural-extramedullary disc material (89).

Further investigations into outcome following surgical or non-surgical management of these rare cases are necessary, in which case accurate classification according to the presence of degenerative and non-degenerative IVD material may help to determine if subclassifying types of IIVDE is clinically useful.

EMBOLIC DISEASE ASSOCIATED WITH THE INTERVERTEBRAL DISC

Fibrocartilaginous Embolic Myelopathy (FCEM)

Fibrocartilaginous embolism to the vasculature of the leptomeninges and spinal cord was first described in 1973 and since that time, has been a topic of speculation and discussion (103). In the first histopathological descriptions of FCEM, the embolized material was examined closely using several histochemical stains leading to its identification as fibrocartilage (96, 103). Embolization of fibrocartilage can affect the arterial and/or the venous sides of the circulation and results in a peracute onset of often dramatically lateralizing paresis or paralysis (98). The condition occurs most commonly in dogs, accounting for 2% of dogs presenting to an emergency clinic for non-ambulatory paraparesis or paraplegia (114), but it also occurs in a range of other species including the cat (95).

Pathophysiology

Of the questions raised about this condition, the first, and most pertinent to this paper, is the unconfirmed source of the embolized fibrocartilage. Although our understanding of the pathophysiology is incomplete, there is a broad consensus that it originates from the underlying intervertebral disc (79, 98, 103). In the first description of the condition it was noted that the annulus fibrosus of a lumbar disc lying beneath the affected spinal cord segments was torn by extruded nuclear material and regions of this torn annulus had identical staining characteristics to the emboli. The author concluded that the annulus fibrosus was the source of the fibrocartilage (103). Subsequently, other authors have proposed it is nucleus pulposus based on histopathology and histochemistry (147–150). However, the normal appearance of the vertebral column on radiography and the intervertebral disks on gross pathology led some authors to conclude that they were unsure of the source (96). More recently, with the widespread use of MRI to image the vertebral column and spinal cord, it has become clear that there are subtle changes in the nucleus pulposus of an IVD close to (frequently just caudal to) the site of the resulting ischaemic myelopathy, lending support to the nucleus as the source of the disc material (**Supplementary Figure 9**) (95, 139). Indeed, in one study evaluating the use of MRI findings to differentiate between ANNPE and ischaemic myelopathy, a reduced volume of nucleus pulposus in an adjacent IVD was reported in 23.6% of cases with an agreed diagnosis of ischaemic myelopathy (such as FCEM) (139). In addition, the similarity of clinical presentations of

medium sized breeds of dog with exercise associated ANNPE and FCEM suggests that peracute extrusion/embolization of nucleus pulposus underlies both conditions.

The second question relates to how the fibrocartilage travels from the intervertebral disc to the leptomeningeal and spinal cord vasculature. In humans a proposed route is herniation of nucleus pulposus into the endplate (Schmorl's nodes) whereby it gains access into the sinusoidal venous channels within the cancellous bone and thus into the basivertebral veins and the spinal cord venous system (151). However, Schmorl's nodes are rare in dogs due to the density of their endplates and have not been described in conjunction with FCEM. Most authors propose extrusion of nucleus pulposus through either healthy annulus fibrosus directly into the internal vertebral venous plexus or spinal arterial vasculature (148, 150), via new blood vessels forming in annulus fibrosus undergoing age related degenerative changes (150), or both (98). The association between FCEM and particular types of exercise that involve jumping and twisting supports the theory that extreme changes in intra-thoracic or abdominal pressure, and therefore intradiscal pressure, result in extrusion of nuclear material directly into the venous or arterial circulation of the spinal cord. Also pertinent to these theories is the marked paucity of reports of FCEM in chondrodystrophic breeds of dog (152–154). It has been hypothesized that the early chondroid metaplasia and ensuing degeneration and calcification of the nucleus pulposus with splitting of the annulus fibrosus results in ready extrusion of nuclear material into the vertebral canal, rather than creating the forces necessary to propel the material into the vasculature.

However, there are numerous reports of cases in which there is no evidence of degenerative changes within the intervertebral disks (96, 98, 103, 155), and exercise or trauma is only reported as an inciting cause in 30% of cases (79). In particular, these theories are less plausible in Irish wolfhounds that develop FCEM as young as 2 months of age, in which it has been proposed that a mismatch between rapidly increasing body weight and the immature IVD results in embolization into the richer arterial supply to the annulus fibrosus of the growing puppy (155). Other unsubstantiated theories for such cases include persistence of embryonal vasculature within the annulus fibrosus and presence of anomalous vessels or arteriovenous fistulae down which fibrocartilage can be embolized (95).

Important histopathological features of FCEM include the presence of numerous embolized vessels, frequently within the leptomeninges, suggesting a shower of embolic material (96, 98, 103, 147, 148). Indeed, the canine spinal cord is resistant to occlusion of single vessels due to its interconnected segmental arrangement and it has been suggested that FCEM will only result when multiple vessels are occluded simultaneously (96). The resulting lesion is one of hemorrhagic ischemic necrosis of the spinal cord that tends to focus on the gray matter, frequently asymmetrically, reflecting the location of the emboli (96, 103).

Clinical Presentation and Diagnosis

The typical clinical presentation is that of a peracute onset of non-painful lateralizing signs in non-chondrodystrophic breeds. In the majority of cases signs do not progress beyond 24 h (79). Signs can be cervical or thoracolumbar, and age of onset ranges from 8 weeks to 14 years with males slightly more likely to be affected than females (79). Perhaps the most familiar presentation is the peracute onset of signs during vigorous exercise in medium to large sized breeds of adult (4–6 year) dog such as the Labrador retriever and the Staffordshire bull terrier (69, 95, 99). Indeed, these dogs account for approximately half of all cases (79), and in some case series as many as 80% of dogs (99). The signalment and history of these dogs shows considerable overlap with dogs suffering from ANNPE, with distinguishing features including a slight breed predisposition (e.g., Border Collies more predisposed to ANNPE and Staffordshire Bull Terriers to FCEM) (69). In addition, dogs with ANNPE are more likely to vocalize at onset, to have spinal hyperesthesia on examination, and are more likely to have cervical lesions (69).

There are numerous reports of FCEM in adult giant breed dogs such as the Great Dane (147). Of particular interest is an apparent predisposition in Irish wolfhound puppies (155). Known as Puppy Paralysis or Drag Leg Syndrome within the breed, dogs present between 2 and 4 months of age with a lateralizing cervical or thoracolumbar myelopathy. In some of these young dogs, onset appears to be associated with exercise or minor trauma. Histopathology confirmed fibrocartilaginous embolisation of leptomeningeal vessels (155, 156).

Small breed dogs are also of interest, accounting for 24% of all cases (79). Within these small breeds the Miniature Schnauzer accounts for nearly 60% of all cases with male dogs more at risk than female (94). An association with exercise is less clear in this breed and signs can be cervical or thoracolumbar.

Clinical suspicion is raised whenever a dog presents with a peracute onset of non-painful, lateralizing myelopathy. Antemortem diagnosis is confirmed through MRI of the spine (**Supplementary Figure 9**) (100, 139, 157). Diagnostic criteria include:

- Focal intramedullary T2 hyperintensity, focused on the gray matter and frequently lateralized.
- Spinal cord lesion overlying vertebral body, not IVD.
- No evidence of extradural material in the region of the lesion.
- Subtle reduction in volume of T2-weighted hyperintense nucleus pulposus signal in the disc caudal to the spinal cord lesion.

CSF analysis can be normal in ∼50% of cases, with variable and non-specific changes in the remaining cases. It should be noted that the presence of fibrocartilage within the vasculature can only be identified post-mortem, so the imaging diagnosis is one of ischemic myelopathy. However, based on histopathology, the most common cause of ischemic myelopathy in dogs is FCEM.

DISCUSSION

Uniform disease definitions and systems of classification are vital tools in order to accurately diagnose patients, as well as to consistently document and report conditions in such a way that allows reliable scientific comparisons and future critique of related research. It is likely that the advancement of techniques such as genetic technology ultimately hold the key to more accurate disease categorization, by completing the path

from genotype to phenotype (158). In the meantime, given the introduction of several new conditions related to the IVD to the veterinary literature in recent years, we have aimed to evaluate current systems of classification and terminology used to ensure consistent recording of IVDD types.

The rapidly expanding language used surrounding types of IVDD in the veterinary literature suggests that a consensus on terminology and classification is required to cover multiple types of IVDD, including those falling outside of the traditional Hansen Type I and II system. Whilst there has been a gradual advancement in the terminology used in the veterinary literature to describe these additional types of IVDD, there remains great inconsistency. This limits the ability to perform high quality research or to accurately compare clinical data, as clinicians and researchers are restricted by the lack of clear disease definitions. An improved understanding of terminology can also facilitate clearer communication between colleagues and clients when discussing IVDD. Increasingly, efforts have been made to adapt terminology used in the veterinary literature to include other recently described types of IVDD, as well as drawing from the human medical field with an emphasis on appropriate anatomical descriptions (10). In **Tables 1**, **2** and throughout this report, we have taken a similar approach to using specific and consistent terminology for each condition. We have aimed to provide a point of reference in matters of IVDD classification moving away from the restrictions of the traditional Hansen Type I and II system, as well as providing our interpretation of the meaning associated with terms currently used in the veterinary literature.

Most systems of disease classification in veterinary and human medicine have origins in historical methods used to document population disease and health statistics, and are usually based on pathological systems of categorization (158). As a result, traditional forms of IVDD classification have focused on degenerative IVDD and the differentiation between Hansen Type I and Hansen Type II IVD, using signalment, clinical presentation and histopathological features (5, 12, 159). Although advanced diagnostic imaging techniques such as MRI have added to the ability to characterize IVD degeneration clinically (12, 13, 126, 160, 161), there are limitations to a system of classification based solely on the determination of degrees of IVD degeneration. For example, acute herniations of an IVD that has features of predominantly fibroid degeneration may provide a challenge with this system of classification. Furthermore, considering recent research suggesting that dog breeds traditionally considered non-chondrodystrophic can also demonstrate features of chondroid metaplasia, it is clear that the distinction of IVDD into the traditional Hansen Type I and Type II labels represents an incomplete picture.

Within the field of human neurology there are numerous examples of disorders that were classified based on histopathological and clinical findings that have been reclassified based on undercovering the genetic basis of the disease. These include for example, the hereditary ataxias, which take into

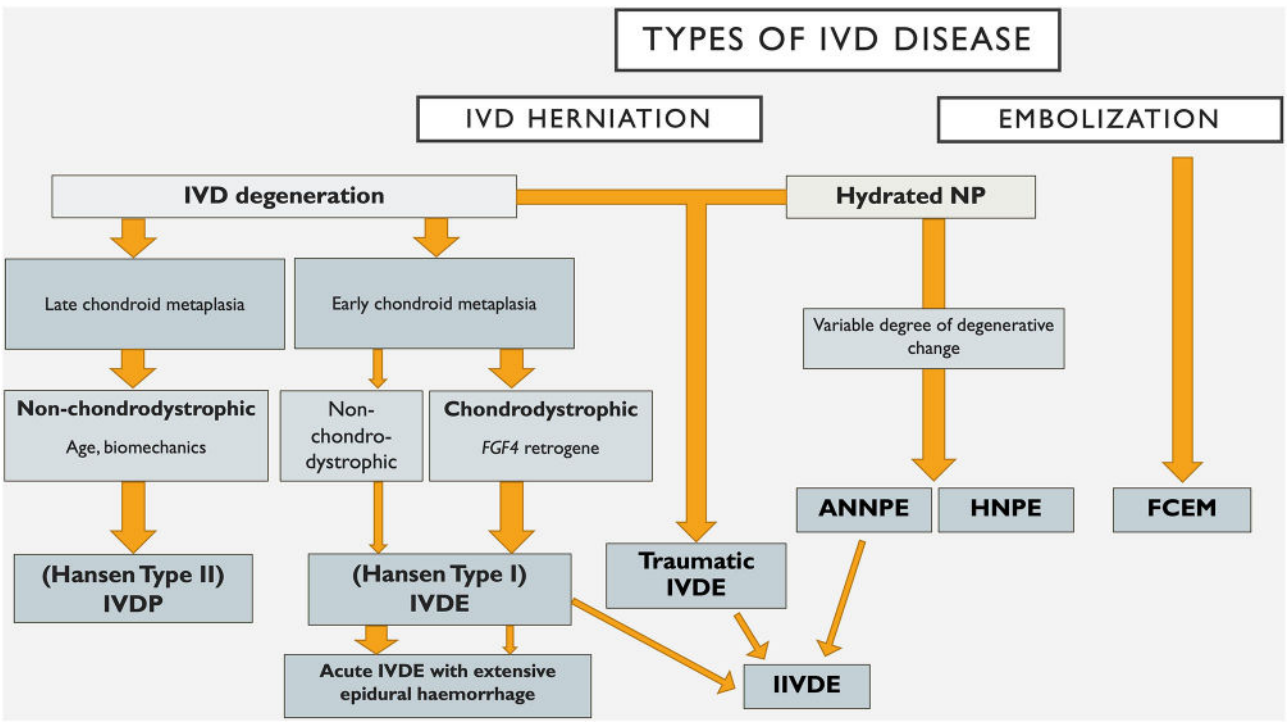

FIGURE 1 | An example of how a classification system for intervertebral disc disease in dogs might evolve. IVD, intervertebral disc; NP, nucleus pulposus; IVDP, intervertebral disc protrusion; IVDE, intervertebral disc extrusion; HNPE, hydrated nucleus pulposus extrusion; ANNPE, acute non-compressive nucleus pulposus extrusion; IIVDE, intradural/intramedullary intervertebral disc extrusion; FCEM, fibrocartilaginous embolic myelopathy.

account the mode of inheritance, the clinical syndrome and the genetic cause (162). Genetic discovery holds similar promise in furthering our understanding of canine IVD degeneration, as evidenced by the identification of the chromosome 12 *FGF4* retrogene as a cause of chondrodystrophy and early, severe chondroid metaplasia with ensuing disc extrusion (15–17). Such advances also enhance our ability to classify IVDD for the purposes of diagnosis, prevention and treatment. An example of how such a system might evolve is provided in **Figure 1**. It is likely that future IVDD classification systems will utilize further genetic findings, such as *FGF4* genotype, to supersede traditional labeling of certain breeds as chondrodystrophic as a means of subcategorising affected dogs. However, the genetic picture is complex and multifactorial (see Dickinson and Bannasch's paper in this series) and in a degenerative disorder such as IVDD, additional environmental influences clearly play a role. The increasing understanding of the complex processes of IVD degeneration, and somewhat contradictory studies on the degree of degenerative changes occurring with types of IVD herniation such as ANNPE and HNPE makes classification based on degenerative processes less clear (9, 14, 81, 85). In the future we anticipate that advanced genetic, diagnostic imaging and histological investigations will build on recent developments and allow subclassification across a wider range of IVD degeneration.

As an alternative to using pathological features of degeneration, we have explored the possibility of using clinical and diagnostic imaging features to classify primary disorders of the IVD. For example, recent human recommendations on lumbar IVDD nomenclature have proposed using morphological characteristics such as the shape of the IVD herniation to distinguish between IVD extrusion and protrusion, regardless of the underlying pathology (11). Another option would be to categorize IVDD primarily into compressive and non-compressive forms, however these classification systems are also open to degrees of interpretation and require separate categories for conditions with unique aetiologic or anatomic features, such as: traumatic IVDE, intradural/intramedullary IVDE (IIVDE) and embolic disease (FCEM). One advantage of such a system would be the incorporation of terminology that ties in with clinical decision-making. For example, traumatic IVD extrusion may necessitate a specific clinical approach that differs from non-traumatic, compressive IVDD, or embolic disease (FCEM). However, by the same note it is important to consider that the treatment for all conditions with spinal cord compression is not necessarily the same. In such cases treatment is typically based on a combination of factors such as severity of clinical signs, owner preference and financial constraints. More specifically, HNPE is often treated medically despite being compressive, largely based on the assumption that the material is well-hydrated and will therefore dissipate over time without intervention (20, 84). It is also important to consider that given the similarity in clinical presentation between some of these conditions, such as ANNPE and FCEM, future research should aim to achieve a better understanding of the correlation between histological, clinical, and imaging features.

Another possible approach is therefore to classify types of IVDD according to their typical clinical presentation. For example, IVD protrusion would be classified as chronic, non-painful and progressive, whereas ANNPE would be classified as peracute, non-painful, and non-progressive. Whilst combinations of clinical features have been shown to be associated with specific forms of IVDD (163), there are still areas of overlap and variations from the typical clinical findings within several of these categories.

As a result, although an ideal system of classification among types of IVDD does not currently exist, consistency across the veterinary literature in the terminology used for each individual form of IVDD would clearly be beneficial. The authors encourage the consistent use of the terms used in the current series of papers clinically and when describing cases in the veterinary literature. Future research efforts should focus on improving our understanding of the underlying pathophysiology of these different types of IVDD, an endeavor which can be facilitated by a clear understanding of terminology used. It is possible that more advanced systems of characterizing IVD degeneration, in combination with advances in genetic investigations, clinical understanding and diagnostic imaging techniques, will allow a more accurate and uniform system of classifying disease of the IVD in the future.

AUTHOR CONTRIBUTIONS

JF and NJO both contributed equally to the design and writing of the manuscript and all CANSORT-SCI members critiqued and approved the final version of the manuscript. All authors contributed to the article and approved the submitted version.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fvets.2020.579025/full#supplementary-material

REFERENCES

1. Frauchiger E, Fankhauser R. *Nervous Diseases of Dogs*. Bern: Verlag Hans Huber (1949).
2. Olsson SE. On disc protrusion in dog (enchondrosis intervertebralis); a study with special reference to roentgen diagnosis and to the value of disc fenestration. *Acta Orthop Scand Suppl.* (1951) 8:1–95. doi: 10.3109/ort.1951.22.suppl-8.01
3. Tillmanns S. *Beiträge zur Enchondrosis Intervertebralis und den Wirbel-Kanaltumoren des Hundes*. Giessen: Ludwigs-Universität Giessen (1939).
4. Hansen HJ. A pathologic-anatomical interpretation of disc degeneration in dogs. *Acta Orthop Scand.* (1951) 20:280–93. doi: 10.3109/17453675108991175
5. Hansen HJ. A pathologic-anatomical study on disc degeneration in dog, with special reference to the so-called enchondrosis intervertebralis. *Acta Orthop Scand Suppl.* (1952) 11:1–117. doi: 10.3109/ort.1952.23.suppl-11.01
6. Olsson SE, Hansen HJ. Cervical disc protrusions in the dog. *J Am Vet Med Assoc.* (1952) 121:361–70.
7. Hansen HJ. Comparative views of the pathology of disk degeneration in animals. *Lab Invest.* (1959) 8:1242–65.
8. Ghosh P, Taylor TK, Braund KG, Larsen LH. A comparative chemical and histochemical study of the chondrodystrophoid and nonchondrodystrophoid canine intervertebral disc. *Vet Pathol.* (1976) 13:414–27. doi: 10.1177/030098587601300603
9. Smolders LA, Bergknut N, Grinwis GC, Hagman R, Lagerstedt AS, Hazewinkel HA, et al. Intervertebral disc degeneration in the dog. Part 2: chondrodystrophic and non-chondrodystrophic breeds. *Vet J.* (2013) 195:292–9. doi: 10.1016/j.tvjl.2012.10.011
10. Levine J, Fingeroth JM. Historical and current nomenclature associated with intervertebral disc pathology. In: Fingeroth JM, Thomas WB, editors. *Advances in Intervertebral Disc Disease in Dogs and Cats*. Hoboken, NJ: Wiley-Blackwell (2015). p. 25–31. doi: 10.1002/9781118940372
11. Fardon DF, Williams AL, Dohring EJ, Murtagh FR, Gabriel Rothman SL, Sze GK. Lumbar disc nomenclature: version 2.0: recommendations of the combined task forces of the North American Spine society, the American society of Spine radiology and the American society of neuroradiology. *Spine J.* (2014) 14:2525–45. doi: 10.1016/j.spinee.2014.04.022
12. Bergknut N, Meij BP, Hagman R, de Nies KS, Rutges JP, Smolders LA, et al. Intervertebral disc disease in dogs - part 1: a new histological grading scheme for classification of intervertebral disc degeneration in dogs. *Vet J.* (2013) 195:156–63. doi: 10.1016/j.tvjl.2012.05.027
13. Kranenburg HJ, Grinwis GC, Bergknut N, Gahrmann N, Voorhout G, Hazewinkel HA, et al. Intervertebral disc disease in dogs - part 2: comparison of clinical, magnetic resonance imaging, and histological findings in 74 surgically treated dogs. *Vet J.* (2013) 195:164–71. doi: 10.1016/j.tvjl.2012.06.001
14. Hansen T, Smolders LA, Tryfonidou MA, Meij BP, Vernooij JCM, Bergknut N, et al. The myth of fibroid degeneration in the canine intervertebral disc: a histopathological comparison of intervertebral disc degeneration in chondrodystrophic and nonchondrodystrophic dogs. *Vet Pathol.* (2017) 54:945–52. doi: 10.1177/0300985817726834
15. Batcher K, Dickinson P, Giuffrida M, Sturges B, Vernau K, Knipe M, et al. Phenotypic effects of FGF4 retrogenes on intervertebral disc disease in dogs. *Genes.* (2019) 10:435. doi: 10.3390/genes10060435
16. Murphy BG, Dickinson P, Marcellin-Little DJ, Batcher K, Raverty S, Bannasch D. Pathologic features of the intervertebral disc in young nova scotia duck tolling retrievers confirms chondrodystrophy degenerative phenotype associated with genotype. *Vet Pathol.* (2019) 56:895–902. doi: 10.1177/0300985819868731
17. Brown EA, Dickinson PJ, Mansour T, Sturges BK, Aguilar M, Young AE, et al. FGF4 retrogene on CFA12 is responsible for chondrodystrophy and intervertebral disc disease in dogs. *Proc Natl Acad Sci USA.* (2017) 114:11476–81. doi: 10.1073/pnas.1709082114
18. De Risio L, Adams V, Dennis R, McConnell FJ. Association of clinical and magnetic resonance imaging findings with outcome in dogs with presumptive acute noncompressive nucleus pulposus extrusion: 42 cases (2000-2007). *J Am Vet Med Assoc.* (2009) 234:495–504. doi: 10.2460/javma.234.4.495
19. Hamilton T, Glass E, Drobatz K, Agnello KA. Severity of spinal cord dysfunction and pain associated with hydrated nucleus pulposus extrusion in dogs. *Vet Comp Orthop Traumatol.* (2014) 27:313–8. doi: 10.3415/VCOT-13-06-0076
20. Beltran E, Dennis R, Doyle V, de Stefani A, Holloway A, de Risio L. Clinical and magnetic resonance imaging features of canine compressive cervical myelopathy with suspected hydrated nucleus pulposus extrusion. *J Small Anim Pract.* (2012) 53:101–7. doi: 10.1111/j.1748-5827.2011.01166.x
21. Windsor RC, Vernau KM, Sturges BK, Kass PH, Vernau W. Lumbar cerebrospinal fluid in dogs with type I intervertebral disc herniation. *J Vet Intern Med.* (2008) 22:954–60. doi: 10.1111/j.1939-1676.2008.0141.x

22. Bergknut N, Smolders LA, Grinwis GC, Hagman R, Lagerstedt AS, Hazewinkel HA, et al. Intervertebral disc degeneration in the dog. Part 1: Anatomy and physiology of the intervertebral disc and characteristics of intervertebral disc degeneration. *Vet J.* (2013) 195:282–91. doi: 10.1016/j.tvjl.2012.10.024
23. Gomes SA, Volk HA, Packer RM, Kenny PJ, Beltran E, De Decker S. Clinical and magnetic resonance imaging characteristics of thoracolumbar intervertebral disk extrusions and protrusions in large breed dogs. *Vet Radiol Ultrasound.* (2016) 57:417–26. doi: 10.1111/vru.12359
24. Olby NJ, Muguet-Chanoit AC, Lim JH, Davidian M, Mariani CL, Freeman AC, et al. A placebo-controlled, prospective, randomized clinical trial of polyethylene glycol and methylprednisolone sodium succinate in dogs with intervertebral disk herniation. *J Vet Intern Med.* (2016) 30:206–14. doi: 10.1111/jvim.13657
25. Scott HW. Hemilaminectomy for the treatment of thoracolumbar disc disease in the dog: a follow-up study of 40 cases. *J Small Anim Pract.* (1997) 38:488. doi: 10.1111/j.1748-5827.1997.tb03303.x
26. Smith JD, Newell SM, Budsberg SC, Bennett RA. Incidence of contralateral versus ipsi-lateral neurological signs associated with lateralised Hansen type I disc extrusion. *J Small Anim Pract.* (1997) 38:495–7. doi: 10.1111/j.1748-5827.1997.tb03305.x
27. Davis GJ, Brown DC. Prognostic indicators for time to ambulation after surgical decompression in nonambulatory dogs with acute thoracolumbar disk extrusions: 112 cases. *Vet Surg.* (2002) 31:513–8. doi: 10.1053/jvet.2002.36015
28. Ruddle TL, Allen DA, Schertel ER, Barnhart MD, Wilson ER, Lineberger JA, et al. Outcome and prognostic factors in nonambulatory Hansen type I intervertebral disc extrusions: 308 cases. *Vet Comp Orthop Traumatol.* (2006) 19:29–34. doi: 10.1055/s-0038-1632970
29. Dennison SE, Drees R, Rylander H, Yandell BS, Milovancev M, Pettigrew R, et al. Evaluation of different computed tomography techniques and myelography for the diagnosis of acute canine myelopathy. *Vet Radiol Ultrasound.* (2010) 51:254–8. doi: 10.1111/j.1740-8261.2010.01667.x
30. Skytte D, Schmökel H. Relationship of preoperative neurologic score with intervals to regaining micturition and ambulation following surgical treatment of thoracolumbar disk herniation in dogs. *J Am Vet Med Assoc.* (2018) 253:196–200. doi: 10.2460/javma.253.2.196
31. Upchurch DA, Renberg WC, Turner HS, McLellan JG. Effect of duration and onset of clinical signs on short-term outcome of dogs with hansen type I thoracolumbar intervertebral disc extrusion. *Vet Comp Orthop Traumatol.* (2020) 33:161–6. doi: 10.1055/s-0039-1700988
32. Ferreira AJA, Correia JHD, Jaggy A. Thoracolumbar disc disease in 71 paraplegic dogs: influence of rate of onset and duration of clinical signs on treatment results. *J Small Anim Pract.* (2002) 43:158–63. doi: 10.1111/j.1748-5827.2002.tb00049.x
33. Brisson BA, Moffatt SL, Swayne SL, Parent JM. Recurrence of thoracolumbar intervertebral disk extrusion in chondrodystrophic dogs after surgical decompression with or without prophylactic fenestration: 265 cases (1995-1999). *J Am Vet Med Assoc.* (2004) 224:1808. doi: 10.2460/javma.2004.224.1808
34. Penning V, Platt SR, Dennis R, Cappello R, Adams V. Association of spinal cord compression seen on magnetic resonance imaging with clinical outcome in 67 dogs with thoracolumbar intervertebral disc extrusion. *J Small Anim Pract.* (2006) 47:644–50. doi: 10.1111/j.1748-5827.2006.00252.x
35. Newcomb B, Arble J, Rochat M, Pechman R, Payton M. Comparison of computed tomography and myelography to a reference standard of computed tomographic myelography for evaluation of dogs with intervertebral disc disease. *Vet Surg.* (2012) 41:207–14. doi: 10.1111/j.1532-950X.2011.00911.x
36. Ingram EA, Kale DC, Balfour RJ. Hemilaminectomy for thoracolumbar Hansen Type I intervertebral disk disease in ambulatory dogs with or without neurologic deficits: 39 cases (2008-2010). *Vet Surg.* (2013) 42:924–31. doi: 10.1111/j.1532-950X.2013.12061.x
37. Olby NJ, Munana KR, Sharp NJ, Thrall DE. The computed tomographic appearance of acute thoracolumbar intervertebral disc herniations in dogs. *Vet Radiol Ultrasound.* (2000) 41:396–402. doi: 10.1111/j.1740-8261.2000.tb01860.x
38. Olby N, Harris T, Burr J, Munana K, Sharp N, Keene B. Recovery of pelvic limb function in dogs following acute intervertebral disc herniations. *J Neurotrauma.* (2004) 21:49–59. doi: 10.1089/089771504772695940
39. Levine JM, Fosgate GT, Chen AV, Rushing R, Nghiem PP, Platt SR, et al. Magnetic resonance imaging in dogs with neurologic impairment due to acute thoracic and lumbar intervertebral disk herniation. *J Vet Intern Med.* (2009) 23:1220–6. doi: 10.1111/j.1939-1676.2009.0393.x
40. Brisson BA, Holmberg DL, Parent J, Sears WC, Wick SE. Comparison of the effect of single-site and multiple-site disk fenestration on the rate of recurrence of thoracolumbar intervertebral disk herniation in dogs. *J Am Vet Med Assoc.* (2011) 238:1593–600. doi: 10.2460/javma.238.12.1593
41. Aikawa T, Fujita H, Kanazono S, Shibata M, Yoshigae Y. Long-term neurologic outcome of hemilaminectomy and disk fenestration for treatment of dogs with thoracolumbar intervertebral disk herniation: 831 cases (2000-2007). *J Am Vet Med Assoc.* (2012) 241:1617–26. doi: 10.2460/javma.241.12.1617
42. Levine GJ, Cook JR, Kerwin SC, Mankin J, Griffin JF, Fosgate GT, et al. Relationships between cerebrospinal fluid characteristics, injury severity, and functional outcome in dogs with and without intervertebral disk herniation. *Vet Clin Pathol.* (2014) 43:437–46. doi: 10.1111/vcp.12165
43. Jeffery ND, Barker AK, Hu HZ, Alcott CJ, Kraus KH, Scanlin EM, et al. Factors associated with recovery from paraplegia in dogs with loss of pain perception in the pelvic limbs following intervertebral disk herniation. *J Am Vet Med Assoc.* (2016) 248:386–94. doi: 10.2460/javma.248.4.386
44. Moore SA, Early PJ, Hettlich BF. Practice patterns in the management of acute intervertebral disc herniation in dogs. *J Small Anim Pract.* (2016) 57:409–15. doi: 10.1111/jsap.12496
45. Jeffery ND, Mankin JM, Ito D, Boudreau CE, Kerwin SC, Levine JM, et al. Extended durotomy to treat severe spinal cord injury after acute thoracolumbar disc herniation in dogs. *Vet Surg.* (2020) 49:884–93. doi: 10.1111/vsu.13423
46. Takahashi F, Honnami A, Toki M, Dosaka A, Fujita Y, Hara Y, et al. Effect of durotomy in dogs with thoracolumbar disc herniation and without deep pain perception in the hind limbs. *Vet Surg.* (2020) 49:860–9. doi: 10.1111/vsu.13409
47. Ito D, Matsunaga S, Jeffery ND, Sasaki N, Nishimura R, Mochizuki M, et al. Prognostic value of magnetic resonance imaging in dogs with paraplegia caused by thoracolumbar intervertebral disk extrusion: 77 cases (2000-2003). *J Am Vet Med Assoc.* (2005) 227:1454–60. doi: 10.2460/javma.2005.227.1454
48. Levine JM, Levine GJ, Kerwin SC, Hettlich BF, Fosgate GT. Association between various physical factors and acute thoracolumbar intervertebral disk extrusion or protrusion in Dachshunds. *J Am Vet Med Assoc.* (2006) 229:370–5. doi: 10.2460/javma.229.3.370
49. Olby NJ, MacKillop E, Cerda-Gonzalez S, Moore S, Munana KR, Grafinger M, et al. Prevalence of urinary tract infection in dogs after surgery for thoracolumbar intervertebral disc extrusion. *J Vet Intern Med.* (2010) 24:1106–11. doi: 10.1111/j.1939-1676.2010.0567.x
50. Muguet-Chanoit AC, Olby NJ, Lim JH, Gallagher R, Niman Z, Dillard S, et al. The cutaneous trunci muscle reflex: a predictor of recovery in dogs with acute thoracolumbar myelopathies caused by intervertebral disc extrusions. *Vet Surg.* (2012) 41:200–6. doi: 10.1111/j.1532-950X.2011.00921.x
51. De Decker S, Gomes SA, Packer RM, Kenny PJ, Beltran E, Parzefall B, et al. Evaluation of magnetic resonance imaging guidelines for differentiation between thoracolumbar intervertebral disk extrusions and intervertebral disk protrusions in dogs. *Vet Radiol Ultrasound.* (2016) 57:526–33. doi: 10.1111/vru.12394
52. Fenn J, Laber E, Williams K, Rousse CA, Early PJ, Mariani CL, et al. Associations between anesthetic variables and functional outcome in dogs with thoracolumbar intervertebral disk extrusion undergoing decompressive hemilaminectomy. *J Vet Intern Med.* (2017) 31:814–24. doi: 10.1111/jvim.14677
53. Olby NJ, Lim JH, Wagner N, Zidan N, Early PJ, Mariani CL, et al. Time course and prognostic value of serum GFAP, pNFH, and S100beta concentrations in dogs with complete spinal cord injury because of intervertebral disc extrusion. *J Vet Intern Med.* (2019) 33:726–34. doi: 10.1111/jvim.15439

54. Davies JV, Sharp NJH. A comparison of conservative treatment and fenestration for thoracolumbar intervertebral disc disease in the dog. *J Small Anim Pract.* (1983) 24:721–9.
55. Jeffery ND. Treatment of acute and chronic thoracolumbar disc disease by 'mini hemilaminectomy'. *J Small Anim Pract.* (1988) 29:611–16.
56. Duval J, Dewey C, Roberts R, Aron D. Spinal cord swelling as a myelographic indicator of prognosis: a retrospective study in dogs with intervertebral disc disease and loss of deep pain perception. *Vet Surg.* (1996) 25:6–12. doi: 10.1111/j.1532-950x.1996.tb01371.x
57. Scott HW, McKee WM. Laminectomy for 34 dogs with thoracolumbar intervertebral disc disease and loss of deep pain perception. *J Small Anim Pract.* (1999) 40:417–22. doi: 10.1111/j.1748-5827.1999.tb03114.x
58. Srugo I, Aroch I, Christopher MM, Chai O, Goralnik L, Bdolah-Abram T, et al. Association of cerebrospinal fluid analysis findings with clinical signs and outcome in acute nonambulatory thoracolumbar disc disease in dogs. *J Vet Intern Med.* (2011) 25:846–55. doi: 10.1111/j.1939-1676.2011.0739.x
59. Yovich JC, Read R, Eger C. Modified lateral spinal decompression in 61 dogs with thoracolurnbar disc protrusion. *J Small Anim Pract.* (1994) 35:351–6.
60. Kinzel S, Wolff M, Buecker A, Krombach GA, Stopinski T, Afify M, et al. Partial percutaneous discectomy for treatment of thoracolumbar disc protrusion: retrospective study of 331 dogs. *J Small Anim Pract.* (2005) 46:479–84. doi: 10.1111/j.1748-5827.2005.tb00276.x
61. Brisson BA. Intervertebral disc disease in dogs. *Vet Clin North Am Small Anim Pract.* (2010) 40:829–58. doi: 10.1016/j.cvsm.2010.06.001
62. Cudia SP, Duval JM. Thoracolumbar intervertebral disk disease in large, nonchondrodystrophic dogs: a retrospective study. *J Am Anim Hosp Assoc.* (1997) 33:456–60. doi: 10.5326/15473317-33-5-456
63. Macias C, McKee WM, May C, Innes JF. Thoracolumbar disc disease in large dogs: a study of 99 cases. *J Small Anim Pract.* (2002) 43:439–46. doi: 10.1111/j.1748-5827.2002.tb00010.x
64. Crawford AH, De Decker S. Clinical presentation and outcome of dogs treated medically or surgically for thoracolumbar intervertebral disc protrusion. *Vet Rec.* (2017) 180:569. doi: 10.1136/vr.103871
65. Crawford AH, De Decker S. Comparison between hemilaminectomy with either anulectomy or partial discectomy for treatment of thoracolumbar intervertebral disc protrusion in dogs. *Vet Comp Orthop Traumatol.* (2018) 31:194–201. doi: 10.1055/s-0038-1639590
66. Guevar J, Olby N. Minimally invasive microsurgical decompression of an intervertebral disc protrusion in a dog. *Vet Surg.* (2020) 49(Suppl. 1):O86–92. doi: 10.1111/vsu.13263
67. Tartarelli CL, Baroni M, Borghi M. Thoracolumbar disc extrusion associated with extensive epidural haemorrhage: a retrospective study of 23 dogs. *J Small Anim Pract.* (2005) 46:485–90. doi: 10.1111/j.1748-5827.2005.tb00277.x
68. Cerda-Gonzalez S, Olby NJ. Fecal incontinence associated with epidural spinal hematoma and intervertebral disk extrusion in a dog. *J Am Vet Med Assoc.* (2006) 228:230–5. doi: 10.2460/javma.228.2.230
69. Fenn J, Drees R, Volk HA, De Decker S. Comparison of clinical signs and outcomes between dogs with presumptive ischemic myelopathy and dogs with acute noncompressive nucleus pulposus extrusion. *J Am Vet Med Assoc.* (2016) 249:767–75. doi: 10.2460/javma.249.7.767
70. Specchi S, Johnson P, Beauchamp G, Masseau I, Pey P. Assessment of interobserver agreement and use of selected magnetic resonance imaging variables for differentiation of acute noncompressive nucleus pulposus extrusion and ischemic myelopathy in dogs. *J Am Vet Med Assoc.* (2016) 248:1013–21. doi: 10.2460/javma.248.9.1013
71. Meij BP, Bergknut N. Degenerative lumbosacral stenosis in dogs. *Vet Clin North Am Small Anim Pract.* (2010) 40:983–1009. doi: 10.1016/j.cvsm.2010.05.006
72. Poncelet L, Heimann M. Intradural vertebral disc herniation in a dog. *Vet Rec.* (2011) 168:486a. doi: 10.1136/vr.c6740
73. Mari L, Behr S, Shea A, Dominguez E, Ricco C, Alcoverro E, et al. Predictors of urinary or fecal incontinence in dogs with thoracolumbar acute non-compressive nucleus pulposus extrusion. *J Vet Intern Med.* (2019) 33:2693–700. doi: 10.1111/jvim.15626
74. Bagley R. Spinal cord enigmas: fibrocartilaginous emboli, arachnoid cyst and others. In: *Proceedings of the 21st Annual American College of Veterinary Internal Medicine Forum.* Charlotte, NC (2003). p. 10–1.
75. Lu D, Lamb CR, Wesselingh K, Targett MP. Acute intervertebral disc extrusion in a cat: clinical and MRI findings. *J Feline Med Surg.* (2002) 4:65–8. doi: 10.1053/jfms.2001.0150
76. Chang Y, Dennis R, Platt SR, Penderis J. Magnetic resonance imaging of traumatic intervertebral disc extrusion in dogs. *Vet Rec.* (2007) 160:795–9. doi: 10.1136/vr.160.23.795
77. Henke D, Gorgas D, Flegel T, Vandevelde M, Lang J, Doherr MG, et al. Magnetic resonance imaging findings in dogs with traumatic intervertebral disk extrusion with or without spinal cord compression: 31 cases (2006-2010). *J Am Vet Med Assoc.* (2013) 242:217–22. doi: 10.2460/javma.242.2.217
78. Griffiths IR. A syndrome produced by dorso-lateral "explosions" of the cervical intervertebral discs. *Vet Rec.* (1970) 87:737–41. doi: 10.1136/vr.87.24.737
79. Bartholomew KA, Stover KE, Olby NJ, Moore SA. Clinical characteristics of canine fibrocartilaginous embolic myelopathy (FCE): a systematic review of 393 cases (1973-2013). *Vet Rec.* (2016) 179:650. doi: 10.1136/vr.103863
80. Chow K, Beatty JA, Voss K, Barrs VR. Probable lumbar acute non-compressive nucleus pulposus extrusion in a cat with acute onset paraparesis. *J Feline Med Surg.* (2012) 14:764–7. doi: 10.1177/1098612X12450110
81. Dolera M, Malfassi L, Marcarini S, Mazza G, Sala M, Carrara N, et al. Hydrated nucleus pulposus extrusion in dogs: correlation of magnetic resonance imaging and microsurgical findings. *Acta Vet Scand.* (2015) 57:58. doi: 10.1186/s13028-015-0151-x
82. De Decker S, Warner AS, Volk HA. Prevalence and breed predisposition for thoracolumbar intervertebral disc disease in cats. *J Feline Med Surg.* (2017) 19:419–23. doi: 10.1177/1098612X16630358
83. Manunta ML, Evangelisti MA, Bergknut N, Grinwis GC, Ballocco I, Meij BP. Hydrated nucleus pulposus herniation in seven dogs. *Vet J.* (2015) 203:342–4. doi: 10.1016/j.tvjl.2014.12.027
84. Borlace T, Gutierrez-Quintana R, Taylor-Brown FE, De Decker S. Comparison of medical and surgical treatment for acute cervical compressive hydrated nucleus pulposus extrusion in dogs. *Vet Rec.* (2017) 181:625. doi: 10.1136/vr.104528
85. Falzone C. Canine acute cervical myelopathy: hydrated nucleus pulposus extrusion or intraspinal discal cysts? *Vet Surg.* (2017) 46:376–80. doi: 10.1111/vsu.12631
86. Konar M, Lang J, Fluhmann G, Forterre F. Ventral intraspinal cysts associated with the intervertebral disc: magnetic resonance imaging observations in seven dogs. *Vet Surg.* (2008) 37:94–101. doi: 10.1111/j.1532-950X.2007.00353.x
87. Kamishina H, Ogawa H, Katayama M, Yasuda J, Sato R, Tohyama K. Spontaneous regression of a cervical intraspinal cyst in a dog. *J Vet Med Sci.* (2010) 72:349–52. doi: 10.1292/jvms.09-0375
88. De Risio L. A review of fibrocartilaginous embolic myelopathy and different types of peracute non-compressive intervertebral disk extrusions in dogs and cats. *Front Vet Sci.* (2015) 2:24. doi: 10.3389/fvets.2015.00024
89. Tamura S, Doi S, Tamura Y, Takahashi K, Enomoto H, Ozawa T, et al. Thoracolumbar intradural disc herniation in eight dogs: clinical, low-field magnetic resonance imaging, and computed tomographic myelography findings. *Vet Radiol Ultrasound.* (2015) 56:160–7. doi: 10.1111/vru.12213
90. Barnoon I, Chai O, Srugo I, Peeri D, Konstantin L, Brenner O, et al. Spontaneous intradural disc herniation with focal distension of the subarachnoid space in a dog. *Can Vet J.* (2012) 53:1191–4.
91. Kitagawa M, Okada M, Kanayama K, Sakai T. Identification of ventrolateral intramedullary intervertebral disc herniation in a dog. *J S Afr Vet Assoc.* (2012) 83:103. doi: 10.4102/jsava.v83i1.103
92. Liptak JM, Allan GS, Krockenberger MB, Davis PE, Malik R. Radiographic diagnosis: intramedullary extrusion of an intervertebral disc. *Vet Radiol Ultrasound.* (2002) 43:272–4. doi: 10.1111/j.1740-8261.2002.tb01002.x
93. McConnell JF, Garosi LS. Intramedullary intervertebral disk extrusion in a cat. *Vet Radiol Ultrasound.* (2004) 45:327–30. doi: 10.1111/j.1740-8261.2004.04062.x
94. Hawthorne J, Wallace L, Fenner W, Waters D. Fibrocartilaginous embolic myelopathy in miniature schnauzers. *J Am Anim Hosp Assoc.* (2001) 37:374–83. doi: 10.5326/15473317-37-4-374

95. De Risio L, Platt SR. Fibrocartilaginous embolic myelopathy in small animals. *Vet Clin North Am Small Anim Pract.* (2010) 40:859–69. doi: 10.1016/j.cvsm.2010.05.003
96. de Lahunta A, Alexander J. Ischemic myelopathy secondary to presumed fibrocartilaginous embolism in nine dogs. *J Am Anim Hosp Assoc.* (1976) 12:37–48.
97. Gilmore R, De Lahunta, A. Necrotizing myelopathy secondary to presumed or confirmed fibrocartilaginous embolism in 24 dogs. *J Am Anim Hosp Assoc.* (1987) 23:373.
98. Cauzinille L, Kornegay JN. Fibrocartilaginous embolism of the spinal cord in dogs: review of 36 histologically confirmed cases and retrospective study of 26 suspected cases. *J Vet Intern Med.* (1996) 10:241–5. doi: 10.1111/j.1939-1676.1996.tb02056.x
99. Gandini G, Cizinauskas S, Lang J, Fatzer R, Jaggy A. Fibrocartilaginous embolism in 75 dogs: clinical findings and factors influencing the recovery rate. *J Small Anim Pract.* (2003) 44:76–80. doi: 10.1111/j.1748-5827.2003.tb00124.x
100. De Risio L, Adams V, Dennis R, McConnell FJ, Platt SR. Association of clinical and magnetic resonance imaging findings with outcome in dogs suspected to have ischemic myelopathy: 50 cases (2000-2006). *J Am Vet Med Assoc.* (2008) 233:129–35. doi: 10.2460/javma.233.1.129
101. Yarrow TG, Jeffery ND. Dura mater laceration associated with acute paraplegia in three dogs. *Vet Rec.* (2000) 146:138–9. doi: 10.1136/vr.146.5.138
102. Theobald A, Volk HA, Dennis R, Berlato D, De Risio L. Clinical outcome in 19 cats with clinical and magnetic resonance imaging diagnosis of ischaemic myelopathy (2000-2011). *J Feline Med Surg.* (2013) 15:132–41. doi: 10.1177/1098612X12463927
103. Griffiths IR. Spinal cord infarction due to emboli arising from the intervertebral discs in the dog. *J Comp Pathol.* (1973) 83:225–32. doi: 10.1016/0021-9975(73)90046-7
104. Coates JR. Intervertebral disk disease. *Vet Clin North Am Small Anim Pract.* (2000) 30:77–110. doi: 10.1016/S0195-5616(00)50004-7
105. McGeady TA. *Veterinary Embryology*. 2nd ed. Chichester; Ames, IA: John Wiley & Sons Inc. (2017). p. 167–95.
106. Bray JP, Burbidge HM. The canine intervertebral disk: part one: structure and function. *J Am Anim Hosp Assoc.* (1998) 34:55–63. doi: 10.5326/15473317-34-1-55
107. Ghosh P, Taylor TK, Braund KG. The variation of the glycosaminoglycans of the canine intervertebral disc with ageing. I. Chondrodystrophoid breed. *Gerontology.* (1977) 23:87–98. doi: 10.1159/000212177
108. Johnson JA, da Costa RC, Allen MJ. Micromorphometry and cellular characteristics of the canine cervical intervertebral discs. *J Vet Intern Med.* (2010) 24:1343–9. doi: 10.1111/j.1939-1676.2010.0613.x
109. Ghosh P, Taylor TK, Braund KG, Larsen LH. The collagenous and non-collagenous protein of the canine intervertebral disc and their variation with age, spinal level and breed. *Gerontology.* (1976) 22:124–34. doi: 10.1159/000212129
110. Bray JP, Burbidge HM. The canine intervertebral disk. Part Two: degenerative changes–nonchondrodystrophoid versus chondrodystrophoid disks. *J Am Anim Hosp Assoc.* (1998) 34:135–44. doi: 10.5326/15473317-34-2-135
111. Bergknut N, Rutges JP, Kranenburg HJ, Smolders LA, Hagman R, Smidt HJ, et al. The dog as an animal model for intervertebral disc degeneration? *Spine.* (2012) 37:351–8. doi: 10.1097/BRS.0b013e31821e5665
112. Ghosh P, Taylor TK, Braund KG. Variation of the glycosaminoglycans of the intervertebral disc with ageing. II. Non-chondrodystrophoid breed. *Gerontology.* (1977) 23:99–109. doi: 10.1159/000212178
113. Fluehmann G, Doherr MG, Jaggy A. Canine neurological diseases in a referral hospital population between 1989 and 2000 in Switzerland. *J Small Anim Pract.* (2006) 47:582–7. doi: 10.1111/j.1748-5827.2006.00106.x
114. Rossi G, Stachel A, Lynch A, Olby N. Intervertebral disc disease and aortic thromboembolism are the most common causes of acute paralysis in dogs and cats presenting to an emergency clinic. *Vet Rec.* (2020) doi: 10.1136/vr.105844. [Epub ahead of print].
115. Stigen O. Calcification of intervertebral discs in the dachshund. A radiographic study of 327 young dogs. *Acta Vet Scand.* (1991) 32: 197–203.
116. Jensen VF, Arnbjerg J. Development of intervertebral disk calcification in the dachshund: a prospective longitudinal radiographic study. *J Am Anim Hosp Assoc.* (2001) 37:274–82. doi: 10.5326/15473317-37-3-274
117. Jensen VF. Asymptomatic radiographic disappearance of calcified intervertebral disc material in the dachshund. *Vet Radiol Ultrasound.* (2001) 42:141–8. doi: 10.1111/j.1740-8261.2001.tb00917.x
118. Cherrone KL, Dewey CW, Coates JR, Bergman RL. A retrospective comparison of cervical intervertebral disk disease in nonchondrodystrophic large dogs versus small dogs. *J Am Anim Hosp Assoc.* (2004) 40:316–20. doi: 10.5326/0400316
119. Mogensen MS, Karlskov-Mortensen P, Proschowsky HF, Lingaas F, Lappalainen A, Lohi H, et al. Genome-wide association study in dachshund: identification of a major locus affecting intervertebral disc calcification. *J Hered.* (2011) 102(Suppl. 1):S81–6. doi: 10.1093/jhered/esr021
120. Packer RM, Hendricks A, Volk HA, Shihab NK, Burn CC. How long and low can you go? Effect of conformation on the risk of thoracolumbar intervertebral disc extrusion in domestic dogs. *PLoS ONE.* (2013) 8:e69650. doi: 10.1371/journal.pone.0069650
121. Jensen VF, Ersboll AK. Mechanical factors affecting the occurrence of intervertebral disc calcification in the dachshund–a population study. *J Vet Med A Physiol Pathol Clin Med.* (2000) 47:283–96. doi: 10.1046/j.1439-0442.2000.00296.x
122. DiVita MM, Bray KY, Williams LM, Traslavina RP. What is your neurologic diagnosis? *J Am Vet Med Assoc.* (2020) 256:981–3. doi: 10.2460/javma.256.9.981
123. Munana KR, Olby NJ, Sharp NJ, Skeen TM. Intervertebral disk disease in 10 cats. *J Am Anim Hosp Assoc.* (2001) 37:384–9. doi: 10.5326/15473317-37-4-384
124. Fadda A, Oevermann A, Vandevelde M, Doherr MG, Forterre F, Henke D. Clinical and pathological analysis of epidural inflammation in intervertebral disk extrusion in dogs. *J Vet Intern Med.* (2013) 27:924–34. doi: 10.1111/jvim.12095
125. Bos AS, Brisson BA, Nykamp SG, Poma R, Foster RA. Accuracy, intermethod agreement, and inter-reviewer agreement for use of magnetic resonance imaging and myelography in small-breed dogs with naturally occurring first-time intervertebral disk extrusion. *J Am Vet Med Assoc.* (2012) 240:969–77. doi: 10.2460/javma.240.8.969
126. Pfirrmann CW, Metzdorf A, Zanetti M, Hodler J, Boos N. Magnetic resonance classification of lumbar intervertebral disc degeneration. *Spine.* (2001) 26:1873–8. doi: 10.1097/00007632-200109010-00011
127. Tidwell AS, Specht A, Blaeser L, Kent M. Magnetic resonance imaging features of extradural hematomas associated with intervertebral disc herniation in a dog. *Vet Radiol Ultrasound.* (2002) 43:319–24. doi: 10.1111/j.1740-8261.2002.tb01011.x
128. Mateo I, Lorenzo V, Foradada L, Munoz A. Clinical, pathologic, and magnetic resonance imaging characteristics of canine disc extrusion accompanied by epidural hemorrhage or inflammation. *Vet Radiol Ultrasound.* (2011) 52:17–24. doi: 10.1111/j.1740-8261.2010.01746.x
129. De Decker S, da Costa RC, Volk HA, Van Ham LM. Current insights and controversies in the pathogenesis and diagnosis of disc-associated cervical spondylomyelopathy in dogs. *Vet Rec.* (2012) 171:531–7. doi: 10.1136/vr.e7952
130. Cooper JJ, Young BD, Griffin JFt, Fosgate GT, Levine JM. Comparison between noncontrast computed tomography and magnetic resonance imaging for detection and characterization of thoracolumbar myelopathy caused by intervertebral disk herniation in dogs. *Vet Radiol Ultrasound.* (2014) 55:182–9. doi: 10.1111/vru.12114
131. Besalti O, Pekcan Z, Sirin YS, Erbas G. Magnetic resonance imaging findings in dogs with thoracolumbar intervertebral disk disease: 69 cases (1997-2005). *J Am Vet Med Assoc.* (2006) 228:902–8. doi: 10.2460/javma.228.6.902
132. Penning VA, Benigni L, Steeves E, Cappello R. Imaging diagnosis–degenerative intraspinal cyst associated with an intervertebral disc. *Vet Radiol Ultrasound.* (2007) 48:424–7. doi: 10.1111/j.1740-8261.2007.00272.x
133. De Decker S, Fenn J. Acute herniation of nondegenerate nucleus pulposus: acute noncompressive nucleus pulposus extrusion and compressive hydrated

nucleus pulposus extrusion. *Vet Clin North Am Small Anim Pract.* (2018) 48:95–109. doi: 10.1016/j.cvsm.2017.08.004
134. Lowrie ML, Platt SR, Garosi LS. Extramedullary spinal cysts in dogs. *Vet Surg.* (2014) 43:650–62. doi: 10.1111/j.1532-950X.2014.12200.x
135. Chiba K, Toyama Y, Matsumoto M, Maruiwa H, Watanabe M, Nishizawa T. Intraspinal cyst communicating with the intervertebral disc in the lumbar spine: discal cyst. *Spine.* (2001) 26:2112–8. doi: 10.1097/00007632-200110010-00014
136. Royaux E, Martle V, Kromhout K, Van der Vekens E, Broeckx BJ, Van Ham L, et al. Detection of compressive hydrated nucleus pulposus extrusion in dogs with multislice computed tomography. *Vet J.* (2016) 216:202–6. doi: 10.1016/j.tvjl.2016.08.012
137. Taylor-Brown FE, De Decker S. Presumptive acute non-compressive nucleus pulposus extrusion in 11 cats: clinical features, diagnostic imaging findings, treatment and outcome. *J Feline Med Surg.* (2017) 19:21–6. doi: 10.1177/1098612X15605150
138. Mari L, Behr S, Shea A, Dominguez E, Johnson PJ, Ekiri A, et al. Outcome comparison in dogs with a presumptive diagnosis of thoracolumbar fibrocartilaginous embolic myelopathy and acute non-compressive nucleus pulposus extrusion. *Vet Rec.* (2017) 181:293. doi: 10.1136/vr.104090
139. Fenn J, Drees R, Volk HA, Decker SD. Inter- and intraobserver agreement for diagnosing presumptive ischemic myelopathy and acute noncompressive nucleus pulposus extrusion in dogs using magnetic resonance imaging. *Vet Radiol Ultrasound.* (2016) 57:33–40. doi: 10.1111/vru.12289
140. Henke D, Vandevelde M, Doherr MG, Stockli M, Forterre F. Correlations between severity of clinical signs and histopathological changes in 60 dogs with spinal cord injury associated with acute thoracolumbar intervertebral disc disease. *Vet J.* (2013) 198:70–5. doi: 10.1016/j.tvjl.2013.04.003
141. Sanders SG, Bagley RS, Gavin PR. Intramedullary spinal cord damage associated with intervertebral disk material in a dog. *J Am Vet Med Assoc.* (2002) 221:1594–6, 74–5. doi: 10.2460/javma.2002.221.1594
142. Kent M, Holmes S, Cohen E, Sakals S, Roach W, Platt S, et al. Imaging diagnosis-CT myelography in a dog with intramedullary intervertebral disc herniation. *Vet Radiol Ultrasound.* (2011) 52:185–7. doi: 10.1111/j.1740-8261.2010.01755.x
143. McKee WM, Downes CJ. Rupture of the dura mater in two dogs caused by the peracute extrusion of a cervical disc. *Vet Rec.* (2008) 162:479–81. doi: 10.1136/vr.162.15.479
144. Hay CW, Muir P. Tearing of the dura mater in three dogs. *Vet Rec.* (2000) 146:279–82. doi: 10.1136/vr.146.10.279
145. Roush J, Douglass J, Hertzke D, Kennedy G. Traumatic dural laceration in a racing greyhound. *Vet Radiol Ultrasound.* (1992) 33:22–4. doi: 10.1111/j.1740-8261.1992.tb01951.x
146. Packer RA, Frank PM, Chambers JN. Traumatic subarachnoid-pleural fistula in a dog. *Vet Radiol Ultrasound.* (2004) 45:523–7. doi: 10.1111/j.1740-8261.2004.04089.x
147. Zaki FA, Prata RG, Kay WJ. Necrotizing myelopathy in five great danes. *J Am Vet Med Assoc.* (1974) 165:1080–4.
148. Zaki FA, Prata RG. Necrotizing myelopathy secondary to embolization of herniated intervertebral disk material in the dog. *J Am Vet Med Assoc.* (1976) 169:222–8.
149. Chick BF. Ischaemic embolic myelopathy in a labrador crossbred dog. *Can Vet J.* (1979) 20:84–6.
150. Hayes MA, Creighton SR, Boysen BG, Holfeld N. Acute necrotizing myelopathy from nucleus pulposus embolism in dogs with intervertebral disk degeneration. *J Am Vet Med Assoc.* (1978) 173:289–5.
151. Feigin I, Popoff N, Adachi M. Fibrocartilagenous venous emboli to the spinal cord with necrotic myelopathy. *J Neuropathol Exp Neurol.* (1965) 24:63–74. doi: 10.1097/00005072-196501000-00006
152. Grunenfelder FI, Weishaupt D, Green R, Steffen F. Magnetic resonance imaging findings in spinal cord infarction in three small breed dogs. *Vet Radiol Ultrasound.* (2005) 46:91–6. doi: 10.1111/j.1740-8261.2005.00019.x
153. Ueno H, Shimizu J, Uzuka Y, Kobayashi Y, Hirokawa H, Ueno E, et al. Fibrocartilaginous embolism in a chondrodystrophoid breed dog. *Aust Vet J.* (2005) 83:142–4. doi: 10.1111/j.1751-0813.2005.tb11620.x
154. Castel A, Olby NJ. Acute change in neurological level following canine intervertebral disc herniation. *J Small Anim Pract.* (2016) 57:220. doi: 10.1111/jsap.12445
155. Junker K, van den Ingh TS, Bossard MM, van Nes JJ. Fibrocartilaginous embolism of the spinal cord (FCE) in juvenile Irish wolfhounds. *Vet Q.* (2000) 22:154–6. doi: 10.1080/01652176.2000.9695046
156. Dyce J, Houlton J. Fibrocartilaginous embolism in the dog. *J Small Anim Pract.* (1993) 34:332–6. doi: 10.1111/j.1748-5827.1993.tb02706.x
157. Abramson CJ, Garosi L, Platt SR, Dennis R, McConnell JF. Magnetic resonance imaging appearance of suspected ischemic myelopathy in dogs. *Vet Radiol Ultrasound.* (2005) 46:225–9. doi: 10.1111/j.1740-8261.2005.00037.x
158. Berlin R, Gruen R, Best J. Systems medicine disease: disease classification and scalability beyond networks and boundary conditions. *Front Bioeng Biotechnol.* (2018) 6:112. doi: 10.3389/fbioe.2018.00112
159. Thompson JP, Pearce RH, Schechter MT, Adams ME, Tsang IK, Bishop PB. Preliminary evaluation of a scheme for grading the gross morphology of the human intervertebral disc. *Spine.* (1990) 15:411–5. doi: 10.1097/00007632-199005000-00012
160. Bergknut N, Grinwis G, Pickee E, Auriemma E, Lagerstedt AS, Hagman R, et al. Reliability of macroscopic grading of intervertebral disk degeneration in dogs by use of the thompson system and comparison with low-field magnetic resonance imaging findings. *Am J Vet Res.* (2011) 72:899–904. doi: 10.2460/ajvr.72.7.899
161. Bergknut N, Auriemma E, Wijsman S, Voorhout G, Hagman R, Lagerstedt AS, et al. Evaluation of intervertebral disk degeneration in chondrodystrophic and nonchondrodystrophic dogs by use of Pfirrmann grading of images obtained with low-field magnetic resonance imaging. *Am J Vet Res.* (2011) 72:893–8. doi: 10.2460/ajvr.72.7.893
162. Jayadev S, Bird TD. Hereditary ataxias: overview. *Genet Med.* (2013) 15:673–83. doi: 10.1038/gim.2013.28
163. Cardy TJ, De Decker S, Kenny PJ, Volk HA. Clinical reasoning in canine spinal disease: what combination of clinical information is useful? *Vet Rec.* (2015) 177:171. doi: 10.1136/vr.102988

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REVIEW
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Emerging and Adjunctive Therapies for Spinal Cord Injury Following Acute Canine Intervertebral Disc Herniation

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Some dogs do not make a full recovery following medical or surgical management of acute canine intervertebral disc herniation (IVDH), highlighting the limits of currently available treatment options. The multitude of difficulties in treating severe spinal cord injury are well-recognized, and they have spurred intense laboratory research, resulting in a broad range of strategies that might have value in treating spinal cord-injured dogs. These include interventions that aim to directly repair the spinal cord lesion, promote axonal sparing or regeneration, mitigate secondary injury through neuroprotective mechanisms, or facilitate functional compensation. Despite initial promise in experimental models, many of these techniques have failed or shown mild efficacy in clinical trials in humans and dogs, although high quality evidence is lacking for many of these interventions. However, the continued introduction of new options to the veterinary clinic remains important for expanding our understanding of the mechanisms of injury and repair and for development of novel and combined strategies for severely affected dogs. This review outlines adjunctive or emerging therapies that have been proposed as treatment options for dogs with acute IVDH, including discussion of local or lesion-based approaches as well as systemically applied treatments in both acute and subacute-to-chronic settings. These interventions include low-level laser therapy, electromagnetic fields or oscillating electrical fields, adjunctive surgical techniques (myelotomy or durotomy), systemically or locally-applied hypothermia, neuroprotective chemicals, physical rehabilitation, hyperbaric oxygen therapy, electroacupuncture, electrical stimulation of the spinal cord or specific peripheral nerves, nerve grafting strategies, 4-aminopyridine, chondroitinase ABC, and cell transplantation.

Keywords: alternative therapies, interventions, dog, intervertebral disc disease, cell transplantation, spinal cord injury, canine

INTRODUCTION

Current treatment for acute canine intervertebral disc herniation (IVDH) can be divided into medical/conservative or surgical management. The decision as to which to pursue depends largely on the severity of neurologic signs. These might be due to either reversible or irreversible damage to the spinal cord itself, with some resulting from tissue ischemia that is difficult to counteract and others resulting from spinal cord compression that are easily reversible. Medical management commonly consists of activity restriction, pain, and anti-inflammatory medications. The goals are: (i) to avoid further disc herniation to minimize additional damage to the spinal cord; (ii) provide pain relief; (iii) allow the extruded disc material to gradually dissipate by phagocytosis over time; (iv) and leave the ruptured disc annulus to seal by fibrosis over time. Surgical intervention is used to alleviate persistent spinal cord compression. The reader is directed to the article, "Current approaches to the management of acute thoracolumbar disc extrusion in dogs" for more information regarding the evidence for the commonly applied treatment options.

Neither medical nor surgical management currently aim to repair the damaged intervertebral disc, nor heal the injured spinal cord, and there are limits to the recovery that can be attained. Severe injuries still result in incomplete recovery and unsatisfactory functional status. Although largely understandable, with restoration of function to spinal cord injured individuals recognized as a holy grail for centuries, this failure has prompted a vast effort in neuroscience research. The aim is to develop strategies that directly target the injury within the spinal cord, limit the extent of secondary injury, facilitate regeneration of axons, or increase compensatory plasticity of the surviving tissue. Many neuroprotective and neuro-regenerative therapies have shown promise in pre-clinical experimental models but few have made it beyond this phase and when they have, repeatedly failed to successfully translate to humans or dogs with naturally-occurring spinal cord injury (SCI) (1, 2). In fact, it has been reported that only about one-third of animal studies for any disease considered to have a high likelihood of translation into human medicine actually progressed to the clinical trial stage and even fewer were associated with any currently available intervention (3, 4). While this should not deter researchers and clinicians from seeking novel treatment options for the injured spinal cord, it does highlight the huge hurdles facing such work and underscores the difficulty of the problem. Importantly, knowledge of what has been done, successful or otherwise, is crucial to broadening our understanding mechanisms of injury and recovery, developing new techniques, or adapting and combining previously suggested treatment modalities for application to clinical populations.

This review will summarize adjunctive or emerging therapies that have been proposed as treatment options for dogs with acute IVDH. We will focus on data available from companion dogs with naturally-occurring SCI but also include data on experimental dogs where relevant. This is the information that we hope will be most useful to veterinary clinicians and might soon be applicable in the neurology clinic. We will focus on therapies that can be applied following acute SCI (within 1 month of injury) but also discuss therapies that might aid in repairing the spinal cord or restoring function in the subacute to chronic patient (> 1 month from injury). We have divided treatments in those delivered at or close to the injury site and those delivered systemically.

ADJUNCTIVE THERAPIES IMPLEMENTED IN THE ACUTE PHASE

Local/Lesion-Based Interventions

A variety of locally administered interventions have been proposed in the treatment of SCI that are applied directly to the lesion site in addition to, or *in lieu* of, decompressive surgery. These include laser therapy, application of electromagnetic fields or oscillating electrical fields, adjunctive surgical techniques, locally-applied spinal cord hypothermia, and cell transplantation. Cell transplantation strategies will be discussed in the subacute to chronic section in the second part of this review.

Laser Therapy

Low-level laser therapy or photobiomodulation has been a reported therapy for various injuries, including SCI. In the nervous system, it has been proposed to enhance neuronal metabolism and sprouting and to decrease glial scar formation and the immune response (5, 6). While not fully understood, the mechanisms of action are reported to include inhibition of NF-kB (which reduces expression of pro-inflammatory mediators) and stimulation of cytochrome oxidase (which might help to optimize oxidative metabolism) (5, 6). In an unblinded, unrandomized prospective study of non-ambulatory paraparetic or paraplegic dogs with IVDH, with or without intact pain perception at enrolment and treated surgically, laser therapy applied post-operatively (for 5 days or until independent ambulation was achieved) was compared to dogs that did not receive additional therapy. The reported time to achieve independent ambulation was shorter in the laser therapy group (3.5 days) compared to untreated control dogs (14 days) (7). However, the characteristics of the laser employed were not detailed making it difficult to try to replicate results. In contrast, a blinded, randomized prospective study evaluating post-operative laser therapy with or without physical rehabilitation in non-ambulatory dogs undergoing surgery for IVDH revealed no difference in recovery (8). Importantly, both studies included a relatively small number of dogs in each treatment group, including few with severe injury, did not incorporate pre-study sample size calculations, and only looked at short-term outcome variables. No adverse events attributable to laser therapy were reported.

Electromagnetic and Electrical Field Therapies

While application of a pulsed electromagnetic field (PEMF) device to the site of injury has been most widely studied in pain and wound repair, PEMF therapy has been reported to reduce back and neck pain in people and possibly improve recovery from SCI in an experimental model in cats (9–13). The mechanism of action of PEMF in pain relief is likely multifactorial and there is

limited evidence in central nervous system injury that it can aid in promoting axonal regeneration or sparing of surviving axons (9, 10). In a recent blinded, randomized prospective clinical trial of paraplegic dogs with absent pain perception secondary to IVDH that underwent surgery, PEMF reduced post-operative incisional pain (as measured by increased mechanical sensory thresholds) compared to sham-treated control dogs. The authors also reported a possible neurologic benefit based on measuring injury severity using plasma GFAP concentration and recovery of proprioceptive placing (14). However, sample size was small and multiple outcomes were evaluated.

Oscillating electrical field therapy, which is suggested to enhance axonal regrowth and improve functional recovery, has been applied to spinal cord-injured animals (15). In paraplegic deep pain negative dogs secondary to IVDH treated surgically, oscillating electrical field therapy was delivered post-operatively via electrodes sutured to the edges of the laminectomy site and attached to an implantable device. Treatment was administered for a variable number of weeks post-operatively and the device and therapy were well-tolerated. Dogs treated with the electrical fields had improved neurologic outcomes at 6 weeks and 6 months after surgery compared with sham-treated dogs (15, 16). Despite initial promise, logistical, and technical issues hindered further development of this treatment modality.

Local Hypothermia

Locally-applied spinal cord hypothermia has been rarely reported as a treatment for SCI in dogs in experimental studies (17–20). Hypothermic conditions (4–6°C) were applied to the spinal cord initiated at 15 min to 4 h after injury and maintained for variable durations ranging from 1 to 18 h. In these studies, hypothermia was reported to improve functional outcome in experimentally-injured dogs compared to untreated controls with a possible additive benefit in combination with other therapies. However, reported drawbacks included extensive technical and personnel demands, the potential for inadvertent damage to spinal cord through prolonged hypothermia and lack of information on long-term outcomes or sequelae (17). This technique has not been reported in dogs with naturally-occurring injury secondary to IVDH but mean body temperature was identified as an exploratory variable worthy of further evaluation in prospective studies in dogs with IVDH (21). Local and systemic hypothermia continue to be investigated in human medicine (22, 23).

Adjunctive Surgical Techniques to Spinal Cord Decompression: Durotomy and Myelotomy

The role and indications for decompressive surgery as well as fenestration as a standalone technique for acute IVDH are outlined in the companion article in this issue, "Current approaches to the management of acute thoracolumbar disc extrusion in dogs." Adjunctive surgical techniques of durotomy and myelotomy are summarized below.

Durotomy has been investigated as a means to decompress a swollen spinal cord, to improve spinal cord blood flow and oxygen delivery and to evaluate for gross myelomalacia as a prognostic indicator (24–27). Durotomy with or without duroplasty has been reported to have positive effects in multiple experimental rodent and human SCI studies; however, reported functional impact is variable, adverse effects are possible, and controlled studies are lacking (24). In experimental studies in dogs, immediate but not delayed (by 2 h) durotomy was reported to enhance recovery rate and overall neurologic outcome (25, 27, 28). In clinical canine patients, durotomy has typically been reserved for severely affected dogs. Blaser et al. demonstrated that durotomy combined with decompressive hemilaminectomy in dogs with IVDH (of varying severity ranging from ambulatory paraparesis to paraplegia with intact pain perception) transiently increased intraoperative spinal cord blood flow, although it returned to normal or lower within 15 min (26). There was no association detected between durotomy and 1-day post-operative neurologic outcome. However, none of the included dogs were those that have the most to benefit from durotomy (i.e., those paralyzed with absent pain perception at presentation), thereby potentially limiting the generalizability of these results. In an additional retrospective study of 48 paraplegic dogs that were deep pain negative secondary to IVDH, no difference was detected in recovery of ambulation between those that did or did not receive a durotomy in conjunction with hemilaminectomy (29), although confounding by severity is a clear possibility in this study.

More recently, contrasting evidence has been provided by Takahashi et al. who reported on 116 paraplegic deep pain negative dogs with thoracolumbar IVDH treated with hemilaminectomy alone ($n = 65$) or hemilaminectomy *plus* durotomy ($n = 51$) (30). A large proportion of dogs recovered following durotomy vs. hemilaminectomy alone (56.9 vs. 38.5%). The low rate of recovery in the non-durotomy group (compared to most published reports of a 50–60% success rate with decompressive surgery) was attributed to inclusion only of cases that had imaging features associated with poor prognosis. Notably, no dogs in the durotomy group compared to 14 in the hemilaminectomy-only group developed progressive myelomalacia. In another recent report, "extended durotomy" of four vertebral lengths centered over the site of herniation was also investigated in 26 consecutive paraplegic dogs that were deep pain negative secondary to thoracolumbar IVDH (31). Of the 26 dogs included in the study, 4 dogs were lost to follow-up while 16/22 remaining dogs recovered independent ambulation within 6 months (with 15/16 also recovering continence) (31). No adverse events were attributable to the extended durotomy; one dog developed progressive myelomalacia. These studies together reinvigorate the discussion as to whether durotomy might be beneficial in dogs with severe injury, especially in preventing development of progressive myelomalacia. Additional information is needed regarding single vs. extended durotomy, the role of duroplasty, patient selection among severely affected dogs, and the risk and functional impact of long-term consequences such as fibrosis that might negatively impact neurologic function.

Dorsal midline myelotomy has been reported as a treatment for SCI to decrease intramedullary pressure, increase the oxygen interface, remove necrotic debris, and release noxious vasoactive

substances trapped in the spinal cord post-injury (24, 32). In an experimental canine model, myelotomy in combination with dimethyl sulfoxide (DMSO) appeared to have an additive benefit on neurologic recovery compared to other experimental treatment combinations, although myelotomy alone was not evaluated (32). In another study on experimental SCI followed by myelotomy, there was immediate improvement in sensory evoked potential amplitude in 2/5 dogs (33), suggesting temporary improvement in conduction, but it is unclear if this is sustained or associated with functional benefit. Myelotomy performed in normal dogs has been associated with extensive gray matter necrosis including destruction of ventral horn motor neurons in some dogs (34). However, clinical impairment from the procedure was generally mild to moderate and improved over several weeks as long as the lumbar intumescence was avoided (34). While a positive effect has been reported in 80% of pre-clinical animal studies, there are no published studies in naturally-occurring injury in dogs and very limited data available in humans (24). The lack of controlled studies is likely attributable to the invasiveness of myelotomy and perceived potential to exacerbate secondary injury and for long-term adverse sequelae.

SYSTEMIC COMPOUND/MEDICATION-BASED THERAPIES

A variety of systemic or "whole dog" interventions have been applied to treat dogs with IVDH. Administration of some type of systemic medication or chemical as a neuroprotective strategy for the treatment of acute IVDH has been reported to be recommended by up to a quarter of specialist veterinarians (35). This varied greatly by treatment type, being highest for steroid administration (34% of boarded surgeons, 23% of boarded neurologists recommended) and <10% for other interventions (35). Adjunctive, non-medication-based therapies typically applied post-operatively were also variably reported as part of an integrated treatment strategy. Physical rehabilitation was most common and recommended by approximately half of treating veterinarians (35).

Corticosteroids

Corticosteroids are a commonly administered adjunctive therapy for the treatment of IVDH in dogs. Methylprednisolone (MPSS) at so-called "shock doses" has received the most attention and been most extensively examined but dexamethasone has also been investigated in dogs (17–19, 32, 36–43). MPSS has been advocated as a neuroprotective treatment for acute SCI through its mitigation of secondary injury primarily through amelioration of lipid peroxidation, other free radical, and oxidative damage and reperfusion injury (44). Although initial results of human clinical trials appeared supportive of use of high dose MPSS for treatment of SCI, subsequent re-analysis of the data cast doubt on the original treatment effect and highlighted risks of adverse effects (45–47). In dogs with IVDH, a benefit for MPSS has not been identified and complications have been reported (36, 42, 43, 48) and the use of MPSS remains controversial (46). The role of corticosteroids in this population is discussed in depth in the companion article "Current approaches to the management of acute thoracolumbar disc extrusion in dogs."

Polyethylene Glycol

Polyethylene glycol (PEG) a hydrophilic polymer capable of fusing cell membranes has been infrequently investigated as a treatment for acute SCI with inconsistent results. In an acute canine spinal cord transection model, immediate application of PEG at the site of injury was determined to be beneficial and to re-establish anatomic continuity (49). In a study of dogs with acute paraplegia with absent pain perception due to IVDH, intravenous PEG administration appeared safe, and associated with modestly improved neurologic status 6–8 weeks after injury and surgery compared to what might be expected in similarly affected dogs not receiving PEG (50). In a more recent clinical trial of acute paraplegic dogs with absent pain perception due to IVDH, no benefit was demonstrated for PEG compared to placebo (36).

Matrix Metalloproteinases

Matrix metalloproteinases (MMPs) are released by cells to degrade the extracellular matrix. MMPs, specifically MMP-9 and MMP-12, have been shown to be upregulated following SCI and are implicated in the deleterious secondary injury cascade. In two prospective studies, Levine et al. evaluated a broad-spectrum MMP inhibitor, GM6001, in dogs treated surgically for IVDH resulting in acute (<48 h) non-ambulatory paraparesis or paraplegia (51, 52). All dogs were treated immediately before decompressive surgery with the compound, GM6001, combined with DMSO ($n = 81$), DMSO alone ($n = 84$), placebo ($n = 41$), or received no treatment ($n = 20$). Transient injection site reactions were common in the GM6001 treated dogs (which could have compromised the blinding) and a subset ($n = 6$) developed self-limiting musculoskeletal signs, but it was otherwise well-tolerated. Treatment with GM6001 with DMSO resulted in improved neurologic recovery compared to placebo but was not different compared to DMSO alone. While efficacy for treatment with this MMP inhibitor was not demonstrated with regard to sensorimotor recovery, it did increased long-term bladder compliance.

Dimethyl Sulfoxide

Dimethyl sulfoxide (DMSO) is most often used as a vehicle to improve drug solubility but has been uncommonly investigated as an intervention for brain and SCI (38, 52). Its purported benefit in central nervous system trauma has been attributed to a reduction in edema, diuretic, anti-inflammatory and vasodilatory effects, and cellular protection from mechanical damage (38, 53). In several studies utilizing an experimental weight drop model, dogs were treated with DMSO (1–4.5 g/kg/d in 40% solution with 0.9% NaCl) alone or in combination with other experimental therapies and compared to control dogs (38, 53–55). In most, DMSO was reported to be beneficial when initially administered within 1 h of induced trauma although one study reported no benefit and no clear synergistic effect was observed by

combining DMSO with dexamethasone or other experimental therapies. As outlined above, DMSO (1 g/kg) was shown to be beneficial compared to placebo in a clinical trial of dogs with IVDH (52). However, Hoerlein et al. also investigated this in acute spinal cord trauma in dogs and found it not to be useful compared to dexamethasone (56). Toxicity due to DMSO was not observed in any of the reported studies and further investigation is warranted regarding its potential therapeutic effect in this population.

Other Compounds/Medications

N-acetylcysteine (NAC) is a precursor of glutathione with potent antioxidant as well as anti-inflammatory and neuronal protective properties that has been proposed as a treatment for acute SCI (57). In a cohort of 70 dogs undergoing surgery for acute IVDH, NAC administered IV prior to decompressive surgery showed no benefit compared to placebo with regard to neurologic outcome or rate or recovery (58). There is only anecdotal reference of veterinarians using other antioxidants such as coenzyme Q10 or vitamin E following SCI in dogs (35, 59). While optimizing nutritional status, weight management, and diets to reduce fecal volume in incontinent dogs are variably implemented as part of post-injury management in dogs, there is no evidence to support specific antioxidant nutraceutical supplementation or nutritional strategies to treat dogs with acute SCI.

There are other rarely reported interventions with limited, mostly experimental evidence in dogs. Analogs of the hypothalamic hormone, thyrotropin releasing hormone (TRH), have been reported to inconsistently improve outcomes after SCI in humans and experimental models (59). In a pilot study of dogs with IVDH, a benefit of a TRH analog was not identified compared to no treatment (60). Crocetin, a carotenoid that increases oxygen diffusion in plasma, was investigated in an experimental weight drop model as a means to counteract local hypoxia and subsequent ischemic necrosis following SCI. Results showed improved neurologic function in crocetin-treated dogs at 4 weeks post-injury compared to control dogs (61). Hyperosmotic agents, mannitol, urea and hypertonic dextrose, have also been evaluated with the goal of reducing swelling (32, 53, 54, 62) but did not appear to improve neurologic recovery compared with control dogs. Improvement in spinal sensory evoked potentials did occur following mannitol infusion in one study (32, 53, 54, 62). Phenytoin, an anticonvulsant, was explored as a SCI treatment based on experimental evidence that it decreases edema of neural tissues through inhibition of antidiuretic hormone and inactivation of catecholamines (40). In an experimental dog model, phenytoin resulted in improved outcome compared to untreated dogs and was at least as effective as dexamethasone, although hypotension and respiratory depression were possible adverse effects (40). Neither reserpine, an alkaloid medication used to treat high blood pressure, nor chlorpromazine, a phenothiazine with various psychiatric and other uses, were effective as treatments for experimentally induced injury in dogs (32, 39). There is no convincing evidence for the use of these compounds in dogs with acute SCI due to IVDH.

SYSTEMIC NON-MEDICATION-BASED THERAPIES

Various systemic, non-medication-based therapies have been advocated for the treatment of acute SCI in dogs including physical rehabilitation, hyperbaric oxygen therapy, and electroacupuncture.

Physical Rehabilitation

Physical rehabilitation in dogs recovering from surgery due to IVDH is being increasingly utilized, recommended by 58% of board-certified surgeons and neurologists surveyed (35, 63). While timing of initiation and specific protocols vary, physical rehabilitation in the neurologic patient typically consists of some combination of passive range of motion, massage, cold or warm packing, assisted balance, standing, coordination and land treadmill, or over-ground walking exercises and aquatic therapy such as underwater treadmill walking or swimming (64). It can be performed on an in-patient or out-patient basis in dogs with specific aspects tailored to patient function (e.g., underwater treadmill walking sessions are typically initiated once motor function is present). Additional specific therapies including therapeutic laser therapy, acupuncture, and neuromuscular electrical stimulation are variably included (63, 64).

For details on the currently available evidence regarding the role of physical rehabilitation in dogs recovering from IVDH, the reader is referred to the companion article "Current approaches to the management of acute thoracolumbar disc extrusion in dogs" in this issue. While there have been relatively few studies performed in this population and the results have been mixed, early post-operative initiation of rehabilitation has been determined to be safe with no associated adverse events or increased post-operative pain (8, 65–70). Inclusion of dogs of variable neurologic severity limits making direct comparisons between studies and conclusions regarding efficacy. Additionally, the role of physical rehabilitation in medically managed presumptive or confirmed IVDH has not been evaluated. Additional investigation of physical rehabilitation in dogs recovering from IVDH is warranted focusing on optimization of protocols (e.g., specific modalities, timing, and duration) and development of validated, objective outcome measures such as the Finnish neurological function testing battery for dogs (FINFUN) (71).

Hyperbaric Oxygen Therapy

Hyperbaric oxygen therapy has been uncommonly reported as a treatment for acute SCI. The proposed mechanism is to increase the tissue partial pressure of oxygen and counteract the adverse effects of spinal cord hypoxia associated with injury (72). There is limited experimental evidence in dogs suggesting a potential benefit of hyperbaric oxygen therapy compared to untreated controls but no additive effect was appreciated when combined with DMSO (54, 72). There are no reports in dogs with IVDH though it is used in clinical cases by some veterinarians (35).

Electroacupuncture

Electroacupuncture has been occasionally reported as a therapy for acute SCI in dogs (73, 74). Its mechanism of action is unknown, but it might have analgesic and anti-inflammatory effects as well as facilitating axonal repair and regrowth (37, 63). In an experimental canine SCI model, electroacupuncture (initiated 48 h post-injury and continued every other day) resulted in improved rate of recovery compared to untreated controls; the benefit appeared synergistic with concurrent MPSS (37). In one retrospective and two prospective case series of dogs with thoracolumbar IVDH of variable severity, electroacupuncture (administered 1–3 ×/week for 1–6 months between studies) was reported to be more effective than decompressive surgery alone for regaining ambulation (75), and was associated with shorter time to walking and a greater proportion of dogs becoming ambulatory compared to medical management alone (73, 74). There was no significant difference in recovery among deep pain negative dogs managed medically with or without electroacupuncture (74). Study limitations for the prospective studies included lack of blinding or randomization, use of historical controls and small sample size within each neurologic grade (74, 75). There is equivocal evidence that electroacupuncture decreases the severity and duration of post-operative pain in dogs with IVDH (74, 76). Electroacupuncture has also been combined with stem cell transplantation in a small group of dogs chronically (> 3 months) deep pain negative following acute IVDH (77). This pilot study showed these interventions were feasible and safe but case numbers in each treatment group were small.

ADJUNCTIVE THERAPIES IMPLEMENTED IN THE SUBACUTE-TO-CHRONIC PHASE

Treatment strategies are also being explored for dogs with permanent impairment following acute SCI. These are typically applied in the subacute-to-chronic stage (> 1 month from the time of injury) and include spinal cord radiation, electrical stimulation of the spinal cord or specific nerves below the injury, nerve grafting, 4-aminopyridine, chondroitinase ABC delivery, and cell transplantation.

Spinal Cord Radiation

Spinal cord radiation has been evaluated in rodents and in an experimental model in Beagles (78). Radiation of the injured cord aims to interfere with the cell cycle to counteract the development of localized chronic inflammation, reduce glial scar formation, and facilitate axonal regrowth and healing (78, 79). In Beagles treated with daily radiation for 2 weeks following spinal cord hemitransection, there was reduced astrocyte and microglial activation, reduced expression of inflammatory mediators, improved long-range axonal regeneration, and improved locomotor recovery (79). This therapy has not been reported in dogs with naturally-occurring injury secondary to IVDH.

Functional Electrical Stimulation

Functional electrical stimulation has not yet been reported in dogs, but a wearable device is being developed that might be useful in dogs with incomplete recovery from acute IVDH (80). Short-term, low-intensity electrical stimulation of the spinal cord with or without stem cell transplantation has been performed in chronically paraplegic dogs (81). While electromyographic changes in pelvic limb muscles implied improvement in motor conduction, further investigation would be necessary to optimize therapy and determine if there is a clinical benefit (81). Additionally, there is experimental evidence and data in dogs with naturally-occurring injury for electrical stimulation of peripheral nerves or nerve roots to aid in restoration of urination and defecation (82–84). Electrical sacral nerve stimulation in dogs is covered in the companion article "Bladder and bowel management in dogs with spinal cord injury" in this issue.

Peripheral Nerve Grafting

Grafting of peripheral nerves from above the injury level (e.g., specific intercostal nerves) into the distal portion of experimentally transected spinal cord has also been performed in dogs with the goal of harnessing the regenerative potential of the peripheral nervous system (85–87). Nerve to nerve or nerve root grafting techniques have also been reported as therapies that aim to restore function while bypassing the spinal cord lesion directly. Toreih demonstrated the feasibility of intercostal to gluteal nerves and ilioinguinal and iliohypogastric to femoral nerves in a dog spinal cord hemisection model (88). Six months following these nerve transfer procedures, there was clinical and electrophysiological evidence of some recovery of hip, gluteal and knee function, though spontaneous improvement is well-known to occur in spinal cord hemisection models and could have resulted in the improvement observed. Vagophrenic nerve anastomosis was also shown to be anatomically feasible in dogs with the ultimate goal to provide a conduit for restoration of respiratory function after severe cervical SCI (89). Nerve anastomoses to reinstate bladder function have also been performed experimentally in dogs (90–94).

Molecular Compounds Given in the Chronic Phase of SCI

Additional compounds that have been investigated in chronically impaired dogs include 4-aminopyridine (4AP) and chondroitinase ABC. 4-aminopyridine is a potassium channel antagonist that has been shown to restore hind limb motor function in some dogs with chronic thoracolumbar SCI (95, 96). This effect is mediated through enhancement of central conduction via anatomically intact axons traversing the site of injury as well as direct synaptic effects (97–99). Response following oral administration is highly variable between individual dogs with a minority regaining independent ambulation (96, 98). Lack of predictable response and narrow therapeutic window have limited widespread use of this medication among chronically paralyzed dogs.

Chondroitinase ABC is an enzyme that degrades chondroitin sulfate proteoglycans which are key components of the glial scar and inhibitors of axonal regeneration following SCI (100). This

has led to active research regarding the use and optimization of chondroitinase ABC to treat SCI (100). A prospective clinical trial of intraspinal injection of a long acting chondroitinase ABC in dogs with naturally occurring severe SCI reported functional improvements compared to sham controls including improved thoracic to pelvic limb coordination and three dogs with restoration of ambulation (101). In an experimental canine model, the combination of chondroitinase ABC with mesenchymal stem cell transplantation was also reported to improve neurologic deficits and enhance neural regeneration (102). However, this study had a small number of dogs, lacked blinding of the observations and the locomotor outcome measure might not reflect voluntary movements (102).

CELL TRANSPLANTATION STRATEGIES

Transplantation of cells into the spinal cord has been investigated in dogs either after creating an experimental spinal cord lesion or after naturally-occurring SCI. This involves predominantly stem cell therapies such as mesenchymal stem cells of various origin, neural stem cells, or bone marrow-derived mononuclear cells but other fully differentiated cells have been used such as olfactory glial cells, olfactory mucosal cells, Schwann cells, or macrophages. Most commonly, the cell transplants are administered via intraparenchymal or intrathecal injection but intravenous delivery has also been reported.

Cell Transplantation for Spinal Cord Repair in Experimental Dogs

Placement and inflation of a ventral epidural balloon has been used to produce experimental compression and contusion of the dog spinal cord (103, 104). Developed in the seventies by investigators such as Kobrine and Griffiths, this technique has the advantage of producing a closed injury, without the need to open the spinal canal via a laminectomy, and causes more vascular injury (ischemia) than weight-drop models (105, 106), although lesions lack reproducibility. Using this model, the effect of canine and human umbilical cord blood-derived mesenchymal stem cells, adipose-derived stem cells (some genetically modified), or bone marrow-derived mesenchymal stem cells (107–117) has been tested. These cell transplants were reported to improve locomotor function, but experimental groups consisted of small numbers (between two and five), observers were not blinded and tail support was used when testing locomotion [which is likely to trigger "involuntary" stepping pelvic movements that are independent from brain connections (118, 119)]. However, histopathological data demonstrated survival of some transplanted cells, albeit with limited integration within the host spinal cord, suggesting that the locomotor improvement could have been due to secretion of trophic or growth factors into the region of injury (107).

A canine hemisection model in which a gel seeded with human neural stem cells was placed immediately into the hemisected spinal cord gap (120, 121) showed better locomotor recovery and more ascending sensory axons in dogs receiving cells alone in one study (121) and no effect in another study (122).

A canine transection model has been reported from groups in China recently, testing the effect of collagen-based biomaterial loaded with human umbilical cord-derived mesenchymal stem cells (123), human placenta-derived mesenchymal stem cells (124), or bone marrow-derived mesenchymal stem cells differentiated into neuron-like cells (125). Recipients of cells had improved locomotor scores compared to controls but remained non-ambulatory and the studies were not blinded. Interestingly, cells survived up to 6 months.

A group from Egypt described a compression/contusion model using a "clip" placed on the spinal cord at the L4 spinal cord segment; neural-induced bone marrow derived stem cells were then injected intrathecally by lumbar puncture 2 weeks after the injury by a blinded investigator (126). The injury initially caused paralysis and loss of pain perception in all dogs, but those receiving the cell transplant had much greater recovery of motor function compared to controls. Further, cells could be found surviving in the lesion at 16 weeks after injection. This work represents a lesion and intervention paradigm that are much closer to clinical injuries than other experimental models.

In summary, there is a growing number of transplantation experiments originating primarily from Korea and China which have generally low power and follow the same experimental pattern with varying cell types. One group in Korea translated their experiment to the clinic by transplanting adipose-derived stem cells (112) to 9 companion dogs with paraplegia and no deep pain (127) but trial design lacked clear inclusion criteria and blinding. Therefore, the utility of these treatments for clinical populations remains to be validated in randomized, blinded studies. A cautious approach has been followed by other laboratories, for example, McMahill et al. at the University of California Davis Medical Center, where they have transplanted canine epidermal neural crest stem into normal canine spinal cord (128) and focused on developing strong outcome measures using cell tracking with magnetic resonance imaging and detailed gait analysis. They showed survival of cells at 3 weeks post-transplantation and are likely now envisaging clinical trials in companion dogs with naturally-occurring lesions. There is also uncertainty as to which cell type, stem cells or other differentiated cells, should be prioritized. For example, two other groups have postulated that transplantation of Schwann cells purified from peripheral nerves or nerve roots could be a repair strategy worth pursuing following SCI in dogs (129, 130). Additionally, transplantation strategies can be leveraged to investigate application of biologics (e.g., chondroitinase ABC) to the lesion site as a means to promote cell survival, regrowth, or mitigate inhibition of axonal regeneration.

Cell Transplantation Within the Spinal Cord in Dogs With Naturally-Occurring Injury

One of the best known cell type studied in dogs is olfactory ensheathing cells, which are not stem cells but fully differentiated cells located within the olfactory mucosa and olfactory bulb, forming an interface between the peripheral and central nervous

systems (131–133). Olfactory ensheathing cells have been reliably cultured for a long time in neuroscience (134), including in dogs (135–138) and have been used in clinical applications (139, 140). They are recognized for their regenerative properties when transplanted within a lesion of the central nervous system. In particular, they are able to form channels guiding axonal regrowth (141) and to remyelinate axons (142). In a randomized controlled trial in dogs with irreversible chronic SCI, autologous olfactory ensheathing cells obtained from the nasal mucosa have been shown to improve thoracic to pelvic limb coordination (143). However, these cells were not able to restore brain-controlled functions such as urinary continence, prompting research into strategies to improve their efficacy. More recently, olfactory ensheathing cells have been engineering to express the chondroitinase ABC enzyme that degrades the glial scar (144), though these have not yet been transplanted into dogs.

In the last decade, there has been an increasing number of publications from Japan, Brazil, Turkey, and India testing different cell transplants in dogs with naturally-occurring injury. First in Japan, two groups reported that autologous bone marrow stromal or mononuclear cell transplants were safe in 7 (145) and 1 (146) dogs with chronic paraplegia and absent deep pain.

Since 2014, institutions in Brazil have reported seven trials testing safety or efficacy of various cell transplants (within the spinal cord or in the sub-arachnoid or epidural space) in small series of companion dogs, sometimes with concomitant spinal cord decompression, or in association with other alternative therapies such as electroacupuncture (77) or low-intensity electrical stimulation (81). The cells tested have been either autologous bone marrow mesenchymal stem cells (147), allogenic fetal bone marrow stem cells (148), allogenic canine adipose tissue-derived mesenchymal stem cells (81, 149, 150) or immature dental pulp stem cells (77, 151).

The follow-up duration in these studies was usually of several months. Taken together, the results suggest an improvement of locomotor function, based on an increase in locomotor scores. However, these cases rarely achieved scores suggesting unassisted ambulation and for those that did walk again, they remained deep pain negative suggesting that the locomotion could have been independent of the therapeutic intervention (i.e., "spinal walking"). Interestingly, in some dogs, there was reported recovery of deep pain (147, 148) but no concomitant recovery of locomotion. These findings could indicate that the transplanted cells have a beneficial effect. However, data are limited and these studies also illustrate the heterogeneity of clinical lesions and the need to increase case numbers to better assess the efficacy of cell transplant techniques.

In Turkey, Besalti et al. transplanted intramedullary neurogenically-induced bone marrow-derived mesenchymal stem cells 42 days after the initial injury (152). They conducted detailed follow-up over 12 months and found that 2 out of 13 dogs recovered somatosensory evoked potentials and magnetic motor evoked potentials, while some other dogs had improved gait scores (6/13) and regained deep pain sensation. Bhat et al. in India also reported a trial in 44 dogs testing bone marrow mesenchymal stem cells without decompressive surgery (153). The authors claimed improved deep pain sensation and locomotion, but the change compared to the control group remained clinically small.

Altogether, the results of various cell transplantation studies in dogs are encouraging, although most studies remain of low power and preliminary. They have proven safety, but the recovery is always limited to a proportion of studied dogs and recovery of one function at a time, either locomotion, continence, or pain perception. This perhaps suggests that other factors than the actual treatment led to the change in function and highlights the severity of lesions and difficulties in repairing them. A consensus on which intervention holds the greatest promise would be useful to then apply in large multicenter trials in dogs, where evaluation of efficacy could be investigated with greater power.

CONCLUSIONS

In conclusion, we have outlined a variety of therapeutic strategies that have been applied to dogs with SCI in both the acute as well as subacute-to-chronic settings. These range from those applied to the spinal cord directly to systemic treatments and with variable goals from repair to compensation. While some techniques are more promising than others, they all serve to highlight the challenges in treating severe SCI and in developing successful treatment options for a heterogeneous clinical population. Moving forward, multimodal approaches to therapy building on conventional treatment options will likely be most successful.

AUTHOR CONTRIBUTIONS

ML, NG, and NJ participated in manuscript conception, preparation, and editing with the first two (ML and NG) contributing equally. The additional members of the CANSORT-SCI consortium contributed to manuscript conception, editing, and review.

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REFERENCES

1. Burns TC, Verfaillie CM. From mice to mind: strategies and progress in translating neuroregeneration. *Eur J Pharmacol.* (2015) 759:90–100. doi: 10.1016/j.ejphar.2015.03.041
2. Jeffery ND, Smith PM, Lakatos A, Ibanez C, Ito D, Franklin RJM. Clinical canine spinal cord injury provides an opportunity to examine the issues in translating laboratory techniques into practical therapy. *Spinal Cord.* (2006) 44:584–93. doi: 10.1038/sj.sc.3101912
3. Bart van der Worp H, Howells DW, Sena ES, Porritt MJ, Rewell S, O'Collins V, et al. Can animal models of disease reliably inform human studies? *PLoS Med.* (2010) 7:e1000245. doi: 10.1371/journal.pmed.1000245
4. Hackam DG, Redelmeier DA. Translation of research evidence from animals to humans. *J Am Med Assoc.* (2006) 296:1731–2. doi: 10.1001/jama.296.14.1731
5. Hamblin MR. Mechanisms and applications of the anti-inflammatory effects of photobiomodulation. *AIMS Biophys.* (2017) 4:337–61. doi: 10.3934/biophy.2017.3.337
6. Hashmi JT, Huang YY, Osmani BZ, Sharma SK, Naeser MA, Hamblin MR. Role of low-level laser therapy in neurorehabilitation. *PMR.* (2010) 2:S292–305. doi: 10.1016/j.pmrj.2010.10.013
7. Draper WE, Schubert TA, Clemmons RM, Miles SA. Low-level laser therapy reduces time to ambulation in dogs after hemilaminectomy: a preliminary study. *J Small Anim Pract.* (2012) 53:465–9. doi: 10.1111/j.1748-5827.2012.01242.x
8. Bennaim M, Porato M, Jarleton A, Hamon M, Carroll JD, Gommeren K, et al. Preliminary evaluation of the effects of photobiomodulation therapy and physical rehabilitation on early postoperative recovery of dogs undergoing hemilaminectomy for treatment of thoracolumbar intervertebral disk disease. *Am J Vet Res.* (2017) 78:195–206. doi: 10.2460/ajvr.78.2.195
9. Crowe MJ, Sun ZP, Battocletti JH, Macias MY, Pintar FA, Maiman DJ. Exposure to pulsed magnetic fields enhances motor recovery in cats after spinal cord injury. *Spine.* (2003) 28:2660–6. doi: 10.1097/01.BRS.0000099385.46102.0D
10. Hattapoglu E, Batmaz I, Dilek B, Karakoç M, Em S, Çevik R. Efficiency of pulsed electromagnetic fields on pain, disability, anxiety, depression, and quality of life in patients with cervical disc herniation: a randomized controlled study. *Turkish J Med Sci.* (2019) 49:1095–101. doi: 10.3906/sag-1901-65
11. Lisi AJ, Scheinowitz M, Saporito R, Onorato A. A pulsed electromagnetic field therapy device for non-specific low back pain: a pilot randomized controlled trial. *Pain Ther.* (2019) 8:1330140. doi: 10.1007/s40122-019-0119-z
12. Sorrell RG, Muhlenfeld J, Moffett J, Stevens G, Kesten S. Evaluation of pulsed electromagnetic field therapy for the treatment of chronic postoperative pain following lumbar surgery: a pilot, double-blind, randomized, sham-controlled clinical trial. *J Pain Res.* (2018) 11:1209–22. doi: 10.2147/JPR.S164303
13. Alzayed KA, Alsaadi SM. Efficacy of pulsed low-frequency magnetic field therapy on patients with chronic low back pain: a randomized double-blind placebo-controlled trial. *Asian Spine J.* (2020) 14:33–42. doi: 10.31616/asj.2019.0043
14. Zidan N, Fenn J, Griffith E, Early PJ, Mariani CL, Muñana KR, et al. The effect of electromagnetic fields on post-operative pain and locomotor recovery in dogs with acute, severe thoracolumbar intervertebral disc extrusion: a randomized placebo-controlled, prospective clinical trial. *J Neurotrauma.* (2018) 35:1–11. doi: 10.1089/neu.2017.5485
15. Borgens RB, Toombs JP, Blight AR, McGinnis ME, Bauer MS, Widmer WR, et al. Effects of applied electric fields on clinical cases of complete paraplegia in dogs. *Restor Neurol Neurosci.* (1993) 5:305–22. doi: 10.3233/RNN-1993-55601
16. Borgens RB, Toombs JP, Breur G, Widmer WR, Waters D, Harbath AM, et al. An imposed oscillating electrical field improves the recovery of function in neurologically complete paraplegic dogs. *J Neurotrauma.* (1999) 16:639–57. doi: 10.1089/neu.1999.16.639
17. Ducker TB, Hamit HF. Experimental treatments of acute spinal cord injury. *J Neurosurg.* (1969) 30:693–7. doi: 10.3171/jns.1969.30.6.0693
18. Hansebout RR, Kuchner EF, Romero Sierra C. Effects of local hypothermia and of steroids upon recovery from experimental spinal cord compression injury. *Surg Neurol.* (1975) 4:531–6.
19. Kuchner EF, Hansebout RR. Combined steroid and hypothermia treatment of experimental spinal cord injury. *Surg Neurol.* (1976) 6:371–6.
20. Wells JD, Hansebout RR. Local hypothermia in experimental spinal cord trauma. *Surg Neurol.* (1978) 10:200–4.
21. Fenn J, Laber E, Williams K, Rousse CA, Early PJ, Mariani CL, et al. associations between anesthetic variables and functional outcome in dogs with thoracolumbar intervertebral disk extrusion undergoing decompressive hemilaminectomy. *J Vet Intern Med.* (2017) 31:814–24. doi: 10.1111/jvim.14677
22. Martirosyan NL, Patel AA, Carotenuto A, Kalani MYS, Bohl MA, Preul MC, et al. The role of therapeutic hypothermia in the management of acute spinal cord injury. *Clin Neurol Neurosurg.* (2017) 154:79–88. doi: 10.1016/j.clineuro.2017.01.002

23. Alkabie S, Boileau AJ. The role of therapeutic hypothermia after traumatic spinal cord injury - a systematic review. *World Neurosurg.* (2016) 86:432–49. doi: 10.1016/j.wneu.2015.09.079
24. Telemacque D, Zhu FZ, Ren ZW, Chen KF, Drepaul D, Yao S, et al. Effects of durotomy vs. myelotomy in the repair of spinal cord injury. *Neural Regen Res.* (2020) 15:1814–20. doi: 10.4103/1673-5374.280304
25. Allen AR. Surgery of experimental lesion of spinal cord equivalent to crush injury of fracture dislocation of spinal column: a preliminary report. *J Am Med Assoc.* (1911) LVII:878–80. doi: 10.1001/jama.1911.04260090100008
26. Blaser A, Lang J, Henke D, Doherr MG, Adami C, Forterre F. Influence of durotomy on laser-doppler measurement of spinal cord blood flow in chondrodystrophic dogs with thoracolumbar disk extrusion. *Vet Surg.* (2012) 41:221–7. doi: 10.1111/j.1532-950X.2011.00909.x
27. Parker AJ, Smith CW. Functional recovery from spinal cord trauma following incision of spinal meninges in dogs. *Res Vet Sci.* (1974) 16:276–9. doi: 10.1016/S0034-5288(18)33725-1
28. Parker AJ, Smith CW. Functional recovery from spinal cord trauma following delayed incision of spinal meninges in dogs. *Res Vet Sci.* (1975) 18:110–2. doi: 10.1016/S0034-5288(18)33641-5
29. Loughin CA, Dewey CW, Ringwood PB, Pettigrew RW, Kent M, Budsberg SC. Effect of durotomy on functional outcome of dogs with type I thoracolumbar disc extrusion and absent deep pain perception. *Vet Comp Orthop Traumatol.* (2005) 18:141–6. doi: 10.1055/s-0038-1632953
30. Takahashi F, Honnami A, Toki M, Dosaka A, Fujita Y, Hara Y, et al. Effect of durotomy in dogs with thoracolumbar disc herniation and without deep pain perception in the hind limbs. *Vet Surg.* (2020) 49:860–9. doi: 10.1111/vsu.13409
31. Jeffery ND, Mankin JM, Ito D, Boudreau CE, Kerwin SC, Levine JM, et al. Extended durotomy to treat severe spinal cord injury after acute thoracolumbar disc herniation in dogs. *Vet Surg.* (2020) 49:884–93. doi: 10.1111/vsu.13423
32. Rucker NC, Lumb W V., Scott RJ. Combined pharmacologic and surgical treatments for acute spinal cord trauma. *Am J Vet Res.* (1981) 42:1138–42.
33. Isu T, Iwasaki Y, Akino M, Abe H. Effect of mannitol administration and myelotomy on acute experimental spinal cord injury: investigation by spinal cord evoked potential. *Neurol Surg.* (1990) 18:267–72.
34. Teague HD, Brasmer TH. Midline myelotomy of the clinically normal canine spinal cord. *Am J Vet Res.* (1978) 39:1584–90.
35. Moore SA, Early PJ, Hettlich BF. Practice patterns in the management of acute intervertebral disc herniation in dogs. *J Small Anim Pract.* (2016) 57:409–15. doi: 10.1111/jsap.12496
36. Olby NJ, Muguet-Chanoit AC, Lim JH, Davidian M, Mariani CL, Freeman AC, et al. A placebo-controlled, prospective, randomized clinical trial of polyethylene glycol and methylprednisolone sodium succinate in dogs with intervertebral disk herniation. *J Vet Intern Med.* (2016) 30:206–14. doi: 10.1111/jvim.13657
37. Yang JW, Jeong SM, Seo KM, Nam TC. Effects of corticosteroid and electroacupuncture on experimental spinal cord injury in dogs. *J Vet Sci.* (2003) 4:97–101. doi: 10.4142/jvs.2003.4.1.97
38. de la Torre JC, Johnson CM, Goode DJ, Mullan S. Pharmacologic treatment and evaluation of permanent experimental spinal cord trauma. *Neurology.* (1975) 25:508–14. doi: 10.1212/WNL.25.6.508
39. Parker AJ, Smith CW. Functional recovery from spinal cord trauma following dexamethazone and chlorpromazine therapy in dogs. *Res Vet Sci.* (1976) 21:246–7. doi: 10.1016/S0034-5288(18)33383-6
40. Gerber AM, Olson WL, Harris JH. Effect of phenytoin on functional recovery after experimental spinal cord injury in dogs. *Neurosurgery.* (1980) 7:472–6. doi: 10.1227/00006123-198011000-00009
41. Coates JR, Sorjonen DC, Simpson ST, Cox NR, Wright JC, Hudson JA, et al. Clinicopathologic EFFECTS of a 21-Aminosteroid compound (U74389G) and high-dose methylprednisolone on spinal cord function after simulated spinal cord trauma. *Vet Surg.* (1995) 24:128–39. doi: 10.1111/j.1532-950X.1995.tb01307.x
42. Laitinen OM, Puerto DA. Surgical decompression in dogs with thoracolumbar intervertebral disc disease and loss of deep pain perception: a retrospective study of 46 cases. *Acta Vet Scand.* (2005) 46:79–85. doi: 10.1186/1751-0147-46-79
43. Levine JM, Levine GJ, Boozer L, Schatzberg SJ, Platt SR, Kent M, et al. Adverse effects and outcome associated with dexamethasone administration in dogs with acute thoracolumbar intervertebral disk herniation: 161 cases (2000-2006). *J Am Vet Med Assoc.* (2008) 232:411–7. doi: 10.2460/javma.232.3.411
44. Hall ED. The neuroprotective pharmacology of methylprednisolone. *J Neurosurg.* (1992) 76:13–22. doi: 10.3171/jns.1992.76.1.0013
45. Evaniew N, Belley-Côté EP, Fallah N, Noonan VK, Rivers CS, Dvorak MF. Methylprednisolone for the treatment of patients with acute spinal cord injuries: a systematic review and meta-analysis. *J Neurotrauma.* (2016) 33:468–81. doi: 10.1089/neu.2015.4192
46. Fehlings MG, Wilson JR, Harrop JS, Kwon BK, Tetreault LA, Arnold PM, et al. Efficacy and safety of methylprednisolone sodium succinate in acute spinal cord injury: a systematic review. *Glob Spine J.* (2017) 7:116S−37S. doi: 10.1177/2192568217706366
47. Khan MF, Burks SS, Al-Khayat H, Levi AD. The effect of steroids on the incidence of gastrointestinal hemorrhage after spinal cord injury: a case-controlled study. *Spinal Cord.* (2014) 52:58–60. doi: 10.1038/sc.2013.122
48. Boag AK, Otto CM, Drobatz KJ. Complications of methylprednisolone sodium succinate therapy in dachshunds with surgically treated intervertebral disc disease. *J Vet Emerg Crit Care.* (2001) 11:105–10. doi: 10.1111/j.1476-4431.2001.tb00076.x
49. Ren S, Liu Z, Kim CY, Fu K, Wu Q, Hou L, et al. Reconstruction of the spinal cord of spinal transected dogs with polyethylene glycol. *Surg Neurol Int.* (2019) 26:59. doi: 10.25259/SNI-73-2019
50. Laverty PH, Leskovar A, Breur GJ, Coates JR, Bergman RL, Widmer WR, et al. A preliminary study of intravenous surfactants in paraplegic dogs: polymer therapy in canine clinical SCI. *J Neurotrauma.* (2004) 21:1767–77. doi: 10.1089/neu.2004.21.1767
51. Levine JM, Cohen ND, Fandel TM, Levine GJ, Mankin J, Griffin JF, et al. early blockade of matrix metalloproteinases in spinal-cord-injured dogs results in a long-term increase in bladder compliance. *J Neurotrauma.* (2017) 34:2656–67. doi: 10.1089/neu.2017.5001
52. Levine JM, Cohen ND, Heller M, Fajt VR, Levine GJ, Kerwin SC, et al. Efficacy of a metalloproteinase inhibitor in spinal cord injured dogs. *PLoS ONE.* (2014) 9:e96408. doi: 10.1371/journal.pone.0096408
53. Kajihara K, Kawanaga H, De la Torre JC, Mullan S. Dimethyl sulfoxide in the treatment of experimental acute spinal cord injury. *Surg Neurol.* (1973) 13:273–6.
54. de la Torre JC, Kawanaga HM, Johnson CM, Goode DJ, Kajihara K, Mullan S. Dimethyl sulfoxide in central nervous system trauma. *Ann N Y Acad Sci.* (1975) 243:362–89. doi: 10.1111/j.1749-6632.1975.tb25377.x
55. Parker AJ, Smith CW. Lack of functional recovery from spinal cord trauma following dimethylsulphoxide and epsilon amino caproic acid therapy in dogs. *Res Vet Sci.* (1979) 27:253–5. doi: 10.1016/S0034-5288(18)32844-3
56. Hoerlein BF, Redding RW, Hoff EJ, McGuire JA. Evaluation of dexamethasone, DMSO, mannitol, and solcoseryl in acute spinal cord trauma. *J Am Anim Hosp Assoc.* (1983) 19:216–25.
57. Zafarullah M, Li WQ, Sylvester J, Ahmad M. Molecular mechanisms of N-acetylcysteine actions. *Cell Mol Life Sci.* (2003) 60:6–20. doi: 10.1007/s000180300001
58. Baltzer WI, McMichael MA, Hosgood GL, Kerwin SC, Levine JM, Steiner JM, et al. Randomized, blinded, placebo-controlled clinical trial of n-acetylcysteine in dogs with spinal cord trauma from acute intervertebral disc disease. *Spine.* (2008) 33:1397–402. doi: 10.1097/BRS.0b013e3181753c37
59. Park EH, White GA, Tieber LM. Mechanisms of injury and emergency care of acute spinal cord injury in dogs and cats. *J Vet Emerg Crit Care.* (2012) 22:160–78. doi: 10.1111/j.1476-4431.2012.00723.x
60. Olby NJ, Burr J, Platt SR, Papich M, Muñana KR, Faden AI. Phase I clinical trial of a thyrotropin-releasing hormone derivative in acute intervertebral disc herniation. In: *Proceedings of the 21st Annual ACVIM Forum*. Charlotte, NC (2008). p. 402.
61. Gainer JV. Use of crocetin in experimental spinal cord injury. *J Neurosurg.* (1977) 46:358–60. doi: 10.3171/jns.1977.46.3.0358
62. Isu T. Spinal cord evoked potential in experimental spinal cord injury–the changes in spinal cord evoked potential following impact injury, and effect of mannitol administration on acute experimental spinal cord injury. *Hokkaido Igaku Zasshi.* (1990) 65:142–51.

63. Frank LR, Roynard PFP. veterinary neurologic rehabilitation: the rationale for a comprehensive approach. *Top Companion Anim Med.* (2018) 33:49–57. doi: 10.1053/j.tcam.2018.04.002
64. Thomas WB, Olby NJ, Sharon L. Neurologic conditions physical rehabilitation of the neurologic patient. In Millis DL, Levine D, Taylor R. editors. *Canine Rehabilitation Physical Therapy*. Philadelphia, PA: WB Saunders (1978). p. 609–27. doi: 10.1016/B978-1-4377-0309-2.00034-X
65. Gallucci A, Dragone L, Menchetti M, Gagliardo T, Pietra M, Cardinali M, et al. Acquisition of involuntary spinal locomotion (spinal walking) in dogs with irreversible thoracolumbar spinal cord lesion: 81 dogs. *J Vet Intern Med.* (2017) 31:492–7. doi: 10.1111/jvim.14651
66. Gordon-Evans WJ, Johnson AL, Knap KE, Griffon DJ. The effect of body condition on postoperative recovery of dachshunds with intervertebral disc disease treated with postoperative physical rehabilitation. *Vet Surg.* (2019) 48:159–63. doi: 10.1111/vsu.13142
67. Hodgson MM, Bevan JM, Evans RB, Johnson TI. Influence of in-house rehabilitation on the postoperative outcome of dogs with intervertebral disk herniation. *Vet Surg.* (2017) 46:566–73. doi: 10.1111/vsu.12635
68. Jeong IS, Piao Z, Rahman MM, Kim S, Kim NS. Canine thoracolumbar intervertebral disk herniation and rehabilitation therapy after surgical decompression: a retrospective study. *J Adv Vet Anim Res.* (2019) 6:394–402. doi: 10.5455/javar.2019.f359
69. Zidan N, Sims C, Fenn J, Williams K, Griffith E, Early PJ, et al. A randomized, blinded, prospective clinical trial of postoperative rehabilitation in dogs after surgical decompression of acute thoracolumbar intervertebral disc herniation. *J Vet Intern Med.* (2018) 32:1133–44. doi: 10.1111/jvim.15086
70. Laura LH, Peter DS. Recovery times for dogs undergoing thoracolumbar hemilaminectomy with fenestration and physical rehabilitation: a review of 113 cases. *J Vet Med Anim Heal.* (2015) 7:278–89. doi: 10.5897/JVMAH2015.0398
71. Boström AF, Hyytiäinen HK, Koho P, Cizinauskas S, Hielm-Björkman AK. Development of the finnish neurological function testing battery for dogs and its intra- and inter-rater reliability. *Acta Vet Scand.* (2018) 60:56. doi: 10.1186/s13028-018-0408-2
72. Kelly DL, Lassiter KR, Vongsvivut A, Smith JM. Effects of hyperbaric oxygenation and tissue oxygen studies in experimental paraplegia. *J Neurosurg.* (1972) 36:425–9. doi: 10.3171/jns.1972.36.4.0425
73. Hayashi AM, Matera JM, Pinto ACBDCF. Evaluation of electroacupuncture treatment for thoracolumbar intervertebral disk disease in dogs. *J Am Vet Med Assoc.* (2007) 231:913–8. doi: 10.2460/javma.231.6.913
74. Han HJ, Yoon HY, Kim JY, Jang HY, Lee B, Choi SH, et al. Clinical effect of additional electroacupuncture on thoracolumbar intervertebral disc herniation in 80 paraplegic dogs. *Am J Chin Med.* (2010) 38:1015–25. doi: 10.1142/S0192415X10008433
75. Joaquim JGF, Luna SPL, Brondani JT, Torelli SR, Rahal SC, De Freitas FP. Comparison of decompressive surgery, electroacupuncture, and decompressive surgery followed by electroacupuncture for the treatment of dogs with intervertebral disk disease with long-standing severe neurologic deficits. *J Am Vet Med Assoc.* (2010) 236:1225–9. doi: 10.2460/javma.236.11.1225
76. Laim A, Jaggy A, Forterre F, Doherr MG, Aeschbacher G, Glardon O. Effects of adjunct electroacupuncture on severity of postoperative pain in dogs undergoing hemilaminectomy because of acute thoracolumbar intervertebral disk disease. *J Am Vet Med Assoc.* (2009) 234:1141–6. doi: 10.2460/javma.234.9.1141
77. Prado C, Fratini P, de Sá Schiavo Matias G, Bocabello RZ, Monteiro J, dos Santos CJ, et al. Combination of stem cells from deciduous teeth and electroacupuncture for therapy in dogs with chronic spinal cord injury: a pilot study. *Res Vet Sci.* (2019) 123:247–51. doi: 10.1016/j.rvsc.2019.01.011
78. Kalderon N. Cell elimination as a strategy for repair in acute spinal cord injury. *Curr Pharm Des.* (2005) 11:1237–45. doi: 10.2174/1381612053507477
79. Zhang Q, Xiong Y, Zhu B, Zhu B, Tian D, Wang W. Low-dose fractionated irradiation promotes axonal regeneration beyond reactive gliosis and facilitates locomotor function recovery after spinal cord injury in beagle dogs. *Eur J Neurosci.* (2017) 46:2507–18. doi: 10.1111/ejn.13714
80. Taghavi N, Luecke GR, Jeffery ND. A wearable body controlling device for application of functional electrical stimulation. *Sensors.* (2018) 18:1251. doi: 10.3390/s18041251
81. Krueger E, Magri LMS, Botelho AS, Bach FS, Rebellato CLK, Fracaro L, et al. Effects of low-intensity electrical stimulation and adipose derived stem cells transplantation on the time-domain analysis-based electromyographic signals in dogs with SCI. *Neurosci Lett.* (2019) 696:38–45. doi: 10.1016/j.neulet.2018.12.004
82. Chang SM, Yu GR, Diao YM, Zhang MJ, Wang SB, Hou CL. Sacral anterior root stimulated defecation in spinal cord injuries: an experimental study in canine model. *World J Gastroenterol.* (2005) 11:1715–8. doi: 10.3748/wjg.v11.i11.1715
83. Walter JS, Wheeler JS, Robinson CJ, Wurster RD. Surface stimulation techniques for bladder management in the spinal dog. *J Urol.* (1989) 141:161–5. doi: 10.1016/S0022-5347(17)40632-X
84. Granger N, Chew D, Fairhurst P, Fawcett JW, Lacour SP, Craggs M, et al. Use of an implanted sacral nerve stimulator to restore urine voiding in chronically paraplegic dogs. *J Vet Intern Med.* (2013) 27:99–105. doi: 10.1111/jvim.12011
85. Turbes CC. Intercostal nerve nerve neurouma (PNS) implantation in spinal cord transection - Enhancement of locomotor recovery. *Biomed Sci Instrum.* (1999) 35:297–302.
86. Cseuz KA, Speakman TJ. Regenerative potential of peripheral nerve implants in the transected spinal cord of dogs. *Surg Forum.* (1962) 13:417–8.
87. Turbes CC. Intercostal nerve neurouma (PNS) implantation in spinal cord bridging spinal cord transection a functional internuncal pathway system result in recovery from paraplegia. *Biomed Sci Instrum.* (2001) 37:137–42.
88. Toreih AA, Sallam AA, Ibrahim CM, Maaty AI, Hassan MM. Intercostal, ilioinguinal, and iliohypogastric nerve transfers for lower limb reinnervation after spinal cord injury: An anatomical feasibility and experimental study. *J Neurosurg Spine.* (2018) 30:268–78. doi: 10.3171/2018.8.SPINE181
89. Wilson AS, Krcek JP. Restoration of function in the paralyzed diaphragm. *Exp Neurol.* (1975) 47:490–502. doi: 10.1016/0014-4886(75)90081-3
90. Tang J, Ma J, Yang L, Huang X, Ge Y, Sui T, et al. The feasibility study of extradural nerve anastomosis technique for canine bladder reinnervation after spinal cord injury. *J Spinal Cord Med.* (2016) 39:679–85. doi: 10.1080/10790268.2016.1209889
91. Lin H, Hou C, Chen A, Xu Z. Reinnervation of atonic bladder after conus medullaris injury using a modified nerve crossover technique in canines. *World Neurosurg.* (2010) 73:582–6. doi: 10.1016/j.wneu.2010.02.020
92. Lin H, Hou C, Chen A, Xu Z. Innervation of reconstructed bladder above the level of spinal cord injury for inducing micturition by contractions of the abdomen-to-bladder reflex arc. *Neurosurgery.* (2010) 66:948–52. doi: 10.1227/01.NEU.0000368386.85578.63
93. Ruggieri MR, Braverman AS, D'Andrea L, Betz R, Barbe MF. Functional reinnervation of the canine bladder after spinal root transection and genitofemoral nerve transfer at one and three months after denervation. *J Neurotrauma.* (2008) 25:401–9. doi: 10.1089/neu.2007.0335
94. Ruggieri MR, Braverman AS, Bernal RM, Lamarre NS, Brown JM, Barbe MF. Reinnervation of urethral and anal sphincters with femoral motor nerve to pudendal nerve transfer. *Neurourol Urodyn.* (2011) 30:1695–704. doi: 10.1002/nau.21171
95. Blight AR, Toombs JP, Bauer MS, Widmer WR. The effects of 4-aminopyridine on neurological deficits in chronic cases of traumatic spinal cord injury in dogs: a phase i clinical trial. *J Neurotrauma.* (1991) 8:103–19. doi: 10.1089/neu.1991.8.103
96. Lim JH, Muguet-Chanoit AC, Smith DT, Laber E, Olby NJ. Potassium channel antagonists 4-aminopyridine and the T-butyl carbamate derivative of 4-aminopyridine improve hind limb function in chronically non-ambulatory dogs; a blinded, placebo-controlled trial. *PLoS ONE.* (2014) 9:e116139. doi: 10.1371/journal.pone.0116139
97. Blight AR. Effect of 4-aminopyridine on axonal conduction-block in chronic spinal cord injury. *Brain Res Bull.* (1989) 22:47–52. doi: 10.1016/0361-9230(89)90126-3
98. Lewis MJ, Laber E, Olby NJ. Predictors of response to 4-aminopyridine in chronic canine spinal cord injury. *J Neurotrauma.* (2019) 36:1428–34. doi: 10.1089/neu.2018.5975
99. Smith KJ, Felts PA, John GR. Effects of 4-aminopyridine on demyelinated axons, synapses and muscle tension. *Brain.* (2000) 123:171–84. doi: 10.1093/brain/123.1.171

100. Muir E, De Winter F, Verhaagen J, Fawcett J. Recent advances in the therapeutic uses of chondroitinase ABC. *Exp Neurol.* (2019) 321:113032. doi: 10.1016/j.expneurol.2019.113032
101. Hu HZ, Granger N, Balakrishna Pai S, Bellamkonda R V., Jeffery ND. Therapeutic efficacy of microtube-embedded chondroitinase ABC in a canine clinical model of spinal cord injury. *Brain.* (2018) 141:1017–27. doi: 10.1093/brain/awy007
102. Lee SH, Kim Y, Rhew D, Kuk M, Kim M, Kim WH, et al. Effect of the combination of mesenchymal stromal cells and chondroitinase ABC on chronic spinal cord injury. *Cytotherapy.* (2015) 17:1374–83. doi: 10.1016/j.jcyt.2015.05.012
103. Lee JH, Choi CB, Chung DJ, Kang EH, Chang HS, Hwang SH, et al. Development of an improved canine model of percutaneous spinal cord compression injury by balloon catheter. *J Neurosci Methods.* (2008) 167:310–6. doi: 10.1016/j.jneumeth.2007.07.020
104. Lim JH, Jung CS, Byeon YE, Wan HK, Yoon JH, Kang KS, et al. Establishment of a canine spinal cord injury model induced by epidural balloon compression. *J Vet Sci.* (2007) 8:89–94. doi: 10.4142/jvs.2007.8.1.89
105. Griffiths IR. Vasogenic edema following acute and chronic spinal cord compression in the dog. *J Neurosurg.* (1975) 42:155–65. doi: 10.3171/jns.1975.42.2.0155
106. Kobrine AI. Blood flow in experimental cord trauma. *J Neurosurg.* (1979) 50:270–1. doi: 10.3171/jns.1979.50.2.0270
107. Lim JH, Byeon YE, Ryu HH, Jeong YH, Lee YW, Wan HK, et al. Transplantation of canine umbilical cord blood-derived mesenchymal stem cells in experimentally induced spinal cord injured dogs. *J Vet Sci.* (2007) 8:275–82. doi: 10.4142/jvs.2007.8.3.275
108. Ryu HH, Kang BJ, Park SS, Kim Y, Sung GJ, Woo HM, et al. Comparison of mesenchymal stem cells derived from fat, bone marrow, Wharton's jelly, and umbilical cord blood for treating spinal cord injuries in dogs. *J Vet Med Sci.* (2012) 74:1617–30. doi: 10.1292/jvms.12-0065
109. Lee SH, Kim Y, Rhew D, Kim A, Jo KR, Yoon Y, et al. Effect of canine mesenchymal stromal cells overexpressing heme oxygenase-1 in spinal cord injury. *J Vet Sci.* (2017) 18:377–86. doi: 10.4142/jvs.2017.18.3.377
110. Park SS, Byeon YE, Ryu HH, Kang BJ, Kim Y, Kim WH, et al. Comparison of canine umbilical cord blood-derived mesenchymal stem cell transplantation times: involvement of astrogliosis, inflammation, intracellular actin cytoskeleton pathways, and neurotrophin-3. *Cell Transplant.* (2011) 20:1867–80. doi: 10.3727/096368911X566163
111. Lee JH, Chung WH, Kang EH, Chung DJ, Choi CB, Chang HS, et al. Schwann cell-like remyelination following transplantation of human umbilical cord blood (hUCB)-derived mesenchymal stem cells in dogs with acute spinal cord injury. *J Neurol Sci.* (2011) 300:86–96. doi: 10.1016/j.jns.2010.09.025
112. Ryu HH, Lim JH, Byeon YE, Park JR, Seo MS, Lee YW, et al. Functional recovery and neural differentiation after transplantation of allogenic adipose-derived stem cells in a canine model of acute spinal cord injury. *J Vet Sci.* (2009) 10:273–84. doi: 10.4142/jvs.2009.10.4.273
113. Kim Y, Jo SH, Kim WH, Kweon OK. Antioxidant and anti-inflammatory effects of intravenously injected adipose derived mesenchymal stem cells in dogs with acute spinal cord injury. *Stem Cell Res Ther.* (2015) 6:229. doi: 10.1186/s13287-015-0236-5
114. Park SS, Lee YJ, Lee SH, Lee D, Choi K, Kim WH, et al. Functional recovery after spinal cord injury in dogs treated with a combination of matrigel and neural-induced adipose-derived mesenchymal Stem cells. *Cytotherapy.* (2012) 14:584–97. doi: 10.3109/14653249.2012.658913
115. Jung DI, Ha J, Kang BT, Kim JW, Quan FS, Lee JH, et al. A comparison of autologous and allogenic bone marrow-derived mesenchymal stem cell transplantation in canine spinal cord injury. *J Neurol Sci.* (2009) 285:67–77. doi: 10.1016/j.jns.2009.05.027
116. Khan IU, Yoon Y, Choi KU, Jo KR, Kim N, Lee E, et al. Therapeutic effects of intravenous injection of fresh and frozen thawed ho-1-overexpressed ad-mscs in dogs with acute spinal cord injury. *Stem Cells Int.* (2019) 2019:8537541. doi: 10.1155/2019/8537541
117. Khan IU, Yoon Y, Kim A, Jo KR, Choi KU, Jung T, et al. Improved healing after the co-transplantation of ho-1 and bdnf overexpressed mesenchymal stem cells in the subacute spinal cord injury of dogs. *Cell Transplant.* (2018) 27:1140–53. doi: 10.1177/0963689718779766
118. Handa Y, Naito A, Watanabe S, Komatsu S, Shimizu Y. functional recovery of locomotive behavior in the adult spinal dog. *Tohoku J Exp Med.* (1986) 148:373–84. doi: 10.1620/tjem.148.373
119. Naito A, Shimizu Y, Handa Y. Analyses of airstepping movement in adult spinal dogs. *Tohoku J Exp Med.* (1990) 162:41–8. doi: 10.1620/tjem.162.41
120. Kim BG, Kang YM, Phi JH, Kim YH, Hwang DH, Choi JY, et al. Implantation of polymer scaffolds seeded with neural stem cells in a canine spinal cord injury model. *Cytotherapy.* (2010) 12:841–5. doi: 10.3109/14653249.2010.501784
121. Lee SH, Chung YN, Kim YH, Kim YJ, Park JP, Kwon DK, et al. Effects of human neural stem cell transplantation in canine spinal cord hemisection. *Neurol Res.* (2009) 31:996–1002. doi: 10.1179/174313209X385626
122. Assina R, Sankar T, Theodore N, Javedan SP, Gibson AR, Horn KM, et al. Activated autologous macrophage implantation in a large animal model of spinal cord injury. *Neurosurg Focus.* (2008) 25:E3. doi: 10.3171/FOC.2008.25.11.E3
123. Li X, Tan J, Xiao Z, Zhao Y, Han S, Liu D, et al. Transplantation of hUC-MSCs seeded collagen scaffolds reduces scar formation and promotes functional recovery in canines with chronic spinal cord injury. *Sci Rep.* (2017) 6:43559. doi: 10.1038/srep43559
124. Han S, Xiao Z, Li X, Zhao H, Wang B, Qiu Z, et al. Human placenta-derived mesenchymal stem cells loaded on linear ordered collagen scaffold improves functional recovery after completely transected spinal cord injury in canine. *Sci China Life Sci.* (2018) 61:2–13. doi: 10.1007/s11427-016-9002-6
125. Wu GH, Shi HJ, Che MT, Huang MY, Wei QS, Feng B, et al. Recovery of paralyzed limb motor function in canine with complete spinal cord injury following implantation of MSC-derived neural network tissue. *Biomaterials.* (2018) 181:15–34. doi: 10.1016/j.biomaterials.2018.07.010
126. El-Kheir WA, Gabr H, Awad MR, Ghannam O, Barakat Y, Farghali HAMA, et al. Autologous bone marrow-derived cell therapy combined with physical therapy induces functional improvement in chronic spinal cord injury patients. *Cell Transplant.* (2014) 24:1813–27. doi: 10.3727/096368913X664540
127. Kim Y, Lee SH, Kim WH, Kweon OK. Transplantation of adipose derived mesenchymal stem cells for acute thoracolumbar disc disease with no deep pain perception in dogs. *J Vet Sci.* (2016) 17:123–6. doi: 10.4142/jvs.2016.17.1.123
128. McMahill BG, Spriet M, Sisó S, Manzer MD, Mitchell G, McGee J, et al. Feasibility study of canine epidermal neural crest stem cell transplantation in the spinal cords of dogs. *Stem Cells Transl Med.* (2015) 4:1173–86. doi: 10.5966/sctm.2015-0018
129. Steffensen N, Lehmbecker A, Gerhauser I, Wang Y, Carlson R, Tipold A, et al. Generation and characterization of highly purified canine schwann cells from spinal nerve dorsal roots as potential new candidates for transplantation strategies. *J Tissue Eng Regen Med.* (2018) 12:e422–37. doi: 10.1002/term.2478
130. Lim JH, Olby NJ. Generation of pure cultures of autologous schwann cells by use of biopsy specimens of the dorsal cutaneous branches of the cervical nerves of young adult dogs. *Am J Vet Res.* (2016) 77:1166–74. doi: 10.2460/ajvr.77.10.1166
131. Doucette R. Olfactory ensheathing cells: potential for glial cell transplantation into areas of CNS injury. *Histol Histopathol.* (1995) 10:503–7.
132. Ramón-Cueto A, Valverde F. Olfactory bulb ensheathing glia: a unique cell type with axonal growth-promoting properties. *Glia.* (1995) 14:163–73. doi: 10.1002/glia.440140302
133. Fawcett JW. Spinal cord repair: from experimental models to human application. *Spinal Cord.* (1998) 36:811–7. doi: 10.1038/sj.sc.3100769
134. Franklin RJM, Barnett SC. Olfactory ensheathing cells and CNS regeneration: the sweet smell of success? *Neuron.* (2000) 28:15–8. doi: 10.1016/S0896-6273(00)00080-5
135. Smith PM, Lakatos A, Barnett SC, Jeffery ND, Franklin RJM. Cryopreserved cells isolated from the adult canine olfactory bulb are capable of extensive remyelination following transplantation into the adult rat CNS. *Exp Neurol.* (2002) 176:402–6. doi: 10.1006/exnr.2002.7936

136. Skinner APC, Pachnicke S, Lakatos A, Franklin RJM, Jeffery ND. Nasal and frontal sinus mucosa of the adult dog contain numerous olfactory sensory neurons and ensheathing glia. *Res Vet Sci.* (2005) 78:9–15. doi: 10.1016/j.rvsc.2004.05.010
137. Ito D, Fujita N, Ibanez C, Sasaki N, Franklin RJM, Jeffery ND. Serum-free medium provides a clinically relevant method to increase olfactory ensheathing cell numbers in olfactory mucosa cell culture. *Cell Transplant.* (2008) 16:1021–7. doi: 10.3727/000000007783472345
138. Wewetzer K, Radtke C, Kocsis J, Baumgärtner W. Species-specific control of cellular proliferation and the impact of large animal models for the use of olfactory ensheathing cells and Schwann cells in spinal cord repair. *Exp Neurol.* (2011) 229:80–7. doi: 10.1016/j.expneurol.2010.08.029
139. Ito D, Carwardine D, Prager J, Wong LF, Kitagawa M, Jeffery N, et al. Methods of olfactory ensheathing cell harvesting from the olfactory mucosa in dogs. *PLoS ONE.* (2019) 14:e0213252. doi: 10.1371/journal.pone.0213252
140. Andrews PJ, Poirrier AL, Lund VJ, Choi D. Safety of human olfactory mucosal biopsy for the purpose of olfactory ensheathing cell harvest and nerve repair: a prospective controlled study in patients undergoing endoscopic sinus surgery. *Rhinol J.* (2016) 54:183–191. doi: 10.4193/Rhin15.365
141. Li Y, Field PM, Raisman G. Repair of adult rat corticospinal tract by transplants of olfactory ensheathing cells. *Science.* (1997) 277:2000–2. doi: 10.1126/science.277.5334.2000
142. Franklin RJM, Gilson JM, Franceschini IA, Barnett SC. Schwann cell-like myelination following transplantation of an olfactory bulb-ensheathing cell line into areas of demyelination in the adult CNS. *Glia.* (1996) 17:217–24. doi: 10.1002/(SICI)1098-1136(199607)17:3<217::AID-GLIA4>3.0.CO;2-Y
143. Granger N, Blamires H, Franklin RJM, Jeffery ND. Autologous olfactory mucosal cell transplants in clinical spinal cord injury: a randomized double-blinded trial in a canine translational model. *Brain.* (2012) 135:3227–37. doi: 10.1093/brain/aws268
144. Carwardine D, Prager J, Neeves J, Muir EM, Uney J, Granger N, et al. Transplantation of canine olfactory ensheathing cells producing chondroitinase ABC promotes chondroitin sulphate proteoglycan digestion and axonal sprouting following spinal cord injury. *PLoS ONE.* (2017) 12:e0188967. doi: 10.1371/journal.pone.0188967
145. Nishida H, Nakayama M, Tanaka H, Kitamura M, Hatoya S, Sugiura K, et al. Safety of autologous bone marrow stromal cell transplantation in dogs with acute spinal cord injury. *Vet Surg.* (2012) 41:437–42. doi: 10.1111/j.1532-950X.2011.00959.x
146. Tamura K, Harada Y, Kunimi M, Takemitsu H, Hara Y, Nakamura T, et al. Autologous bone marrow mononuclear cell transplant and surgical decompression in a dog with chronic spinal cord injury. *Exp Clin Transplant.* (2015) 13:100–5. doi: 10.6002/ect.2013.0237
147. Penha EM, Meira CS, Guimarães ET, Mendonça MVP, Gravely FA, Pinheiro CMB, et al. Use of autologous mesenchymal stem cells derived from bone marrow for the treatment of naturally injured spinal cord in dogs. *Stem Cells Int.* (2014) 2014:437521. doi: 10.1155/2014/437521
148. Sarmento C, Rodrigues M, Bocabello R, Mess A, Miglino M. Pilot study: bone marrow stem cells as a treatment for dogs with chronic spinal cord injury. *Regen Med Res.* (2014) 2:9. doi: 10.1186/2050-490X-2-9
149. Escalhão CCMI, Ramos IP, Hochman-Mendez C, Brunswick THK, Souza SAL, Gutfilen B, et al. Safety of allogeneic canine adipose tissue-derived mesenchymal stem cell intraspinal transplantation in dogs with chronic spinal cord injury. *Stem Cells Int.* (2017) 2017:3053759. doi: 10.1155/2017/3053759
150. Bach FS, Rebelatto CLK, Fracaro L, Senegaglia AC, Fragoso FYI, Daga DR, et al. Comparison of the efficacy of surgical decompression alone and combined with canine adipose tissue-derived stem cell transplantation in dogs with acute thoracolumbar disk disease and spinal cord injury. *Front Vet Sci.* (2019) 6:383. doi: 10.3389/fvets.2019.00383
151. Feitosa MLT, Sarmento CAP, Bocabello RZ, Beltrão-Braga PCB, Pignatari GC, Giglio RF, et al. Transplantation of human immature dental pulp stem cell in dogs with chronic spinal cord injury. *Acta Cir Bras.* (2017) 32:540–9. doi: 10.1590/s0102-865020170070000005
152. Besalti O, Aktas Z, Can P, Akpinar E, Elcin AE, Elcin YM. The use of autologous neurogenically-induced bone marrow-derived mesenchymal stem cells for the treatment of paraplegic dogs without nociception due to spinal trauma. *J Vet Med Sci.* (2016) 78:1465–73. doi: 10.1292/jvms.15-0571
153. Bhat IA, Sivanarayanan TB, Somal A, Pandey S, Bharti MK, Panda BSK, et al. An allogenic therapeutic strategy for canine spinal cord injury using mesenchymal stem cells. *J Cell Physiol.* (2019) 234:2705–18. doi: 10.1002/jcp.27086

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REVIEW
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Diagnostic Imaging in Intervertebral Disc Disease

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Imaging is integral in the diagnosis of canine intervertebral disc disease (IVDD) and in differentiating subtypes of intervertebral disc herniation (IVDH). These include intervertebral disc extrusion (IVDE), intervertebral disc protrusion (IVDP) and more recently recognized forms such as acute non-compressive nucleus pulposus extrusion (ANNPE), hydrated nucleus pulposus extrusion (HNPE), and intradural/intramedullary intervertebral disc extrusion (IIVDE). Many imaging techniques have been described in dogs with roles for survey radiographs, myelography, computed tomography (CT), and magnetic resonance imaging (MRI). Given how common IVDH is in dogs, a thorough understanding of the indications and limitations for each imaging modality to aid in diagnosis, treatment planning and prognosis is essential to successful case management. While radiographs can provide useful information, especially for identifying intervertebral disc degeneration or calcification, there are notable limitations. Myelography addresses some of the constraints of survey radiographs but has largely been supplanted by cross-sectional imaging. Computed tomography with or without myelography and MRI is currently utilized most widely and have become the focus of most contemporary studies on this subject. Novel advanced imaging applications are being explored in dogs but are not yet routinely performed in clinical patients. The following review will provide a comprehensive overview on common imaging modalities reported to aid in the diagnosis of IVDH including IVDE, IVDP, ANNPE, HNPE, and IIVDE. The review focuses primarily on canine IVDH due to its frequency and vast literature as opposed to feline IVDH.

Keywords: herniation, extrusion, protrusion, computed tomography, magnetic resonance imaging, intervertebral disc

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INTRODUCTION

Intervertebral disc disease (IVDD) is the most common spinal cord disease of dogs, being responsible for 2.3–3.7% of admissions to veterinary hospitals (1, 2). The diagnosis of all IVDD forms is based on imaging, with the imaging techniques evolving over the years.

Even though IVDD was first reported in a paraplegic Dachshund by Dexler in 1896, it was truly well-characterized by Olsson and Hansen in the early 1950s (3). In 1952, Hansen classified IVDD into acute, or Hansen type I intervertebral disc extrusion (IVDE), and chronic, or Hansen type II intervertebral disc protrusion (IVDP) (4). Over the years and primarily with the widespread use of magnetic resonance imaging (MRI), newer forms began to be recognized, such as acute

non-compressive nucleus pulposus extrusion (ANNPE), hydrated nucleus pulposus extrusion (HNPE) and intradural/intramedullary intervertebral disc extrusion (IIVDE) (5, 6). In addition, intervertebral disc (IVD) degeneration and protrusion are also associated with other important spinal conditions such as cervical spondylomyelopathy and degenerative lumbosacral stenosis (7, 8). A study with 677,000 dogs suggested that the overall prevalence of all intervertebral disc degeneration-related diseases was 27.8% (1).

The most commonly described clinical form of intervertebral disc herniation (IVDH) is IVDE, which leads to acute spinal pain and variable degrees of paresis up to paralysis. Radiography was the most widely used method for the diagnosis of IVDD starting in the 1950s and extending until the 1980s (4, 9–11).

Myelography started to be used in the 1960s and 1970s (12, 13), but publications utilizing this technique in dogs remained variable until the early to mid 1990s (14–16). Starting in the 1990s, published case series began to have all dogs confirmed with myelography (17), which then became the norm in the publications from the 2000s onward (18). It was also in the 2000s that magnetic resonance imaging (MRI) and computed tomography (CT) started to become routinely used in referral hospitals. Since that time, most publications have focused on descriptive and comparative studies involving multiple modalities, typically either CT or MRI, or both.

A 2016 study looked at the practice patterns of diplomates of the American College of Veterinary Internal Medicine in the specialty of Neurology and American College of Veterinary Surgeons for the diagnosis of IVDH in North America. Among the board-certified neurologists, MRI was the most commonly used technique (75%), whereas among the board-certified surgeons, CT with or without concurrent myelography was the most commonly used imaging modality (58%). Approximately 28% of board-certified surgeons used MRI. Myelography alone was chosen as the most commonly used modality by 14% of board-certified surgeons, and <1% of board-certified neurologists (19). The imaging preferences in other countries or continents are unknown, although, overall, CT scanners are far more widely available than MRI.

As the saying goes "the best treatment is a correct diagnosis," in order to accurately diagnose all forms of IVDD and institute appropriate treatment, correct identification of affected disc sites, extension and lateralization are crucial for treatment planning. In this manuscript, we will review all imaging modalities used to presumptively or accurately diagnose all forms of IVDD.

IMAGING DIAGNOSIS OF ACUTE INTERVERTEBRAL DISC EXTRUSION (IVDE)

Hansen type I intervertebral disc extrusion (IVDE) is characterized by chondroid degeneration of the gelatinous nucleus pulposus with transformation to hyaline cartilage and mineralization. Ultimately this process leads to rupture of the annulus fibrosus and herniation of calcified nucleus pulposus into the vertebral canal and/or intervertebral foramen (4, 20). Disc herniation can be a misleading term because it is not the entire disc that herniates but primarily parts of the nucleus pulposus and/or the ruptured annulus. The extruded disc material will cause myelopathic (proprioceptive ataxia, paresis, plegia) and radiculopathic (pain) clinical signs (21).

RADIOGRAPHY

Orthogonal radiographic projections, lateral and ventrodorsal, are routinely used in the diagnostic investigation of cases suspected of IVDE. The use of general anesthesia has been recommended in order to obtain diagnostic radiographs (22). However, this is very rarely, if ever, done in clinical practice and truly is an unnecessary recommendation. Patients can be properly positioned for radiographs under sedation. Radiographs are a screening test in the diagnostic approach of spinal cases, therefore general anesthesia should be reserved for techniques that can provide a definitive diagnosis of compressive lesions such as myelography, CT or MRI.

Radiographic changes supportive of IVDE are narrowing of the disc space, narrowing of the articular facets, narrowing and/or increased opacity of the intervertebral foramen, presence of mineralized disc material within the vertebral canal and vacuum phenomenon (22, 23). A popular finding, mineralization (also known as calcification) of the disc space *in situ*, is a controversial one. Intervertebral disc mineralization is supportive of intervertebral disc degeneration but not disc extrusion. Radiographic detection of calcification requires significant IVD mineralization to be present, since many discs documented to have mineralization on histopathology were not apparent radiographically (24). Interestingly, a recent study found that neither CT nor low-field MRI had a higher intra- or interobserver agreement compared to radiographs for the detection of disc calcification (25). Studies have shown that disc calcification at 2 years of age was a significant predictor of disc herniation later in life (26) and also a risk factor for recurrent herniation following surgery (27, 28). However, it has been reported that just as many extrusions occurred at intervertebral discs with radiographic evidence of calcification as those without it (29). Additionally, in a prospective MRI study, none of the 65 calcified discs corresponded with the actual site of disc extrusion (30). It must be kept in mind that discs can show evidence of calcification and decalcification naturally, as demonstrated in longitudinal studies in Dachshunds (31). Therefore, using intervertebral disc calcification as a diagnostic criterion would not be appropriate.

In the thoracolumbar region, survey radiographs typically have a reported sensitivity ranging between 51 and 61% (32–36), with only one study reporting a much higher sensitivity at 94.7% using digital radiographs (37). Importantly, radiographic findings can be *suggestive* of thoracolumbar IVDE but are never diagnostic and can also be fraught with significant interobserver variation (23). Radiographs are even less sensitive in the cervical region. Among radiographic changes, intervertebral disc

mineralization and narrowing of the affected disc space had the highest correlation with myelography (38). A study evaluated the diagnostic accuracy of radiographs in the diagnosis of cervical IVDE and IVDP using 4 raters (2 radiologists and 2 surgeons) and found an overall accuracy of only 35% (31.3–40.6%). When a suspected abnormal disc space was detected, a higher agreement was found but remained low, at only 58% (53–67%) (39).

MYELOGRAPHY

Myelography is a radiographic technique in which spinal radiographs are obtained following the injection of a radiopaque contrast agent into the subarachnoid space. It is widely available because it can be performed in any place with an X-ray machine (40, 41). Myelography has been largely replaced by cross-sectional imaging in countries where CT and MRI are routinely available for veterinary patients.

In order to achieve a diagnostic myelographic study, several technical aspects are important. Non-ionic, iodinated, water-soluble contrast agents such as iohexol, iopamiron, or iotrolan, with concentrations ranging from 180 to 240 mg/ml, ideally at body temperature, should be used (42). Injection sites can be either the cerebellomedullary cistern or lumbar region, although lumbar myelography is superior, especially for investigating the thoracolumbar region (21, 43). Lumbar myelography is also overall, safer than cisternal myelography, though it is technically more challenging and is more frequently associated with leakage of contrast into the epidural space. The site of injection site should be the ventral subarachnoid space at L5-6 (ideally). The L4-5 space should be avoided since it is the center of the lumbosacral intumescence in most dogs. In the cerebellomedulary cistern, the dorsal subarachnoid space should be used. Dose of contrast agents ranges 0.3–0.45 ml/kg body weight and should not exceed a total volume >8 ml independent of the dog's size (44). Larger volumes should only be used if the initial injection was not diagnostic. Following injection, lateral, ventrodorsal, +/- oblique projections should be acquired in all cases (45).

Myelographic patterns of IVDE are those characteristics of an extradural compression. In order to facilitate differentiation from IVDP, the following criteria have been proposed for the diagnosis of IVDE: (1) thinning and deviation of the contrast columns, (2) thinning of the contrast columns is mild to severe or contrast columns are discontinuous, (3) thinning of the contrast columns is diffuse and beyond the boundaries of the affected disc, and (4) asymmetrical distribution of contrast column thinning cranial or caudal to the affected disc (**Figure 1**) (46). It is important to evaluate the ventrodorsal and/or oblique views to determine the presence of axial deviation of the contrast to determine lateralization of the lesion and guide the surgical approach (21, 45, 47). It has been proposed that extensive intramedullary patterns were associated with a poor outcome (48); however, this was not confirmed by a different study (49). Extensive spinal cord swelling with evidence of contrast medium infiltration into the spinal cord has been reported as an indication of myelomalacia (50). Myelography can yield false negative results in cases of IVDE, primarily with lateral or foraminal extrusions, with these cases requiring CT or MRI (51).

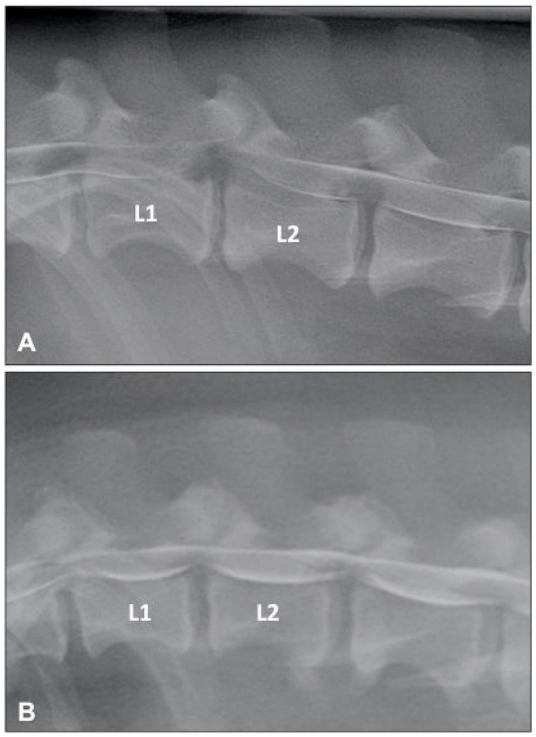

FIGURE 1 | (A) Lateral myelogram of a 5-year old Basset Hound with an L1-L2 IVDE. Thinning of the contrast columns is severe and the ventral contrast column is almost discontinued at L1-L2. Thinning of the contrast column is beyond the boundaries of the affected disc and distributed asymmetrical with a longer dorsal deviation of the ventral contrast column caudal to the affected disc. **(B)** Lateral myelogram of a 9-year old Basset Hound with IVDPs between L1-L2 and L2-L3. Thinning of the contrast columns is only mild, dorsal deviation of the ventral contrast column is centered over the affected disc spaces and distribution of contrast thinning is cranio-caudally symmetrical over the disc spaces.

Estimates of diagnostic accuracy of myelography for the diagnosis and site of IVDE have ranged from 72 to 99% (37, 45, 52, 53). Determination of lateralization is less accurate, especially when compared to CT or MRI. The reported sensitivity for lateral localization ranges from 49 to 83%, although the precision can be greatly improved with the combination of ventrodorsal and oblique views (15, 17, 30, 37, 45, 53, 54).

Myelography is an invasive procedure and is associated with some inherent risks, primarily temporary deterioration of neurologic status, and post-myelographic seizures (55, 56). Even though the incidence of post-myelographic seizures was as high as 21.4% in a study with 183 dogs (55), another study with 503 dogs found a incidence of only 3% (56). An important reason for this 7-fold difference relates to the volume of contrast medium administered. In the first study, the mean total volume of iohexol was 9.1 and 16.8 mL in dogs without and with seizures (55), whereas in the second study it was, 4.5 and 11.7 mL, in dogs without and with seizures, respectively (56). Importantly, the

dose per kg was not associated with seizures in either study. It is therefore recommended to carefully assess the total volume required, and avoid using total volumes higher than 8 mL, even in large or giant dogs. Another key point to minimize the risk of seizures is the use of lumbar injections, since the risk of seizure was seven times higher with cerebellomedulary cistern injections in both studies (55, 56). Due to the risk of post-myelographic seizures, patients should be monitored in an ICU for at least 12 h post-myelography, which can significantly increase the expense associated with this procedure.

While myelography has limitations and has largely been superseded by cross-sectional imaging, it remains a reasonable imaging option for diagnosing IVDE. Importantly, in a study of 107 dogs assessing the choice of myelography or MRI for diagnosis of IVDE, patient outcomes using myelography were not inferior compared to MRI (57).

COMPUTED TOMOGRAPHY

Computed tomography (CT) is a very important modality in the diagnostic work-up of dogs and cats suspected of having IVDD (37, 52, 58, 59). Tomography by definition is simply the depiction of a section of the patient free from superimposition of overlaying structures (60). As conventional radiographs represent variations in tissue absorption of x-rays in a linear direction, physical structures are superimposed. Computed tomography transmits x-rays to detectors through the patient, around a single axis of rotation; the data is processed by a computer and creates the image as a slice, free of superimposition. Tissue physical density is measured relative to the density of water and assigned a numerical value called a CT number or Hounsfield unit (HU) (60).

There are 3 possible techniques to achieve a diagnosis of IVDE using CT: non-contrast CT, intravenous contrast CT (CT-angiography), and subarachnoid contrast CT (CT-myelography). CT-myelography requires ∼25 to 50% of the volume of contrast medium compared to conventional myelography, and it is therefore much safer. In all of these CT modalities, transverse images can be reformatted into other imaging planes, primarily when using multidetector CTs which allow multiplanar reformatting in different spatial planes (37, 58, 61, 62). Advantages of CT relative to MRI include that it is more widely available, less costly and much faster. It is also possible to carry out CT studies routinely under sedation. Most, but not all, cases of IVDE can be diagnosed with non-contrast CT. In this context, CT can be used as a convenient, quick screening test for spinal cases where the neurosurgical caseload is high. For dogs in which non-contrast CT alone is insufficient, MRI or CT-myelography can then be utilized for a diagnosis. Median examination times in a study were 4, 8, and 32 min, for helical CT, conventional CT, and myelography, respectively (37). These times were achieved with a single slice helical CT. With newer systems ranging from 16 to 128 slices, scanning times are much shorter, often <1 min.

Most investigations relating to IVDD have been dedicated to non-contrast CT. The normal intervertebral disc is of uniform soft tissue opacity in CT images, with no visible distinction between the nucleus pulposus and annulus fibrosus (63). The spinal cord, cerebrospinal fluid, and meninges are of similar tissue physical density and cannot be clearly discriminated without the introduction of intra-thecal contrast media (64). This combination of structures is referred to as the thecal sac in plain CT images. Epidural fat surrounds the thecal sac and is less dense than soft tissue, so it appears darker gray. This difference in tissue density allows discrimination of the outer margins of the thecal sac. Calcified disc material is visible in non-contrast enhanced CT images because it has a higher physical (radiographic) density than adjacent soft tissues and fat (65).

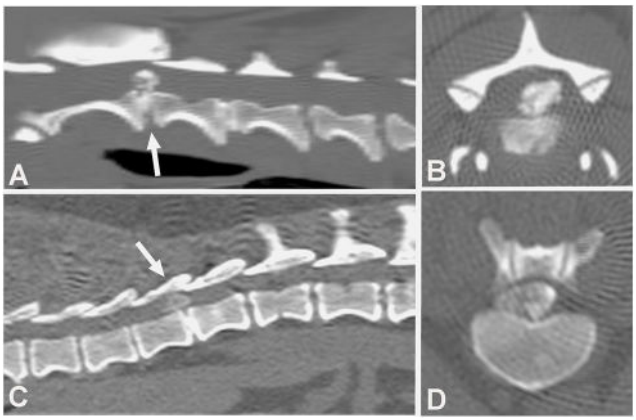

FIGURE 2 | Computed tomographic (CT) images showing the appearance of mineralized intervertebral disc extrusion in the vertebral canal. **(A)** Sagittal reconstructed non-contrast CT image showing a hyperattenuating mass suggestive of extruded disc material into the vertebral canal between C2-3 (arrow). The intervertebral disc space also mineralization at C2-3. **(B)** Transverse non-contrast CT image showing a large hyperattenuating mass into the vertebral canal at the intervertebral disc level C2-3. **(C)** Sagittal reconstructed non-contrast CT image showing hyperattenuating material into the vertebral canal between T1-12 (arrow). **(D)** Transverse non-contrast CT image showing a large hyperattenuating mass disc extrusion occupying most of the vertebral canal at the intervertebral disc level T11-12.

The CT characteristics of acute IVDE include hyperattenuating material within the vertebral canal, loss of epidural fat, and distortion of the spinal cord (**Figure 2**). A case series of 23 surgically confirmed thoracolumbar IVDE categorized the CT patterns of herniation into 3 groups: acutely extruded mineralized nucleus pulposus; acute extrusion of nucleus pulposus with hemorrhage; and chronic mineralized nucleus pulposus (65). The acutely extruded, mineralized nucleus pulposus group had heterogenous, hyperattenuating (mean 219 HU) extradural masses causing severe spinal cord compression. The acute extrusion of nucleus pulposus with hemorrhage group was found to have herniated material causing less compression, extending over multiple vertebral spaces, and that was only slightly distinguishable from the spinal cord (mean 59 HU). Since the spinal cord itself cannot be directly visualized on plain CT, spinal cord compression in both groups of dogs was inferred from loss of the hypoattenuating epidural space. In the chronic, mineralized nucleus pulposus, the extruded

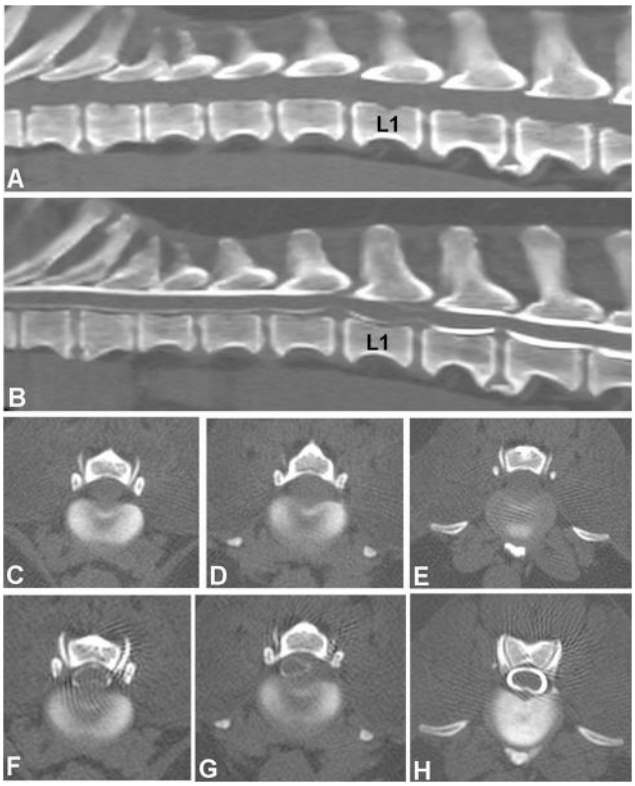

FIGURE 3 | Images of a 10-year old female spayed Beagle presented with an acute onset of non-ambulatory paraparesis and spinal pain. **(A)** Sagittal non-contrast CT image revealing areas of spondylosis and no obvious compressive lesion. **(B)** CT-myelography shows areas of ventral subarachnoid space compression at T13-L1, L1-2, and L2-3. Thinning of dorsal and ventral contrast columns is noted between L1-2. **(C–E)** images show transverse non-contrast CT images representing T13-L1, L1-2, and L2-3 intervertebral levels, respectively. Peridural fat is lost at L1-2 level. **(F–H)** CT-myelographic images at levels T13-L1, L1-2, and L2-3. Ventral, lateral and dorsal spinal cord compressions are noted at L1-2. Surgical decompression and biopsy confirmed that the compressive material was extruded disc material.

material was described as extremely hyperattenuating (mean 745 HU) and more homogenous (65). In a study with 111 dogs with thoracolumbar IVDE, a 3–11% increase in mean certainty scores for correct diagnoses was found when multi-planar reconstruction (MPR) CT images were used (mainly oblique transverse and curved dorsal MPR views) compared to 2D CT images (61).

Several studies compared the sensitivity of non-contrast CT to myelography and/or CT-myelography for the identification of IVDE in the thoracolumbar spine. A large retrospective study evaluated non-contrast CT and myelography in 182 dogs of varying breeds (58). Both methods had similar sensitivities, 81.8% for CT and 83.6% for myelography, though CT was better for chronically affected dogs (58). The same percentage of 81.8% was reported in another study using non-contrast CT (66). In this study, CT-myelography was required to confirm the diagnosis in the remaining 18.9% of cases (66). In a case series of 19 dogs, agreement with surgery was found in 94.7, 100, and 94.7% of cases using helical CT, conventional CT or myelography, respectively (37). In a prospective study using CT-myelography as the gold standard, non-contrast CT was found to be more sensitive than conventional myelography with a longitudinal and lateral localization accuracy of 91 and 94%, compared to 64 and 74% (**Figure 3**) (54).

A retrospective study suggested that CT-angiography was as diagnostic as CT-myelography in dogs with IVDE, with sensitivities of 97 and 94%, respectively (67). This finding was not corroborated by other studies. CT-angiography was evaluated in a prospective study and had the lowest sensitivity of all CT techniques (lower even than plain CT), at only 53% (52), whereas in another study it did not add any additional information compared to non-contrast CT (68). The diagnostic accuracy of CT for IVDH was 88–90% in two prospective studies where it was compared to MRI (69, 70).

A prospective study of 46 dogs with acute cervical and thoracolumbar myelopathies mimicking the diagnostic work-up of any acute spinal case, compared four modalities: non-contrast CT, CT-angiography, myelography, and CT-myelography, using as gold standard either surgery or necropsy (52). Almost 80% of dogs had extradural lesions and most had IVDE. Overall diagnostic sensitivity for all techniques was 66% for non-contrast CT, 53% for CT-angiography, 79% for myelography, and 97% for CT-myelography. Non-contrast CT had a sensitivity of 91% for IVDE, though it was not able to identify the associated spinal cord swelling in more severely affected dogs noted with other modalities. This led the authors suggest that CT-myelography should be the technique of choice for paralyzed patients to allow wider decompressive procedures (52).

A retrospective study of 555 dogs with thoracolumbar myelopathy investigated parameters that could be used to predict the need for additional imaging beyond non-contrast CT (68). The vast majority of the population had IVDE (94.6%, 525 dogs) and were chondrodystrophic (81.1%, 455 dogs), therefore the population studied was biased toward the likely diagnosis of IVDE. Additional imaging (either myelography, CT-myelography or MRI) was required in 17.4% of non-chondrodystrophic breeds, but in only 3.6% of Dachshunds. Timing of imaging was the second most important factor leading to additional imaging, with only 4.8% of dogs scanned during business hours requiring additional imaging. This number almost tripled (13.6%), when dogs were imaged out-of-hours. Patient preselection for CT or MRI explains this difference, as MRI was not available out-of-hours at the investigator's institution (68).

Based on all the available information, it is possible to conclude that non-contrast CT is as diagnostic as myelography for the localization of IVDE in chondrodystrophic dogs. It can be reliably used in the diagnostic investigation of cases suspected of having IVDD, primarily small breed, young dogs, emphasizing the point that case selection is key when electing a diagnostic work-up with non-contrast CT. The selection of non-contrast CT should also be undertaken with the acceptance that further imaging, either CT-myelography or MRI, might be required should no lesion or a lesion not matching the neurolocalization be identified (37, 52, 54, 59, 65, 68, 70, 71).

MAGNETIC RESONANCE IMAGING

MRI is considered the gold standard for diagnostic imaging in IVDD in both humans and veterinary patients. Manipulation of the various imaging characteristics of tissues allows the multiple anatomical structures within the vertebral column to be distinguished, including the supporting ligamentous structures, synovial joints, bone marrow, nerve roots, spinal cord parenchyma, cerebrospinal fluid, epidural fat, and the layers of the IVD (60). Images can be acquired in multiple planes without repositioning the patient, which is an advantage because image quality is always the same. However, the more planes, the longer the examination, a disadvantage compared to CT. Also, whereas myelography might be required in conjunction with CT for the diagnosis of IVDD, this is not necessary with MRI because of the ability to alter tissue contrast by applying different acquisition sequences. Thus, the risks associated with myelography are avoided. A key point is that IVDE can go undetected on myelography or CT, but the likelihood of a false negative result with MRI is much lower (30, 69, 70, 72).

There is substantial variation in MRI quality in veterinary practice with both low and high field MRIs available. Due to upfront and maintenance costs, low-field (0.2–0.4 T) units currently greatly outnumber high-field ones, primarily in private referral practices (73). While low field units offer diagnostic quality in the vast majority of studies, the compact magnet design of some veterinary scanners may not allow scanning or have significant quality issues in the caudal cervical or cranial thoracic spine of very large or giant breed dogs (73). Similarly, these scanners have a small maximal field of view, that may require patient repositioning when examining larger regions, thus making it more time consuming than with high field MRI systems. Spinal studies in very small dogs or cats (<3 kg) can also be of questionable diagnostic quality, as many low-field systems cannot have slices thinner than 3 mm. Enlarging the field of view (FOV), increasing slice thickness, and increasing the number of acquisitions will increase the signal to noise ratio (which means improved image quality), but these adjustments reduce image resolution and prolong anesthesia and magnet time (59). High field MRI units typically range from 1.0 to 3.0 T, with the most popular unit in veterinary hospitals being the 1.5 T. In general, the higher the field strength, the higher the signal to noise ratio and the faster the imaging times, to a limit (71).

As the diagnostic quality and accuracy of MRI for the diagnosis of IVDD are dependent on several technical aspects, a brief overview of these factors is warranted. For a comprehensive review of technical aspects, the reader is referred to other sources (74–76).

POSITIONING

Patients are typically positioned in dorsal recumbency. It is extremely important to achieve a straight spinal alignment to allow comparison of multiple intervertebral sites on sagittal images. Sandbags and foam wedges are very useful for this purpose (71). The high prevalence of vertebral body malformation, leading to kyphosis and scoliosis in brachycephalic screw-tailed dogs, like the French bulldog, can make straight spinal alignment impossible to be achieved (77).

ACQUISITION PLANES

The sagittal plane is used initially to localize specific intervertebral regions to be investigated with transverse slices. The FOV of the sagittal plane in low field MRIs can be small, so determination of the affected intervertebral disc(s) level can be challenging in the thoracolumbar region without reliable cranial and caudal vertebral references. A strategy commonly used is the identification of celiac and mesenteric arteries. In one study the celiac artery and mesenteric arteries were ventral to the first lumbar vertebra in 71 and 97% of dogs, respectively (78). In another study where they were under the body of L1 in only 59.6% of dogs, with an additional 11.2 and 20.2% under the intervertebral discs T13-L1 and L1-2, respectively (79). As such, their location is variable and they do not represent an accurate vertebral landmark. The dorsal plane can be very useful in these instances since the ribs and transverse processes can be easily identified. In cases where dorsal images are not available, sagittal (parasagittal) images can be used (79). Rib articulations and lumbar transverse processes can be easily distinguished based on different shapes and heights relative to the vertebral canal (79). The transverse plane is essential for circumferential assessment of spinal cord compression and lateralization. Selection of transverse sections is typically based on the identification of compressive sites on sagittal images. The reliability of sagittal T2-weighted (T2W) images alone for the determination of compressive disc lesions was determined to range from 81.4 to 89.0%. This means that without acquiring transverse images, a compressive lesion would have been missed in 11–18.6% of dogs, with almost 10% being considered clinically relevant (80). It is therefore recommended to always obtain transverse images at least encompassing the disc spaces immediately cranial and caudal to the supposedly affected disc, or the entire spinal region if multiple areas appear abnormal (80).

IMAGE SEQUENCES

While there are variations regarding preferences for sequences and planes, it is generally agreed that T2W and T1W images in both sagittal and transverse plane should be routine (**Figure 4**). The importance of transverse T1W images is typically minimized, although they are very useful to help distinguish extradural compression caused by extruded nuclear material, typically hypointense, from soft tissue masses and hemorrhage. Similarly, post-contrast T1W images, although not routinely used, increased the ability to determine the site and side of IVDE in one study (30). Ultrafast heavily T2W, short T1 inversion recovery (STIR), and gradient echo sequences (GRE, also known as T2* or fast field echo—FFE) are also routinely acquired (**Figure 4**). A low-field MRI study looked at sagittal STIR images compared to sagittal T2W and found no difference (81). However, the parallel evaluation of the paired sagittal T2W and STIR series yielded a higher sensitivity than using either

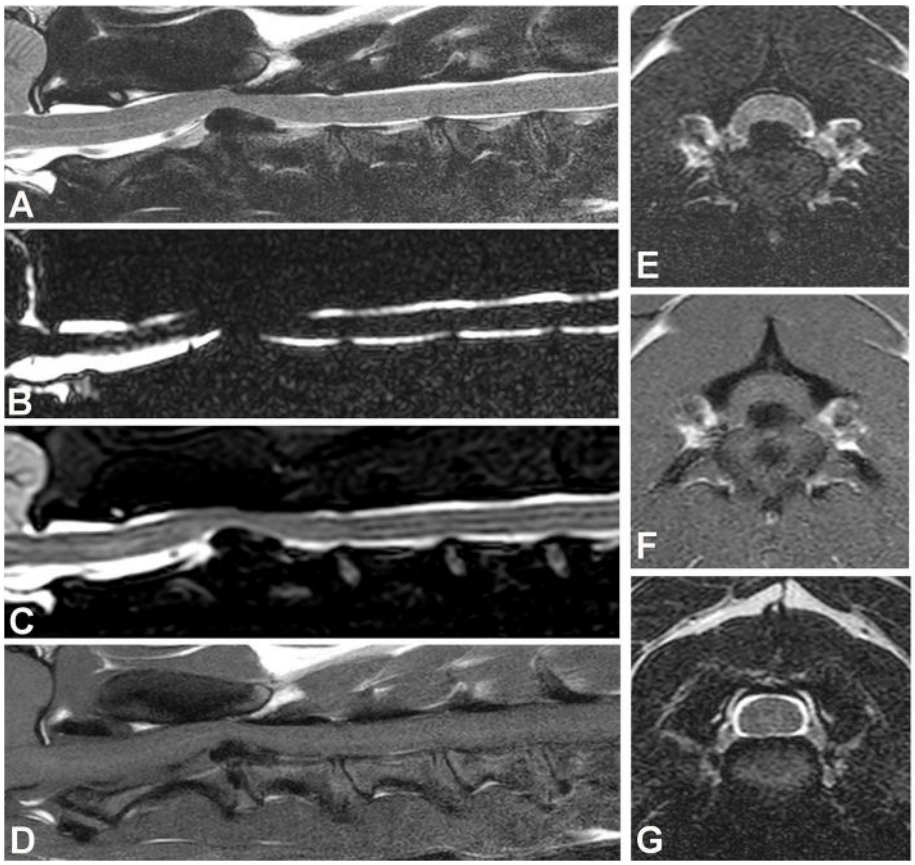

FIGURE 4 | Images of a dog with cervical intervertebral disc extrusion. **(A)** Sagittal T2W image. **(B)** Sagittal HASTE image. **(C)** Sagittal STIR image. **(D)** Sagittal T1W image. All sagittal images show ventral extradural compression in the ventral aspect of the vertebral canal at the intervertebral level C2-3. Note how the compressive material is hypointense in all images, though image resolution varies with different sequences. The signal of intervertebral discs *in situ* also varies in all sequences. **(E)** Transverse T2W image showing a large hypointense mass causing spinal cord compression at the C2-3 level. **(F)** Transverse T1W image showing the hypointense compressive material at C2-3. **(G)** Transverse T2W image showing the C3-4 level with no spinal cord compression.

sagittal screening series in isolation (81). Heavily T2W images, also called "myelo-MRI" or HASTE (Half-Fourier acquisition single-shot turbo spin echo) images, can be used to highlight the cerebrospinal fluid signal and rapidly identify an area of interest (**Figure 4**). They are very fast sequences, so a valid addition in MRI spinal protocols, though two studies showed that they are not as reliable as sagittal T2W images for identification of compressive IVDD lesions (80, 82). They are useful in distinguishing acute from chronic cases of IVDH and as a predictor of progressive myelomalacia.

MRI OF INTERVERTEBRAL DISC DEGENERATION

MRI allows clear visualization of the intervertebral disc. The nucleus pulposus and annulus fibrosus are best appreciated on T2W images. In the healthy intervertebral disc, both the nucleus pulposus and the inner part of the annulus fibrosus are hyperintense on T2W images, appearing as a hyperintense ellipsoid area on sagittal images (83). The degree of brightness of the nucleus pulposus in the T2 signal correlates with the proteoglycan concentration but not with water or collagen concentration (84). Annular and nuclear disc material cannot be distinguished in T2W images of degenerative IVDs due to biochemical changes in the extracellular matrix of the nucleus. A lower water content and a shift of proteoglycan composition of the nucleus pulposus results in a lower signal intensity in T2W images, making annular and nuclear material iso-intense to each other (84, 85). It has been long proposed that IVDE results from IVD degeneration (4). It is important to emphasize that disc degeneration is a very common finding in clinically normal dogs, and *per se*, does not lead to clinical signs, except in uncommon cases of discogenic spinal pain (71, 86).

In human medicine, the Pfirrmann system is the most widely used system for grading IVD degeneration on the basis of sagittal T2W MRI findings (87). It is based on a system for grading gross pathological changes in IVDs proposed by Thompson et al., which is the most commonly used criterion-referenced standard in human medicine (88). The Pfirrmann system has been validated in dogs, showing good correlation with age and chondrodystrophic breeds (89), as well as the Thompson

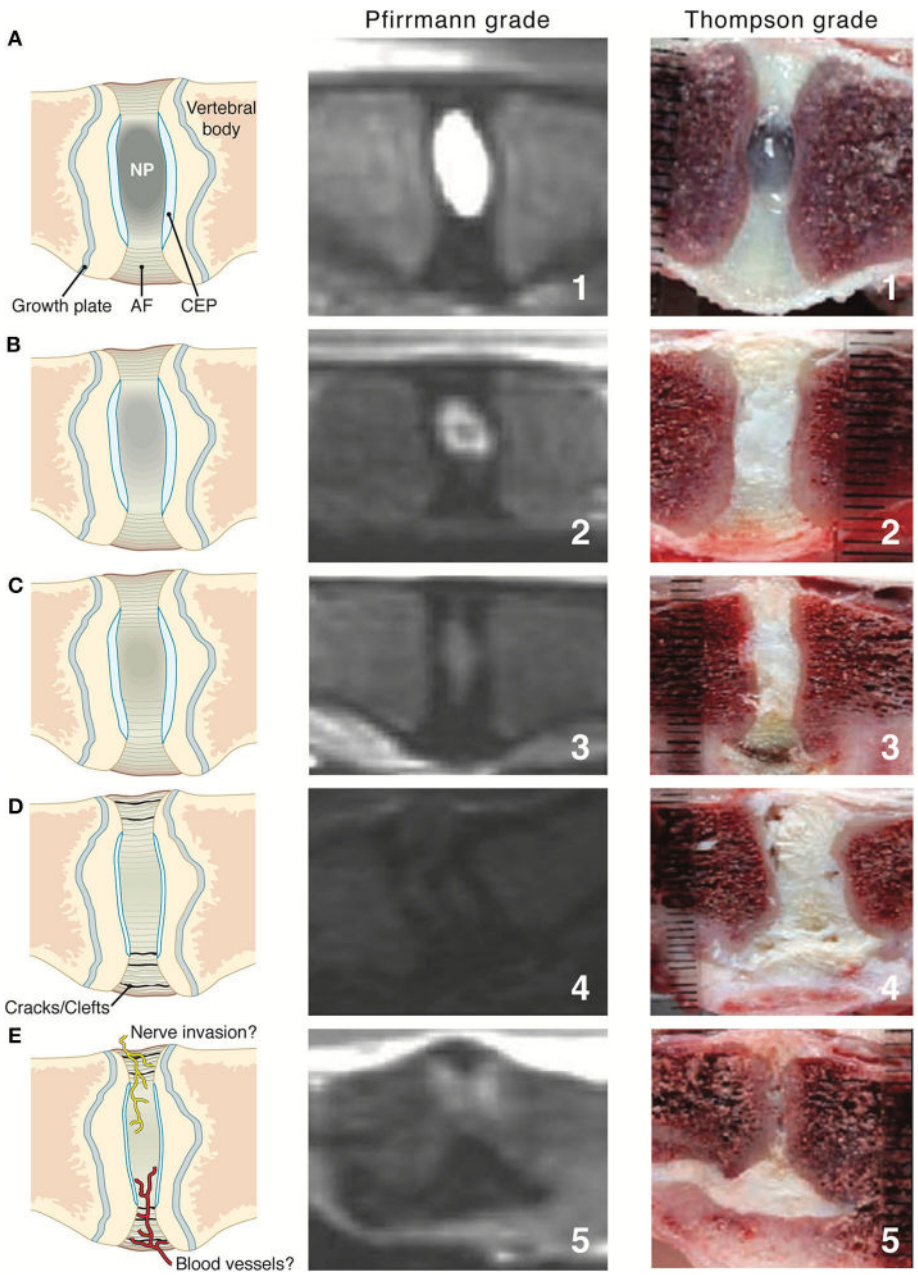

FIGURE 5 | Intervertebral disc (IVD) maturation from young to early and late stage IVDD where the first column shows illustrative representations throughout the stages **(A–E)**, middle column shows Pfirrmann grading via T2W MR images and last column showing Thompson grading of canine IVD. AF, annulus fibrosus; CEP, cartilaginous end plate; NP, nucleus pulposus. Pfirrmann grade and Thompson grade images adapted with permission from Bergknut et al. (90) and Thompson et al. (91).

macroscopic grading scheme for disc degeneration (**Figure 5**) (20, 90). A study looked at a modified Pfirrmann grading system using images from a novel T1 weighted FFE-sequence images compared with T2W images (92). It was concluded that T2W images should remain the sequence of choice to grade IVD degeneration (92).

The initiating factor for IVDD in humans is thought to be a loss of diffusional capacity of the vertebral endplate blood vessels that provide nutrition for the nucleus pulposus (93).

Endplate changes have also been thought to be associated with canine IVDD (94, 95), and were recently investigated in dogs with and without IVDD (96). The most common abnormal endplate change was hyperintensity on T1W and T2W images (97.2% of dogs). These changes were significantly associated with the presence of IVDD in the adjacent disc. Dogs with vertebral

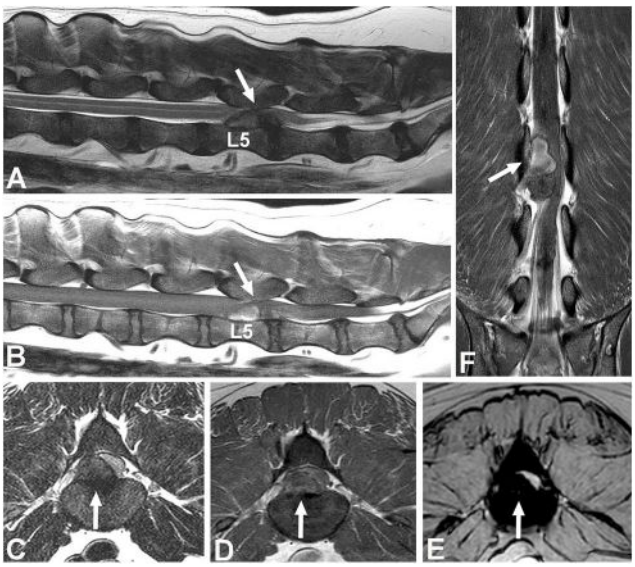

FIGURE 6 | Images of a female spayed, 6-year-old, mixed breed dog with an acute onset of paraparesis, spinal pain, and fecal and urinary incontinence. **(A)** Sagittal T2W image showing a large mass with mixed signal intensity between L5-6 (arrow). **(B)** Sagittal T1W image showing that the cranial aspect of the mass is hyperintense (arrow). **(C)** Transverse T2 image showing severe spinal cord compression caused by a large hypointense mass between L5-6 (arrow). **(D)** Transverse T1W contrast-enhanced image showing mild heterogeneous contrast enhancement of the hypointense lesion. **(E)** Gradient echo image showing marked hypointensity. **(F)** Dorsal T1W contrast-enhanced image showing contrast enhancement of the cranial aspect of the compressive lesion. Note the length of the compressive lesion. Surgical decompression and biopsy confirmed that the compressive material was extruded intervertebral disc with hemorrhage.

endplate changes anywhere in the vertebral column were not, however, more likely than dogs without vertebral endplate to have IVDD (96).

MRI FINDINGS IN INTERVERTEBRAL DISC EXTRUSION

MR imaging features of IVDE include extradural compression of the spinal cord centered over or near the intervertebral disc space. This mass effect caused by the extruded material causes compression and/or displacement of the spinal cord, seen on T2W images as displacement or loss of the hyperintense signal associated with subarachnoid and epidural spaces. Extruded nucleus pulposus is typically identified as a hypointense mass within the epidural space on T1W and T2W images (**Figure 4**). It can be characterized as dispersed if it is not associated with the affected intervertebral space and spread throughout the epidural space, or non-dispersed if it remains in contact with the affected IVD (94). The MRI features of IVDE in cats are similar to dogs, with the difference that most reported cats have had disc extrusions in the lumbar vertebral column, as opposed to the caudal thoracic and thoracolumbar (T12-13, T13-L1) as seen in small breed dogs (21, 97–99).

Epidural hemorrhage associated with IVDE can result in a wide range of signal intensities, including signal void, thus a diagnosis of IVDE should not rely on one pattern of signal intensity (**Figure 6**) (100, 101). Gradient echo sequences can confirm the presence of hemorrhage.

The degree of spinal cord compression can be categorized based on morphologic compression scales (102). A common categorization is based on the percentage of reduction in spinal cord diameter, graded as mild (<25%), moderate (25 to 50%), or severe (>50%) (102, 103). Morphometric estimates of cross-sectional area of either spinal cord or extruded material offer even more precise information (104–106). The degree of spinal cord compression seen on transverse MR images in cases of IVDE was associated with the severity of pre-operative neurological signs in the cervical spine (107), but not in the thoracolumbar region (94, 108, 109).

Contrast enhancement of extradural compressive material can be seen in dogs with IVDH, primarily in dogs with IVDE (60%) compared with 16% of dogs with IVDP (110, 111). Various patterns of enhancement can be seen, including homogeneous, heterogeneous, and even peripheral enhancement patterns (110). These findings can be misinterpreted as a granulomatous or neoplastic lesion, thus it is important to be aware of these contrast patterns to avoid misdiagnosing IVDE as another condition (71). Meningeal enhancement adjacent to the extruded disc material was also noted in up to 40% of dogs (111).

Paravertebral muscle signal changes were seen in 36% of dogs with IVDE or ANNPE in one study. These changes are characterized by an edematous pattern that is hyperintense on T2W and iso- or hypointense on T1W sequences and are best visualized on T2W fat-suppressed sequences. No signal void is seen on T2*W GRE and a variable degree of contrast enhancement is seen in 45% of dogs. Paravertebral signal alterations are more commonly seen in disc extrusions caudal from L1 and in animals with a more severe neurological grade. Histopathology typically does not reveal specific abnormalities and the underlying pathomechanism might be related to ischemia, muscle spasm or denervation edema (112).

In dogs with acute thoracolumbar disc extrusion, areas of spinal cord hyperintensity on T2W images can be observed (**Figure 7**). These spinal cord hyperintensities have been shown to correlate with the severity of neurologic signs at presentation (94, 109, 113). The specific pathologic processes associated with hyperintensity on T2W images are not fully known, but have been thought to involve necrosis, myelomalacia, intramedullary hemorrhage, inflammation, and edema (114–116). The prognostic value of spinal cord hyperintensity on T2W images is subject of considerable controversy, with newer studies (117, 118), questioning older findings (109, 113, 119). Interestingly, a recent low-field MRI study of IVDE found spinal cord hyperintensity on T2W images in 18% of dogs in the preoperative period. MRI was repeated after decompressive surgery (immediately after surgery in most dogs) and the number of dogs with hyperintensity almost doubled (34%) (105). The relationship between spinal cord hyperintensity and prognosis of IVDD can be found in the article "Prognostic factors in acute spinal cord injury" in this issue.

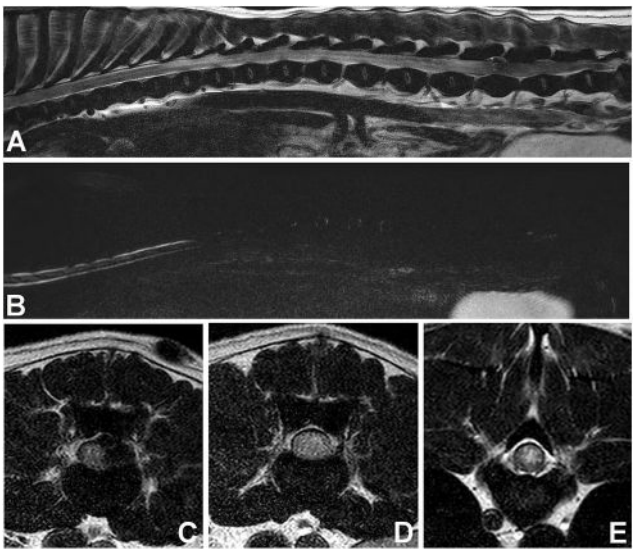

FIGURE 7 | Images of female spayed, 13-year-old Cocker Spaniel with an acute onset of paraplegia. **(A)** Sagittal T2W image showing a large area of diffuse spinal cord hyperintensity. **(B)** Sagittal HASTE image showing diffuse spinal cord swelling. **(C)** Transverse T2W image at the L5-6 level showing lateralized spinal cord compression and spinal cord hyperintensity. **(D)** Transverse T2W image at the L3-4 level showing diffuse spinal cord hyperintensity and apparent swelling. **(E)** Transverse T2W image at T9-10 level showing centrally located spinal cord hyperintensity. Myelomalacia and intervertebral disc extrusion at L5-6 were confirmed at post-mortem examination.

Two prospective studies compared non-contrast CT (helical multislice systems) vs. MRI (1.0 T systems) for the diagnosis of thoracolumbar IVDH, using surgery as the gold standard. The first study had 44 dogs and found that the sensitivity of MRI was 98.5%, compared with 88.6% for CT for identifying the site of IVDH (69). In another study with 40 dogs with confirmed IVDE, a lesion was detected using MRI in all dogs, whereas CT did not allow identification of a lesion in 4 out of 40 dogs (10%) (70). It can be concluded based on these studies that MR imaging has a sensitivity approximately 10% higher than non-contrast CT for the diagnosis of thoracolumbar IVDE. This 10% difference was also seen when MRI was prospectively compared to myelography (30). The overall diagnostic accuracy of MRI in dogs with IVDE is therefore between 98.5 and 100% (30, 69, 70, 72).

While MRI has been reported to be more accurate than CT in discriminating IVDE from IVDP, this distinction can still be difficult based on MRI (69). In order to assist with this differentiation, criteria have been proposed and are discussed in the IVDP section below.

MRI AND SURGERY

MRI findings have been compared to actual surgical findings by assessing the cranio-caudal extent of extruded disc material using calipers intraoperatively (72). In two thirds of dogs, authors observed 100% agreement between MR imaging and surgical findings, but only 50% agreement in the remaining third of dogs (72). A prospective study compared surgical planning using MRI and CT in a population of 40 dogs with IVDE. Hemilaminectomy planning varied in about 50% of cases between MRI and CT (43.5–66.6%). In all cases where planning differed, a significantly larger hemilaminectomy defect (a greater number of articular facets removed) was planned using MRI compared to CT (70). MRI is also a very useful modality to investigate patients in the post-operative period (105, 120–122). MR imaging can differentiate residual extruded nucleus from hemorrhage, based on signal intensity and gradient echo characteristics (101, 120). Gelatin sponges can also be easily differentiated from residual disc material based on their sharply demarcated shape and hyperintensity on T2W images (120). MRI was also very accurate (100%) for identification of late recurrent disc extrusion either at the same or different site (mean interval between initial surgery and recurrence 404.5 days) in a study comparing it to myelography (122).

A recent prospective low-field MR study evaluated the agreement between surgeon's perception of spinal cord decompression and residual disc on postoperative MR images acquired immediately after surgery in most cases (105). In most instances in which the MRI results differed from the surgeon's perception, the degree of surgical decompression was perceived as satisfactory by the surgeon, but was revealed as unsatisfactory via MRI. This was observed primarily in large disc extrusions (105). The clinical outcome of those dogs with unsatisfactory spinal cord decompression was worse than those with adequate spinal cord decompression (105). Another prospective high-field MRI study found residual spinal cord compression in 44% of dogs treated with mini-hemilaminectomy (120). The median percentage of residual material was 7.7% of the preoperative volume, and the mean degree of residual spinal cord compression was 10.9%, compared with 37% before surgery. The volume of residual disc in this study did not appear to have an influence on outcome (120).

MRI FEATURES OF PROGRESSIVE MYELOMALACIA

Progressive myelomalacia (PMM) is one of the worst complications seen in dogs with IVDE. It is reported to occur in ∼10–17.5% of paraplegic dogs with absent pain perception (123, 124), and as high as 33% in French Bulldogs (125). Even though it is a clinical diagnosis in many cases, it is important to recognize its MRI features. MR imaging of PMM shows severe spinal cord swelling, more easily appreciated in the heavily T2 sequences (HASTE, myelo-MRI), along with diffuse parenchymal hyperintensity on T2W images over several spinal cord segments (**Figure 7**). On transverse T2W images, the hyperintensity is centered in the gray matter. FLAIR images will also reveal diffuse hyperintensity, whereas GRE images will show diffuse hypointensity (126, 127). Since myelomalacia is a form of hemorrhagic necrosis of the spinal cord, depending on when MRI studies are performed, it is possible to also observe hypointensity in the T2W images (126). A study proposed that

diffuse spinal cord hyperintensity 6 times longer than the body of L2 vertebral body was suggestive of PMM (126), however this MRI pattern was seen in only 45% of dogs in another study (128). Others suggested that an area of hyperintensity 4.57 times longer than the body of L2, or of loss of cerebrospinal fluid signal on HASTE (MR-myelo) sequences 7.4 times longer than the body of L2 were at higher risk for PMM (**Figure 7**) (129, 130). Importantly, dogs can develop PMM without the presence of spinal cord hyperintensity on MRI (129).

INTERVERTEBRAL DISC PROTRUSION

In contrast to IVDE, the degenerative process in IVDP is much slower, with concurrent changes in the annulus fibrosus and nucleus pulposus. The annulus progressively loses its structural integrity, which allows the nucleus pulposus to move dorsally into the weakened annulus fibrosus. This will cause gradual protrusion of the dorsal annulus fibrosus into the vertebral canal, ultimately resulting in chronic progressive spinal cord compression. The pathophysiology, classification and clinical presentation of animals with IVDP is discussed in more detail in a companion article of this issue by Fenn et al.

Although IVDP is a common spinal disorder (131), only a few studies have focused on the specific imaging characteristics of this type of IVDD (46, 132). There are, however, indications that thoracolumbar IVDE and IVDP might be associated with differences in recommended surgical techniques, surgical complications, and outcomes after surgical and medical management (46, 133–135). Differentiation between IVDD subtypes is therefore clinically important.

Similar to IVDE, spinal radiographs cannot be used to confirm a diagnosis of IVDP. Common radiographic abnormalities seen in dogs with IVDP include vertebral endplate sclerosis (67%), and spondylosis deformans (47%) with narrowing of the intervertebral disc space occurring less commonly (25%) (46). Although spondylosis deformans has been associated with the presence of IVDP, this radiographic finding is also commonly observed in clinically normal animals. Spondylosis deformans can therefore not be considered a reliable indicator for IVDP or IVDD in general (136). Additionally, survey radiographs can be normal in dogs with IVDP but are often performed to look for other diseases with similar clinical presentation, such as vertebral neoplasia.

Until two decades ago, myelography was probably the most commonly used imaging modality to diagnose spinal conditions in veterinary medicine (16). Myelographic criteria have been reported to differentiate between IVDE and IVDP (46). The criteria for IVDP were (1) thinning and dorsal or dorsolateral deviation of the contrast columns, (2) thinning of the contrast columns is mild, (3) thinning of the contrast columns is focal and centered on the cranial and caudal boundaries of the affected disc, and (4) there is a symmetrical distribution of contrast column thinning cranial and caudal to the affected disc (**Figure 1**). This compares to the myelographic criteria for IVDE previously listed in the IVDE section. It was reported that application of these myelographic criteria allowed reliable differentiation between IVDE and IVDP when compared to intraoperative findings (46). For reasons outlined above including diagnostic limitations and procedural risks, myelography has now largely been replaced by more advanced diagnostic imaging modalities, such as CT and MRI (71).

Although CT is widely used to diagnose acute and chronic IVDE, there are no studies that have specifically focused on the use of CT to diagnose IVDP. The herniated material in IVDP is not mineralized and is therefore difficult to visualize on non-contrast CT. Spinal cord compression can further not be directly visualized on non-contrast CT and it is therefore likely that it cannot reliably be used to obtain a diagnosis of IVDP (59). Although fibrotic disc protrusions can occasionally be seen on CT, it is important to consider that a large number of dogs with IVDP have multiple affected intervertebral discs and that IVDH can also occur in dogs without clinical signs, further complicating the interpretation of CT findings in this population. While non-contrast CT failed to identify any of 16 cases of non-mineralized IVDP in one study, CT-myelography was able to diagnosis all of these cases (52). However, CT-myelography is associated with similar safety limitations as myelography. The CT-myelographic appearance of IVDP is typically characterized by a ventral extradural spinal cord compression with soft tissue density characteristics, overlying and not exceeding the intervertebral disc space. Chronic spinal cord compression can also result in irreversible spinal cord pathology and spinal cord atrophy (137). A circumferential widening of the subarachnoid space with a more triangular shaped spinal cord has been suggested to represent spinal cord atrophy (138). Although this is considered a negative prognostic indicator in human medicine, the prognostic role of this imaging finding is currently unclear in dogs with chronic spinal cord compressions (139).

MRI has become the imaging modality of choice for most spinal disorders in veterinary medicine (71). Although the MRI characteristics of IVDD have extensively been reported, only a few studies have focused on IVDP (132, 140). One study identified four MRI variables that could be considered independent predictors of thoracolumbar IVDE vs. IVDP (**Figure 8**) (132):

- Midline instead of lateralized intervertebral disc herniation was associated with a diagnosis of IVDP
- Partial instead of complete loss of the hyperintense signal of the nucleus pulposus was associated with a diagnosis of IVDP
- The presence of a single intervertebral disc herniation instead of multiple intervertebral disc herniations was associated with IVDE
- Herniated material dispersed beyond the boundaries of the intervertebral disc space was associated with a diagnosis of IVDE.

In a later study by the same group, using these four criteria significantly improved diagnostic accuracy and interobserver agreement of MRI to differentiate thoracolumbar IVDD subtypes (140). It is currently unknown if these proposed MRI variables are also useful for cervical IVDH. It is worth mentioning that IVDP can be lateralized in both cervical and thoracolumbar regions.

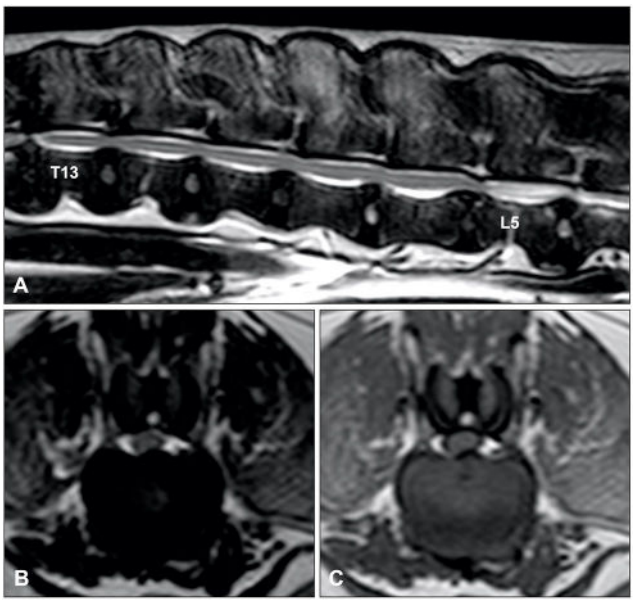

FIGURE 8 | Sagittal T2-weighted **(A)**, and transverse T2-weighted **(B)** and T1-weighted MR **(C)** images of a 10-year old Dalmatian with multiple IVDPs between T13 and L6. The nucleus pulposus of most intervertebral discs has maintained some T2-weighted hyperintense signal, multiple discs are affected, intervertebral disk herniations are contained within the boundaries of the intervertebral disc space, and have resulted in midline spinal cord compression.

A common MRI finding in animals with chronic intervertebral disc herniations is focal T2W intraparenchymal intensity changes at the site of spinal cord compression (132). As previously suggested, the exact meaning of these abnormalities is unclear, but intraparenchymal intensity changes are likely to represent a wide spectrum of reversible and potentially irreversible pathological changes, such as edema, gliosis, and malacia (141). Although it has been suggested that intraparenchymal hyperintensity can aid in differentiation of clinically relevant from irrelevant disc-associated spinal cord compressions (104, 142), it is currently unclear if specific subtypes of intraparenchymal intensity changes can be used as prognostic indicators (**Figure 9**) (143). The high sensitivity of MRI can also complicate interpretation of MRI studies as intervertebral disc degeneration, herniation and even spinal cord compression can be observed on MRI studies of clinically normal, especially older, dogs (71, 104, 144). This underlines that abnormalities observed on MRI studies should always be correlated with results of thorough clinical and neurologic examinations, and that accurate interpretation of MRI studies requires experience and expertise.

Intervertebral disc protrusion can also be a prominent component of more complex and multifactorial neurological syndromes, such as degenerative lumbosacral stenosis and disc-associated cervical spondylomyelopathy (8, 143). Although a detailed description of these neurological syndromes is beyond the scope of this article and strict diagnostic criteria are lacking, it is common for dogs with these neurological syndromes to have a combination of bony and soft-tissue abnormalities contributing to vertebral canal stenosis. Additional abnormalities that can be observed on diagnostic imaging studies of dogs with degenerative lumbosacral stenosis include ligamentum flavum hypertrophy, articular process hypertrophy, telescoping of the sacral dorsal lamina, transitional vertebra, osteochondrosis, and vertebral malalignment (8). Additional abnormalities that can be seen in dogs with disc-associated cervical spondylomyelopathy include ligamentum flavum hypertrophy, craniodorsal vertebral tilting, abnormally shaped vertebral body, funnel shaped vertebral canal and articular process hypertrophy (143).

HERNIATION OF NONE OR MINIMALLY DEGENERATIVE NUCLEUS PULPOSUS EXTRUSION

Since MRI has become more widely available in veterinary medicine, additional types of IVDH have been recognized, which are characterized by sudden herniation of non-degenerative or minimally degenerative nucleus pulposus. The two most common types of extrusion of non-degenerative or minimally degenerative nucleus pulposus are acute non-compressive nucleus pulposus extrusion (ANNPE) and hydrated nucleus pulposus extrusion (HNPE) (145). Another type of IVDH that can be associated with herniation of non-degenerative nucleus pulposus is intradural/intramedullary intervertebral disc extrusion (IIVDE). It should however be noted that IIVDE has also been reported after IVDE (146). In contrast to IVDE and IVDP, herniation associated with ANNPE and HNPE occurs without advanced degeneration and dehydration of the intervertebral disc. Acute herniation of non-degenerative, well-hydrated nucleus pulposus material is predominantly associated with contusive spinal cord injury and has an uncertain role for sustained spinal cord compression. Acute non-compressive nucleus pulposus extrusion and HNPE are therefore associated with distinct pathophysiology, clinical presentation, diagnostic imaging findings, and recommended treatment options compared to that of compressive IVDH (145). The classification, terminology and clinical presentation of different types of IVDH are discussed in detail in a companion article in this research topic by Fenn et al. ("Canine Intervertebral Disc Disease: The Current State of Knowledge"). A diagnosis of ANNPE, HNPE, and IIVDE is based on a combination of compatible clinical signs and diagnostic imaging findings. Magnetic resonance imaging is considered the imaging modality of choice for ANNPE and HNPE, while CT-myelography has been considered particularly sensitive for diagnosis of IIVDE (6, 147, 148).

ACUTE NON-COMPRESSIVE NUCLEUS PULPOSUS EXTRUSION

Acute non-compressive nucleus pulposus extrusion is characterized by a sudden extrusion of non- or minimally degenerative nucleus pulposus without residual spinal cord

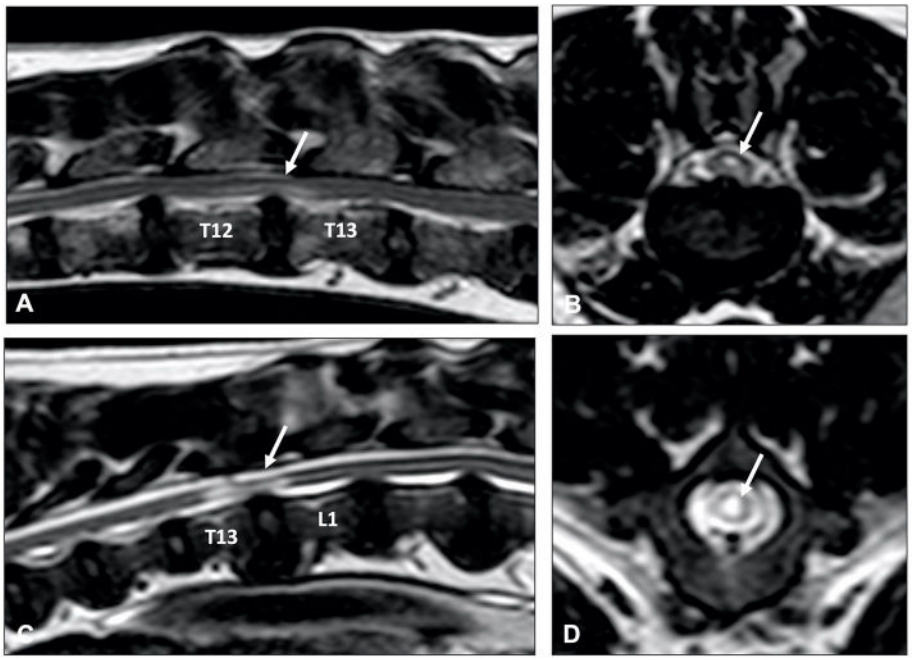

FIGURE 9 | Sagittal **(A,C)** and transverse **(B,D)** T2-weighted MR images of a 9-year old Labrador retriever **(A,B)** and an 11-year old Poodle **(C,D)** with thoracolumbar IVDP. A poorly demarcated, diffuse intraparenchymal hyperintensity is seen overlying the affected intervertebral disc (arrow) in the Labrador retriever, while a well-demarcated and bright intraparenchymal hyperintensity is seen in the Poodle (arrow). It is currently unclear if this difference in intraparenchymal intensity characteristics is associated with differences in clinical presentation or outcome.

compression. This condition has been reported with various terms over the years, namely Hansen Type III IVD herniation (even though Hansen did not report it), traumatic IVD extrusion, IVD explosion, missile discs, and high-velocity low-volume IVD extrusion. Affected animals typically present with a peracute onset of possibly strongly lateralized clinical signs, often during strenuous exercise or trauma. Severity of clinical signs should not progress after the initial 24 h and sustained spinal hyperesthesia should not be present.

Survey radiographs can be normal or reveal a narrowed intervertebral disc space in animals with ANNPE. This radiographic finding is not specific and does not help differentiating ANNPE from other types of IVDH. Myelography can reveal a small focal extradural compressive lesion overlying an intervertebral disc space, with an adjacent intramedullary pattern caused by spinal cord swelling (**Figure 10**) (149). Although it is unclear how specific these findings are for a diagnosis of ANNPE, one study suggested that myelography could reliably be used to differentiate between ANNPE and IVDE (150). An almost perfect interobserver agreement ($\kappa = 0.8$) was reached to make a presumptive diagnosis of ANNPE (instead of IVDE) using myelography. The myelographic studies of all dogs with ANNPE demonstrated an intramedullary pattern and an additional extradural pattern was seen in approximately half of dogs. The degree of spinal cord swelling was not associated with severity of clinical signs or outcome (150). Although no specific CT or CT-myelography findings have been reported in dogs with ANNPE, it can be hypothesized that these imaging modalities will show similar abnormalities as survey radiography and myelography, respectively. As mentioned above, ANNPE can be associated with external trauma, such as road traffic accidents or falls from a height. It has been suggested that onset of clinical signs is associated with external trauma in 40% of dogs and more

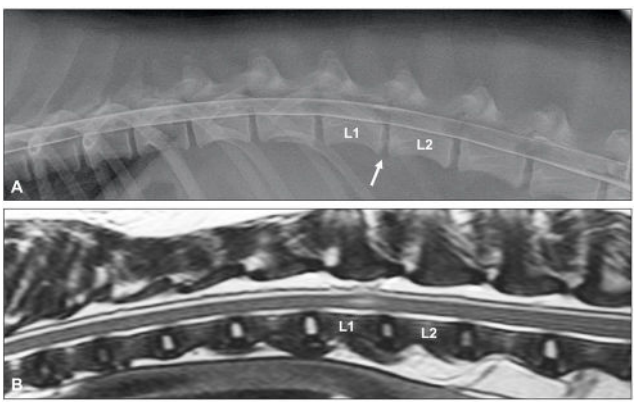

FIGURE 10 | Lateral myelogram **(A)** and sagittal T2-weighted image **(B)** of a 4-year old Soft-Coated Wheaten terrier with an L1-L2 ANNPE. **(A)** Although the L1-L2 intervertebral disc space is narrowed (arrow), the myelographic pattern appears to be normal. **(B)** A focal intramedullary hyperintense lesion just cranial from the L1-L2 disc space can be observed. The homogenously hyperintense nucleus pulposus has a reduced volume and the intervertebral disc space is narrowed.

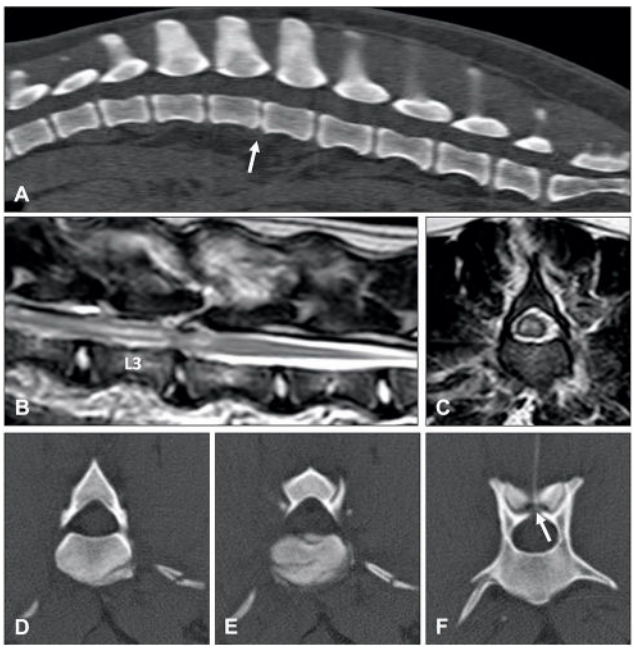

FIGURE 11 | ANNPE can occur after external trauma. CT can be used to exclude or characterize concurrent vertebral fractures and luxations. **(A)** Sagittal reconstructed CT study of a 6-year old Boxer who became paraplegic immediately after a road traffic accident. Although a narrowed intervertebral disc space (arrow) is in itself not sufficient to confirm a diagnosis of ANNPE, the CT study was able to exclude the presence of a vertebral fracture and luxation. Sagittal **(B)** and transverse **(C)** T2-weighted MR images and transverse CT images at the levels of caudal L3 **(D)**, the caudal vertebral endplate of L3 **(E)** and cranial L4 **(F)** of a 4-year old Border collie who ran into a tree. Although the MRI **(A,B)** reveals abnormalities suggestive for ANNPE, there are also abnormalities suggestive for external trauma, such as diffuse and bilateral hyperintensities in the paravertebral muscles. **(C–E)** The CT study revealed multiple fractures and **(D)** a mildly increased distance between the cranial and caudal articular processes.

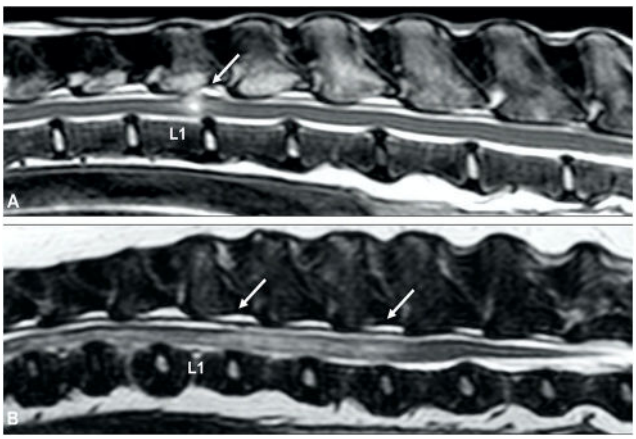

FIGURE 12 | T2-weighted sagittal MR images of **(A)** an 8-year old cross breed with ANNPE and **(B)** a 4-year old Staffordshire Bull terrier with ischemic myelopathy. **(A)** Focal hyperintensity can be seen overlying the L1-L2 intervertebral disc. The L1-L2 intervertebral disc space is narrowed and has a reduced volume of T2W hyperintense nucleus pulposus. **(B)** A relatively well-demarcated longitudinal hyperintensity can be seen between L1 and L4.

than 70% of cats with ANNPE (147, 151). This observation has two important clinical consequences: (1) ANNPE and vertebral fracture and luxation are two important differential diagnoses in animals suffering spinal cord dysfunction immediately after external trauma, and (2) animals can have concurrent ANNPE and vertebral fracture and luxation after external trauma has occurred (**Figure 11**). CT is considered the diagnostic modality of choice for animals with suspected vertebral fracture and luxation (152, 153). This diagnostic imaging technique can therefore be utilized in an emergency setting to exclude or characterize a vertebral fracture and luxation after a witnessed or suspected external trauma, but it might miss the presence of ANNPE (**Figure 11**).

MRI is considered the imaging modality of choice for ANNPE. The following MRI characteristics have been proposed to make a presumptive diagnosis of ANNPE in dogs (147):

- Focal intramedullary, often well-demarcated, spinal cord T2W hyperintensity, which is typically isointense on T1W sequences
- The lesion is located over an intervertebral disc space and is often lateralized
- The nucleus pulposus has a homogenous T2W hyperintense signal and a reduced volume. This is associated with a narrowed intervertebral disc space
- There is a small volume of extradural material dorsal to the affected intervertebral disc space, with minimal to no spinal cord compression. This can be associated with signal changes in the epidural space.

Other less commonly observed MRI changes can include a cleft in the dorsal part of the annulus fibrosus. Mild local enhancement of the meninges or epidural material after IV administration of gadolinium-based contrast has been reported (154), though this is usually not present (147, 150). Additionally, although more commonly observed in dogs with IVDE, signal alterations in the paravertebral muscles can also occasionally be seen in dogs with ANNPE (112). Paravertebral signal changes are occasionally seen with ANNPE and appear similar to what is described for IVDE (112).

The clinical presentation of animals with ANNPE is almost identical to that of animals with ischemic myelopathy, which is most commonly caused by fibrocartilaginous embolic myelopathy. Differentiating between both conditions is important because there are indications that the long-term outcome, and especially the prevalence of fecal incontinence, might be different between dogs with ANNPE and ischemic myelopathy (155, 156). Similar to ANNPE, specific MRI characteristics have been reported for ischemic myelopathy, which include a focal, relatively well-demarcated, possibly lateralized, longitudinal T2W hyperintense intramedullary lesion primary affecting the gray matter. The length of the lesion is usually longer than one vertebral body length (**Figure 12**) (157). Although it is possible to presumptively differentiate these two

conditions using MRI, there is considerable overlap between MRI appearance of dogs with ANNPE and ischemic myelopathy. Studies evaluating the interobserver agreement to differentiate ANNPE and ischemic myelopathy based on MRI characteristics have produced variable results reporting moderate ($\kappa = 0.56$) to perfect ($\kappa = 1$) interobserver agreement (158, 159). A lesion overlying the intervertebral disc, lesion lateralization, reduced nucleus pulposus volume, presence of extradural signal changes, meningeal enhancement and a non-longitudinal directional pattern of hyperintensity on T2W images have been associated with a diagnosis of ANNPE instead of ischemic myelopathy. A lesion overlying the vertebral body and greater length of an intramedullary hyperintensity were associated with a diagnosis of ischemic myelopathy instead of ANNPE (155, 159). A limitation to the literature on the subject is the lack of histopathologic confirmation of imaging findings.

Abnormalities observed on MRI in dogs with ANNPE have also been associated with the functional outcome (147, 160). Larger lesions observed on transverse sections have been associated with an unsuccessful outcome (147, 160). More information regarding MRI and outcome is presented in the manuscript "Prognostic factors in acute spinal cord injury" in this issue.

HYDRATED NUCLEUS PULPOSUS EXTRUSION

Hydrated nucleus pulposus extrusion (HNPE) is characterized by sudden extrusion of minimally to non-degenerative nucleus pulposus. Well-hydrated, gelatinous, extradural material can be identified in the vertebral canal, which is associated with varying degrees of spinal cord compression (148). Although HNPE can occur in the thoracolumbar region, it has a predilection for the cervical vertebral column (145). The most common clinical presentation consists therefore of acute onset non-ambulatory tetraparesis. Cervical hyperesthesia is not commonly observed. In contrast to dogs with ANNPE, onset of clinical signs is typically not associated with vigorous exercise or external trauma (161).

Magnetic resonance imaging is the diagnostic modality of choice and specific, almost pathognomonic, MRI abnormalities have been described (**Figure 13**) (148). The typical MRI appearance of HNPE is characterized by:

- A narrowed intervertebral disc space with a reduced volume of hyperintense nucleus pulposus (148)
- Ventral, midline, extradural compressive material, which is homogenously hyperintense on T2W sequences and isointense in all sequences to normal, non-degenerative nucleus pulposus lying immediately dorsal to the affected intervertebral disc
- The extruded material can have a characteristic bilobed or "seagull appearance." This typical shape is possibly explained by the location of the compressive material just ventral to the intact dorsal longitudinal ligament (162).
- A T2W hyperintense intraparenchymal lesion can be present in the overlying spinal cord and extruded nucleus pulposus can demonstrate a degree of contrast enhancement (148, 162).

Similar to ANNPE, MRI findings have been linked to the likelihood of neurological recovery in dogs with HNPE. Although T2W hyperintense intraparenchymal lesions are present in less than half of dogs with HNPE (163, 164), the length of such lesions was negatively associated with the likelihood for functional recovery within a time period of 9 days (164).

Although MRI is preferred, one study evaluated the usefulness of CT to diagnose cervical HNPE (165). Non-contrast CT did not reveal any specific abnormalities, however CT-angiography did allow visualization of cervical HNPE as a well-demarcated hypodense extradural compressive lesion with rim enhancement immediately dorsal to the intervertebral disc space. Contrast enhanced CT had a sensitivity of 91% and specificity of 100% to differentiate between cervical HNPE and IVDE (165).

INTRADURAL/INTRAMEDULLARY INTERVERTEBRAL DISC EXTRUSION

Intradural/intramedullary intervertebral disk extrusion (IIVDE) is the least common type of herniation of nucleus pulposus. This type of disc extrusion is characterized by intradural or intramedullary extrusion of calcified and dehydrated or minimally degenerate and hydrated intervertebral disc material (6, 146). The variation in hydration status of extruded material in dogs with IIVDE can result in variable clinical signs and imaging findings. Affected dogs can have a similar clinical presentation as dogs with ANNPE and ischemic myelopathy (146) or can present with a similar presentation as dogs with IVDE (6). Similar to ANNPE, MRI in animals with IIVDE caused by minimally degenerate nucleus pulposus can demonstrate a narrowed intervertebral disc space, decreased volume of homogenously hyperintense nucleus pulposus, and a T2W hyperintensity dorsal to the affected intervertebral disc space. Specific MRI findings in animals with IIVDE include a linear tract (predominantly hyperintense on T2W images, iso to hypointense on T1W images and hypointense on T2*-weighted GRE images) extending from the intervertebral disc into the spinal cord parenchyma (**Figure 14**) (146). Mild enhancement of the tissue adjacent to the tract can be observed on T1W images after IV administration of gadolinium-based contrast medium. Similar to IVDE, MRI in dogs with IIVDE caused by dehydrated and calcified nucleus pulposus can demonstrate a narrowed and homogenously hypointense intervertebral disc space with a hypointense mass dorsal to the affected intervertebral disc space, resulting in obvious spinal cord compression. Hyperintense lesions on T2W images, possibly indicating leakage of CSF, can be observed surrounding the herniated material. CT-myelography has been demonstrated to be particularly useful for diagnosing IIVDE and has been suggested to be more sensitive than low-field MRI for this purpose (6). CT-myelography can reveal extruded intervertebral disc material within the subarachnoid space with focal accumulation of contrast media within the subarachnoid space. The filling defect associated with the intradural location of the lesion can appear relative small in size predicted by the accumulation of contrast medium (6). It has been suggested that

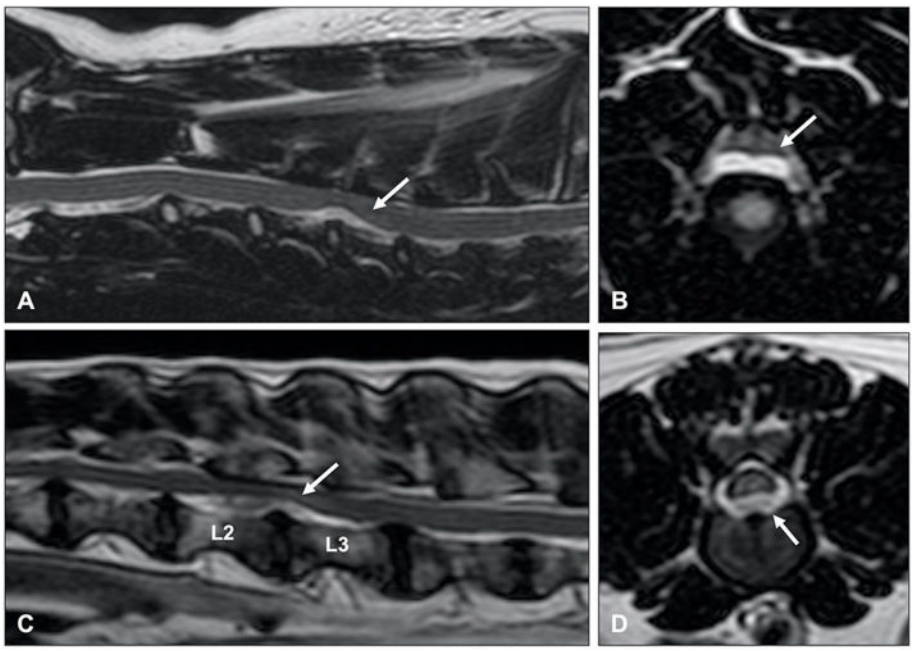

FIGURE 13 | T2-weighted **(A)** sagittal and **(B)** transverse MR images of a 6-year old Border collie with a C4-C5 HNPE. The intervertebral disc space is narrowed and contains a reduced volume of hyperintense nucleus pulposus. There is a ventral extradural homogenously hyperintense compressive lesion. **(B)** The compressive material is midline and has a bilobed, "seagull" appearance. T2-weighted **(C)** sagittal and **(D)** transverse images of 7-year old Beagle with acute onset paraplegia. Although HNPE occurs most often in the cervical region, this dog was diagnosed with an L2-L3 HNPE.

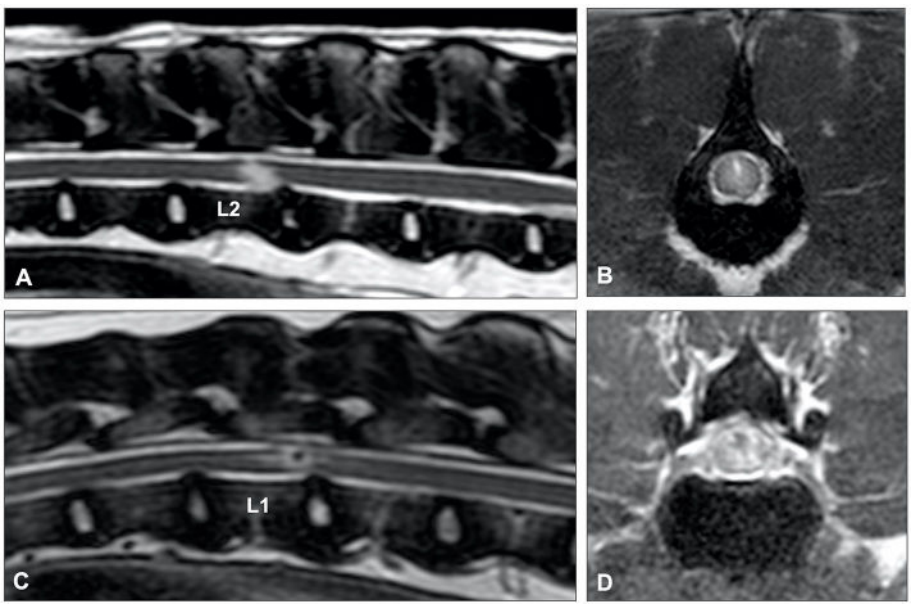

FIGURE 14 | (A) T2-weighted sagittal and **(B)** BALT GRAD (T2-weighted thin-slice gradient echo) transverse MR images of an 8-year old Greyhound with an L2-L3 IIVDE. **(A)** The intervertebral disc has a decreased volume of homogenously hyperintense nucleus pulposus. A ventrocaudal to dorsocranial intramedullary linear hyperintensity, starting from the L2-L3 intervertebral disc can be seen. **(B)** Hyperintense linear tract can be seen through the spinal cord. **(C)** T2-weighted sagittal and **(D)** BALT GRAD transverse MR images of a 6-year old Beagle with an L1-L2 IIVDE. The intramedullary lesion has a hypointense center, suggestive for intraparenchymal hemorrhage.

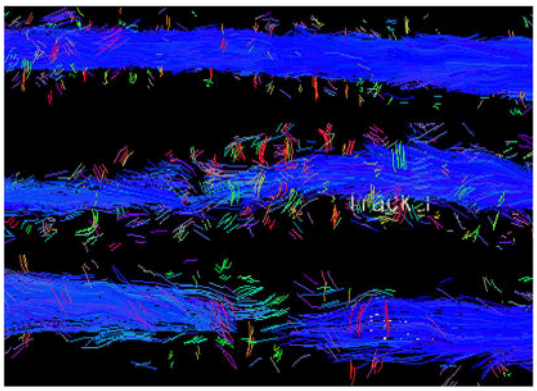

FIGURE 15 | Tractography of the spinal cord from a normal dog (top) compared to dogs with SCI showing moderate fiber disruption (middle) and complete trans-lesional discontinuity (bottom). Blue depicts cranial to caudal oriented fibers.

visualization of extradural leakage, suggestive of a dural tear, can be improved by applying traction to the head (166).

NOVEL ADVANCED IMAGING TECHNIQUES IN IVDD

Diffusion Tensor Imaging and Tractography

While conventional advanced imaging (MRI, CT, less commonly, myelography) are indispensable in the diagnosis of canine IVDH, there is a growing role for specialized applications of MRI including diffusion tensor imaging (DTI) in evaluating this population.

Diffusion tensor imaging is a variation on diffusion weighted imaging that relies on the strength and direction of cellular diffusion of water molecules to create images (167–169). The movement of water molecules varies by tissue type and is altered by pathology allowing DTI to provide quantitative microstructural information. Quantitative analysis typically includes calculation of the fractional anisotropy (FA) and apparent diffusion coefficient (ADC) or mean diffusivity (MD). Fractional anisotropy approximates the degree of directional dependence within a region from isotropic (0) to completely anisotropic along a single axis, providing information on white matter integrity. Apparent diffusion coefficient or MD relate to the magnitude of diffusion within a given region measured as the rate of water motion and reflect global tissue architecture. Axial diffusivity (AD) and radial diffusivity (RD) are performed less commonly and refer to diffusivity in the direction of or perpendicular to fiber tracts, respectively. Axial diffusivity and RD have been inconsistently reported to be associated with axonal injury and demyelination, respectively (169–172). The quantitative information on diffusivity within each voxel can then be combined to create a visual representation of white matter tracts called tractography (**Figure 15**) (173, 174).

Spinal cord DTI is useful to quantify disruption of the normally highly anisotropic white matter tracts and can detect microstructural changes such as axonal damage, demyelination, Wallerian degeneration and loss of tissue architecture (169). In experimental rodent models and human SCI patients, DTI has been shown to be more sensitive to pathologic changes than conventional MRI sequences (174–179). Additionally, quantitative indices and tractography have also been variably associated with injury severity, spinal cord integrity and long-term functional outcome in people and rodent models of SCI (176–185).

Diffusion Tensor Imaging of the Spinal Cord in Dogs

The feasibility of spinal cord DTI has been established in neurologically normal dogs (186–189). Healthy Beagles and Dachshunds predominated (186, 188, 189), but Hobert et al. included dogs of various ages, breeds and body sizes (187). No association was identified between FA or ADC values and body size or the cranial to caudal location along the spinal cord; age was not specifically investigated (187). These studies provide useful protocol and scan time information and broad reference values for quantitative variables in the uninjured cervical and thoracolumbar spinal cord.

Diffusion tensor imaging has been reported in dogs with experimental injury and various naturally-occurring myelopathies (118, 188, 190–196). Among experimental models, DTI indices have been correlated with specific histologic changes after injury, supporting its potential utility as a non-invasive measure of spinal cord microstructure (193, 195, 196). In dogs with naturally-occurring SCI, the majority of which were due to IVDE, quantitative analysis and tractography were able to distinguish between normal controls and dogs with SCI and between acute and chronic injury (118, 188, 194). In general, FA is reported to be decreased within and adjacent to an area of injury compared to controls and is further decreased in chronic relative to acute injury (118, 188, 191, 192). Values of ADC/MD within and adjacent to the lesion are more variable, likely influenced by the variable imaging timing across studies (118, 188, 191, 192, 194). Additionally, ADC/MD values can be decreased in acute SCI but become significantly elevated (relative to acute injury or healthy control dogs) in the chronic setting (118, 191, 192). Consistent with studies in people and experimental models, DTI in dogs has also been able to detect abnormal areas that appear macroscopically normal on T2W sequences, improving delineation of the regional extent of the SCI beyond what is visible with standard MRI (192).

Diffusion tensor imaging has been explored in dogs as a potential non-invasive biomarker and prognostic indicator (118, 192, 197). In dogs with paraplegia secondary to IVDE who underwent decompressive surgery, FA was not shown to be a better predictor of functional outcome compared to initial assessment of pain perception status (118). However, for dogs imaged in the chronic timeframe after prior acute injury, higher FA was associated with greater pelvic limb motor function, suggesting an association between diffusivity, structure and function (192). Additionally, among paraplegic dogs with or without pain perception secondary to IVDE, MD might be useful

to detect acute injury severity and to predict outcome (197). Tractography has also been shown to detect loss of spinal cord integrity including two dogs with IVDE (**Figure 15**) (192). Based on the small number of current studies, the utility of DTI and tractography to quantify injury severity and predict prognosis remains unclear. Further evaluation focusing on prospective studies in a larger number of dogs with IVDH is warranted.

In spite of the potential advantages, DTI faces several technical and logistical considerations that generally impede widespread use in dogs with SCI. DTI also requires specialized software, technical expertise and extensive post-processing further limiting its utility in real-time, clinical decision making for dogs with acute IVDH. Standardizing and optimizing acquisition and processing protocols will be important in order for DTI to be incorporated into routine clinical use in SCI patients. Despite limitations, DTI is worthy of continued development to complement standard MRI studies in cases of acute and chronic IVDH in dogs.

Additional MRI-Based Spinal Cord Imaging Techniques

Other novel advanced imaging techniques have been evaluated in the spinal cord of people including magnetic resonance spectroscopy, magnetization transfer imaging, myelin water fraction imaging and functional MRI (178, 198–201). These imaging modalities have been reported for the canine brain, in dogs with cervical spondylomyelopathy and in an experimental canine disc degeneration model but have not yet been investigated in dogs with IVDH (104, 202, 203). Future studies in canine IVDH should be considered to assess their clinical and translational utility in this population.

CONCLUSION

This review outlined the available imaging modalities used in the diagnosis of all forms of IVDD including IVDE, IVDP, ANNPE, HNPE, and IIVDE. Radiographs remain a screening tool but have limitations, especially with regard to differentiating between IVDD subtypes and offering a definitive diagnosis. Myelography, CT, or MRI are all viable ways to diagnose IVDD, with CT and MRI largely having supplanted myelography in routine clinical practice. While there are pros and cons for both CT and MRI, patient selection is the most important factor when choosing the appropriate imaging modality to maximize the likelihood of achieving a definitive diagnosis. Non-contrast CT is a quick and economical choice that is highly likely to be successful in an acutely non-ambulatory, chondrodystrophic dog where there is high suspicion for IVDE, with CT-myelography required in selected cases. Magnetic resonance imaging is considered the gold standard imaging modality for acute and chronic spinal cases due to its ability to allow spinal cord visualization and the diagnosis of IVDE, IVDP, ANNPE, HNPE, or IIVDE. There are a growing number of studies of DTI in dogs with IVDD that offer a means to assess microstructural lesion characteristics and spinal cord integrity. It is likely that additional, novel spinal cord imaging techniques will be developed for application in dogs with IVDD. Overall, diagnostic imaging is indispensable in the diagnosis of IVDD and a keen knowledge of the advantages and limitations of the various imaging modalities is crucial to maximize the diagnostic information obtained.

AUTHOR CONTRIBUTIONS

RC, SD, ML, and HV contributed to manuscript concept, preparation, and editing. The additional members of the CANSORT-SCI contributed to manuscript concept, editing, and review. All authors contributed to the article and approved the submitted version.

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REFERENCES

1. Bergknut N, Egenvall A, Hagman R, Gustas P, Hazewinkel HAW, Meij BP, et al. Incidence of intervertebral disk degeneration-related diseases and associated mortality rates in dogs. *J Am Vet Med Assoc.* (2012) 240:1300–9. doi: 10.2460/javma.240.11.1300
2. Priester WA. Canine intervertebral disc disease – occurrence by age, breed, and sex among 8,117 cases. *Theriogenology.* (1976) 6:293–303. doi: 10.1016/0093-691X(76)90021-2
3. Olsson SE. On disc protrusion in dog (enchondrosis intervertebralis); a study with special reference to roentgen diagnosis and to the value of disc fenestration. *Acta Orthop Scand Suppl.* (1951) 8:1–95 doi: 10.3109/ort.1951.22.suppl-8.01
4. Hansen H. A pathologic-anatomical study on disc degeneration in the dog. *Acta Orthop Scand Suppl.* (1952) 11:1–117. doi: 10.3109/ort.1952.23.suppl-11.01
5. Konar M, Lang J, Fluhmann G, Forterre F. Ventral intraspinal cysts associated with the intervertebral disc: magnetic resonance observations in seven dogs. *Vet Surg.* (2008) 37:94–101. doi: 10.1111/j.1532-950X.2007.00353.x
6. Tamura S, Doi S, Tamura Y, Takahashi K, Enomoto H, Ozawa T, et al. Thoracolumbar intradural disc herniation in eight dogs: clinical, low-field magnetic resonance imaging, and computed tomographic myelographic findings. *Vet Radiol Ultrasound.* (2015) 56:160–7. doi: 10.1111/vru.12213
7. da Costa RC. Cervical spondylomyelopathy (Wobbler syndrome) in dogs. *Vet Clin North Am Small Anim Pract.* (2010) 40:881–913. doi: 10.1016/j.cvsm.2010.06.003
8. Meij BP, Bergknut N. Degenerative lumbosacral stenosis in dogs. *Vet Clin North Am Small Anim Pract.* (2010) 40:983–1009. doi: 10.1016/j.cvsm.2010.05.006
9. Hoerlein BF. Intervertebral disc protrusions in the dog. III. Radiological diagnosis. *Am J Vet Res.* (1953) 14:275–86.
10. Butterworth SJ Denny HR. Follow-up study of 100 cases with thoracolumbar disc protrusions treated by lateral fenestration. *J Small Anim Pract.* (1991) 32:443–7. doi: 10.1111/j.1748-5827.1991.tb00983.x
11. Lee R. Interpretation of radiographs: inter-vertebral disc lesions in the dog. *J Small Anim Pract.* (1973) 14:111–2 doi: 10.1111/j.1748-5827.1973.tb06901.x
12. Bullock LP, Zook BC. Myelography in dogs, using water-soluble contrast mediums. *J Am Vet Med Assoc.* (1967) 151:321–7.
13. Morgan JP, Suter PF, Holliday TA. Myelography with water-soluble contrast medium. Radiographic interpretation of disc herniation in dogs. *Acta Radiol Suppl.* (1972) 319:217–30. doi: 10.1080/05678067209174022
14. Jeffery ND. Treatment of acute and chronic thoracolumbar disc disease by 'mini hemilaminectomy'. *J Small Anim Pract.* (1988) 29:611–16. doi: 10.1111/j.1748-5827.1988.tb02181.x
15. Muir P, Johnson KA, Manley PA, Dueland RT. Comparison of hemilaminectomy and dorsal laminectomy for thoracolumbar intervertebral disc extrusion in dachshunds. *J Small Anim Pract.* (1995) 36:360–7. doi: 10.1111/j.1748-5827.1995.tb02950.x
16. Sande RD. Radiography, myelography, computed tomography, and magnetic resonance imaging of the spine. *Vet Clin North Am Small Anim Pract.* (1992) 22:811–31. doi: 10.1016/S0195-5616(92)50078-X
17. Scott HW. Hemilaminectomy for the treatment of thoracolumbar disc disease in the dog: a follow-up study of 40 cases. *J Small Anim Pract.* (1997) 38:488–94. doi: 10.1111/j.1748-5827.1997.tb03303.x
18. Ferreira AJA, Correia JHD, Jaggy A. Thoracolumbar disc disease in 71 paraplegic dogs: influence of rate of onset and duration of clinical signs on treatment results. *J Small Anim Pract.* (2002) 43:158–63. doi: 10.1111/j.1748-5827.2002.tb00049.x
19. Moore SA, Early PJ, Hettlich BF. Practice patterns in the management of acute intervertebral disc herniation in dogs. *J Small Anim Pract.* (2016) 57:409–15. doi: 10.1111/jsap.12496
20. Kranenburg HJ, Grinwis GC, Bergknut N, Gahrmann N, Voorhout G, Hazewinkel HAW, et al. Intervertebral disc disease in dogs - part 2: comparison of clinical, magnetic resonance imaging, and histological findings in 74 surgically treated dogs. *Vet J.* (2013) 195:164–71. doi: 10.1016/j.tvjl.2012.06.001
21. Brisson BA. Intervertebral disc disease in dogs. *Vet Clin North Am Small Anim Pract.* (2010) 40:829–58. doi: 10.1016/j.cvsm.2010.06.001
22. Burk RL. Problems in the radiographic interpretation of intervertebral disc disease in the dog. *Probl Vet Med.* (1989) 1:381–401.
23. Lamb CR, Nichols A, Targett M, Mannion P. Accuracy of survey radiographic diagnosis of intervertebral disc protrusion in dogs. *Vet Radiol Ultrasound.* (2002) 3:222–8. doi: 10.1111/j.1740-8261.2002.tb00994.x
24. Stigen O Kolbjornsen O. Calcification of intervertebral discs in the dachshund: a radiographic and histopathological study of 20 dogs. *Acta Vet Scand.* (2007) 49:39–45. doi: 10.1186/1751-0147-49-39
25. Rosenblatt AJ, Lappalainen AK, James NA, Webster NSL, Caraguel CGB. Scorer and modality agreement for the detection of intervertebral disc calcification in Dachshunds. *Acta Vet Scand.* (2018) 60:62. doi: 10.1186/s13028-018-0416-2
26. Jensen VF, Beck S, Christensen KA, Arnbjerg J. Quantification of the association between intervertebral disk calcification and disk herniation in dachshunds. *J Am Vet Med Assoc.* (2008) 233:1090–5. doi: 10.2460/javma.233.7.1090
27. Mayhew PD, McLear RC, Ziemer LS, Culp WTN, Russell KN, Shofer FS, et al. Risk factors for recurrence of clinical signs associated with thoracolumbar intervertebral disk herniation in dogs: 229 cases (1994-2000). *J Am Vet Med Assoc.* (2004) 225:1231–6. doi: 10.2460/javma.2004.225.1231
28. Brisson BA, Holmberg DL, Parent JP, Sears WC, Wick SE. Comparison of the effect of single-site and multiple-site disk fenestration on the rate of recurrence of thoracolumbar intervertebral disk herniation in dogs. *J Am Vet Med Assoc.* (2011) 238:1593–600. doi: 10.2460/javma.238.12.1593
29. Rohdin C, Jeserevic J, Viitmaa R, Cizinauskas S. Prevalence of radiographic detectable intervertebral disc calcifications in dachshunds surgically treated for disc extrusion. *Acta Vet Scand.* (2010) 52:24. doi: 10.1186/1751-0147-52-24
30. Bos AS, Brisson BA, Nykamp SG, Poma R, Foster RA. Accuracy, intermethod agreement, and inter-reviewer agreement for use of magnetic resonance imaging and myelography in small-breed dogs with naturally occurring first-time intervertebral disk extrusion. *J Am Vet Med Assoc.* (2012) 240:969–77. doi: 10.2460/javma.240.8.969
31. Jensen VF. Asymptomatic radiographic disappearance of calcified intervertebral disc material in the dachshund. *Vet Radiol Ultrasound.* (2001) 42:141–8. doi: 10.1111/j.1740-8261.2001.tb00917.x
32. Yovich JC, Read R, Eger C. Modified lateral spinal decompression in 61 dogs with thoracolumbar disc protrusion. *J Small Anim Pract.* (1994) 35:351–6. doi: 10.1111/j.1748-5827.1994.tb01715.x
33. Brown NO, Helphrey ML Prata RG. Thoracolumbar disk disease in the dog: a retrospective analysis of 187 cases. *J Am Anim Hosp Assoc.* (1977) 13:665–72. doi: 10.1111/j.1752-1688.1977.tb02007.x
34. Schulz KS, Walker M, Moon M, Waldron D, Slater M, McDonald DE. Correlation of clinical, radiographic, and surgical localization of intervertebral disc extrusion in small-breed dogs: a prospective study of 50 cases. *Vet Surg.* (1998) 27:105–11. doi: 10.1111/j.1532-950X.1998.tb00105.x
35. McKee WM. A comparison of hemilaminectomy (with concomitant disk fenestration) and dorsal laminectomy for the treatment of thoracolumbar disk protrusion in dogs. *Vet Rec.* (1992) 130:296–300. doi: 10.1136/vr.130.14.296
36. Kirberger RM, Roos CJ, Lubbe AM. The radiological diagnosis of thoracolumbar disc disease in the dachshund. *Vet Radiol Ultrasound.* (1992) 33:255–61. doi: 10.1111/j.1740-8261.1992.tb00140.x
37. Hecht S, Thomas WB, Marioni-Henry K, Echandi RL, Matthews AR, Adams WH. Myelography vs. computed tomography in the evaluation of acute thoracolumbar intervertebral disk extrusion in chondrodystrophic dogs. *Vet Radiol Ultrasound.* (2009) 50:353–9. doi: 10.1111/j.1740-8261.2009.01549.x
38. Morgan PW, Parent J, Holmberg DL. Cervical pain secondary to intervertebral disc disease in dogs; radiographic findings and surgical implications. *Prog Vet Neurol.* (1993) 4:76–80.
39. Somerville ME, Anderson SM, Gill PJ, Kantrowitz BJ, Stowater JL. Accuracy of localization of cervical intervertebral disk extrusion or protrusion using survey radiography in dogs. *J Am Anim Hosp Assoc.* (2001) 37:563–72. doi: 10.5326/15473317-37-6-563

40. Roberts RE, Selcer BA. Myelography and epidurography. *Vet Clin North Am Small Anim Pract.* (1993) 23:307–29. doi: 10.1016/S0195-5616(93)50030-X
41. Widmer WR, Blevins WE. Veterinary myelography: a review of contrast media, adverse effects, and technique. *J Am Anim Hosp Assoc.* (1991) 27:163–77.
42. Shimizu J, Yamada K, Kishimoto M, Iwasaki T, Miyake Y. The difference of contrast effects of myelography in normal dogs: comparison of iohexol (180 mgI/ml), iohexol (240 mgI/ml) and iotrolan (240 mgI/ml). *J Vet Med Sci.* (2008) 70:659–63. doi: 10.1292/jvms.70.659
43. Olby NJ Houlton JEF. Correlation of plain radiographic and lumbar myelographic findings with surgical findings in thoracolumbar disc disease. *J Small Anim Pract.* (1994) 35:345–50. doi: 10.1111/j.1748-5827.1994.tb01713.x
44. Dewey CW, da Costa RC, Ducote JM. Neurodiagnostics. In: Dewey CW, da Costa RC, editors. *Practical Guide to Canine and Feline Neurology,* 3rd ed. Ames, IA: Wiley (2016). p. 61–9.
45. Gibbons SE, Macias C, De Stefani A, Pinchbeck GL, McKee WM. The value of oblique versus ventrodorsal myelographic views for lesion lateralisation in canine thoracolumbar disc disease. *J Small Anim Pract.* (2006) 47:658–62. doi: 10.1111/j.1748-5827.2006.00067.x
46. Macias C, McKee WM, May C, Innes JF. Thoracolumbar disc disease in large dogs: a study of 99 cases. *J Small Anim Pract.* (2002) 43:439–46. doi: 10.1111/j.1748-5827.2002.tb00010.x
47. Bos AS, Brisson BA, Holmberg DL, Nykamp SG. Use of the ventrodorsal myelographic view to predict lateralization of extruded disk material in small-breed dogs with thoracolumbar intervertebral disk extrusion: 104 cases (2004-2005). *J Am Vet Med Assoc.* (2007) 230:1860–65. doi: 10.2460/javma.230.12.1860
48. Duval J, Dewey C, Roberts R, Aron D. Spinal cord swelling as a myelographic indicator of prognosis: a retrospective study in dogs with intervertebral disc disease and loss of deep pain perception. *Vet Surg.* (1996) 25:6–12. doi: 10.1111/j.1532-950X.1996.tb01371.x
49. Scott HW, McKee WM. Laminectomy for 34 dogs with thoracolumbar intervertebral disc disease and loss of deep pain perception. *J Small Anim Pract.* (1999) 40:417–22. doi: 10.1111/j.1748-5827.1999.tb03114.x
50. Lu D, Lamb CR, Targett MP. Results of myelography in seven dogs with myelomalacia. *Vet Radiol Ultrasound.* (2002) 43:326–30. doi: 10.1111/j.1740-8261.2002.tb01012.x
51. Bersan E, McConnell F, Trevail R, Behr S, De Decker S, Volk HA, et al. Cervical intervertebral foraminal disc extrusion in dogs: clinical presentation, MRI characteristics and outcome after medical management. *Vet Rec.* (2015) 176:597. doi: 10.1136/vr.102851
52. Dennison SE, Drees R, Rylander H, Yandell BS, Milovancev M, Pettigrew R, et al. Evaluation of different computed tomography techniques and myelography for the diagnosis of acute canine myelopathy. *Vet Radiol Ultrasound.* (2010) 51:254–8. doi: 10.1111/j.1740-8261.2010.01667.x
53. Tanaka H, Nakayama M, Takase K. Usefulness of myelography with multiple views in diagnosis of circumferential location of disc material in dogs with thoracolumbar intervertebral disc herniation. *J Vet Med Sci.* (2004) 66:827–33. doi: 10.1292/jvms.66.827
54. Newcomb B, Arble J, Rochat M, Pechman R, Payton M. Comparison of computed tomography and myelography to a reference standard of computed tomography myelography for evaluation of dogs with intervertebral disc disease. *Vet Surg.* (2012) 41:207–14. doi: 10.1111/j.1532-950X.2011.00911.x
55. Barone G, Ziemer LS, Shofer FS, Steinberg SA. Risk factors associated with development of seizures after use of iohexol for myelography in dogs: 182 cases (1998). *J Am Vet Med Assoc.* (2002) 220:1499–502. doi: 10.2460/javma.2002.220.1499
56. da Costa RC, Parent JM, Dobson H. Incidence of and risk factors for seizures after myelography performed with iohexol in dogs: 503 cases (2002-2004). *J Am Vet Med Assoc.* (2011) 238:1296–300. doi: 10.2460/javma.238.10.1296
57. Parry AT, Harris A, Upjohn MM, Chandler K, Lamb CR. Does choice of imaging modality affect outcome in dogs with thoracolumbar spinal conditions? *J Small Anim Pract.* (2010) 51:312–7 doi: 10.1111/j.1748-5827.2010.00936.x
58. Israel SK, Levine JM, Kerwin SC, Levine GJ, Fosgate GT. The relative sensitivity of computed tomography and myelography for identification of thoracolumbar intervertebral disk herniations in dogs. *Vet Radiol Ultrasound.* (2009) 50:247–52. doi: 10.1111/j.1740-8261.2009.01528.x
59. Robertson I, Thrall DE. Imaging dogs with suspected disc herniation: pros and cons of myelography, computed tomography, and magnetic resonance. *Vet Radiol Ultrasound.* (2011) 52:S81–84. doi: 10.1111/j.1740-8261.2010.01788.x
60. Tidwell AS Jones JC. Advanced imaging concepts: a pictorial glossary of CT and MRI technology. *Clin Tech Small Anim Pract.* (1999) 14:65–111. doi: 10.1016/S1096-2867(99)80008-5
61. King JB, Jones JC, Rossmeisl JH Jr, Harper TA, Lanz OI, Were SR. Effect of multi-planar CT image reformatting on surgeon diagnostic performance for localizing thoracolumbar disc extrusions in dogs. *J Vet Sci.* (2009) 10:225–32. doi: 10.4142/jvs.2009.10.3.225
62. Ricciardi M, Campanella A, Grieco G, Zammit R. Usefulness of spinal unenhanced computed tomography and CT-myelography in the age of multidetector CT technology and magnetic resonance imaging - preliminary considerations. *Open Vet J.* (2018) 8:265–81. doi: 10.4314/ovj.v8i3.6
63. Lancourt JE, Glenn WV Jr, Wiltse LL. Multiplanar computerized tomography in the normal spine and in the diagnosis of spinal stenosis. A gross anatomic-computerized tomographic correlation. *Spine.* (1979) 4:379–90. doi: 10.1097/00007632-197907000-00006
64. Gellad F, Rao KC, Joseph PM, Vigorito RD. Morphology and dimensions of the thoracic cord by computer-assisted metrizamide myelography. *Am J Neuroradiol.* (1983) 4:614–17. doi: 10.1097/00004728-198302000-00176
65. Olby NJ, Muñana KR, Sharp NJ, Thrall DE. The computed tomographic appearance of acute thoracolumbar intervertebral disc herniations in dogs. *Vet Radiol Ultrasound.* (2000) 41:396–402. doi: 10.1111/j.1740-8261.2000.tb01860.x
66. Shimizu J, Yamada K, Mochida K, Kako T, Muroya N, Teratani Y, et al. Comparison of the diagnosis of intervertebral disc herniation in dogs by CT before and after contrast enhancement of the subarachnoid space. *Vet Rec.* (2009) 165:200–2. doi: 10.1136/vr.165.7.200
67. Schroeder R, Pelsue DH, Park RD, Gasso D, Bruecker KA. Contrast-enhanced CT for localizing compressive thoracolumbar intervertebral disc extrusion. *J Am Anim Hosp Assoc.* (2011) 47:203–9. doi: 10.5326/JAAHA-MS-5550
68. Emery L, Hecht S, Sun X. Investigation of parameters predicting the need for diagnostic imaging beyond computed tomography in the evaluation of dogs with thoracolumbar myelopathy: retrospective evaluation of 555 dogs. *Vet Radiol Ultrasound.* (2018) 59:147–54. doi: 10.1111/vru.12576
69. Cooper JJ, Young BD, Griffin JF IV, Fosgate GT, Levine JM. Comparison between noncontrast computed tomography and magnetic resonance imaging for detection and characterization of thoracolumbar myelopathy caused by intervertebral disk herniation in dogs. *Vet Radiol Ultrasound.* (2014) 55:182–9. doi: 10.1111/vru.12114
70. Noyes JA, Thomovsky SA, Chen AV, Owen TJ, Fransson BA, Carbonneau KJ, et al. Magnetic resonance imaging versus computed tomography to plan hemilaminectomies in chondrodystrophic dogs with intervertebral disc extrusion. *Vet Surg.* (2017) 46:1025–31. doi: 10.1111/vsu.12700
71. da Costa RC, Samii VF. Advanced imaging of the spine in small animals. *Vet Clin North Am Small Anim Pract.* (2010) 40:765–90. doi: 10.1016/j.cvsm.2010.05.002
72. Naude SH, Lambrechts NE, Wagner WM, Thompson PN. Association of preoperative magnetic resonance imaging findings with surgical features in dachshunds with thoracolumbar intervertebral disk extrusion. *J Am Vet Med Assoc.* (2008) 232:702–8. doi: 10.2460/javma.232.5.702
73. Konar M, Lang J. Pros and cons of low-field magnetic resonance imaging in veterinary practice. *Vet Radiol Ultrasound.* (2011) 52(Suppl. 1):S5–14. doi: 10.1111/j.1740-8261.2010.01780.x
74. Dennis R. Optimal magnetic resonance imaging of the spine. *Vet Radiol Ultrasound.* (2011) 52(Suppl. 1):S72–80. doi: 10.1111/j.1740-8261.2010.01787.x
75. Dennis R. Optimized technique: Spine. In: Mai W, editor. *Diagnostic MRI in Dogs and Cats*. Boca Raton, FL: CRC Press (2018) 106–129.
76. Mai W. Normal MRI spinal anatomy, degenerative disc disease and disc herniation. In: Mai W, editor. *Diagnostic MRI in Dogs and Cats*. Boca Raton, FL: CRC Press. (2018) 413–446.

77. Inglez de Souza MCCM, Ryan R, Ter Haar G, Packer RMA, Volk HA, De Decker S. Evaluation of the influence of kyphosis and scoliosis on intervertebral disc extrusion in French bulldogs. *BMC Vet Res.* (2018) 14:5. doi: 10.1186/s12917-017-1316-9
78. Dennis R. Assessment of location of the celiac and cranial mesenteric arteries relative to the thoracolumbar spine using magnetic resonance imaging. *Vet Radiol Ultrasound.* (2005) 46:388–90. doi: 10.1111/j.1740-8261.2005.00070.x
79. Yeamans CL, Haley A, Gutierrez-Quintana R, Penderis J. Surgical anatomical landmarks of the thoracolumbar vertebral column on magnetic resonance imaging in dogs. *Anat Histol Embryol.* (2016) 45:140–4. doi: 10.1111/ahe.12184
80. Guillem Gallach R, Suran J, Cáceres AV, Reetz JA, Brown DC, Mai W. Reliability of T2-weighted sagittal magnetic resonance images for determining the location of compressive disk herniation in dogs. *Vet Radiol Ultrasound.* (2011) 52:479–86. doi: 10.1111/j.1740-8261.2011.01833.x
81. Housley D, Caine A, Cherubini G, Taeymans O. Evaluation of T2-weighted versus short-tau inversion recovery sagittal sequences in the identification and localization of canine intervertebral disc extrusion with low-field magnetic resonance imaging. *Vet Radiol Ultrasound.* (2017) 58:433–43. doi: 10.1111/vru.12500
82. Mankin JM, Hecht S, Thomas WB. Agreement between T2 and haste sequences in the evaluation of thoracolumbar intervertebral disc disease in dogs. *Vet Radiol Ultrasound.* (2012) 53:162–6. doi: 10.1111/j.1740-8261.2011.01894.x
83. Seiler G, Häni H, Scheidegger J, Busato A, Lang J. Staging of lumbar intervertebral disc degeneration in nonchondrodystrophic dogs using low-field magnetic resonance imaging. *Vet Radiol Ultrasound.* (2003) 44:179–84. doi: 10.1111/j.1740-8261.2003.tb01268.x
84. Pearce RH, Thompson JP, Bebault GM, Flak B. Magnetic resonance imaging reflects the chemical changes of aging degeneration in the human intervertebral disk. *J Rheumatol Suppl.* (1991) 27:42–43.
85. Benneker LM, Heini PF, Anderson SE, Alini M, Ito K. Correlation of radiographic and MRI parameters to morphological and biochemical assessment of intervertebral disc degeneration. *Eur Spine J.* (2005) 14:27–35. doi: 10.1007/s00586-004-0759-4
86. Fingeroth JM, Melrose J. Discogenic Pain (signs associated with disc degeneration but without herniation): does it occur? In: Fingeroth JM, Thomas WB, editors. *Advances in Intervertebral Disc Disease in Dogs and Cats*. Ames, IA: Wiley (2015). p. 127–31.
87. Pfirrmann CW, Metzdorf A, Zanetti M, Hodler J, Boos N. Magnetic resonance classification of lumbar intervertebral disc degeneration. *Spine.* (2001) 26:1873–8. doi: 10.1097/00007632-200109010-00011
88. Thompson JP, Pearce RH, Schechter MT, Adams ME, Tsang IK, Bishop PB. Preliminary evaluation of a scheme for grading the gross morphology of the human intervertebral disc. *Spine.* (1990) 15:411–15. doi: 10.1097/00007632-199005000-00012
89. Bergknut N, Auriemma E, Wijsman S, Voorhout G, Hagman R, Lagerstedt A, et al. Evaluation of intervertebral disk degeneration in chondrodystrophic and nonchondrodystrophic dogs by use of Pfirrmann grading of images obtained with low-field magnetic resonance imaging. *Am J Vet Res.* (2011) 72:893–8. doi: 10.2460/ajvr.72.7.893
90. Bergknut N, Grinwis G, Pickee E, Auriemma E, Lagerstedt AS, Hagman R, et al. Reliability of macroscopic grading of intervertebral disk degeneration in dogs by use of the Thompson system and comparison with low-field magnetic resonance imaging findings. *Am J Vet Res.* (2011) 72:899–904. doi: 10.2460/ajvr.72.7.899
91. Thompson K, Moore S, Tang S, Wiet M, Purmessur D. The chondrodystrophic dog: a clinically relevant intermediate-sized animal model for the study of intervertebral disc-associated spinal pain. *JOR Spine.* (2018) 1:e1011. doi: 10.1002/jsp2.1011
92. Kunze K, Stein VM, Tipold A. Evaluation of the canine intervertebral disc structure in turbo spin echo-T2 and fast field echo-T1 sequences in magnetic resonance imaging. *Front Vet Sci.* (2019) 6:68. doi: 10.3389/fvets.2019.00068
93. Kepler CK, Ponnappan RK, Tannoury CA, Risbud MV, Anderson DG. The molecular basis of intervertebral disc degeneration. *Spine J.* (2013) 13:318–30. doi: 10.1016/j.spinee.2012.12.003
94. Besalti O, Pekcan Z, Sirin YS, Erbas G. Magnetic resonance imaging findings in dogs with thoracolumbar intervertebral disk disease: 69 cases (1997-2005). *J Am Vet Med Assoc.* (2006) 228:902–8. doi: 10.2460/javma.228.6.902
95. Gendron K, Doherr MG, Gavin P, Lang J. Magnetic resonance imaging characterization of vertebral endplate changes in the dog. *Vet Radiol Ultrasound.* (2012) 53:50–56. doi: 10.1111/j.1740-8261.2011.01861.x
96. Deards E, Clements DN, Schwarz T. MRI signal changes and their association with intervertebral disc disease in canine vertebral endplates. *Ir Vet J.* (2019) 72:12. doi: 10.1186/s13620-019-0148-2
97. Farrell M, Fitzpatrick N. Feline intervertebral disc disease. In Fingeroth JM, Thomas WB, editors. *Advances in Intervertebral Disc Disease in Dogs and Cats*. Ames, IA: Wiley (2015). p. 36–40.
98. De Decker S, Warner AS, Volk HA. Prevalence and breed predisposition for thoracolumbar intervertebral disc disease in cats. *J Feline Med Surg.* (2017) 19:419–23. doi: 10.1177/1098612X16630358
99. Hamilton-Bennett SE, Behr S. Clinical presentation, magnetic resonance imaging features, and outcome in 6 cats with lumbar degenerative intervertebral disc extrusion treated with hemilaminectomy. *Vet Surg.* (2019) 48:556–62. doi: 10.1111/vsu.13182
100. Tidwell AS, Specht A, Blaeser L, Kent M. Magnetic resonance imaging features of extradural hematomas associated with intervertebral disc herniation in a dog. *Vet Radiol Ultrasound.* (2002) 43:319–24. doi: 10.1111/j.1740-8261.2002.tb01011.x
101. Mateo I, Lorenzo V, Foradada L, Munoz A. Clinical, pathologic, and magnetic resonance imaging characteristics of canine disc extrusion accompanied by epidural hemorrhage or inflammation. *Vet Radiol Ultrasound.* (2011) 52:17–24. doi: 10.1111/j.1740-8261.2010.01746.x
102. Martin-Vaquero P, da Costa RC. Magnetic resonance imaging features of great danes with and without clinical signs of cervical spondylomyelopathy. *J Am Vet Med Assoc.* (2014) 245:393–400. doi: 10.2460/javma.245.4.393
103. Provencher M, Habing A, Moore SA, Cook L, Phillips G, da Costa RC. Kinematic magnetic resonance imaging for evaluation of disc-associated cervical spondylomyelopathy in doberman pinschers. *J Vet Intern Med.* (2016) 30:1121–8. doi: 10.1111/jvim.13981
104. da Costa RC, Parent JM, Partlow G, Dobson H, Holmberg DL, Lamarre J. Morphologic and morphometric magnetic resonance imaging features of doberman pinschers with and without clinical signs of cervical spondylomyelopathy. *Am J Vet Res.* (2006) 67:1601–12. doi: 10.2460/ajvr.67.9.1601
105. Tirrito F, Cozzi F, Bonaldi M, Conti S, Lombardo R. Agreement of surgeon's perception of the effectiveness of spinal cord decompression with findings on postoperative magnetic resonance imaging for dogs surgically treated for intervertebral disk extrusion. *J Am Vet Med Assoc.* (2020) 256:210–19. doi: 10.2460/javma.256.2.210
106. Martin-Vaquero P, da Costa RC, Lima CG. Cervical spondylomyelopathy in great danes: a magnetic resonance imaging morphometric study. *Vet J.* (2014) 201:64–71. doi: 10.1016/j.tvjl.2014.04.011
107. Ryan TM, Platt SR, Llabres-Diaz FJ, McConnell JF, Adams VJ. Detection of spinal cord compression in dogs with cervical intervertebral disc disease by magnetic resonance imaging. *Vet Rec.* (2008) 163:11–5. doi: 10.1136/vr.163.1.11
108. Penning V, Platt SR, Dennis R, Cappello R, Adams V. Association of spinal cord compression seen on magnetic resonance imaging with clinical outcome in 67 dogs with thoracolumbar intervertebral disc extrusion. *J Small Anim Pract.* (2006) 47:644–50. doi: 10.1111/j.1748-5827.2006.00252.x
109. Levine JM, Fosgate GT, Chen AV, Rushing R, Nghiem PP, Platt SR, et al. Magnetic resonance imaging in dogs with neurologic impairment due to acute thoracic and lumbar intervertebral disk herniation. *J Vet Intern Med.* (2009) 23:1220–6. doi: 10.1111/j.1939-1676.2009.0393.x
110. Freeman AC, Platt SR, Kent M, Howerth E, Holmes SP. Magnetic resonance imaging enhancement of intervertebral disc disease in 30 dogs following chemical fat saturation. *J Small Anim Pract.* (2012) 53:120–25. doi: 10.1111/j.1748-5827.2011.01174.x
111. Suran JN, Durham A, Mai W, Seiler GS. Contrast enhancement of extradural compressive material on magnetic resonance imaging. *Vet Radiol Ultrasound.* (2011) 52:10–6. doi: 10.1111/j.1740-8261.2010.01734.x

112. Trampus P, Goepfert C, Welle M, Henke D, Forterre F, Schweizer-Gorgas D. Magnetic resonance imaging signal alterations in paraspinal muscles in dogs with acute thoracolumbar intervertebral disk extrusion. *Front Vet Sci.* (2018) 5:16. doi: 10.3389/fvets.2018.00016
113. Boekhoff TM, Flieshardt C, Ensinger EM, Fork M, Kramer S, Tipold A. Quantitative magnetic resonance imaging characteristics: evaluation of prognostic value in the dog as a translational model for spinal cord injury. *J Spinal Disord Tech.* (2012) 25:E81–87. doi: 10.1097/BSD.0b013e31823f2f55
114. Falconer JC, Narayana PA, Bhattacharjee MB, Liu SJ. Quantitative MRI of spinal cord injury in a rat model. *Magn Reson Med.* (1994) 32:484–91. doi: 10.1002/mrm.1910320410
115. Schouman-Claeys E, Frija G, Cuenod CA, Begon D, Paraire F, Martin V. MR imaging of acute spinal cord injury: results of an experimental study in dogs. *Am J Neuroradiol.* (1990) 11:959–65.
116. Flanders AE, Spettell CM, Friedman DP, Marino RJ, Herbison GJ. The relationship between the functional abilities of patients with cervical spinal injury and the severity of damage revealed by MR imaging. *Am J Neuroradiol.* (1999) 20:926–34.
117. Otamendi AJ, Boudreau B, Gilmour L, Griffin J, Levine J, Jeffery ND. Imaging findings at 3T and clinical outcome in deep-pain negative dogs with intervertebral disc herniation. *J Vet Intern Med Proc ACVIM Forum.* (2017) 31:1262.
118. Wang-Leandro A, Siedenburg JS, Hobert MK, Dziallas P, Rohn K, Stein VM, et al. Comparison of preoperative quantitative magnetic resonance imaging and clinical assessment of deep pain perception as prognostic tools for early recovery of motor function in paraplegic dogs with intervertebral disk herniations. *J Vet Intern Med.* (2017) 31:842–8. doi: 10.1111/jvim.14715
119. Ito D, Matsunaga S, Jeffery ND, Sasaki N, Nishimura R, Mochizuki M, et al. Prognostic value of magnetic resonance imaging in dogs with paraplegia caused by thoracolumbar intervertebral disc extrusion: 77 cases (2000–2003). *J Am Vet Med Assoc.* (2005) 227:1454. doi: 10.2460/javma.2005.227.1454
120. Huska JL, Gaitero L, Brisson BA, Nykamp S, Thomason J, Sears WC. Presence of residual material following mini-hemilaminectomy in dogs with thoracolumbar intervertebral disc extrusion. *Can Vet J.* (2014) 55:975–80.
121. Forterre F, Gorgas D, Dickomeit M, Jaggy A, Lang J, Spreng D. Incidence of spinal compressive lesions in chondrodystrophic dogs with abnormal recovery after hemilaminectomy for treatment of thoracolumbar disc disease: a prospective magnetic resonance imaging study. *Vet Surg.* (2010) 39:165–72. doi: 10.1111/j.1532-950X.2009.00633.x
122. Reynolds D, Brisson BA, Nykamp SG. Agreement between magnetic resonance imaging, myelography, and surgery for detecting recurrent, thoracolumbar intervertebral disc extrusion in dogs. *Vet Comp Orthop Traumatol.* (2013) 26:12–8. doi: 10.3415/VCOT-12-05-0067
123. Olby NJ, Muguet-Chanoit AC, Lim JH, Davidian M, Mariani CL, Freeman AC, et al. A placebo-controlled, prospective, randomized clinical trial of polyethylene glycol and methylprednisolone sodium succinate in dogs with intervertebral disk herniation. *J Vet Intern Med.* (2016) 30:206–14. doi: 10.1111/jvim.13657
124. Olby N, Levine J, Harris T, Munana K, Skeen T, Sharp N. Long-term functional outcome of dogs with severe injuries of the thoracolumbar spinal cord: 87 cases (1996-2001). *J Am Vet Med Assoc.* (2003) 222:762–9. doi: 10.2460/javma.2003.222.762
125. Aikawa T, Shibata M, Asano M, Hara Y, Tagawa M, Orima H. A comparison of thoracolumbar intervertebral disc extrusion in French bulldogs and dachshunds and association with congenital vertebral anomalies. *Vet Surg.* (2014) 43:301–7. doi: 10.1111/j.1532-950X.2014.12102.x
126. Okada M, Kitagawa M, Ito D, Itou T, Kanayama K, Sakai T. Magnetic resonance imaging features and clinical signs associated with presumptive and confirmed progressive myelomalacia in dogs: 12 cases (1997–2008). *J Am Vet Med Assoc.* (2010) 237:1160–5. doi: 10.2460/javma.237.10.1160
127. Platt SR, McConnell J, Bestbier M. Magnetic resonance imaging characteristics of ascending hemorrhagic myelomalacia in a dog. *Vet Radiol Ultrasound.* (2006) 47:78–82. doi: 10.1111/j.1740-8261.2005.00109.x
128. Castel A, Olby NJ, Mariani CL, Muñana KR, Early PJ. Clinical characteristics of dogs with progressive myelomalacia following acute intervertebral disc extrusion. *J Vet Intern Med.* (2017) 31:1782–9. doi: 10.1111/jvim.14829
129. Balducci F, Canal S, Contiero B, Bernardini M. Prevalence and risk factors for presumptive ascending/descending myelomalacia in dogs after thoracolumbar intervertebral disk herniation. *J Vet Intern Med.* (2017) 31:498–504. doi: 10.1111/jvim.14656
130. Gilmour LJ, Jeffery ND, Miles K, Riedesel E. Single-shot turbo spin echo pulse sequence findings in dogs with and without progressive myelomalacia. *Vet Radiol Ultrasound.* (2017) 58:197–205. doi: 10.1111/vru.12463
131. Cardy TJ, De Decker S, Kenny PJ, Volk HA. Clinical reasoning in canine spinal disease: what combination of clinical information is useful? *Vet Rec.* (2015) 177:171. doi: 10.1136/vr.102988
132. Gomes SA, Volk HA, Packer RM, Kenny PJ, Beltran E, De Decker S. Clinical and magnetic resonance imaging characteristics of thoracolumbar intervertebral disk extrusions and protrusions in large breed dogs. *Vet Radiol Ultrasound.* (2016) 57:417–26. doi: 10.1111/vru.12359
133. Crawford AH, De Decker S. Clinical presentation and outcome of dogs treated medically or surgically for thoracolumbar intervertebral disc protrusion. *Vet Rec.* (2017) 180:569. doi: 10.1136/vr.103871
134. Moissonnier P, Meheust P, Carozzo C. Thoracolumbar lateral corpectomy for treatment of chronic disk herniation: technique description and use in 15 dogs. *Vet Surg.* (2004) 33:620–8. doi: 10.1111/j.1532-950X.2004.04085.x
135. Jeffery ND, Harcourt-Brown TR, Barker AK, Levine JM. Choices and decisions in decompressive surgery for thoracolumbar intervertebral disk herniation. *Vet Clin North Am Small Anim Pract.* (2018) 48:169–86. doi: 10.1016/j.cvsm.2017.08.014
136. Levine GJ, Levine JM, Walker MA, Pool RR, Fosgate GT. Evaluation of the association between spondylosis deformans and clinical signs of intervertebral disk disease in dogs: 172 cases (1999-2000). *J Am Vet Med Assoc.* (2006) 228:96–100. doi: 10.2460/javma.228.1.96
137. Vandevelde M, Higgins RJ, Oevermann A. Trauma. *Veterinary Neuropathology: Essentials of Theory and Practice.* Chichester: Wiley-Blackwell (2012). p. 81–91.
138. Sharp NJ, Cofone M, Robertson ID, De Carlo A, Smith GK, Thrall DE. Computed tomography in the evaluation of caudal cervical spondylomyelopathy of the doberman pinscher. *Vet Radiol Ultrasound.* (1995) 36:100–8. doi: 10.1111/j.1740-8261.1995.tb00224.x
139. Yu YL, Du Boulay GH, Stevens JM, Kendall BE. Computed tomography in cervical spondylotic myelopathy and radiculopathy: visualization of structures, myelographic comparison, cord measurements, and clinical utility. *Neuroradiology.* (1986) 28:221–36. doi: 10.1007/BF00548196
140. De Decker S, Gomes SA, Packer RM, Kenny PJ, Beltran E, Parzefall B, et al. Evaluation of magnetic resonance imaging guidelines for differentiation between thoracolumbar intervertebral disk extrusion and intervertebral disk protrusion in dogs. *Vet Radiol Ultrasound.* (2016) 57:526–33. doi: 10.1111/vru.12394
141. Suri A, Chabbra RP, Mehta VS, Gaikwad S, Pandey RM. Effect of intramedullary signal changes on the surgical outcome of patients with cervical spondylotic myelopathy. *Spine J.* (2003) 3:33–45. doi: 10.1016/S1529-9430(02)00448-5
142. De Decker S, Gielen IM, Duchateau L, Lang J, Dennis R, Corzo NM, et al. Intraobserver and interobserver agreement for results of low field magnetic resonance imaging in dogs with and without clinical signs of disk associated Wobbler syndrome. *J Am Vet Med Assoc.* (2011) 238:74–80. doi: 10.2460/javma.238.1.74
143. De Decker S, Da Costa RC, Volk HA, Van Ham L. Current insights and controversies in the pathogenesis and diagnosis of disc-associated cervical spondylomyelopathy in dogs. *Vet Rec.* (2012) 171:531–7. doi: 10.1136/vr.e7952
144. De Decker S, Gielen IM, Duchateau L, Van Soens I, Bavegems V, Bosmans T, et al. Low field magnetic resonance imaging findings of the caudal portion of the cervical region in clinically normal doberman pinschers and foxhounds. *Am J Vet Res.* (2010) 71:428–34. doi: 10.2460/ajvr.71.4.428
145. De Decker S, Fenn J. Acute herniation of nondegenerate nucleus pulposus: acute noncompressive nucleus pulposus extrusion and compressive hydrated nucleus pulposus extrusion. *Vet Clin North Am Small Anim Pract.* (2018) 48:95–109. doi: 10.1016/j.cvsm.2017.08.004
146. De Risio L. A review of fibrocartilaginous embolic myelopathy and different types of peracute non-compressive intervertebral disk extrusions in dogs and cats. *Front Vet Sci.* (2015) 18:24. doi: 10.3389/fvets.2015.00024
147. De Risio L, Adams V, Dennis R, McConnell FJ. Association of clinical and magnetic resonance imaging findings with outcome in

dogs with presumptive acute noncompressive nucleus pulposus extrusion: 42 cases (2000–2007). *J Am Vet Med Assoc.* (2009) 234:495–504. doi: 10.2460/javma.234.4.495
148. Beltran E, Dennis R, Doyle V, de Stefani A, Holloway A, De Risio L. Clinical and magnetic resonance imaging features of canine compressive cervical myelopathy with suspected hydrated nucleus pulposus extrusion. *J Small Anim Pract.* (2012) 53:101–7. doi: 10.1111/j.1748-5827.2011.01166.x
149. McKee WM, Downes CJ, Pink JJ, Gemmill TJ. Presumptive exercise-associated peracute thoracolumbar disc extrusion in 48 dogs. *Vet Rec.* (2010) 166:523–8. doi: 10.1136/vr.b4823
150. Ros C, de la Fuente C, Rodenas S, Novellas R, Viu J, Añor S. Myelographic and low-field magnetic resonance imaging findings in dogs with presumptive acute hydrated non-compressive nucleus pulposus extrusion. *Vet Rec.* (2017) 181:594–9. doi: 10.1136/vr.104201
151. Taylor-Brown FE, De Decker S. Presumptive acute non-compressive nucleus pulposus extrusion in 11 cats: clinical features, diagnostic imaging findings, treatment and outcome. *J Feline Med Surg.* (2015) 19:21–26. doi: 10.1177/1098612X15605150
152. Gallastegui A, Davies E, Zwingenberger AL, Nykamp S, Rishniw M, Johnson PJ. MRI has limited agreement with CT in the evaluation of vertebral fractures of the canine trauma patient. *Vet Radiol Ultrasound.* (2019) 60:533–42. doi: 10.1111/vru.12785
153. Kinns J, Mai W, Seiler G, Zwingenberger A, Johnson V, Cáceres A, et al. Radiographic sensitivity and negative predictive value for acute canine spinal trauma. *Vet Radiol Ultrasound.* (2006) 47:563–70. doi: 10.1111/j.1740-8261.2006.00186.x
154. Chang Y, Dennis R, Platt SR, Penderis J. Magnetic resonance imaging of traumatic intervertebral disc extrusion in dogs. *Vet Rec.* (2007) 160:795–9. doi: 10.1136/vr.160.23.795
155. Fenn J, Drees R, Volk HA, De Decker S. Comparison of clinical signs and outcomes between dogs with presumptive ischemic myelopathy and dogs with acute noncompressive nucleus pulposus extrusion. *J Am Vet Med Assoc.* (2016) 249:767–75. doi: 10.2460/javma.249.7.767
156. Mari L, Behr S, Shea A, Dominguez E, Johnson PJ, Ekiri A, et al. Outcome comparison in dogs with a presumptive diagnosis of thoracolumbar fibrocartilaginous embolic myelopathy and acute non-compressive nucleus pulposus extrusion. *Vet Rec.* (2017) 181:293. doi: 10.1136/vr.104090
157. De Risio L, Adams V, Dennis R, McConnell F, Platt S. Magnetic resonance imaging findings and clinical associations in 52 dogs with suspected ischemic myelopathy. *J Vet Intern Med.* (2007) 21:1290–8. doi: 10.1111/j.1939-1676.2007.tb01951.x
158. Fenn J, Drees R, Volk HA, De Decker S. Inter- and intraobserver agreement for diagnosing presumptive ischemic myelopathy and acute noncompressive nucleus pulposus extrusion in dogs using magnetic resonance imaging. *Vet Radiol Ultrasound.* (2016) 57:33–40. doi: 10.1111/vru.12289
159. Specchi S, Johnson P, Beauchamp G, Masseau I, Pey P. Assessment of interobserver agreement and use of selected magnetic resonance imaging variables for differentiation of acute noncompressive nucleus pulposus extrusion and ischemic myelopathy in dogs. *J Am Vet Med Assoc.* (2016) 248:1013–21. doi: 10.2460/javma.248.9.1013
160. Mari L, Behr S, Shea A, Dominguez E, Ricco C, Alcoverro E, et al. Predictors of urinary or fecal incontinence in dogs with thoracolumbar acute non-compressive nucleus pulposus extrusion. *J Vet Intern Med.* (2019) 33:2693–700. doi: 10.1111/jvim.15626
161. Hamilton T, Glass E, Drobatz K, Agnello KA. Severity of cord dysfunction and pain associated with hydrated nucleus pulposus extrusion in dogs. *Vet Comp Orthop Traumatol.* (2014) 27:313–18. doi: 10.3415/VCOT-13-06-0076
162. Dolera M, Malfassi L, Marcarini S, Mazza G, Sala M, Carrara N, et al. Hydrated nucleus pulposus extrusion in dogs: correlation of magnetic resonance imaging and microsurgical findings. *Acta Vet Scand.* (2015) 57:58. doi: 10.1186/s13028-015-0151-x
163. Borlace T, Gutierrez-Quintana R, Taylor-Brown FE, De Decker S. Comparison of medical and surgical treatment for acute cervical compressive hydrated nucleus pulposus extrusion in dogs. *Vet Rec.* (2017) 181:625. doi: 10.1136/vr.104528
164. Nessler J, Flieshardt C, Tünsmeyer J, Dening R, Tipold A. Comparison of surgical and conservative treatment of hydrated nucleus pulposus extrusion in dogs. *J Vet Intern Med.* (2018) 32:1989–95. doi: 10.1111/jvim.15304
165. Royaux E, Martle V, Kromhout K, van der Vekens E, Broeckx BJG, van Ham L, et al. Detection of compressive hydrated nucleus pulposus extrusion in dogs with mulitslice computed tomotgraphy. *Vet J.* (2016) 216:202–6. doi: 10.1016/j.tvjl.2016.08.012
166. McKee WM, Downes CJ. Rupture of the dura mater in two dogs caused by the peracute extrusion of a cervical disc. *Vet Rec.* (2008) 162:479–81. doi: 10.1136/vr.162.15.479
167. Basser PJ, Mattiello J, LeBihan D. MR diffusion tensor spectroscopy and imaging. *Biophys J.* (1994) 66:259–67. doi: 10.1016/S0006-3495(94)80775-1
168. Ito R, More S, Melhem ER. Diffusion tensor brain imaging and tractography. *Neuroimaging Clin N Am.* (2002) 12:1–19. doi: 10.1016/S1052-5149(03)00067-4
169. Vedantam A, Jirjis MB, Schmit BD, Wang MC, Ulmer JL, Kurpad SN. Diffusion tensor imaging of the spinal cord: insights from animal and human studies. *Neurosurgery.* (2014) 74:1–8. doi: 10.1227/NEU.0000000000000171
170. Budde MD, Kim JH, Liang HF, Schmidt RE, Russel JH, Cross AH, et al. Toward accurate diagnosis of white matter pathology using diffusion tensor imaging. *Magn Reson Med.* (2007) 57:688–95. doi: 10.1002/mrm.21200
171. Herrera JJ, Chacko T, Narayana PA. Histological correlation of diffusion tensor imaging metrics in experimental spinal cord injury. *J Neurosci Res.* (2008) 86:443–7. doi: 10.1002/jnr.21481
172. Sundberg LM, Herrera JJ, Narayana PA. *In vivo* longitudinal MRI and behavioral studies in experimental spinal cord injury. *J Neurotrauma.* (2010) 27:1753–67. doi: 10.1089/neu.2010.1369
173. Basser PJ, Pajevic S, Pierpaoli C, Duda J, Aldroubi A. *In vivo* fiber tractography using DT-MRI data. *Magn Reson Med.* (2000) 44:625–32. doi: 10.1002/1522-2594(200010)44:4<625::AID-MRM17>3.0.CO;2-O
174. Ducreux D, Fillard P, Facon D, Ozanne A, Lepeintre J, Renoux J, et al. Diffusion tensor magnetic resonance imaging and fiber tracking in spinal cord lesions: current and future indications. *Neuroimaging Clin N Am.* (2007) 17:137–47. doi: 10.1016/j.nic.2006.11.005
175. Demir A, Ries M, Vital JM, Dehais J, Arne P, Caille JM, et al. Diffusion-weighted MR imaging with apparent diffusion coefficient and apparent diffusion tensor maps in cervical spondylotic myelopathy. *Radiology.* (2003) 229:37–43. doi: 10.1148/radiol.2291020658
176. Facon D, Ozanne A, Fillard P, Lepeintre JF, Tournoux-Facon C, Ducreux D. MR diffusion tensor imaging and fiber tracking in spinal cord compression. *Am J Neuroradiol.* (2005) 26:1587–94.
177. Kelley BJ, Harel NY, Kim CY, Papademetris X, Coman D, Wang X, et al. Diffusion tensor imaging as a predictor of locomotion function after experimental spinal cord injury and recovery. *J Neurotrauma.* (2014) 31:1363–73. doi: 10.1089/neu.2013.3238
178. Martin AR, Alexsanderek I, Cohen-Adad J, Tarmohamed Z, Tetreault L, Smith N, et al. Translating state-of-the-art spinal cord MRI techniques to clinical use: a systematic review of clinical studies utilizing DTI, MR, MWF, MRS, and fMRI. *Neuroimage.* (2016) 10:192–238. doi: 10.1016/j.nicl.2015.11.019
179. Peterson JA, Wilm BJ, von Meyenburg J, Schubert M, Seifert B, Najafi Y, et al. Chronic cervical spinal cord injury: DTI correlates with clinical and electrophysiological measures. *J Neurotrauma.* (2012) 29:1556–66. doi: 10.1089/neu.2011.2027
180. Cheran S, Shanmuganathan K, Zhuo J, Mirvis SE, Aarabi B, Alexander MT, et al. Correlation of MR diffusion tensor imaging parameters with ASIA motor scores in hemorrhagic and nonhemorrhagic acute spinal cord injury. *J Neurotrauma.* (2011) 28:1881–92. doi: 10.1089/neu.2010.1741
181. Ellingson BM, Ulmer JL, Kurpad SN, Schmit BD. Diffusion tensor MR imaging in chronic spinal cord injury. *Am J Neuroradiol.* (2008) 29:1976–82 doi: 10.3174/ajnr.A1272
182. Endo T, Suzuki S, Utsunomiya A, Uenohara H, Tominaga T. Prediction of neurological recovery using apparent diffusion coefficient in cases of incomplete spinal cord injury. *Neurosurgery.* (2011) 68:329–36. doi: 10.1227/NEU.0b013e3182031ce7
183. Koskinen E, Brander A, Hakulinen U, Luoto T, Helminen M, Ylinen A, et al. Assessing the state of chronic spinal cord injury using diffusion tensor imaging. *J Neurotrauma.* (2013) 30:1587–95. doi: 10.1089/neu.2013.2943

184. Schwartz ED, Duda J, Shumsky JS, Cooper ET, Gee J. Spinal cord diffusion tensor imaging and fiber tracking can identify white matter tract disruption and glial scar orientation following lateral funiculotomy. *J Neurotrauma.* (2005) 22:1388–98. doi: 10.1089/neu.2005.22.1388
185. Vedantam A, Eckardt G, Wang MC, Schmit BD, Kurpad SN. Clinical correlates of high cervical fractional anisotropy in acute cervical spinal cord injury. *World Neurosurg.* (2015) 83:824–8 doi: 10.1016/j.wneu.2013.09.017
186. Griffin JF, Cohen ND, Young BD, Eichelberger BM, Padua A, Purdy D, et al. Thoracic and lumbar spinal cord diffusion tensor imaging in dogs. *J Magn Reson Imaging.* (2013) 37:632–41. doi: 10.1002/jmri.23862
187. Hobert MK, Stein VM, Dziallas P, Ludwig DC, Tipold A. Evaluation of normal appearing spinal cord by diffusion tensor imaging, fiber tracking, fractional anisotropy, and apparent diffusion coefficient measurement in 13 dogs. *Acta Vet Scand.* (2013) 55:36. doi: 10.1186/1751-0147-55-36
188. Pease A, Miller R. The use of diffusion tensor imaging to evaluate the spinal cord in normal and abnormal dogs. *Vet Radiol Ultrasound.* (2011) 52:492–7. doi: 10.1111/j.1740-8261.2011.01837.x
189. Yoon H, Park NW, Ha YM, Kim J, Moon WJ, Eom K. Diffusion tensor imaging of white and grey matter within the spinal cord of normal beagle dogs: sub-regional differences of the various diffusion parameters. *Vet J.* (2016) 215:110–7. doi: 10.1016/j.tvjl.2016.03.018
190. da Costa RC, Armstrong J, Choi S, Martin-Vaquero P, Dewey B, et al. Diffusion tensor imaging in dogs with and without cervical spondylomyelopathy. *J Vet Intern Med Proc ACVIM Forum.* (2014) 28:976–1134. doi: 10.1111/jvim.12361
191. Konishi Y, Satoh H, Kuroiwa Y, Kusaka M, Yamashita A, Asada Y, et al. Application of fiber tractography and diffusion tensor imaging to evaluate spinal cord diseases in dogs. *J Vet Med Sci.* (2017) 79:418–24. doi: 10.1292/jvms.16-0504
192. Lewis MJ, Yap PT, McCullough S, Olby NJ. The relationship between lesion severity characterized by diffusion tensor imaging and motor function in chronic canine spinal cord injury. *J Neurotrauma.* (2017) 35:500–7. doi: 10.1089/neu.2017.5255
193. Liu C, Yang D, Li J, Li D, Yang M, Sun W, et al. Dynamic diffusion tensor imaging of spinal cord contusion: a canine model. *J Neuro Res.* (2018) 96:1093–103. doi: 10.1002/jnr.24222
194. Wang-Leandro A, Hobert MK, Kramer S, Rohn K, Stein VM, Tipold A, et al. The role of diffusion tensor imaging as an objective tool for the assessment of motor function recovery after paraplegia in a naturally-occurring large animal model of spinal cord injury. *J Transl Med.* (2018) 16:258. doi: 10.1186/s12967-018-1630-4
195. Yoon H, Kim J, Moon WJ, Nahm SS, Zhao J, Kim HM, et al. Characterization of chronic axonal degeneration using diffusion tensor imaging in canine spinal cord injury: a quantitative analysis of diffusion tensor imaging parameters according to histopathological differences. *J Neurotrauma.* (2017) 34:3041–50. doi: 10.1089/neu.2016.4886
196. Yoon H, Moon WJ, Nahm SS, Kim J, Eom K. Diffusion tensor imaging of scarring, necrosis, and cavitation based on histopathological findings in dogs with chronic spinal cord injury: evaluation of multiple diffusion parameters and their correlations with histopathological findings. *J Neurotrauma.* (2018) 35:1387–97. doi: 10.1089/neu.2017.5409
197. Lewis MJ, Early PJ, Mariani CL, Munana KR, Olby NJ. Influence of duration of injury on diffusion tensor imaging in acute canine spinal cord injury. *J Neurotrauma.* (2020). doi: 10.1089/neu.2019.6786. [Epub ahead of print].
198. Cohen-Adad J. Functional magnetic resonance imaging of the spinal cord: current status and future developments. *Semin Ultrasound CT MRI.* (2017) 38:176–86. doi: 10.1053/j.sult.2016.07.007
199. Filippi M, Rocca MA. Magnetization transfer magnetic resonance imaging of the brain, spinal cord, and optic nerve. *J Am Soc Exper Neurother.* (2007) 4:401–13. doi: 10.1016/j.nurt.2007.03.002
200. Laule C, Moore GRW. Myelin water imaging to detect demyelination and remyelination and its validation in pathology. *Brain Pathol.* (2018) 28:750–64. doi: 10.1111/bpa.12645
201. Wyss PO, Hock A, Kollias S. The application of human spinal cord magnetic resonance spectroscopy to clinical studies: a review. *Semin Ultrasound CT MRI.* (2017) 38:153–62. doi: 10.1053/j.sult.2016.07.005
202. Chen C, Jia Z, Han Z, Gu T, Li W, Li H, et al. Quantitative T2 relaxation time and magnetic transfer ratio predict endplate biochemical content of intervertebral disc degeneration in a canine model. *BMC Musculoskel Disord.* (2015) 16:157. doi: 10.1186/s12891-015-0610-6
203. Vite CH, Cross JR. Correlating magnetic resonance findings with neuropathology and clinical signs in dogs and cats. *Vet Radiol Ultrasound.* (2011) 52:S23–31. doi: 10.1111/j.1740-8261.2010.01782.x

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REVIEW
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Current Insights Into the Pathology of Canine Intervertebral Disc Extrusion-Induced Spinal Cord Injury

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Spinal cord injury (SCI) in dogs is commonly attributed to intervertebral disc extrusion (IVDE). Over the last years substantial progress was made in the elucidation of factors contributing to the pathogenesis of this common canine disease. A detailed understanding of the underlying histopathological and molecular alterations in the lesioned spinal cord represents a prerequisite to translate knowledge on the time course of secondary injury processes into the clinical setting. This review summarizes the current state of knowledge of the underlying pathology of canine IVDE-related SCI. Pathological alterations in the spinal cord of dogs affected by IVDE-related SCI include early and persisting axonal damage and glial responses, dominated by phagocytic microglia/macrophages. These processes are paralleled by a pro-inflammatory microenvironment with dysregulation of cytokines and matrix metalloproteinases within the spinal cord. These data mirror findings from a clinical and therapeutic perspective and can be used to identify biomarkers that are able to more precisely predict the clinical outcome. The pathogenesis of progressive myelomalacia, a devastating complication of SCI in dogs, is not understood in detail so far; however, a fulminant and exaggerating secondary injury response with massive reactive oxygen species formation seems to be involved in this unique neuropathological entity. There are substantial gaps in the knowledge of pathological changes in IVDE with respect to more advanced and chronic lesions and the potential involvement of demyelination. Moreover, the role of microglia/macrophage polarization in IVDE-related SCI still remains to be investigated. A close collaboration of clinical neurologists and veterinary pathologists will help to facilitate an integrative approach to a more detailed understanding of the molecular pathogenesis of canine IVDE and thus to identify therapeutic targets.

Keywords: spinal cord injury, IVDE, extrusion, macrophage, immunohistochemistry, axonal damage, macrophage polarization, cytokine

INTRODUCTION

Spinal cord injury (SCI) in dogs can be caused by either extrinsic or intrinsic forces. Though extrinsic traumatic forces such as road accidents, which make up the majority of human cases of severe SCI, do also occur in pet dogs, intervertebral disc extrusion (IVDE) is by far the most common cause for SCI in dogs (1). IVDE-induced SCI accounts for up to 2% of all diseases in dogs (2–4) and represents one of the most common diagnoses made by veterinary neurologists. In a study conducted in Switzerland with a referral hospital population of nearly 3,500 dogs with central nervous system (CNS) diseases included, IVDE represented the most common diagnosis, followed by epilepsy and other neurodegenerative diseases (5).

Due to high standards in clinical management, the prognosis of IVDE-induced SCI of mild to moderate severity is generally good; however, 40–50% of dogs with severe SCI secondary to IVDE (those who are paraplegic with absent nociception) do not recover the ability to ambulate and may be euthanized because of the condition, even with the highest standard of care. The post-mortem examination of such cases provides an opportunity to gain basic insights into the pathology and pathogenetic basis of this clinically important disease. Besides its doubtless high veterinary relevance, IVDE-induced SCI in dogs moreover shares striking similarities with human traumatic SCI (6). Similar to most cases of traumatic SCI in humans, IVDE-induced SCI is caused by a relative contribution of both compressive and contusive forces caused by structures anatomically located ventral to the spinal cord (6–8). This is in contrast to most experimental rodent models for SCI, which commonly rely on purely concussive injuries caused by dorsal weight drop or complete cord transection (6–9). Moreover, compared to rodents, the canine spinal cord more closely resembles the size of the human counterpart (8). Lastly, and probably most important, canine IVDE is a spontaneous disease with a high inter-individual variability (6, 7). Based on these similarities, canine IVDE has developed into an acknowledged translational animal model that may add the missing heterogeneity to experimental investigations in rodent models of SCI (6, 8). Consequently, veterinary clinical studies in canine SCI may help to translate findings from experimental rodent studies into the clinically relevant, naturally occurring disease (7, 8, 10–12).

An enormous body of literature exists on the morphologic and molecular pathology of experimental SCI, with comparatively less data on naturally occurring cases of human SCI. It is highly likely that canine IVDE-induced SCI shares many of these pathological features reported in both human traumatic SCI and experimental animal models. The present summary focuses on data that have been gained in (histo-) pathological studies on naturally occurring IVDE-induced SCI in dogs, referring to only a few studies on experimental SCI, where the knowledge of IVDE-induced canine SCI is only fragmentary or absent. Starting with a brief overview on canine intervertebral disc disease, the major focus of this paper is to provide an overview of the pathologic events in the injured canine spinal cord with reference to therapeutic implications where applicable.

Basic Pathological Mechanisms of Canine Intervertebral Disc Degeneration

Degeneration of the intervertebral disc is commonly the prerequisite for later IVDE-induced SCI; i.e., IVD degeneration represents an important predisposing factor for the disc herniation into the vertebral canal. Early pioneer studies by Hansen (1952) (13) have extensively described the pathological changes during IVD degeneration and details of intervertebral disc anatomy and degeneration are reviewed elsewhere in this issue (Fenn et al.). Briefly, in chondrodystrophic dog breeds such as the dachshund, beagle, and Pekingese, the nucleus pulposus of multiple intervertebral discs undergoes progressive chondroid metaplasia beginning in juvenile individuals (2, 6). Initial degenerative changes are completed as early as 1 year of age (2, 4). Due to their familial predisposition, chondrodystrophic breeds are much more likely to develop disc herniation than non-chondrodystrophic breeds, as they are prone to premature senescence of the nucleus pulposus (6, 14). Among chondrodystrophic breeeds, French Bulldogs have gained enormously in popularity. Recent studies suggest that French bulldogs are prone to various neurological diseases with IVDE ranging on top of the neurological diseases in this breed (15). In contrast to other breeds, cervical location of IVDE seems to be more common in French bulldogs (15). The reasons for the relatively high level of predisposition for neurological diseases in this breed remain speculative; however, besides chondrodystrophy, excessive inbreeding might represent one factor that contributes to predisposition of French Bulldogs to neurological disease development (15). For a more detailed review of the genetic factors involved in canine IVDD the reader is referred to Dickinson et al. in this edition. Recent independent genome-wide association analyses for skeletal dysplasia and IVDE identified a highly expressed *FGF4* retrogene on CFA12, which is associated with both IVDE and chondrodystrophy (16). The nucleus pulposus is replaced by hyaline cartilage. The latter progressively degenerates and calcifies in the late stage of IVD degeneration (2, 17). In dachshunds with acute disc herniation, histopathology reveals that the majority of extruded disc material is calcified, even in the absence of radiographically visible calcification (18). More recent histopathological studies propose a grading scheme for intervertebral disc degeneration based on an assortment of parameters. These include morphology of the annulus fibrosus, chondrocyte metaplasia of the annulus fibrosus, tears and cleft formations, chondrocyte proliferation within the nucleus pulposus, presence of notochordal cells in the nucleus pulposus, matrix staining of nucleus pulposus with Alcian Blue/Picrosirius Red, endplate morphology, new bone formation, and subchondral bone sclerosis (19). In this study, glycosaminoglycan content and total histological score showed high correlation.

In contrast to chondrodystrophic breeds, the intervertebral disc of non-chondrodystrophic dogs has historically been believed to undergo an age-dependent and slowly progressing

fibroblastic metaplasia of both the annulus fibrosus and nucleus pulposus (2, 4, 6), which may represent a non-hereditary wear-and-tear-phenomenon. This traditional concept, i.e., chondroid metaplasia of the nucleus pulposus in chondrodystrophic breeds, and fibrous metaplasia in non-chondrodystrophic dogs, has been recently been disputed by studies suggesting that IVDD in chondrodystrophic and non-chondrodystrophic breeds is more similar than previously believed (20). In fact, chondroid metaplasia is observed in both chondrodystrophic and non-chondrodystrophic dogs and fibrocytes were not seen in the nucleus pulposus in any of the investigated discs in a recent study, thus challenging this original "chondrodystrophic" and "non-chondrodystrophic" paradigm in canine IVDD (20).

Clinical Spinal Cord Injury Caused by Intervertebral Disc Herniation

The clinical presentation of IVDE in dogs spans a spectrum ranging from neck or back pain to severe spinal cord injury with loss of sensory and motor function caudal to the lesion. While several clinical grading systems have been employed throughout the literature to quantify severity of injury, the most commonly used is a version of the modified Frankel scale. This scale ranges from paraplegia with absent superficial and deep pain sensation to normal dogs. Injury severity, as measured in this way, correlates well with prognosis for recovery after surgical decompression where dogs with grade 0 injuries experience return of unassisted ambulation and fecal and urinary continence in 50–60% of cases (21, 22).

The pathogenesis of canine IVD degeneration and IVDE has been recently reviewed in detail (14) and is also covered in detail in other articles in this issue. Using Hansen's descriptors, chondrodystrophic breeds are predisposed to Hansen type I herniation (IVDE) whereas the non-chondrodystrophic breeds are more prone to type II herniation (2, 17, 23). The vast majority of research focusses on IVDE, as it is the most common type and often induces the most severe lesions. Rapid extrusion of nucleus pulposus results in compressive and contusive injury to the spinal cord. Though IVDE induces a mixed contusive/compressive force to the respective spinal cord segment (6–8, 14), the extent of each varies both within the individual patient and with the type of herniation observed. Since Hansen type I disc extrusions typically occur acutely and with substantial force, they generally cause considerably more severe trauma to the respective spinal cord segments as compared to Hansen type II disc protrusions, which are less severe and lead to more slowly developing forces applied to the spinal cord (i.e., focus on the compressive part of the force) (2, 6, 13, 14, 23).

In chondrodystrophic breeds, approximately 75% of intervertebral disc herniations are found at the level of the Th 12 to L2 (13). Intervertebral disc herniations at the cervical level are less common, accounting for approximately 14 to 35 percent of all intervertebral disc herniations (4, 6, 23).

Some cases can clinically not be classified into either Hansen type I or type II, as a proportion of non-chondrodystrophic dogs may develop acute clinical signs with rapid onset, while few dogs with Hansen type I herniation (extrusion) may develop slowly progressing signs (14). Other types of herniation have been described and besides the aforementioned forms, in which IVDE is the sequela of IVD degeneration, non-degenerate physiological disc material may be herniated into the vertebral canal and/or spinal cord by extrinsic traumatic forces (traumatic disc prolapse) (13, 14, 24). Various other forms of IVD disease are covered in detail in the article of Fenn et al. in this Issue.

Irrespective of the exact type, herniation of the intervertebral disc typically occurs in the dorsal direction, i.e., into the vertebral canal (14). Monocytes and macrophages are found in extruded disc material, and there is activation of extracellular signal-regulated kinase p38 (25). Moreover, similar to IVDE in humans, canine thoracolumbar IVDE is associated with elevated gene and protein expression of key cytokines such as IL-6 and TNF-α and down-regulated expression of IL-1β (25).

PATHOLOGY OF CANINE INTERVERTEBRAL DISC HERNIATION-INDUCED SPINAL CORD INJURY

General Morphology

Most of the data on pathological lesions in the spinal cord derive from individuals with acute to subacute severe SCI (paraplegic with and without pain perception, respectively), with a considerable lack of histopathological descriptions on more chronic lesions and less severe injuries, which is due to the fact that, in a non-experimental set-up of studies on a naturally occurring spontaneous disease, material for histopathological investigations most commonly derives from euthanized individuals with an acute onset of severe clinical signs and a poor prognosis. Thus, pathological descriptions are somewhat biased, and one should consider them mostly mirroring extreme cases of a wide spectrum of time course and lesion severity, respectively.

IVDE causes considerable, though highly variable, pathological alterations within the respective spinal cord segments and at distant sites within the neuraxis. Upon necropsy, dorsal removal of vertebral laminae exposes the vertebral canal and degenerate intervertebral disc material may be detected within the vertebral canal in close proximity and often firmly attached to the contused and compressed spinal cord segment. Macroscopic alterations in the spinal cord itself may range from no detectable changes, to discoloration, grossly obvious hemorrhage, severe spinal cord and dural laceration, or spinal cord atrophy in long standing cases. The histological changes observed in dogs with IVDE-associated SCI are relatively similar to histopathological alterations in spinal cords of humans affected by SCI, underlining the role of canine IVDE as a translational animal model that may allow extrapolation of findings to naturally occurring human cases.

Histopathological alterations in the spinal cord of dogs with SCI have been detailed as early as 1978 (26). In general, lesions are highly variable, and may consist of variable degrees of necrosis and hemorrhage in acute stages (26); **Figure 1**. Ultrastructurally, hemorrhages, axonal spheroid formation, glial cell swelling,

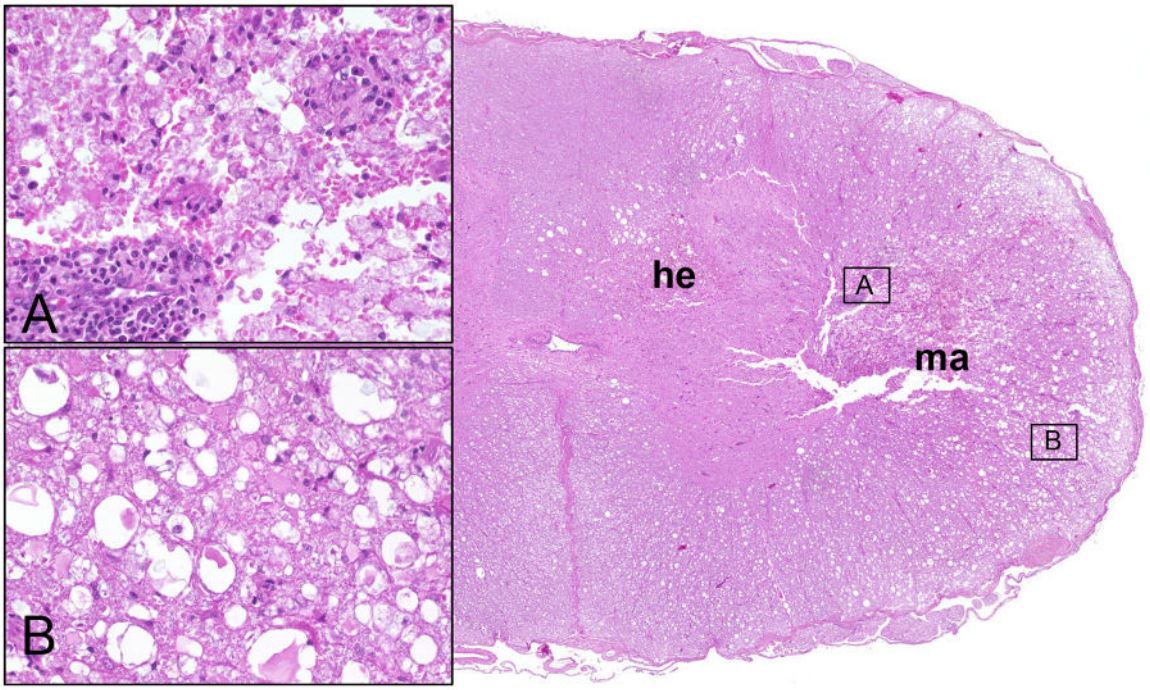

FIGURE 1 | Male Dachshund with type I intervertebral disc herniation (acute extrusion). Overview (right side) of HE stained spinal cord transversal section with hemorrhage (he) accentuated within the gray matter and white matter malacia (ma). Inset upper left **(A)**: moderate perivascular cuffing of mononuclear leukocytes and focal disintegration of neuroparenchyma (necrosis, malacia). Inset lower left **(B)**: moderate to severe white matter vacuolation within the ventrolateral funiculus, characterized by multiple dilated myelin sheaths that contain hypereosinophilic swollen axons (spheroids). 20x magnification in insets.

white matter edema, and demyelination are observed in cases of naturally occurring canine SCI (9). Moreover, remyelination in the advanced disease by both oligodendrocytes and Schwann cells was shown using electron microscopy (9). Depending on the severity of the initial trauma, secondary injury processes may finally culminate into liquefactive necrosis (malacia) of the spinal cord segment and glial scarring with variable involvement of neuroparenchymal cavitation and cyst formation. Chronic intramedullary lesions/cavitations are associated with severe initial SCI and negative clinical outcome (27). Ascending and descending myelomalacia is a devastating complication in a proportion of dogs with SCI and will be discussed at the end of this chapter.

Clinical neurological grades of dogs affected by thoracolumbar IVDE-induced SCI correlate with the extent of white matter damage (28). Of interest, however, is the notable observation that clinical injury severity does not always correlate with severity of histopathologic lesions, underscoring the need for further studies of pathological features of canine IVDE-induced SCI (28). Additionally, some clinical signs such as duration of clinical signs, Schiff-Sherrington posture, loss of reflexes and pain on spinal palpation are not associated with the histopathological severity of spinal cord damage (28). These results suggest that some clinical signs are rather associated with functional neurological disturbances such as conduction block due to energy depletion or failure, that are not necessarily reflected by histopathological alterations.

Thus, both the immune response and axonal pathology are pivotal hallmarks of SCI (6). Consequently, these pathogenetic factors have been proposed to serve as major targets for future therapies (6, 29–32) and a detailed understanding of the underlying pathology during canine IVDE-induced SCI is a basis essential to the development of such therapeutic interventions (6).

Axonal and Myelin Pathology in Canine IVDE-Induced SCI

Axonal damage is a central hallmark of all forms of endogenous or exogenous traumatic CNS injury and various studies have characterized the underlying molecular pathogenesis of axonal degeneration and regeneration in traumatic brain and spinal cord injury in detail (33). As axonal damage may be the most obvious pathological correlate of clinical motor deficits, it is not surprising that axonal damage is a consistent histopathological feature of canine IVDE-induced SCI. In histopathology, axonal damage generally appears as axonal swelling and the occurrence of hypereosinophilic spherical enlarged axons (spheroids, **Figure 1**) within dilated myelin sheaths. Sharing many pathogenetic features with Wallerian degeneration, axonal damage is not restricted to the lesion center at the site of disc herniation but may also be seen in various spinal cord segments cranial and caudal to the initial lesion site.

Ultrastructurally, axoplasmic changes in spinal cords from dogs suffering from IVDE-induced SCI are relatively similar to

the ultrastructural axonal changes seen in experimental SCI in rodents and monkeys (9, 34–39). Following compressive injury to the spinal cord in rats there is periaxonal space formation, myelin disruption and granular disintegration of neurofilaments (35). Moreover, organelle accumulation and giant axons may occur (35). Contusion SCI in rhesus monkeys similarly leads to axonal accumulation of dense bodies, vesicular structures, multivesicular bodies, and organelles (36). Axoplasmic vesicles, mitochondria, and electron-dense bodies are observed within reactive axonal enlargements (37). Though variable, all of the above mentioned features are also observed ultrastructurally in dogs with IVDE-induced SCI (6, 9, 34).

Deficits in both fast anterograde axonal transport mechanisms and axonal neurofilament phosphorylation have been implicated in the pathogenesis of axonal damage in canine IVDE-induced SCI (6, 34). Using immunohistochemistry, β-amyloid precursor protein (APP) is not detectable in healthy axons due to fast axonal transport under physiological circumstances. However, there is fast accumulation of APP, if axonal transport is disturbed due to pathological conditions (40). Consequently, APP is a well-established immunohistological marker for axonal damage and has been previously used to detect damaged axons in experimental SCI in dogs caused by inflated angioplasty balloons, where its expression correlates with severity and duration of compression (41, 42). Similarly, experimental SCI in rodents and spontaneous SCI in people are both associated with strong axonal expression of APP (43–45).

In dogs with naturally occurring IVDE-induced SCI, APP is detectable in the lesion epicenter of both acutely and subacutely injured dogs (**Figure 2**) (6, 34). However, axonal APP expression can also be observed up to 3 cm caudal to the lesion epicenter during the subacute phase of injury, suggesting progressive spatial spread of disturbances in fast axonal transport (6, 34). Similarly, experimental rodent models and naturally occurring SCI in people leads to axonal APP-expression distant to the lesion epicenter (43, 45). These findings indicate that axonopathy is not simply and solely attributed to the initial primary injury but rather a timely and spatially progressive phenomenon reflecting secondary injury mechanisms (6).

In parallel to axonal APP-immunoreactivity, previous immunohistochemical studies on canine IVDE-induced SCI revealed enhanced axonal expression of non-phosphorylated neurofilaments (n-NF) in axons of dogs with acute and subacute SCI (6, 34); **Figure 2**. Moreover, similar to APP, n-NF-expression has been noted in axons several centimeters apart from the lesion center (34). However, in contrast to APP, which is mainly detected in swollen axons, n-NF immunopositivity was also seen in several axons with normal diameters (**Figure 2**) (6, 34). This implies that both markers might label, at least in part, distinct axonal pathological processes (6). In traumatic brain injury in rats, neurofilament compaction in axons has previously been reported to occur independently from APP-immunoreactivity (46). Thus, neurofilament alterations and disturbed axoplasmic transport might in part represent differing pathological phenomena (6).

In addition to traumatic CNS injury, enhanced axonal n-NF and APP-expression has been demonstrated in several animal models of demyelinating disease in various species including some dog studies (47–50), suggesting that altered neurofilament phosphorylation and disturbances in fast axonal transport represent conserved phenomena of axonopathy irrespective of the underlying disease entity.

Though axonal damage predominates, evidence for intrinsic axonal regeneration attempts has been reported in dogs with IVDE-induced SCI in terms of axonal expression of growth-associated protein 43 (GAP-43) (34). GAP-43 immunoreactivity was noted in a small proportion of axons in dogs with acute and subacute SCI, which vas verified by immune-electron microscopy. Ultrastructurally, immunoreaction was noted in swollen axons lacking dense body accumulation, but filled with large numbers of mitochondria (34). Axons express GAP-43 during development and regeneration (51). Live imaging on individual axons in experimental SCI have shown early axonal regeneration attempts (52); however, functional restoration seems to be insufficient. It is proposed that regenerating axons during SCI may fail to navigate to a proper target (52). This might in part be attributed to the expression of regeneration-inhibiting molecules such as Nogo and MAG, and pharmacological modulation of these molecules is believed to represent a promising target to facilitate axonal regeneration in terms of functional restoration (53, 54). Several further experimental therapeutic approaches aim to facilitate these intrinsic regenerative responses (29). In fact, transplanting regeneration promoting cells into the spinal cord of rodents with SCI has shown to enhance axonal GAP-43 immunoreactivity, which was associated with an improved clinical outcome (55–57). Moreover, facilitation of alternatively activated anti-inflammatory macrophages is paralleled by increased axonal expression of GAP-43 and improved locomotor recovery in spinal cord lesioned mice (58).

Myelin pathology, though a focus of experimental SCI work, has not been extensively reported in canine SCI. Though there is reduced immunoreactivity of myelin basic protein (MBP) in the white matter of dogs with subacute IVDE-induced SCI (34), this has rather been attributed to myelin edema and myelin sheath swelling than true demyelination. In an ultrastructural study of canine SCI, including various causes such as fractures, subluxations, and IVDE, demyelinated axons were observed within 2 weeks after initial injury and, interestingly, in advanced disease stages, both Schwann cell and oligodendrocyte remyelination was observed (9). Moreover, subtle partial and paranodal myelin abnormalities were seen ultrastructurally. This evidence for delayed myelin loss in canine IVDE-induced SCI is mirrors the situation in human spinal cord injury. Naturally occurring SCI in humans is associated with delayed and long-lasting myelin loss (59, 60). Morphologically detectable myelin abnormalities are generally observed subsequent to early axonal damage, thus recapitulating the principle processes during Wallerian degeneration. Moreover, demyelination in canine SCI might in part also reflect pathomechanisms referred to as the "inside-out theory" in neurodegenerative diseases (6, 61). Though this concept is controversial, it suggests that axonal damage functions as a mechanism triggering secondary demyelination (47, 61). Several lines of evidence indicate

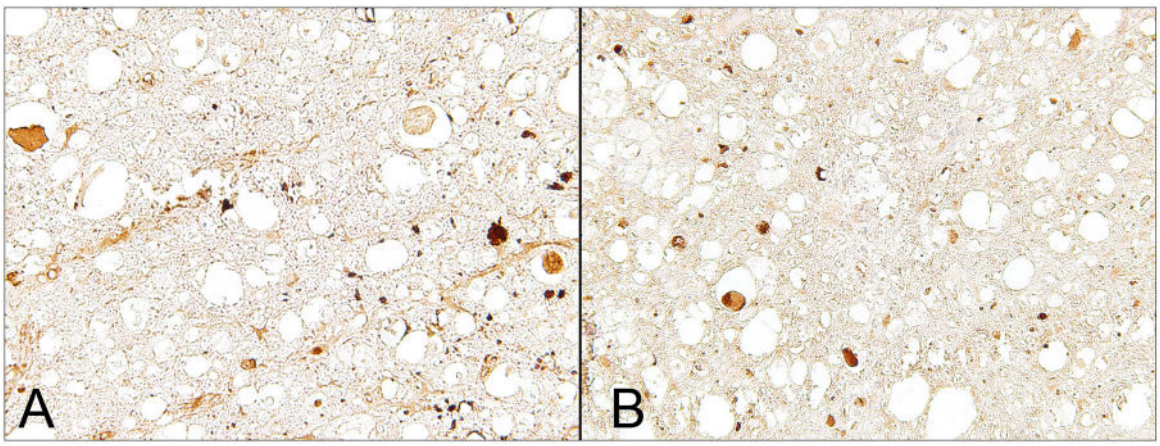

FIGURE 2 | Male Dachshund with type I intervertebral disc herniation (acute extrusion). Immunohistochemical detection of axonal damage. **(A)** Beta-APP accumulates within swollen axons indicating disruption of the fast axonal transport machinery. **(B)** Non-phosphorylated neurofilament (nNF), another marker for axonal damage, is detected within numerous swollen axons but is also expressed by axons with a normal appearing diameter. 40x magnification.

similarities in terms of this triggering function of primary axonopathy between neurodegenerative and viral CNS diseases on the one side and SCI on the other side (6, 61). In a clinical context, dogs with thoracolumbar IVDE with loss of ambulation had higher MBP concentration within the CSF compared with control dogs, suggesting that elevated MBP levels within the CSF are associated with poor clinical outcome (62).

Based on the assumption that demyelination is an event that occurs relatively late in the progress of secondary injury, investigations on chronic cases of canine IVDE-induced SCI are urgently needed. Evidence that demyelination does occur in chronically injured dogs is for instance based on clinical trials. 4-Aminopyridine (4-AP) is a compound known to improve function in demyelinating conditions. Dogs with spinal cord injury treated with 4-AP show significant improvement in supported stepping scores (63) suggesting that demyelination plays a role in advanced and long standing cases. As mentioned above, pathological data on naturally occurring canine SCI are primarily based on dogs with acute to subacute IVDE-induced SCI but there is little information upon the histopathology of chronic cases (14, 64). Similar to experimental data and lesion pathology in human SCI, chronic cases of canine IVDE-induced SCI are characterized by progressive white and gray matter loss with or without cyst formation and progressive replacement by extensive gliosis (14). In an MRI-study on chronic SCI in dogs, intramedullary cavitation and cyst formation was reported (27). Histopathologically, chronic lesions were characterized by gray matter-accentuated malacia, severe gliosis, and variable infiltration of phagocytic gitter cells (27). Multiple axonal spheroids can be detected, suggesting ongoing axonal damage. Myelin sheaths within the white matter showed dilatation and occasional myelinophages within dilated myelin sheaths. Some cases exhibited pan-myelomalacia with complete loss of organotypic structure, replaced by diffuse extensive gliosis (27). Mirroring overall neuroparenchymal loss of both gray and white matter, macroscopic changes of the chronically injured canine spinal cord may include hour-glass shaped atrophy of the respective spinal cord segment (27). Similarly, in experimentally induced SCI in dogs histological analyses at 12 weeks after SCI revealed amorphous cavities in the gray matter with spread to the white matter with caudally accentuated spatial spread up to 1 cm apart from the epicenter (65).

Inflammation and Glial Cell Reactions in Canine IVDE-Induced SCI

In severe acute cases of canine SCI, the first cell type that arises are neutrophils, and increased cell numbers of neutrophils are commonly detected within the CSF of dogs with IVDE-induced SCI. Histopathologically, neutrophils are commonly associated with areas of hemorrhage (66). In parallel, there is infiltration of MAC-387-positive monocyte-derived macrophages and variable perivascular leukocyte cuffing (**Figures 1**, **3**). Cellular reactive changes begin to be more obvious in subacute cases, in which there is a phagocytic response that is impressively dominated by microglia/macrophages (66). MHC class II expressing microglia/macrophages have also been reported as the predominating cell type in human SCI (**Figure 3**) (67), whereas lymphocytes seem to play a subordinate role (67, 68). In dogs, microglial cells have been analyzed in detail in various neurological diseases such as canine distemper virus infection and SCI (11, 69, 70). In healthy dogs, canine microglia derived from the spinal cord show a relatively higher capacity of phagocytosis and generation of reactive oxygen species (ROS) as compared to cells derived from the healthy brain (70). Dogs with SCI reveal enhanced microglial expression of surface molecules such as B7-1, B7-2, MHC class II, CD1c, ICAM 1, CD14, CD44, and CD45, as determined by flow cytometry (11). Besides, phagocytosis and ROS generation of microglia are elevated in dogs with SCI (11).

Extensive research on microglia/macrophages is similarly done in experimental laboratory studies of SCI and manipulation of the response of these cells is regarded as a promising

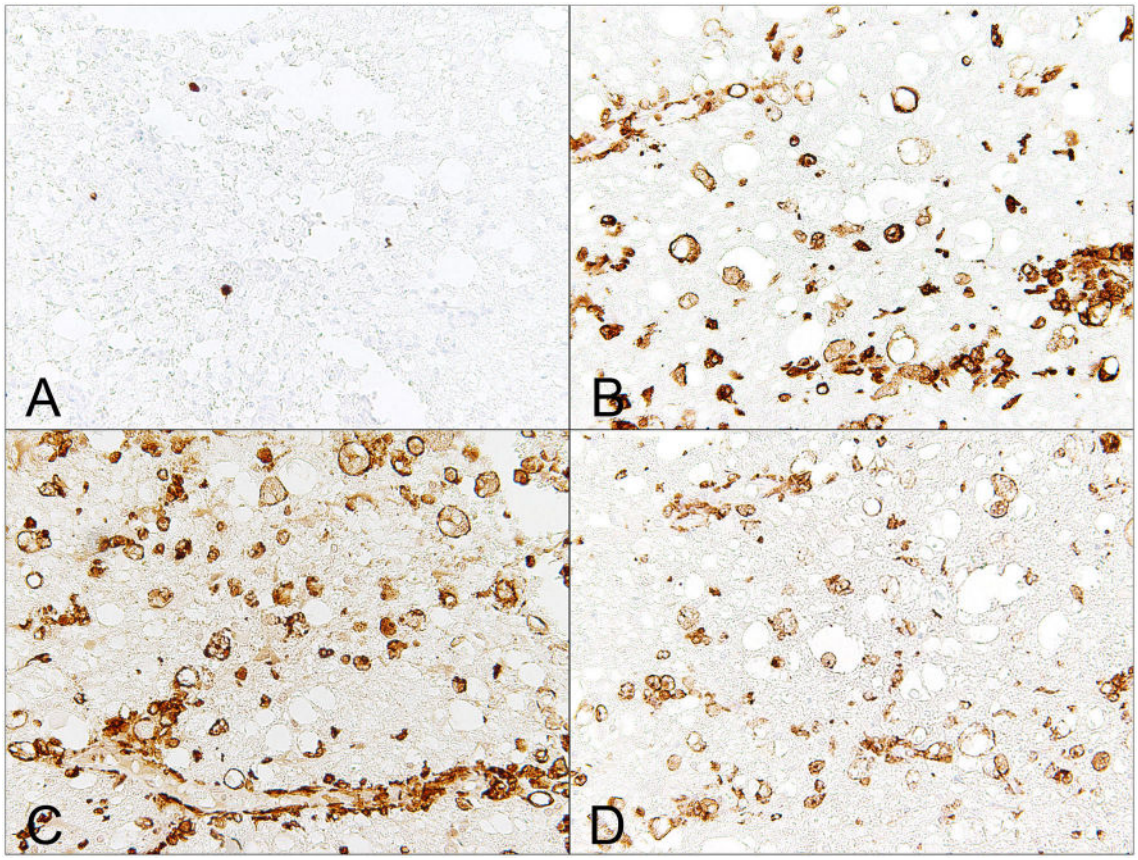

FIGURE 3 | Male Dachshund with type I intervertebral disc herniation (acute extrusion). Immunohistochemical detection of macrophages, which are a dominating immune cell population involved in secondary injury mechanisms. **(A)** Mac387, a clone that detects myeloid/histiocyte antigen, only detects relatively few, monocyte-like blood born macrophages. **(B)** There is severe up-regulation of MHC class II on phagocytic gitter cells. **(C)** Similarly, Iba-1, a pan-macrophage marker, labels numerous phagocytic microglia/macrophages within the affected white matter. **(D)** CD204, a marker that has been proposed to mainly detect M2-polarized macrophages, labels several microglia/macrophages within the white matter and within dilated, optically empty myelin sheaths (myelinophagia). 40x magnification.

field in the development of new therapeutic approaches. Based on a relatively novel basic, but very simplified concept that microglia/macrophages may be polarized into either pro-inflammatory and neurotoxic (M1-) cells or alternatively activated, anti-inflammatory and regeneration promoting (M2-) cells, a bulk of experimental research has been conducted focusing on the role of these cells in SCI. Pioneer studies on rodent SCI revealed that SCI is characterized by an early and persisting M1-dominated macrophage response (71). The fact that this polarized M1-response overwhelms a relatively sparse M2-macrophage response has led to the idea that shifting this phenomenon toward a regeneration-promoting M2-dominated response might be a rewarding research target for therapies in SCI (71).

Whether this macrophage polarization also occurs in the context of clinically relevant naturally occurring canine SCI has not been investigated to date. However, several lines of evidence indicate that the microglia/macrophage response is similarly associated with a polarization of macrophages toward a pro-inflammatory phenotype. Subacute canine IVDE-induced SCI is associated with a dominating response of MHC class II-expressing phagocytic microglia/macrophages that is paralleled by a pro-inflammatory microenvironment (66, 72). Moreover, microglia/macrophages are a pivotal source of ROS, tissue degrading metalloproteinases and neurotoxic mediators.

Detection of M1- and M2-macrophages *in situ* relies on immunohistochemical markers and there is a well-established panel of such antibodies for the distinctive detection of these cells in laboratory rodent tissue. However, the markers routinely used for the detection of rodent M1- and M2-macrophages cannot simply be transferred to other species. The nitric oxide and arginase metabolism of macrophages is a commonly used basis for the detection of rodent M1- and M2 macrophages. Consequently, arginase (Arg)1 and inducible nitric oxide synthase (iNOS) are the prototype markers to detect rodent M2 and M1-macrophages in tissue sections, respectively (71). However, there are considerable species differences, especially in the context of NO metabolism of macrophages and these well-established markers are not necessarily adequate to detect human and canine macrophages (73). Thus, development of

a panel of antibodies that enables the detection of canine M1- and M2-macrophages in tissue sections is highly needed. Recently, canine polarized macrophages have been characterized *in vitro*. Unstimulated (M0), M1- (GM-CSF, LPS, IFNγ-stimulated) and M2- (M-CSF, IL-4-stimulated)-polarized canine blood-derived macrophages showed distinct ultrastructural morphologies (73, 74). Interestingly, immunofluorescence using standard literature-based prototype-antibodies against CD16, CD32, iNOS, MHC class II for the detection of M1-macrophages and CD163, CD206, and arginase-1 for the detection of M2-macrophages demonstrated that solely CD206 was an appropriate marker that discriminated M2-macrophages from both other phenotypes (73, 74). In the same study, a global microarray analysis was performed and revealed changes in the transcriptome of polarized canine macrophages and similar to the results on the protein level, there were only minor overlaps in the gene sets of the dog compared to prototype markers of murine and human macrophages (73, 74). The transcriptome data of these canine macrophages might represent a basis for the subsequent development of immunohistochemical markers for the distinction between canine M1- and M2-macrophages, respectively, that are highly needed to classify the microglia/macrophage phenotype in the naturally injured canine spinal cord.

As mentioned above, data on naturally occurring chronic IVDE-induced lesions are extremely sparse. Glial scar formation is a common finding in experimental and naturally occurring human SCI. Similarly, extensive glial proliferation (gliosis) has been reported in dogs with chronic IVDE-induced SCI (27). Experimental SCI in dogs 12 weeks post injury is similarly characterized by severe astrogliosis as revealed by enhanced immunoreactivity for GFAP with spatial spread, mainly in the caudal direction (65).

Ependymal cells have recently been highlighted to participate in the cellular reaction following canine SCI. Due to its function as a source for neural precursors the spinal ependymal layer is believed to possess regenerative capacity and consequently represents another field of growing research, especially in the context of SCI (75, 76). Immunohistochemistry revealed increased numbers of GFAP-positive cells in acute IVDE-induced SCI in dogs with SCI at the lesion epicenter and additionally at sites proximal to the lesion center (76). It is proposed that the spinal ependymal layer may have the capacity of astrocytic differentiation during naturally occurring SCI in dogs. Besides enhanced GFAP-immunoreactivity of the spinal ependymal layer, acute IVDE-induced SCI is also characterized by altered E-cadherin expression patterns, indicating that a loss of cellular polarity could promote ependymal cell migration to the injury site (76).

The Need for Non-invasive Biomarkers in Canine IVDE Induced SCI

From a clinical perspective, a non-invasive biomarker that is able to predict clinical outcome, particularly in dogs with the most severe SCI, is highly needed. Multiple studies have assessed molecules in the CSF or serum, based on the hypothesis that the concentration of such metabolites is associated with injury severity and outcome, respectively. The results of these studies are also interesting from a pathological point of view, as clinically detected elevated levels of serum and CSF molecules may also be assessed in pathological analyses on post-mortem tissue such as immunohistochemistry and RT-qPCR methods. Vice versa, evidence from pathological studies may be extrapolated to clinical settings as enhanced expression of molecules detected via histopathological methods or molecular biology on post mortem tissue might develop new hypotheses in the search for novel biomarkers in a clinical setting. Thus, research on biomarkers for IVDE-induced SCI is an interesting field in which pathology and clinical neurology obviously benefit from each other.

Previously assessed candidate biomarkers in canine IVDE-induced SCI in CSF and serum, respectively, include metalloproteinases, neuronal/axonal cytoskeletal molecules, inflammatory cell counts, acute phase proteins, cytokines, arachidonic acid metabolites, and glial cytoskeletal components.

Enhanced MMP-9 activity in the CSF of the lumbar spine has been reported to indicate severe SCI with poor prognosis (77). Similarly, microtubule-associated protein tau, detected by ELISA in cisternal CSF, is associated with unsuccessful outcome in paraplegic dogs suffering from thoracolumbar or cervical IVDE (78). Serum levels of phosphorylated neurofilament heavy chain (pNF-H) are associated with severity of thoracolumbar IVDE and may predict an unfavorable prognosis (79). Increased cisternal CSF total nucleated cell count correlates with injury severity; however, the investigated CSF characteristics did not differentiate IVDE-induced SCI from other spinal cord diseases (80). The CSF concentrations of the acute phase proteins C-reactive protein and haptoglobin are associated with IVDE-induced injury severity; however, not correlating with 42 d motor outcome (81). The concentration of the arachidonic acid metabolites PLA2 and PGE2 in the CSF are higher in dogs with SCI compared to control dogs, while LCT4 concentration is lower in dogs with SCI than that in control dogs (82). Moreover, the concentration of PGE2 positively correlates with increased severity of SCI. Within the 1st days of IVDE-induced SCI, serum levels of GFAP and S100β rapidly rise, while pNF-H showed a later peak at 14 days post injury (83). Moreover, serum GFAP levels during the first 3 days can be used as a biomarker to predict recovery in severe SCI (83).

Matrix Metalloproteinases in Canine IVDE-Induced SCI

Matrix metalloproteinases (MMPs) have been shown to participate in the pathogenesis of canine IVDE-induced SCI in several studies. MMPs play a pleiotropic role in various neurologic diseases. They are involved in both axonal degeneration and regrowth and their signaling is crucial for postinjury reorganization and synaptic stabilization (84). Besides, MMPs are pivotal mediators of secondary injury and promote disruption of the blood-brain and blood-spinal cord barrier (84). In parallel, their signaling is necessary for healing processes such as angiogenesis, but on the other hand MMP expression promotes formation of a regeneration-inhibitory glial scar. Thus, MMPs play an important pathogenetic role during SCI.

Especially, the gelatinases MMP-2 and MMP-9 show time-dependent expression during SCI in both experimental and naturally occurring SCI (68). MMP-9 knock-out mice show less expression of regeneration inhibiting molecules when compared to wild-type mice with SCI (85). MMP-9 has thus gained much attention as a therapy target, as modulation of its expression might reduce glial scarring following SCI (85). In acute human SCI, MMP-9 is expressed by neutrophils in areas with hemorrhage as revealed by immunohistochemistry (68). Rapidly enhanced expression of MMP-9 in experimental contusion SCI in rodents is associated with an inappropriate function of the blood-spinal cord barrier as well as in inflammation and locomotor recovery (86). Compared to wild type mice, there is improved locomotor recovery in MMP-9 knock-out mice (86, 87).

Following experimental SCI in rodents, there is also upregulation of MMP-2. However, this upregulation is delayed when compared to MMP-9 (88, 89). In contrast to MMP-9, deficiency in the expression of MMP-2 is associated with impairment of locomotion in experimental SCI in mice (88). Thus, it is proposed that MMP-2 rather plays a beneficial role following SCI, in part by at regulating function that seems to target axonal plasticity and white matter sparing (88).

Dysregulation of the gelatinases MMP-2 and MMP-9 has been reported in previous studies on canine IVDE-induced SCI by means of RT-qPCR on spinal cord tissue of dogs (6, 34). While MMP-9 transcripts were up-regulated in dogs with acute SCI, MMP-2 exhibited a transient downregulation in the acute disease phase as compared to spinal cord tissue of neurologically healthy dogs (6, 34). Similarly, MMP-9 activity is increased in the CSF and serum of dogs with acute IVDD as revealed by zymography (90). Interestingly, elevated MMP-9 levels are associated with a poor outcome in dogs with IVDE-induced SCI (77, 90). Based on these observations and the hypothesis of a detrimental role of early MMP-9 signaling in dogs with IVDE-induced SCI, a randomized, blinded, placebo-controlled study was initiated to assess efficacy of the broad spectrum MMP-inhibitor GM6001 (91). In this study, dogs received GM6001 dissolved in dimethyl sulfoxide (DMSO), DMSO alone, or saline. GM6001 reduced serum MMP-9 activity compared to the other two groups (91). Interestingly, dogs treated with saline had significantly lower functional scores than dogs receiving DMSO or GM6001, demonstrating that there was no independent effect of GM6001 (91). The authors conclude that DMSO might have therapeutic effects in the acutely injured spinal cord. Similarly, recent clinical trials using the same agent, GM6001, demonstrated higher bladder compliance in dogs treated with GM6001 and DMSO as compared to controls (92). However, there were transient greater adverse events in GM6001-treated dogs compared to those treated with the vehicle control, and again, there was no difference in motor scores between dogs treated with GM6001 and DMSO vs. dogs treated with DMSO alone (92).

Cytokines in Canine IVDE-Induced SCI

The cerebrospinal fluid of dogs with acute, surgically treated, thoracolumbar IVDE has been assessed regarding expression of interleukin (IL)-2,−6,−7,−8,−10,−15,−18, granulocyte macrophage colony stimulating factor (GMCSF), interferon gamma (IFN-γ), keratinocyte chemoattractant-like (KC-like) protein, IFN-γ-inducible protein-10 (IP-10), monocyte chemotactic protein-1 (MCP-1), and tumor necrosis factor alpha (TNF-α) (93). Using a bioplex system, IL-8 concentration was found to be significantly higher in SCI cases than healthy controls and negatively correlated with the duration of SCI (93). Moreover, the MCP-1 concentration demonstrated to be negatively associated with 42-days post-injury outcome (93). Similarly, an early upregulation of pro-inflammatory cytokine mRNA (IL-6, IL-8 and TNF) has been noted in spinal cord tissue of dogs with acute IVDE-induced SCI (1–4 days post IVDE) using RT-qPCR of mRNA extracted from affected spinal cord tissue (66). IL-8 mRNA upregulation was also found in dogs with more than 4 days post IVDE suggesting a prolonged role of this pro-inflammatory cytokine in the pathogenesis of canine IVDE-induced SCI (66, 93). While IL-10 showed no differences in expression in either control dogs or dogs with SCI, expression of TGF-β showed up-regulation exclusively in spinal cord tissue of dogs with subacute SCI for more than 4 days. It is concluded that acute IVDE-induced SCI in dogs is dominated by a pro-inflammatory microenvironment (66, 72). The previous findings on cytokine expression in canine IVDE-induced SCI largely mirror findings in human cases of SCI and experimental SCI in rodents. For instance, several pro-inflammatory cytokines including IL-6 and IL-8 have also been reported to be upregulated in the CSF of humans affected by SCI (94). Interestingly, IL-8 levels within the CSF of people with SCI positively correlate with injury severity (94, 95). The delayed expression of TGF-β in dogs with IVDE-induced SCI is in concordance with experimental SCI in rats (96). TGF-β reduces the lesion volume and is associated with decreased numbers of macrophages in experimental rat SCI (96, 97)

Taken together, there is dysregulated cytokine expression with a lack or delay of anti-inflammatory cytokines and a dominance of pro-inflammatory cytokines during acute canine IVDE-induced SCI. These factors are thus believed to contribute to the lesion development and secondary injury processes in canine IVDE-induced SCI (6).

Further demonstrating that pro-inflammatory processes predominate in acute IVDE-induced SCI in dogs, there is significant dysregulation of acute phase proteins in the CSF of dogs with IVDE-induced SCI. Concentrations of C-reactive protein (CRP), haptoglobin (Hp), alpha-1-glycoprotein, and serum amyloid A were measured in a previous study (81). Interestingly, compared with healthy control dogs, Hp concentrations were higher in the CSF of affected dogs (81). Moreover, the authors reported that higher concentrations of CRP and Hp were associated the severity of injury; however, CSF APP concentrations and 42 d motor outcome did not reveal significant correlation (81).

ASCENDING AND DESCENDING MYELOMALACIA

A small but significant proportion of dogs affected by IVDE may develop one of the most disastrous complications, progressive

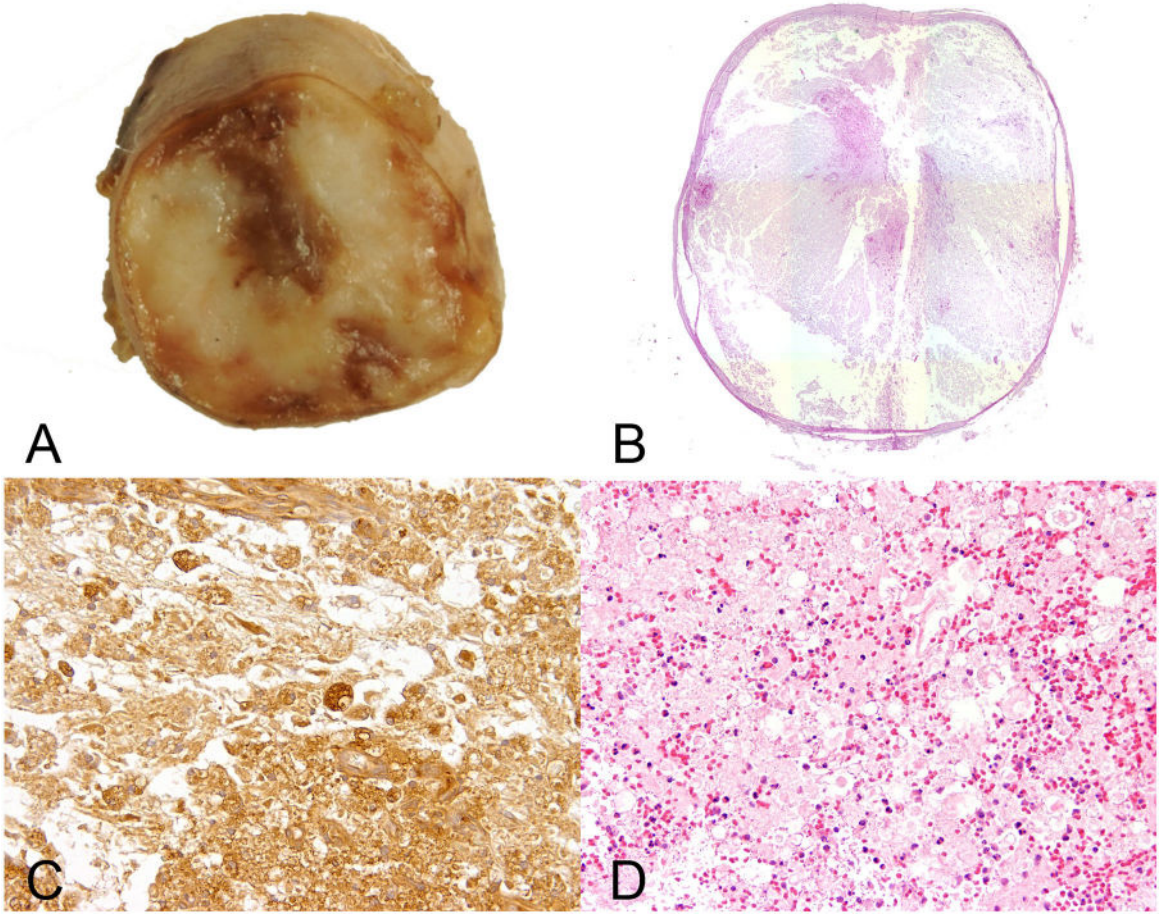

FIGURE 4 | Male 6 years old Yorkshire Terrier with progressive myelomalacia (PMM) following acute intervertebral disc extrusion. In PMM the shown lesions are not restricted to the initial site of spinal cord injury but extend several centimeters into cranial and caudal direction (ascending and descending malacia). **(A)** Gross picture of a transversal section of the formalin fixed spinal cord with complete disintegration of spinal cord neuroparenchyma and hemorrhage. **(B)** The HE stained overview of the transversal section shows polio- and leukomyelomalacia with complete loss of cellular details and loss of distinction between white and gray matter. **(C)** Multiple foamy microglia/macrophages labeled by the lectin of Bandeiraea simplicifolia 1 have infiltrated the lesion and remove cellular debris. 40x magnification. **(D)** There is severe extravasation of erythrocytes within the white and gray matter (hemorrhage), associated with infiltration of viable and degenerate neutrophils adjacent to areas of white matter damage with spheroids and myelin vacuolation. 10x magnification.

myelomalacia [PMM; (98)]. PMM is a unique entity, observed in both humans and dogs with severe injuries to the spinal cord and distinct from the initial SCI event. Though PMM can be observed following various forms of SCI including external trauma such as fractures, IVDE represents the most common initial type of SCI in dogs with subsequent PMM. The condition is characterized by progressive hemorrhagic necrosis of the spinal cord that diffusely ascends and/or descends over many spinal cord segments (99). PMM often develops early during the time course of IVDE and most dogs with PMM are euthanized within 3 days after onset of signs due to progressive respiratory paralysis (100). Considerable efforts have been undertaken to identify risk factors that are associated with this typically fatal condition. The prevalence of PMM is as low as 2% in the overall dog population with thoracolumbar IVDE, but severely elevated in paraplegic dogs that lack pain perception (101). In fact, the prevalence of PMM rises up to 10–12% in paraplegic dogs with absent deep nociception (21, 102). It appears that French Bulldogs may possess a breed predisposition to develop the devastating condition and the condition is more commonly diagnosed in dogs with extensive hyperintensity of the spinal cord on T2 weighed magnetic resonance imaging, dogs < 6 years of age, dogs with L5-6 disc herniations, and dogs with a rapidly progressive onset of clinical signs (103). A comprehensive recent study on 45 dogs with PMM identified IVDE at the lumbar intumescence as a strong risk factor that was associated with PMM (104). Moreover, surgery performed more than 12 h after loss of ambulation was also positively and treatment with corticosteroids was negatively associated with the development of PMM (104). Serum levels of GFAP have also been proposed as a biomarker for PMM. In

one study, of which seven dogs had detectable levels of serum GFAP, 6 developed PMM (105). Sensitivity and specificity of the GFAP to predict PMM were reported to be 75 and 97.7%, respectively (105).

While there have been some advances in the identification of risk factors associated with the disease, knowledge on the pathogenesis of PMM is strikingly sparse. In a histologic study, endothelin-1 (ET-1) immunoreactivity was noted in astrocytes, macrophages, and neurons, but only rarely in endothelial cells (106). At the lesion epicenter of spinal cord hemorrhage, ET-1 immunoreactivity was significantly higher in astrocytes and lower in neurons than in non-affected control dogs. Moreover, there was higher astrocytic and neuronal ET-1 immunoreactivity in spinal cord segments remote from the epicenter than in the center itself. The authors conclude that elevated ET-1 expression over multiple spinal cord segments after IVDE might be involved in the pathogenesis of PMM (106).

Histopathologic alterations of PMM are generally characterized by severe liquefactive necrosis of the spinal cord that extends over several segments (**Figure 4**). It is proposed that PMM represents a form of exuberant and dysregulated secondary injury response (99). The affected spinal cord tissue shows extensive hemorrhage and necrosis in both the gray and white matter with disruption of myelin and necrotic and chromatolytic neurons as well as prominent swollen endothelial cells lining remaining blood vessels (**Figure 4**) (99). Parenchymal and meningeal blood vessels have been reported to be necrotic with perivascular deposition of fibrin (98). Moreover, some vessels may contain thrombi (98). Severe lesions are characterized by an amorphous mixture of tissue debris, macrophages, and blood (106). Variably, intervertebral disc material may be detected in proximity to the meninges. The necrotic processes are accompanied by a reactive inflammatory response with neutrophils predominating due to the acute nature of the pathologic alterations. Moreover, lesions are characterized by infiltration of CD18-positive phagocytic microglia/macrophages (99). Hemorrhagic and necrotic debris may also be detected within the central canal in spinal cord segments remote from the lesion epicenter (107). In fact, intramedullary and subdural hemorrhages are significantly associated with the degree of white and gray matter damage, and the progressive nature of PMM is in part thought to be linked to high intramedullary pressure (107).

Oxidative stress is proposed to be involved in the pathogenesis of PMM, evidenced by the fact that PMM is associated with elevated levels of 8-isoprostanes and acrolein with concurrent reduction in endogenous anti-oxidation of glutathione in the CSF and urine of dogs suffering from the disease (99). The authors propose that the pathological condition of PMM represents an extreme case of secondary injury, in which the physiological defense systems are unable to terminate the progression of oxidative injury (99). Moreover, decreased anti-oxidation is associated with increased phagocytosis at the lesion center (99), suggesting that macrophages that occur during PMM might play a detrimental role in the process. The role of macrophages, and especially their potential polarization toward a neurotoxic phenotype, has not been addressed in detail so far and might represent a promising field for future studies. However, infiltration of neutrophils and macrophages has so far been regarded as a bystander phenomenon, that is not initiating the progression of PMM by itself (99).

CONCLUSIONS AND FUTURE DIRECTIONS

Conclusively, in parallel to the ongoing and growing focus on IVDE as a translational clinical animal model for SCI, there is a growing number of publications, investigating pathologic processes that occur following herniation of the intervertebral disc into the vertebral canal. Morphologic, axonal, glial and immune responses largely mirror changes seen in other animal models for SCI and the human disease; however, despite its high prevalence in veterinary clinical neurology, relatively little is known on the exact time course of secondary injury processes in the canine spinal cord affected by SCI. As immune processes, and here, especially the role of microglia/macrophages, is a rapidly growing field of experimental SCI research and a promising target for novel therapeutic approaches, further focus on the role of this cell population in this clinically relevant SCI model appears highly interesting for future histopathological and molecular studies. This will involve the establishment of immunohistochemical markers that are distinctive in the detection of different canine macrophage polarization stages (i.e., M1 and M2 macrophages). Moreover, integration of clinical and pathologic data in order to get detailed insights into the time course of immune responses and axonopathy, will provide an opportunity to improve the prediction of outcome and identify potential therapy targets. The same is true for the search of biomarkers, where close integrative collaboration of basic pathology science and research in the clinical setting will profit from each other in order to identify predictive factors influencing the course and outcome of IVDE-induced SCI.

The considerable paucity of pathologic data on chronic and advanced disease stages demonstrates the necessity of pathological investigations of such cases. This involves routine sampling of spinal cord tissue during necropsy, also from cases without an acute neurologic disease history from the side of pathology and rigorous communication of anamnestic data, as a considerable number of dogs might undergo necropsy due to other acute diseases where spinal cord alterations subsequent to IVDE years ago may be overseen.

Lastly, the pathogenesis of PMM as a devastating complication of canine SCI, is incompletely understood. There are surprisingly few studies on the pathology of this exacerbated form of secondary injury. The ongoing development of a panel of immunohistochemical methods for the detection of secondary injury processes such as immune and glial response and axonal damage including molecular alterations in research of canine IVDE induced SCI provides a promising tool for investigations on PMM. This will help to identify commonalities and differences and potentially contribute to the identification of predictive biomarkers and more detailed understanding of the pathogenesis of PMM.

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REFERENCES

1. Olby N. The pathogenesis and treatment of acute spinal cord injuries in dogs. *Vet Clin North Am Small Anim Pract.* (2010) 40:791–807. doi: 10.1016/j.cvsm.2010.05.007
2. Bray JP, Burbidge HM. The canine intervertebral disk. Part two: degenerative changes—nonchondrodystrophoid versus chondrodystrophoid disks. *J Am Anim Hosp Assoc.* (1998) 34:135–44. doi: 10.5326/15473317-34-2-135
3. Webb AA, Ngan S, Fowler JD. Spinal cord injury I: a synopsis of the basic science. *Can Vet J.* (2010) 51:485–92.
4. Coates JR. Intervertebral disk disease. *Vet Clin North Am Small Anim Pract.* (2000) 30:77–110. doi: 10.1016/S0195-5616(00)50004-7
5. Fluehmann G, Doherr MG, Jaggy A. Canine neurological diseases in a referral hospital population between 1989 and 2000 in Switzerland. *J Small Anim Pract.* (2006) 47:582–7. doi: 10.1111/j.1748-5827.2006.00106.x
6. Spitzbarth I. *A Morphological and Molecular Investigation on the Spatio-Temporal Development of Spinal Cord Lesions Following Spinal Injury in Dogs*. Zugl: Hannover, Tierärztl (2012). p. 88.
7. Levine JM, Levine GJ, Porter BF, Topp K, Noble-Haeusslein LJ. Naturally occurring disk herniation in dogs: an opportunity for pre-clinical spinal cord injury research. *J Neurotrauma.* (2011) 28:675–88. doi: 10.1089/neu.2010.1645
8. Jeffery ND, Smith PM, Lakatos A, Ibanez C, Ito D, Franklin RJ. Clinical canine spinal cord injury provides an opportunity to examine the issues in translating laboratory techniques into practical therapy. *Spinal Cord.* (2006) 44:584–93. doi: 10.1038/sj.sc.3101912
9. Smith PM, Jeffery ND. Histological and ultrastructural analysis of white matter damage after naturally-occurring spinal cord injury. *Brain Pathol.* (2006) 16:99–109. doi: 10.1111/j.1750-3639.2006.00001.x
10. Boekhoff TM, Flieshardt C, Ensinger E-M, Fork M, Kramer S, Tipold A. Quantitative magnetic resonance imaging characteristics: evaluation of prognostic value in the dog as a translational model for spinal cord injury. *J Spinal Disord Tech.* (2012) 25:E81–7. doi: 10.1097/BSD.0b013e31823f2f55
11. Boekhoff TM, Ensinger E-M, Carlson R, Bock P, Baumgärtner W, Rohn K, et al. Microglial contribution to secondary injury evaluated in a large animal model of human spinal cord trauma. *J Neurotrauma.* (2012) 29:1000–11. doi: 10.1089/neu.2011.1821
12. Kwon BK, Hillyer J, Tetzlaff W. Translational research in spinal cord injury: a survey of opinion from the SCI community. *J Neurotrauma.* (2010) 27:21–33. doi: 10.1089/neu.2009.1048
13. Hansen HJ. A pathologic-anatomical study on disc degeneration in dog, with special reference to the so-called enchondrosis intervertebralis. *Acta Orthop Scand Suppl.* (1952) 11:1–117. doi: 10.3109/ort.1952.23.suppl-11.01
14. Jeffery ND, Levine JM, Olby NJ, Stein VM. Intervertebral disk degeneration in dogs: consequences, diagnosis, treatment, and future directions. *J Vet Intern Med.* (2013) 27:1318–33. doi: 10.1111/jvim.12183
15. Mayousse V. Prevalence of neurological disorders in French bulldog: a retrospective study of 343 cases (2002–2016). *BMC Vet Res.* (2017) 13:212. doi: 10.1186/s12917-017-1132-2
16. Brown EA, Dickinson PJ, Mansour T, Sturges BK, Aguilar M, Young AE, et al. FGF4 retrogene on CFA12 is responsible for chondrodystrophy and intervertebral disc disease in dogs. *Proc Natl Acad Sci USA.* (2017) 114:11476–81. doi: 10.1073/pnas.1709082114

17. Simpson ST. Intervertebral disc disease. *Vet Clin North Am Small Anim Pract.* (1992) 22:889–97. doi: 10.1016/S0195-5616(92)50081-X
18. Stigen O, Ciasca T, Kolbjornsen O. Calcification of extruded intervertebral discs in dachshunds: a radiographic, computed tomographic and histopathological study of 25 cases. *Acta Vet Scand.* (2019) 61:13. doi: 10.1186/s13028-019-0448-2
19. Bergknut N, Meij BP, Hagman R, Nies KS de, Rutges JP, Smolders LA, et al. Intervertebral disc disease in dogs - part 1: a new histological grading scheme for classification of intervertebral disc degeneration in dogs. *Vet J.* (2013) 195:156–63. doi: 10.1016/j.tvjl.2012.05.027
20. Hansen T, Smolders LA, Tryfonidou MA, Meij BP, Vernooij JC, Bergknut N, et al. The myth of fibroid degeneration in the canine intervertebral disc: a histopathological comparison of intervertebral disc degeneration in chondrodystrophic and nonchondrodystrophic dogs. *Vet Pathol.* (2017) 54:945–52. doi: 10.1177/0300985817726834
21. Olby N, Levine J, Harris T, Muñana K, Skeen T, Sharp N. Long-term functional outcome of dogs with severe injuries of the thoracolumbar spinal cord: 87 cases (1996-2001). *J Am Vet Med Assoc.* (2003) 222:762–9. doi: 10.2460/javma.2003.222.762
22. Langerhuus L, Miles J. Proportion recovery and times to ambulation for non-ambulatory dogs with thoracolumbar disc extrusions treated with hemilaminectomy or conservative treatment: a systematic review and meta-analysis of case-series studies. *Vet J.* (2017) 220:7–16. doi: 10.1016/j.tvjl.2016.12.008
23. Brisson BA. Intervertebral disc disease in dogs. *Vet Clin North Am Small Anim Pract.* (2010) 40:829–58. doi: 10.1016/j.cvsm.2010.06.001
24. Decker S de, Fenn J. Acute herniation of nondegenerate nucleus pulposus: acute noncompressive nucleus pulposus extrusion and compressive hydrated nucleus pulposus extrusion. *Vet Clin North Am Small Anim Pract.* (2018) 48:95–109. doi: 10.1016/j.cvsm.2017.08.004
25. Monchaux M, Forterre S, Spreng D, Karol A, Forterre F, Wuertz-Kozak K. Inflammatory processes associated with canine intervertebral disc herniation. *Front Immunol.* (2017) 8:1681. doi: 10.3389/fimmu.2017.01681
26. Griffiths IR. Spinal cord injuries: a pathological study of naturally occurring lesions in the dog and cat. *J Comp Pathol.* (1978) 88:303–15. doi: 10.1016/0021-9975(78)90033-6
27. Alisauskaite N, Spitzbarth I, Baumgärtner W, Dziallas P, Kramer S, Dening R, et al. Chronic post-traumatic intramedullary lesions in dogs, a translational model. *PLoS ONE.* (2017) 12:e0187746. doi: 10.1371/journal.pone.0187746
28. Henke D, Vandevelde M, Doherr MG, Stockli M, Forterre F. Correlations between severity of clinical signs and histopathological changes in 60 dogs with spinal cord injury associated with acute thoracolumbar intervertebral disc disease. *Vet J.* (2013) 198:70–5. doi: 10.1016/j.tvjl.2013.04.003
29. Webb AA, Ngan S, Fowler D. Spinal cord injury II: Prognostic indicators, standards of care, and clinical trials. *Can Vet J.* (2010) 51:598–604.
30. Jeffery ND, Lakatos A, Franklin RJ. Autologous olfactory glial cell transplantation is reliable and safe in naturally occurring canine spinal cord injury. *J Neurotrauma.* (2005) 22:1282–93. doi: 10.1089/neu.2005.22.1282
31. Gensel JC, Donnelly DJ, Popovich PG. Spinal cord injury therapies in humans: an overview of current clinical trials and their potential effects on intrinsic CNS macrophages. *Expert Opin Ther Targets.* (2011) 15:505–18. doi: 10.1517/14728222.2011.553605
32. Wewetzer K, Radtke C, Kocsis J, Baumgärtner W. Species-specific control of cellular proliferation and the impact of large animal models for the use of olfactory ensheathing cells and Schwann cells in spinal cord repair. *Exp Neurol.* (2011) 229:80–7. doi: 10.1016/j.expneurol.2010.08.029
33. Egawa N, Lok J, Washida K, Arai K. Mechanisms of axonal damage and repair after central nervous system injury. *Transl Stroke Res.* (2017) 8:14–21. doi: 10.1007/s12975-016-0495-1
34. Bock P, Spitzbarth I, Haist V, Stein VM, Tipold A, Puff C, et al. Spatio-temporal development of axonopathy in canine intervertebral disc disease as a translational large animal model for nonexperimental spinal cord injury. *Brain Pathol.* (2013) 23:82–99. doi: 10.1111/j.1750-3639.2012.00617.x
35. Anthes DL, Theriault E, Tator CH. Characterization of axonal ultrastructural pathology following experimental spinal cord compression injury. *Brain Res.* (1995) 702:1–16. doi: 10.1016/0006-8993(95)01028-6
36. Bresnahan JC. An electron-microscopic analysis of axonal alterations following blunt contusion of the spinal cord of the rhesus monkey (Macaca mulatta). *J Neurol Sci.* (1978) 37:59–82. doi: 10.1016/0022-510X(78)90228-9
37. Lampert PW. A comparative electron microscopic study of reactive, degenerating, regenerating, and dystrophic axons. *J Neuropathol Exp Neurol.* (1967) 26:345–68. doi: 10.1097/00005072-196707000-00001
38. Rosenberg LJ, Wrathall JR. Quantitative analysis of acute axonal pathology in experimental spinal cord contusion. *J Neurotrauma.* (1997) 14:823–38. doi: 10.1089/neu.1997.14.823
39. Gomes-Leal W, Corkill DJ, Picanco-Diniz CW. Systematic analysis of axonal damage and inflammatory response in different white matter tracts of acutely injured rat spinal cord. *Brain Res.* (2005) 1066:57–70. doi: 10.1016/j.brainres.2005.10.069
40. Coleman M. Axon degeneration mechanisms: commonality amid diversity. *Nat Rev Neurosci.* (2005) 6:889–98. doi: 10.1038/nrn1788
41. Purdy PD, Duong RT, White CL, Baer DL, Reichard RR, Pride GL, et al. Percutaneous translumbar spinal cord compression injury in a dog model that uses angioplasty balloons: MR imaging and histopathologic findings. *Am J Neuroradiol.* (2003) 24:177–84.
42. Purdy PD, White CL, Baer DL, Frawley WH, Reichard RR, Pride GL, et al. Percutaneous translumbar spinal cord compression injury in dogs from an angioplasty balloon: MR and histopathologic changes with balloon sizes and compression times. *AJNR Am J Neuroradiol.* (2004) 25:1435–42.
43. Li GL, Farooque M, Holtz A, Olsson Y. Changes of beta-amyloid precursor protein after compression trauma to the spinal cord: an experimental study in the rat using immunohistochemistry. *J Neurotrauma.* (1995) 12:269–77. doi: 10.1089/neu.1995.12.269
44. Ahlgren S, Li GL, Olsson Y. Accumulation of beta-amyloid precursor protein and ubiquitin in axons after spinal cord trauma in humans: immunohistochemical observations on autopsy material. *Acta Neuropathol.* (1996) 92:49–55. doi: 10.1007/s004010050488
45. Cornish R, Blumbergs PC, Manavis J, Scott G, Jones NR, Reilly PL. Topography and severity of axonal injury in human spinal cord trauma using amyloid precursor protein as a marker of axonal injury. *Spine.* (2000) 25:1227–33. doi: 10.1097/00007632-200005150-00005
46. Stone JR, Singleton RH, Povlishock JT. Intra-axonal neurofilament compaction does not evoke local axonal swelling in all traumatically injured axons. *Exp Neurol.* (2001) 172:320–31. doi: 10.1006/exnr.2001.7818
47. Tsunoda I, Kuang L-Q, Libbey JE, Fujinami RS. Axonal injury heralds virus-induced demyelination. *Am J Pathol.* (2003) 162:1259–69. doi: 10.1016/S0002-9440(10)63922-3
48. Kreutzer M, Seehusen F, Kreutzer R, Pringproa K, Kummerfeld M, Claus P, et al. Axonopathy is associated with complex axonal transport defects in a model of multiple sclerosis. *Brain Pathol.* (2012) 22:454–71. doi: 10.1111/j.1750-3639.2011.00541.x
49. Seehusen F, Baumgärtner W. Axonal pathology and loss precede demyelination and accompany chronic lesions in a spontaneously occurring animal model of multiple sclerosis. *Brain Pathol.* (2010) 20:551–9. doi: 10.1111/j.1750-3639.2009.00332.x
50. Spitzbarth I, Lempp C, Kegler K, Ulrich R, Kalkuhl A, Deschl U, et al. Immunohistochemical and transcriptome analyses indicate complex breakdown of axonal transport mechanisms in canine distemper leukoencephalitis. *Brain Behav.* (2016) 6:e00472. doi: 10.1002/brb3.472
51. Schwab ME, Bartholdi D. Degeneration and regeneration of axons in the lesioned spinal cord. *Physiol Rev.* (1996) 76:319–70. doi: 10.1152/physrev.1996.76.2.319
52. Kerschensteiner M, Schwab ME, Lichtman JW, Misgeld T. *In vivo* imaging of axonal degeneration and regeneration in the injured spinal cord. *Nat Med.* (2005) 11:572–7. doi: 10.1038/nm1229
53. Bareyre FM, Haudenschild B, Schwab ME. Long-lasting sprouting and gene expression changes induced by the monoclonal antibody IN-1 in the adult spinal cord. *J Neurosci.* (2002) 22:7097–110. doi: 10.1523/JNEUROSCI.22-16-07097.2002
54. GrandPre T, Li S, Strittmatter SM. Nogo-66 receptor antagonist peptide promotes axonal regeneration. *Nature.* (2002) 417:547–51. doi: 10.1038/417547a
55. Tobias CA, Shumsky JS, Shibata M, Tuszynski MH, Fischer I, Tessler A, et al. Delayed grafting of BDNF and NT-3 producing fibroblasts into the

injured spinal cord stimulates sprouting, partially rescues axotomized red nucleus neurons from loss and atrophy, and provides limited regeneration. *Exp Neurol.* (2003) 184:97–113. doi: 10.1016/S0014-4886(03)00394-7
56. Kamada T, Koda M, Dezawa M, Anahara R, Toyama Y, Yoshinaga K, et al. Transplantation of human bone marrow stromal cell-derived schwann cells reduces cystic cavity and promotes functional recovery after contusion injury of adult rat spinal cord. *Neuropathology.* (2011) 31:48–58. doi: 10.1111/j.1440-1789.2010.01130.x
57. Andrews MR, Stelzner DJ. Evaluation of olfactory ensheathing and schwann cells after implantation into a dorsal injury of adult rat spinal cord. *J Neurotrauma.* (2007) 24:1773–92. doi: 10.1089/neu.2007.0353
58. Guerrero AR, Uchida K, Nakajima H, Watanabe S, Nakamura M, Johnson WE, et al. Blockade of interleukin-6 signaling inhibits the classic pathway and promotes an alternative pathway of macrophage activation after spinal cord injury in mice. *J Neuroinflammation.* (2012) 9:40. doi: 10.1186/1742-2094-9-40
59. Buss A, Brook GA, Kakulas B, Martin D, Franzen R, Schoenen J, et al. Gradual loss of myelin and formation of an astrocytic scar during wallerian degeneration in the human spinal cord. *Brain.* (2004) 127:34–44. doi: 10.1093/brain/awh001
60. Buss A, Pech K, Merkler D, Kakulas BA, Martin D, Schoenen J, et al. Sequential loss of myelin proteins during wallerian degeneration in the human spinal cord. *Brain.* (2005) 128:356–64. doi: 10.1093/brain/awh355
61. Tsunoda I, Fujinami RS. Inside-out versus outside-in models for virus induced demyelination: axonal damage triggering demyelination. *Springer Semin Immunopathol.* (2002) 24:105–25. doi: 10.1007/s00281-002-0105-z
62. Levine GJ, Levine JM, Witsberger TH, Kerwin SC, Russell KE, Suchodolski J, et al. Cerebrospinal fluid myelin basic protein as a prognostic biomarker in dogs with thoracolumbar intervertebral disk herniation. *J Vet Intern Med.* (2010) 24:890–6. doi: 10.1111/j.1939-1676.2010.0531.x
63. Lim J-H, Muguet-Chanoit AC, Smith DT, Laber E, Olby NJ. Potassium channel antagonists 4-aminopyridine and the T-butyl carbamate derivative of 4-aminopyridine improve hind limb function in chronically non-ambulatory dogs; a blinded, placebo-controlled trial. *PLoS ONE.* (2014) 9:e116139. doi: 10.1371/journal.pone.0116139
64. Moore SA, Granger N, Olby NJ, Spitzbarth I, Jeffery ND, Tipold A, et al. Targeting translational successes through CANSORT-SCI: using pet dogs to identify effective treatments for spinal cord injury. *J Neurotrauma.* (2017) 34:2007–18. doi: 10.1089/neu.2016.4745
65. Liu C-B, Yang D-G, Zhang X, Zhang W-H, Li D-P, Zhang C, et al. Degeneration of white matter and gray matter revealed by diffusion tensor imaging and pathological mechanism after spinal cord injury in canine. *CNS Neurosci Ther.* (2019) 25:261–72. doi: 10.1111/cns.13044
66. Spitzbarth I, Bock P, Haist V, Stein VM, Tipold A, Wewetzer K, et al. Prominent microglial activation in the early proinflammatory immune response in naturally occurring canine spinal cord injury. *J Neuropathol Exp Neurol.* (2011) 70:703–14. doi: 10.1097/NEN.0b013e3182270f8e
67. Schmitt AB, Buss A, Breuer S, Brook GA, Pech K, Martin D, et al. Major histocompatibility complex class II expression by activated microglia caudal to lesions of descending tracts in the human spinal cord is not associated with a T cell response. *Acta Neuropathol.* (2000) 100:528–36. doi: 10.1007/s004010000221
68. Fleming JC, Norenberg MD, Ramsay DA, Dekaban GA, Marcillo AE, Saenz AD, et al. The cellular inflammatory response in human spinal cords after injury. *Brain.* (2006) 129:3249–69. doi: 10.1093/brain/awl296
69. Stein VM, Baumgärtner W, Kreienbrock L, Zurbriggen A, Vandevelde M, Tipold A. Canine microglial cells: stereotypy in immunophenotype and specificity in function? *Vet Immunol Immunopathol.* (2006) 113:277–87. doi: 10.1016/j.vetimm.2006.05.011
70. Ensinger E-M, Boekhoff TM, Carlson R, Beineke A, Rohn K, Tipold A, et al. Regional topographical differences of canine microglial immunophenotype and function in the healthy spinal cord. *J Neuroimmunol.* (2010) 227:144–52. doi: 10.1016/j.jneuroim.2010.07.017
71. Kigerl KA, Gensel JC, Ankeny DP, Alexander JK, Donnelly DJ, Popovich PG. Identification of two distinct macrophage subsets with divergent effects causing either neurotoxicity or regeneration in the injured mouse spinal cord. *J Neurosci.* (2009) 29:13435–44. doi: 10.1523/JNEUROSCI.3257-09.2009
72. Spitzbarth I, Baumgärtner W, Beineke A. The role of pro- and anti-inflammatory cytokines in the pathogenesis of spontaneous canine CNS diseases. *Vet Immunol Immunopathol.* (2012) 147:6–24. doi: 10.1016/j.vetimm.2012.04.005
73. Heinrich F, Lehmbecker A, Raddatz BB, Kegler K, Tipold A, Stein VM, et al. Morphologic, phenotypic, and transcriptomic characterization of classically and alternatively activated canine blood-derived macrophages *in vitro*. *PLoS ONE.* (2017) 12:e0183572. doi: 10.1371/journal.pone.0183572
74. Heinrich F. *In vitro Phenotypical and Transcriptomic Characterization of Canine Macrophages*. (Dissertation). Hannover.
75. Moore SA. The spinal ependymal layer in health and disease. *Vet Pathol.* (2016) 53:746–53. doi: 10.1177/0300985815618438
76. Moore SA, Oglesbee MJ. Spinal cord ependymal responses to naturally occurring traumatic spinal cord injury in dogs. *Vet Pathol.* (2015) 52:1108–17. doi: 10.1177/0300985814560235
77. Nagano S, Kim SH, Tokunaga S, Arai K, Fujiki M, Misumi K. Matrix metalloprotease-9 activity in the cerebrospinal fluid and spinal injury severity in dogs with intervertebral disc herniation. *Res Vet Sci.* (2011) 91:482–5. doi: 10.1016/j.rvsc.2010.09.009
78. Roerig A, Carlson R, Tipold A, Stein VM. Cerebrospinal fluid tau protein as a biomarker for severity of spinal cord injury in dogs with intervertebral disc herniation. *Vet J.* (2013) 197:253–8. doi: 10.1016/j.tvjl.2013.02.005
79. Nishida H, Nakayama M, Tanaka H, Kamishina H, Izawa T, Hatoya S, et al. Evaluation of serum phosphorylated neurofilament subunit NF-H as a prognostic biomarker in dogs with thoracolumbar intervertebral disc herniation. *Vet Surg.* (2014) 43:289–93. doi: 10.1111/j.1532-950X.2014.12144.x
80. Levine GJ, Cook JR, Kerwin SC, Mankin J, Griffin JF, Fosgate GT, et al. Relationships between cerebrospinal fluid characteristics, injury severity, and functional outcome in dogs with and without intervertebral disk herniation. *Vet Clin Pathol.* (2014) 43:437–46. doi: 10.1111/vcp.12165
81. Anderson KM, Welsh CJ, Young C, Levine GJ, Kerwin SC, Boudreau CE, et al. Acute phase proteins in cerebrospinal fluid from dogs with naturally-occurring spinal cord injury. *J Neurotrauma.* (2015) 32:1658–65. doi: 10.1089/neu.2015.3895
82. Russell RL, Levine JM, Jeffery ND, Young C, Mondragon A, Lee B, et al. Arachidonic acid pathway alterations in cerebrospinal fluid of dogs with naturally occurring spinal cord injury. *BMC Neurosci.* (2016) 17:31. doi: 10.1186/s12868-016-0269-4
83. Olby NJ, Lim J-H, Wagner N, Zidan N, Early PJ, Mariani CL, et al. Time course and prognostic value of serum GFAP, pNFH, and S100beta concentrations in dogs with complete spinal cord injury because of intervertebral disc extrusion. *J Vet Intern Med.* (2019) 33:726–34. doi: 10.1111/jvim.15439
84. Trivedi A, Noble-Haeusslein LJ, Levine JM, Santucci AD, Reeves TM, Phillips LL. Matrix metalloproteinase signals following neurotrauma are right on cue. *Cell Mol Life Sci.* (2019) 76:3141–56. doi: 10.1007/s00018-019-03176-4
85. Hsu J-YC, Bourguignon LY, Adams CM, Peyrollier K, Zhang H, Fandel T, et al. Matrix metalloproteinase-9 facilitates glial scar formation in the injured spinal cord. *J Neurosci.* (2008) 28:13467–77. doi: 10.1523/JNEUROSCI.2287-08.2008
86. Noble LJ, Donovan F, Igarashi T, Goussev S, Werb Z. Matrix metalloproteinases limit functional recovery after spinal cord injury by modulation of early vascular events. *J Neurosci.* (2002) 22:7526–35. doi: 10.1523/JNEUROSCI.22-17-07526.2002
87. Busch SA, Horn KP, Silver DJ, Silver J. Overcoming macrophage-mediated axonal dieback following CNS injury. *J Neurosci.* (2009) 29:9967–76. doi: 10.1523/JNEUROSCI.1151-09.2009
88. Hsu J-YC, McKeon R, Goussev S, Werb Z, Lee J-U, Trivedi A, et al. Matrix metalloproteinase-2 facilitates wound healing events that promote functional recovery after spinal cord injury. *J Neurosci.* (2006) 26:9841–50. doi: 10.1523/JNEUROSCI.1993-06.2006
89. Zhang H, Chang M, Hansen CN, Basso DM, Noble-Haeusslein LJ. Role of matrix metalloproteinases and therapeutic benefits of their inhibition in spinal cord injury. *Neurotherapeutics.* (2011) 8:206–20. doi: 10.1007/s13311-011-0038-0
90. Levine JM, Ruaux CG, Bergman RL, Coates JR, Steiner JM, Williams DA. Matrix metalloproteinase-9 activity in the cerebrospinal fluid and serum of

dogs with acute spinal cord trauma from intervertebral disk disease. *Am J Vet Res.* (2006) 67:283–7. doi: 10.2460/ajvr.67.2.283
91. Levine JM, Cohen ND, Heller M, Fajt VR, Levine GJ, Kerwin SC, et al. Efficacy of a metalloproteinase inhibitor in spinal cord injured dogs. *PLoS ONE.* (2014) 9:e96408. doi: 10.1371/journal.pone.0096408
92. Levine JM, Cohen ND, Fandel TM, Levine GJ, Mankin J, Griffin JF, et al. Early blockade of matrix metalloproteinases in spinal-cord-injured dogs results in a long-term increase in bladder compliance. *J Neurotrauma.* (2017) 34:2656–67. doi: 10.1089/neu.2017.5001
93. Taylor AR, Welsh CJ, Young C, Spoor E, Kerwin SC, Griffin JF, et al. Cerebrospinal fluid inflammatory cytokines and chemokines in naturally occurring canine spinal cord injury. *J Neurotrauma.* (2014) 31:1561–9. doi: 10.1089/neu.2014.3405
94. Kwon BK, Stammers AM, Belanger LM, Bernardo A, Chan D, Bishop CM, et al. Cerebrospinal fluid inflammatory cytokines and biomarkers of injury severity in acute human spinal cord injury. *J Neurotrauma.* (2010) 27:669–82. doi: 10.1089/neu.2009.1080
95. Stammers AT, Liu J, Kwon BK. Expression of inflammatory cytokines following acute spinal cord injury in a rodent model. *J Neurosci Res.* (2012) 90:782–90. doi: 10.1002/jnr.22820
96. Tyor WR, Avgeropoulos N, Ohlandt G, Hogan EL. Treatment of spinal cord impact injury in the rat with transforming growth factor-beta. *J Neurol Sci.* (2002) 200:33–41. doi: 10.1016/S0022-510X(02)00113-2
97. Genovese T, Esposito E, Mazzon E, Di Paola R, Caminiti R, Bramanti P, et al. Absence of endogenous interleukin-10 enhances secondary inflammatory process after spinal cord compression injury in mice. *J Neurochem.* (2009) 108:1360–72. doi: 10.1111/j.1471-4159.2009.05899.x
98. Griffiths IR. The extensive myelopathy of intervertebral disc protrusions in dogs ('the ascending syndrome'). *J Small Anim Pract.* (1972) 13:425–38. doi: 10.1111/j.1748-5827.1972.tb06870.x
99. Marquis A, Packer RA, Borgens RB, Duerstock BS. Increase in oxidative stress biomarkers in dogs with ascending-descending myelomalacia following spinal cord injury. *J Neurol Sci.* (2015) 353:63–9. doi: 10.1016/j.jns.2015.04.003
100. Castel A, Olby NJ, Mariani CL, Munana KR, Early PJ. Clinical characteristics of dogs with progressive myelomalacia following acute intervertebral disc extrusion. *J Vet Intern Med.* (2017) 31:1782–9. doi: 10.1111/jvim.14829
101. Balducci F, Canal S, Contiero B, Bernardini M. Prevalence and risk factors for presumptive ascending/descending myelomalacia in dogs after thoracolumbar intervertebral disk herniation. *J Vet Intern Med.* (2017) 31:498–504. doi: 10.1111/jvim.14656
102. Aikawa T, Fujita H, Kanazono S, Shibata M, Yoshigae Y. Long-term neurologic outcome of hemilaminectomy and disk fenestration for treatment of dogs with thoracolumbar intervertebral disk herniation: 831 cases (2000-2007). *J Am Vet Med Assoc.* (2012) 241:1617–26. doi: 10.2460/javma.241.12.1617
103. Aikawa T, Shibata M, Asano M, Hara Y, Tagawa M, Orima H. A comparison of thoracolumbar intervertebral disc extrusion in French bulldogs and dachshunds and association with congenital vertebral anomalies. *Vet Surg.* (2014) 43:301–7. doi: 10.1111/j.1532-950X.2014.12102.x
104. Castel A, Olby NJ, Ru H, Mariani CL, Munana KR, Early PJ. Risk factors associated with progressive myelomalacia in dogs with complete sensorimotor loss following intervertebral disc extrusion: a retrospective case-control study. *BMC Vet Res.* (2019) 15:433. doi: 10.1186/s12917-019-2186-0
105. Sato Y, Shimamura S, Mashita T, Kobayashi S, Okamura Y, Katayama M, et al. Serum glial fibrillary acidic protein as a diagnostic biomarker in dogs with progressive myelomalacia. *J Vet Med Sci.* (2013) 75:949–53. doi: 10.1292/jvms.12-0483
106. Mayer D, Oevermann A, Seuberlich T, Vandevelde M, Casanova-Nakayama A, Selimovic-Hamza S, et al. Endothelin-1 immunoreactivity and its association with intramedullary hemorrhage and myelomalacia in naturally occurring disk extrusion in dogs. *J Vet Intern Med.* (2016) 30:1099–111. doi: 10.1111/jvim.14364
107. Henke D, Gorgas D, Doherr MG, Howard J, Forterre F, Vandevelde M. Longitudinal extension of myelomalacia by intramedullary and subdural hemorrhage in a canine model of spinal cord injury. *Spine J.* (2016) 16:82–90. doi: 10.1016/j.spinee.2015.09.018

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REVIEW
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Bladder and Bowel Management in Dogs With Spinal Cord Injury

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Spinal cord injury in companion dogs can lead to urinary and fecal incontinence or retention, depending on the severity, and localization of the lesion along the canine nervous system. The bladder and gastrointestinal dysfunction caused by lesions of the autonomic system can be difficult to recognize, interpret and are easily overlooked. Nevertheless, it is crucial to maintain a high degree of awareness of the impact of micturition and defecation disturbances on the animal's condition, welfare and on the owner. The management of these disabilities is all the more challenging that the autonomic nervous system physiology is a complex topic. In this review, we propose to briefly remind the reader the physiology of micturition and defecation in dogs. We then present the bladder and gastrointestinal clinical signs associated with sacral lesions (i.e., the L7-S3 spinal cord segments and nerves) and supra-sacral lesions (i.e., cranial to the L7 spinal cord segment), largely in the context of intervertebral disc herniation. We summarize what is known about the natural recovery of urinary and fecal continence in dogs after spinal cord injury. In particular we review the incidence of urinary tract infection after injury. We finally explore the past and recent literature describing management of urinary and fecal dysfunction in the acute and chronic phase of spinal cord injury. This comprises medical therapies but importantly a number of surgical options, some known for decades such as sacral nerve stimulation, that might spark some interest in the field of spinal cord injury in companion dogs.

Keywords: bladder, urinary and fecal incontinence, spinal cord injury, autonomic, dysfunction, canine, dog, sacral implant

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INTRODUCTION

Spinal cord injury can cause irreversible locomotor and autonomic dysfunction including urinary and fecal incontinence. The two functions of the bladder and bowel are *storage* and *voiding*. After severe spinal cord injury, both of these are impaired as a result of altered sensation and altered voluntary control amongst others. People with thoracic and thoracolumbar spinal cord injury value restoration of bladder and bowel control by far over regaining walking (1, 2), which signifies the great social and psychological consequences of urinary and fecal incontinence. In dogs, the impact of urinary and fecal incontinence on quality of life is not clear but lack of recovery of continence affects owners and increases the time and demands required to care for dogs (3). It seems that the focus of owners is regaining locomotion, and that urinary or fecal incontinence,

whether persistent or temporary, are forgotten or "managed" dysfunctions that will either pass or will need to be accepted as permanent. Further, the problem for owners caring for a dog with urinary and fecal incontinence varies greatly depending on the localization of the inciting injury: (i) with low lumbar lesions, the incontinence is often permanent and consistent, therefore more problematic, due to loss of sphincter tone, which likely results in a greater demand for euthanasia; (ii) with "supra-sacral" lesions, there is retention urinary incontinence characterized by preserved or increased sphincter tone and involuntary bladder contractions (i.e., reflex incontinence) and fecal incontinence characterized by inability to control defecation once the defecation reflex has been triggered; therefore the owner's challenges are typically those of caring for a dog where manual emptying of the bladder is difficult, there is regular emission of small volumes of urine and there is sudden urge fecal incontinence (4). The treatment goals to address these difficulties are very different, sometimes requiring stimulating a function sometimes requiring blocking a function, and this adds to the complexity of managing bladder and bowel dysfunction in animals.

In this paper, we will review briefly the physiology of micturition and defecation, the characteristics of urinary and fecal incontinence after spinal cord injury as well as their recovery, and we will assess the extent to which these events occur in spinal cord-injured dogs. We will provide state of the art management recommendations for bladder and bowel dysfunction in the acute (from the day of injury to approximately a month) and chronic (the weeks to months after injury) phases of spinal cord injury in dogs and discuss these related to lesion level. We will also identify aspects of autonomic dysfunction that are currently unclear in spinal cord injured dogs and provide opportunities for further studies. This could be critical to better understanding and manipulating these systems and therefore providing avenues for development of therapeutic strategies for dogs and humans via translational mechanisms.

PHYSIOLOGY OF MICTURITION

Here we provide a brief overview of the physiology of micturition; for a detailed review on anatomy and physiology the reader is referred elsewhere (5–8).

Regulation of urination involves a complex neural control system in the brain, spinal cord, and peripheral autonomic ganglia that coordinates the activity of smooth and striated muscles of bladder and urethral outlet. Spinal storage mechanisms are regulated by circuitry in the rostral brain stem and initiate reflex voiding. Input from the forebrain triggers voluntary voiding by modulating the brainstem circuitry.

The switch between storage and voiding is mediated by a long-loop spino-bulbo-spinal voiding reflex which has its rostral terminus in the brainstem. During urine storage, as the bladder fills, bladder afferent signals increase in strength until they exceed a certain threshold in the brainstem. In the absence of any controlling influences, the pontine micturition center is activated, the urethral sphincter relaxes, the bladder contracts, and reflex voiding occurs (8). When the bladder is empty, urine storage resumes. Cortical modulation of this circuitry underlies the voluntary control of voiding. In addition, neurons in other subcortical areas (e.g., the *nucleus subceruleus*, *reticularis pontis oralis*, cerebellum, hypothalamus, medullary raphe nucleus) exert direct or indirect modulatory influences on the voiding reflex (8).

Micturition involves the parasympathetic system (pelvic motor and sensory neurons and nerves), sympathetic system (hypogastric motor and sensory neurons and nerves) and somatic pathways (from the brain to the sacral spinal cord segments and pudendal nerve). Sensory input on bladder filling is transmitted via the pelvic nerve from the detrusor (smooth) muscle to sacral spinal cord segments (9, 10) and via ascending spinal cord tracts to the pontine reticular formation. The pontine reticular formation is in turn responsible for the micturition reflex through activation of parasympathetic influence (pelvic nerve) and reduction of sympathetic influence (hypogastric) and inhibition of sphincter muscle contraction (pudendal) (8, 11). This constitutes the detrusor reflex. Sensory input also travels to the cerebrum which strongly influences detrusor muscle activation during micturition in normal animals (8, 12). Furthermore, the cerebellum has an inhibitory influence on detrusor muscle activation. Altogether these systems allow for full voluntary bladder emptying.

Urine storage is facilitated by the urethral sphincter. Stretch at that level is sensed by the spindle cells of the striated muscles of the urethral sphincter and transmitted via the pudendal nerve to the sacral spinal cord segments (L7-S3; motoneurons of the pudendal nerve are located within Onuf's nucleus in the ventral horn of the sacral spinal cord). Once the stretch threshold has been reached there is a direct (monosynaptic) motor response via the pudendal nerve activating the external urethral sphincter composed of striated muscle fibers leading to sphincter closure. This motor response also occurs when there is firing from the pelvic nerve afferents allowing continence in situations when there is sudden increase in bladder pressure (e.g., when jumping, barking etc.). The external component of the urethral sphincter is also under influence of the cortex, allowing voluntary contraction or inhibition, and the pelvic nerve, which during detrusor muscle contraction inhibits pudendal nerve firing (8).

Finally, storage of urine is facilitated by the hypogastric nerve, which originates from the L1 to L4 spinal cord segments in the dog. The alpha-adrenergic component synapses to the bladder trigone, neck and proximal urethra and causes contraction. The beta-adrenergic component synapses to the detrusor muscle and allows smooth muscle relaxation. The hypogastric nerve feeds back to the brain information on bladder overdistension and bladder pain signals during inflammatory processes such as urinary tract infection (8), thereby providing a protective mechanism which may be lacking after spinal cord injury. During voiding, pelvic nerve neurons inhibit hypogastric neurons.

PHYSIOLOGY OF DEFECATION

Here we provide a brief overview of the physiology of defecation; for a detailed review on physiology and pathophysiology after

spinal cord injury the reader is referred elsewhere (13, 14) and De Lahunta and Glass also provide an overview in companion animals in their textbook (6).

The normal neurophysiological control of the gastrointestinal system is dependent on local enteric circuits, autonomic input through the parasympathetic and sympathetic nervous systems, and higher cortical processes that serve to control timing of elimination. The local regulation of many gastrointestinal reflex functions is governed by enteric neurocircuitry that is capable of independent secretory and motor (propulsive) reflexes as well as regulating the homeostatic requirements of the gastrointestinal tissues such as blood flow (13, 15). Parasympathetic innervation to the descending colon and rectum arises within the sacral spinal cord segments and travels by way of the pelvic nerve, promoting motility. Gastrointestinal sensory input is derived by way of the hypogastric nerve originating in the lumbar spinal cord segments (L1 to L4 in dogs) and hypogastric nerve stimulation elevates internal anal sphincter pressure and inhibits the descending colon and rectum. Sympathetic input is largely inhibitory of motor and secretory processes and provokes vasoconstriction. Transmission and perception of visceral nociceptive stimuli is generally considered to be relayed through the sympathetic splanchnic nerves and terminates within the spinal cord. Voluntary control and closure of the external anal sphincter muscle is through the pudendal nerve of which its motoneurons are located within Onuf's nucleus. This system is under cortico-thalamic and brainstem control through ascending (dorsal, dorsolateral, and ventrolateral white matter tracts) spinal tracts and descending reticulo-spinal tracts.

During the storage phase, there is autonomic unconscious closure of the internal (hypogastric nerve control) and external (pudendal nerve control) anal sphincters while the descending colon and rectum are inhibited (hypogastric nerve control). When the rectum is full, there is involuntary relaxation of the internal anal sphincter (recto-sphincteric reflex) and defecation can then occur when decided (16). This involves cortically driven relaxation of the external anal sphincter (pudendal nerve control), contraction of abdominal wall muscles and relaxation of pelvic wall muscles.

CONSEQUENCES OF SPINAL CORD INJURY ON MICTURITION

We make a distinction through the rest of the text between urinary incontinence defined as involuntary loss of urine and urinary retention defined as the inability to voluntary void.

Supra-Sacral Lesions

Lesions cranial to the L7 spinal cord segment result in preservation of the sacral spinal cord segments but disruption of the ascending and descending pathways to and from the brain. This loss of supraspinal input results in impaired control over detrusor muscle activation and relaxation and loss of bladder fullness sensation and voluntary micturition. The pelvic and pudendal nerves remain functional; therefore, the external urethral sphincter remains closed. This condition is often referred to as "upper motor neuron" bladder, and clinically, the manifestation is urine retention with a large distended bladder, often felt as tensed or firm on palpation, that is difficult to express. Some neurologists have suggested presence of "small unsynchronized" bladder contractions (5). Therefore, there is incontinence either due to bladder overflow or intermittent emission of urine from involuntary bladder contractions. These animals are at risk for bladder rupture either through excess filling against a closed sphincter or as a complication during manual bladder expression, although this is rare.

Sacral Lesions

Sacral lesions result in loss of bladder innervation from the pelvic and pudendal nerves which causes loss of detrusor muscle function and urethral sphincter tone. It results in excessive bladder distension that feels flaccid on manual palpation and there is typically constant leakage of urine due to poor sphincter function. This is often referred as "lower motor neuron" bladder. The incontinence can occur because of overflow or simply when the pressure in the bladder exceeds that of the weakened urethral sphincter but clinically, the bladder is often quite large. The animal is frequently soiled with urine and this causes major difficulties in nursing care of these patients in hospital and also at home.

In some instances of focal sacral lesions, sphincter tone can be preserved resulting in a combination of flaccid detrusor muscle with sphincter tone that is difficult to overcome and resulting bladder distension. While this is most commonly seen in cats with "tail pull" injuries that damage the pelvic and pudendal nerve roots, it can occur with dogs that have suffered a focal sacral lesion due to acute non-compressive nucleus pulposus extrusions or fibrocartilaginous emboli (17). The exact mechanisms causing this type of dysfunction remain unclear, although there is a suggestion that this could be mediated by the preserved hypogastric structures and internal urethral function.

Is Spinal Shock Associated With Bladder Atony?

Following transection of the spinal cord in experimental dogs (between T8 and T12), a state of spinal shock occurs immediately and leads to complete urinary retention and areflexia of the detrusor for a period of 2–6 weeks (18, 19) before reflex detrusor activity returns. Spinal shock is also known to occur in some companion dogs after "natural" severe spinal cord injury (20, 21) and leads to loss of spinal reflexes, sensation and muscle tone below the lesion for a period of time, usually ∼24–48 h. It is not clear in companion dogs whether spinal shock in cases with severe, acute spinal cord injury also leads to prolonged loss of sacral reflexes and bladder atony and there is limited data on that topic. Atalan et al. (22) described a gradual return of normal residual volume in 25 dogs recovering from spinal cord injury. Levine et al. (23) showed that bladder compliance, capacity, and residual volume were higher in 20 dogs with natural occurring acute spinal cord injury from the day of presentation to 3, 7, and 42 days after spinal cord injury than their control dogs but the presence or absence of spinal shock was not mentioned; however, the control group consisted of

normal experimental dogs and not companion dogs and it is unknown whether these dogs have similar bladder function to house trained dogs. The finding of Levine at al. that bladder tone is increased in acute spinal cord injury (23) is opposite to the finding in experimental dogs where the bladder is atonic in the acute phase after spina cord transection (18, 19). However, this clinical trial recruited dogs with Hansen type 1 intervertebral disc herniation in which spinal shock is less common than in more severe cases, moreover, the majority had only moderately severe injuries, again making spinal shock unlikely. In another study on thoracolumbar acute non-compressive nucleus pulposus extrusion, a weak association was found between spinal shock and the odds of remaining faecally incontinent, although this likely reflects an association between spinal shock and severity of injury (24). Interestingly, bladder atony at the time of spinal shock can be improved with functional electrical stimulation of sacral nerve roots in experimental dogs (25, 26) when delivered early in the disease process. Levine et al. (23) also showed that early pharmacological intervention with sub-cutaneous injection of a matrix metalloproteinase inhibitor could improve bladder compliance. These findings form interesting observations on which we can build further studies. In particular, we need to define whether bladder atony occurs in the most severe cases of canine spinal cord injury (i.e., with complete loss of sensori-motor function and loss of deep pain). We then need to study the sustainability of the effect of early bladder interventions (in particular functional electrical stimulation) on continence, particularly in those cases that will not recover deep pain.

CONSEQUENCES OF SPINAL CORD INJURY ON GASTROINTESTINAL FUNCTION

With upper motor neuron lesions (i.e., supra-sacral) the external anal sphincter remains closed and feces accumulate. The animal occasionally drops feces without awareness when the pressure in the colon and rectum increases and causes involuntary reflex evacuation. Fecal incontinence is largely a feature of thoraco-lumbar (T3-L3) lesions causing paraplegia since traumatic cervical injuries in dogs severe enough to cause fecal incontinence are extremely rare (4, 27). Companion dogs with only moderate gait deficits and concomitant fecal incontinence have been infrequently reported and shown to have discrete lesions in the dorsal aspect of the spinal cord. These cases are rarely diagnosed with intervertebral disc herniation (28, 29). The most prevalent gastrointestinal co-morbidity following spinal cord injury in humans is "neurogenic bowel", which is frequently described as colonic dysfunction that presents as reduced colonic contractions and transit, constipation, disordered evacuation reflexes and potential overflow incontinence. In dogs, constipation is rarely a problem, but it is encountered in cats following spinal cord injury more commonly (30). Also, in humans and rat there is evidence for development of esophageal and gastric impairment of function after upper spinal cord injury (with increased incidence of heartburn and esophageal chest pain chronically) (31). In dogs, subclinical gastroduodenal ulceration has been reported in only one study with a prevalence as high as 76%, raising awareness of that possible complication in the acute phase of intervertebral disc extrusion (32). Finally, the development of autonomic dysreflexia, a condition in which a noxious visceral stimulation, often accompanying severe constipation or bladder distension, triggers a life-threatening increase in sympathetic discharge below the injury level and systemic hypertension, has so far not been shown to occur in dogs. In humans and rodents this syndrome is seen to occur secondary to severe cervical and high thoracic lesion and can be fatal but high thoracic lesions are rare in dogs, so it is difficult to know if autonomic dysreflexia exists or not in dogs.

Fecal incontinence is a more common problem in dogs that suffer acute contusive or vascular (i.e., "non-compressive") lesions of the spinal cord, such as acute nucleus pulposus extrusion or fibro-cartilaginous embolism, respectively (24, 33) compared to compressive lesions. This might be because parenchymal lesions are more centrally located within the spinal cord or perhaps resulting from dilation of the spinal canal. This is seen in dogs with chronic compression of the spinal cord from sub-arachnoid diverticulum, such as in Pugs where fecal incontinence is common. The prognosis for dogs with acute contusive or vascular lesions is further discussed in the treatment section below.

In cases with spinal cord injury caused by intervertebral disc extrusion (i.e., a "compressive" lesion) in the T3-L3 spinal cord segments, ∼40% of owners of dogs that have recovered from paraplegia with loss of deep pain report that their dog's fecal continence is not as good as it was prior to their injury (4, 27). They report two types of fecal incontinence. In the first type, called sudden urge incontinence here, the dog becomes acutely aware of the need to defecate, makes a dash for a suitable location and is unable to prevent the defecation reflex. The second type occurs when the dog drops stool without apparently being aware of it. Both of these forms of incontinence can happen to differing degrees in dogs that are also able to defecate voluntarily. For most owners, this is not a major complaint, but for a small subset the severity of the problem results in euthanasia of the dog whether the dog has recovered motor function or not (4).

With lower motor neuron lesions (i.e., sacral), the anal sphincter is weak or absent and this causes constant leakage of feces. This is a much more serious problem to manage the dog in hospital and at home and can lead to megacolon in the author's experience.

SPONTANEOUS RECOVERY OF URINARY AND FECAL CONTINENCE AFTER SPINAL CORD INJURY

Possible Mechanisms Underlying Recovery

Reorganization of the micturition reflex following spinal cord injury is dependent in part on the plasticity of bladder afferent pathways and the unmasking of reflexes triggered by unmyelinated, capsaicin-sensitive, C-fiber bladder afferent neurons. Plasticity of bladder afferent neurons is associated with

morphologic, chemical, and electrical changes, which appears to be mediated in part by neurotrophic factors released at the level of spinal cord and the peripheral target organs (34). Upregulation of anti-inflammatory mediators and neuroprotective molecules is likely to play an important role in the plasticity of bladder afferent pathways as well as reorganization of synaptic connections in the spinal cord (35). In rats, poor voiding efficiency at 4 and 8 weeks after spinal cord injury was coincident with upregulation of pro-inflammatory cytokines (IL-1α, IL-1β, IL-2 IL-5, IL-6, IL-18, and TNF-α), chemokines (CX3CL-1, CCL2, CXCL-1, CXCL2, CXCL-10) and downregulation of anti-inflammatory cytokines IL-4 and IL-13, whereas spontaneous recovery of voiding function at 12 weeks was associated with maximum expression of anti-inflammatory cytokine IL-1018, neurotrophin BDNF and CXCL-5 as well as the neuroprotective leptin19 in bladder (16).

Similarly, the negative impact of inflammation in the recovery process might be greater than previously thought, opening some possible research avenues to look at controlling this inflammatory response. Multiorgan dysfunction following spinal cord injury has been recognized to start minutes to weeks after spinal cord injury with both visceral and somatic tissues affected, including the cardiovascular, pulmonary, renal, skeletomuscular and hepatic systems (36). Also, neuroendocrine changes along the hypothalamic-pituitary-adrenal axis elevate circulating macrophage migration inhibitory factor. It is likely that the systemic inflammatory response that is initiated at the spinal cord injury site, spills over to the circulation, contributing further to acute-phase hepatic pro-inflammatory release.

Clinical Characteristics of the Recovery

With intervertebral disc herniation causing compression within the T3-L3 spinal cord segments (upper motor neuron lesions) and when dogs have present (intact) deep pain sensation in the pelvic limbs, recovery of urination usually occurs concomitantly with return of motor function (5, 22, 23) and the prognosis for recovery of urinary continence is good (17). When deep pain sensation is absent at the time of injury, the prognosis for recovery is less certain (4, 27) and ∼50–60% of dogs eventually regain urination along with deep pain and walking. Of these dogs recovering, ∼30% of owners report less than perfect urinary continence. In particular, they report that they cannot leave their pet without access to somewhere to urinate for as long as they could before the injury and find that the excitement of their return to the house will trigger involuntary urination (4). Further, 30% (27/93) to 59% (48/81) of dogs that do not recover pain perception after intervertebral disc herniation can develop "spinal" walking (i.e., an automatic gait lacking presumably brain control) but none of these spinal walking dogs fully recover either urinary or fecal continence (4, 27, 37). Urinary continence has not been evaluated in detail in these dogs, but it is clear that reflex voiding is present with varying degrees of success and efficiency of bladder emptying, starting in the weeks after injury. Animals with no deep pain can also develop ascending/descending myelomalacia in the days after spinal cord injury (38, 39); in a subset of cases, there is only descending myelomalacia (or the ascending lesions stop progressing and the front limb function is spared while there is descending myelomalacia) and this can lead to destruction of the sacral spinal cord segments and lower motor neuron incontinence and, in our experience, rectal prolapse, with a poor prognosis.

The prognosis for recovery of urinary continence in dogs with non-compressive T3-L3 spinal cord injury seems similar to those with a compressive injury, with 91–98% of dogs with hind limb dysfunction and present (intact) deep pain regaining urinary continence (24, 40, 41). However, interestingly, 15–23% of dogs remain faecally incontinent in the chronic phase of recovery (24, 40, 41), suggesting that the injury perhaps affects spinal cord tracts differently to acute "compressive" intervertebral disc herniation. Indeed, contusive and vascular lesions induce more centrally located damage (as evident on MRI) than is induced by compressive intervertebral disc extrusion, likely affecting the descending inhibitory control of the defecation reflex. The possibility of persistent fecal incontinence should be conveyed to owners of these cases.

With intervertebral disc herniation causing compression of the lumbo-sacral spinal cord segments (L4 to S3) (lower motor neuron lesions) or in dogs with non-compressive lesions of these segments, data on recovery are scarce. A recent study looked at the recovery rate within the first 3 weeks after injury (17). Dogs over 15 kg with compressive lesions and lower motor neuron bladder or dogs with non-compressive lesions and upper motor neuron bladder (e.g., dogs with lesions of the lumbar spinal cord segments rather than sacral) recovered urinary continence in ∼60–70% of the cases. However, dogs with non-compressive lesions and lower motor neuron bladder (i.e., lesions likely extending from the lumbar spinal cord segments to the sacral segments) never recovered continence in that study, although this only represented 5 dogs. Finally, in 13 dogs with absent deep pain and a lesion within the L4-S3 spinal cord segments, only one dog recovered continence, signaling a poor prognosis for this category of dogs.

Secondary lesions of the lower urinary tract in the acute and then chronic phases of spinal cord injury are poorly described in companion dogs. There is a clear occurrence of urinary tract infections (bacterial cystitis, pyelonephritis) that is discussed alongside the management methods to drain the bladder in the following parts of this article. But the histopathological changes of the bladder (e.g., bladder fibrosis or loss of the neuro-muscular junction at the detrusor level) or kidneys (e.g., vesico-ureteral reflux and hydronephrosis) are not described in companion dogs although there are data in experimental dogs (42, 43).

MANAGEMENT OF URINARY RETENTION AND INCONTINENCE IN THE ACUTE PHASE AND EARLY WEEKS AFTER SPINAL CORD INJURY

Diligent management of retention and overflow incontinence in the acute phase of spinal cord injury is paramount. It involves preventing bladder over-distention and ensuring bladder emptying but also managing the dog as a whole and implementing hygiene measures that will prevent complications. The spinal patient might be especially susceptible to developing

complications such as decubital ulcers and urinary tract infection because of the combination of neurogenic incontinence (leading to increased moisture on skin), recumbency [leading to reduced tissue perfusion and pressure >60 mmHg in risk zones such as scapula-humeral articulation, the greater trochanter and the thirteenth rib (44)] and reduced immunity after spinal cord injury [which is known to occur in people from decentralization of the autonomic nervous system (45)] potentially sustained by poor nutrition. Although these factors have not been clearly studied and established as "risks" in dogs after spinal cord injury, they are in people and for the time being, it appears logical to consider these as such. Guidelines for the management of urinary retention and incontinence in the acute phase are proposed in **Table 1**.

Mechanical Techniques to Manage Bladder Emptying

There are three proposed methods employed to empty the bladder in dogs following spinal cord injury: (i) manual emptying; (ii) intermittent aseptic catheterization; and (iii) placement of a indwelling catheter.

Manual emptying is the preferred method because it is quick, simple, cost effective and non-invasive. However, it is not always feasible if the patient is in pain, not cooperative or it might appear a challenging task for untrained staff. Care should be taken in cases with concomitant trauma to the abdomen and therefore potentially trauma to the bladder. A distended bladder might also be painful, rendering manual expression even more difficult. Although manual expression usually allows to empty "some" urine from the bladder, it's success can be extremely variable. It has been shown that on average only ∼50% of the urine is removed by manual emptying, therefore leaving a large residual volume for some dogs, while it is efficient for others (47). Assessing residual volume with ultrasound might therefore be useful, especially in animals that are recovering to see how well they are voiding and try to determine if the urine in the cage is overflow or voiding. The impact of leaving a large residual volume in the bladder is not known. The presence of hematuria in 50% of cases managed this way suggests trauma to the bladder wall but whether this is due to the manual palpation or an effect urine stasis/large residual volume is not known (48). However, it remains likely that this technique is sufficient to prevent bladder over-distension, at least.

Intermittent sterile catheterization is also simple, guarantees an almost complete bladder emptying. But it is more invasive, carries a risk of introducing an infection in the bladder, can potentially cause urethral inflammation and stricture [as known in humans (49)] if done frequently, and might also appear challenging to untrained staff in particular in female dogs. In female dogs, the clinicians' preference might also often be to leave indwelling catheter in place rather than repeating sterile catheterization.

Placement of an indwelling catheter has similar advantages and disadvantages with the previous method but can lead to bladder mucosa minor trauma and bleeding (silicone Foley catheters are preferred), therefore blocking the catheter. It is usually used for a short time during hospitalization because it will be difficult for the owner to manage it at home. The use of a closed bag system and long-term placement of the catheter removes the need for repeated catheterization and is comfortable for the nursing team and the dog, also reducing the risk of urine scald especially for large dogs.

The prevalence of urinary tract infection in the acute phase of thoraco-lumbar spinal cord injury was first assessed in 2006 by Stiffler et al. in 92 cases (managed by manual emptying or catheterization) and found 15% of dogs with a urinary tract infection before surgery and between 12 and 20% of dogs with a urinary tract infection in the 7 days post-operatively, despite antibiotic prophylaxis (50). Non-ambulatory females and dogs with intra-operative hypothermia (<35°C) were at higher risk of developing a urinary tract infection. A year later, Bubenik et al. (51) described that 40% (44 of 105 dogs) of dogs catheterized after spinal cord injury had a urinary tract infection and the odds of developing one increased after each day spent in hospital. In that study, dogs had an indwelling urinary catheter managed with a closed catheter system (as opposed to intermittent clean/sterile) and had to have had a urine culture submitted when the catheter was removed, suggesting a possible bias toward those dogs with a clinically suspected infection but not urine culture performed. The concomitant use of antibiotics with an indwelling catheter was also found to increase the risk of urinary tract infection by 454% (OR, 4.54; 95% CI, 1.83–11.27) (51), an important finding signaling that the systematic use of antibiotics in this situation should be strictly avoided. Administration of dexamethasone within 48 h before surgery also constitutes an increased risk of developing a urinary tract infection in cases with acute spinal cord injury (52). Bubenik and Hosgood also looked at the risk of developing a urinary tract infection with each of the three methods described above to empty the bladder (53). There was no significance difference between the three methods (16% with manual emptying, 32% with indwelling catheter and 8% with intermittent catheterization), but the duration of bladder management dictated the risk of developing a urinary tract infection. In this study, there were many more infections in dogs with indwelling catheters than control dogs, which likely reflected the longer duration of therapy in dogs with indwelling catheters than dogs managed with manual expression or intermittent catheterization. This risk could be managed by earlier removal of the indwelling catheter. Olby et al. (48) followed dogs for 3 months after injury and found that 10 of 25 dogs developed a urinary tract infection during this time period, with most occurring between weeks 1 and 6. Of these cases, many had occult infections, now termed "bacteriuria", of questionable clinical significance. During this study, owners were asked to monitor their dog's urine with dipsticks with the hope of identifying an easy method to alert owners to a developing urinary tract infection. This attempt was a failure, with most owners finding it extremely challenging to monitor urine in this way. The same group also found no effect of cranberry extract given in the acute phase of spinal cord injury on the occurrence of bacteriuria in a randomized control trial in dogs (54).

The reader is referred to previous reviews for a more in depth discussion around detection and treatment of urinary

TABLE 1 | Acute phase of spinal cord injury—in hospital management (within 1 month of injury).

Checks/parameters	Bladder size	Bladder emptying	Feces emission
Frequency	Every 4 h; and check fur is dry	Every 8 h or pending bladder size check	Suprasacral lesions: no need for rectal emptying but clean fur and skin if needed with checks every 8 h Sacral lesions: likely weak sphincters and constant fecal incontinence requiring frequent checks (e.g., every 4 h) and bath fur and skin then dry
Method	Ultrasound > manual palpation	Manual bladder expression > indwelling Foley catheter > intermittent sterile catheterisation; culture urine at removal of indwelling catheter or if suspicion of urinary tract infection: active urine sediment on urinalysis defined as > 5 white blood cell/high power field ± bacteriuria (>10^5 CFU/mL), pyuria, urine cloudiness and foul smell, pyrexia [see (46) for further information]	Inspection of animal
Cut-off/recommendations	Bladder volume estimation from ultrasound = L × W × ([DL + DT]/2) × 0.625 where L is longitudinal bladder length, W is transverse bladder width, DL is longitudinal bladder depth, and DT is transverse bladder depth [see (22)]; emptying proposed if estimated volume > 10 mL/kg	Consider factors that might impair manual bladder expression (e.g., non-cooperative patient, other soft tissue injuries, pain, untrained staff) or factors that might predispose to pressure sores if there is urine leakage (e.g., dogs with long fur)—in these instances consider indwelling Foley catheter	Dedicated neurology ward with shower station

Checks/parameters	Feeding plan	Skin and bed check	Change of recumbency	Blood pressure
Frequency	Check body weight daily; offer food early on with meals every 8 h of a highly digestible (gastrointestinal) diet	Every 6 h	Every 6 h	Twice daily in cases with high thoracic lesions; autonomic dysreflexia reported in humans and experimental rats
Method	By mouth; offer food and water by bringing bowls to the animal's head because recumbent animals might not reach bowls in larger cages; avoid lateral recumbency after a meal	Inspection of animal in particular pressure points/bony prominences at risk such as scapula-humeral articulation, the greater trochanter and the thirteenth rib [see (44)]	Trained staff; ideally two members of staff	Cuff or Doppler measures
Cut-off/recommendations	Monitor urine analysis and biochemistry values in case of unexplained anorexia; search possible concomitant diseases (e.g., hypothyroidism, hypertension, protein losing nephropathy)	Provide absorbent bed pads such as those used for puppy toilet training; provide pressure-relieving mat; static pressure relieving mats generally insufficient; safe pressure in risk zones <60 mmHg [see (44)]	Provide whole body harness with handle, and washable sling support; remove when dogs in cage; See harness details in **Table 3**	Investigate if > 160 mmHg

tract infections (55) and some guidelines are also presented by Baigi et al. (46). Briefly, a urinary tract infection is suspected if there is an active urine sediment, defined as > 5 white blood cell/high power field ± bacteriuria (>10^5 CFU/mL), pyuria, urine cloudiness and foul smell and/or pyrexia.

Pharmacological Interventions for Bladder Management

Pharmacological interventions will vary depending on the localization of the inciting injury and it is imperative to consider carefully the characteristics of the clinical signs when choosing medications. **Table 2** present some possible medications for neurogenic bladder management.

With a lesion of supra-sacral segments and in the acute phase of spinal cord injury, the clinician often faces a bladder that does not contract and is difficult to express due to persistent or increased sphincter tone. In that scenario, it would be ideal to be able to stimulate detrusor contractions with parasympathomimetic drugs such as bethanechol of carbachol, although this would also reduce the ability of the bladder to store urine. Bethanechol should only be administered in combination with medication that reduce sphincter tone such as alpha-sympatholytic drugs. However, bethanechol does not seem to change either urethral or bladder pressure in normal

TABLE 2 | Possible drugs suggested to act on the lower urinary tract physiology during neurogenic dysfunction.

Drug	Class/action	Dose	Common side effects
Bethanechol	Parasympathomimetic/increases detrusor contractility	1.25–25 mg/kg p.o. q8h in dogs; 0.625–5 mg/cat p.o. q8h in cats; (titrate dose upwards to avoid side effects)	Vomiting, diarrhea, salivation, bradycardia
Oxybutynin	Muscarinic receptor antagonists/decrease detrusor contractility	0.5 mg p.o. q8-12h	Anticholinergic signs: reduced gastro-intestinal motility, dry mouth, tachycardia
Mirabegron	β3 adreno-receptor agonists/decrease detrusor contractility	<0.3 mg/kg p.o. q24h (side effects seen in dogs with a single dose of 0.3 mg/kg p.o.)	Tachycardia, arrythmias, erythema, vomiting, destruction of the zygomatic salivary gland
Phenylpropanolamine	Sympathomimetic/increase urethral tone	1.5 mg/kg p.o. q8-12h in dogs and cats	Hypertension, urine retention
Prazosin	Alpha 1-sympatholytic/decrease urethral resistance	1 mg/dog p.o. q8-12h under 15 kg and 2 mg/dog p.o. q8-12h above 15 kg in dogs; 0.25–1 mg/cat p.o. q8-12h in cats	Hypotension (syncope), salivation, sedation
Phenoxybenzamine	Non-specific alpha-sympatholytic/decrease urethral resistance	0.25–1 mg/kg p.o. q8-24h for minimum 5 days in dogs; 0.5–1 mg/kg p.o. q12h for minimum 5 days in cats	Hypotension (syncope, weakness)
Diazepam	Benzodiazepine/decrease urethral resistance	0.25–1 mg/kg p.o. q8-12h in dogs; Avoid in cats	Sedation, hepatocellular necrosis in cats
Dantrolene	Calcium release inhibitor in muscle/decrease urethral resistance	1–5 mg/kg p.o. q8h in dogs; 0.5–2 mg/kg p.o. q8h in cats	Rare—weakness, hepatoxicity, vomiting, hypotension

dogs (56) and is reported in a textbook as ineffective in a fully are flexic bladder (5) and therefore does not present a clear benefit for supra-sacral lesions. Parasympathomimetic molecules can also potentially cause increased motility of the gastrointestinal tract and diarrhea, which can render patient management difficult. Therefore, bethanechol should be kept as a second option if alpha-sympatholytic drugs have failed to help first. Indeed, alpha-sympatholytic drugs can be used in supra-sacral lesions to reduce sphincter tone which is typically increased or at least persistent and impairs voiding in particular when manual bladder expression is attempted. Alpha-sympatholytic drugs (e.g., prazosin, alfuzosin, and phenoxybenzamine) can be used in that respect and effectively target and relax the smooth urethral sphincter in normal dogs (57) but can cause hypotension. Further, striated muscle relaxants (e.g., diazepam, dantrolene, and baclofen) can be used to reduce the external urethral sphincter and assist urine voiding. Although combining a sympatholytic molecule and a muscle relaxant is attractive (and this might facilitate manual bladder expression), the utility of combining prazosin or diazepam in the urinary incontinence management of dogs with T3-L3 intervertebral disc herniation has recently been tested and the authors found no effect of these molecules on the urinary incontinence or duration of hospitalization (58). This was a retrospective study on 71 dogs with various injury grades and therefore of low power. It also seems clinically that drugs to facilitate manual evacuation (e.g., prazosin, diazepam) are often no longer needed after a few weeks.

Terazosin, a long-acting selective α-1 adrenoreceptor blocking molecule has been used to treat vesico-sphincter dyssynergia in spinal cord-injured male humans and reduced bladder outlet obstruction (59) but causes side effects such as collapse and has not been reported in clinical papers since 2002. In dogs, terazosin has been used to treat vesico-urethral reflex dyssynergia but not in the context of spinal cord injury and showed side effects in 93% of the cases (60). Similarly, tamsulosin is also a α-1 adrenoreceptor blocking molecule with higher affinity that terazosin (61) but has not been trialed after spinal cord injury in dogs.

When faced with an atonic bladder (either through a L7-S3 lesion causing a lower motor neuron bladder or because the bladder has been overstretched), the clinician has few options and there is no clear pharmacological method to reduce constant urinary leakage, which is problematic. Bethanechol can be trialed to improve bladder contraction but there is no clinical evidence to back this and the efficacy is poor in the author's experience.

MANAGEMENT OF URINARY INCONTINENCE IN THE CHRONIC PHASE

Guidelines for the management of urinary incontinence in the chronic phase are proposed in **Table 3**.

Mechanical Interventions

These are very much the same as in the acute phase although the different methods of bladder emptying have not been compared and their impact on occurrence of urinary tract infection has not been studied. In our experience, it seems that most owners will eventually manage bladder emptying by manual expression well and very few opt for repeated sterile catheterisation. We know from a study of 26 dogs (3) that the management of the incontinence (urinary and fecal) in the chronic phase (> 3 months) of spinal cord injury requires owners to spend a

TABLE 3 | Chronic phase of spinal cord injury—in hospital management (weeks to months after injury).

Checks/parameters	Bladder emptying	Feces emission
Frequency	Every 6 h; take dog first thing in the morning	Suprasacral lesions: no need for rectal emptying but clean fur and skin if needed with checks every 8 h Sacral lesions: likely weak sphincters and constant fecal incontinence requiring frequent checks (e.g., every 4 h) and bath fur and skin then dry
Method	Manual bladder expression > intermittent sterile catheterisation; possible implantation of a sacral anterior root stimulator for cases with T3-L3 spinal cord lesions and an upper motor neuron bladder	Feces emission usually spontaneous; light perineal stimulation (either digital wearing a disposable glove, or with a Q-tip) possible to trigger defecation; the sacral anterior root stimulator available for dogs with T3-L3 lesions will allow rectum emptying
Recommendations	Avoid indwelling Foley catheterisation as home method of management; Owners should monitor for the following signs and contact their veterinarian for a urine culture if any are seen: 1. A change in urination frequency, or a change in level of continence. 2. Pain on manual expression or during urination. 3. Foul odor to urine. 4. An increasing in licking of penis/vulva. 5. An increase in drinking. Possible cystometry every 6 months to screen for bladder overactivity (leading possibly to vesico-ureteral reflux) or bladder atony.	Regular drying; avoid wet fur; do not use talk powder; in female, inspect vulva carefully twice daily; diapers overnight can keep contamination down but prefer avoiding their use if possible

Checks/parameters	Feeding plan	Skin and bed check	Change of recumbency, mobility
Frequency	Check body weight weekly in the first 3 months after spinal cord injury	Every 6 h	Every 4–6 h
Method	By mouth; offer food and water by bringing bowls to the animal's head because recumbent animals might not reach bowls in house if not able to move freely; avoid lateral recumbency after a meal	Inspection of animal in particular pressure points/bony prominences at risk such as scapula-humeral articulation, the greater trochanter and the thirteenth rib [see (44)]	Walking, physiotherapy, rehabilitation plan to be adapted to each animal pending the type of spinal cord injury and surgery the animal has received (presence/absence of implant, extent of spinal cord decompression, lesion level)
Recommendations	Low residue diet that reduces stool volume and creates firm stools; acid-suppression with for example proton-pump inhibitors if regurgitations, reflux are observed; possible use of probiotics; water intake should be of minimum 50 mL/kg	Provide absorbent bed pads such as those used for puppy toilet training; provide pressure-relieving mat; static pressure relieving mats generally insufficient; safe pressure in risk zones <60 mmHg [see (44)]	Provide whole body harness with handle (the top part of the harness should rest on the whither, the ventral part on the sternum, with the connecting front straps wrapping around the distal cervical region, avoiding contact with the shoulder joint, and the caudal straps resting caudally to the triceps muscle without interfering with the armpit or rubbing on the caudal shoulder muscles during the caudal phase of the stride); Provide washable sling support

median of ∼3 h per week (ranging from 0 to 16 h) whereas the time required to manage mobility issues is a median of 10 h (ranging from 1 to 30 h), perhaps reflecting that incontinence is not a major problem for the owner, who tends to focus on locomotion. Further, in this study, 77% of owners felt that the effort in caring for their pet was worthwhile, although this is a population of owners who had decided to keep their dog alive in the chronic phase of spinal cord injury. Bauer et al. made similar observations in 30 chronically paralyzed and incontinent dogs where 60% of owners reported suspected urinary tract infections (although this could have been simply hematuria) but felt that this was not a problem and 35% reported them as "very infrequent" (62).

More recently, a high rate of bacteriuria was identified in dogs with chronic (>3 months) spinal cord injury (46). The most common isolate was *E. Coli*: 35 of 47 dogs had at least 1 positive urine culture and 13 dogs had recurrent bacteriuria. Fever and cloudy urine were not associated with infection, whereas pyuria was. Interestingly, of 35 dogs for which long term survival data was available, eight had died and only one was euthanized because of inability to empty the bladder. But no death seemed directly related to urinary tract dysfunction such as pyelonephritis or septicemia. This is in contrast with people with chronic spinal cord injury in which the rate of urinary tract infection is also high, ranging from 10 to 68%, but this is associated with a death rate of ∼9%, perhaps reflecting the long

duration of survival (decades) after the injury compared to a few years with dogs (63, 64). Although the data remain scarce in dogs and from a population of dogs referred to specialist hospitals, the above observations raise interesting questions. In particular, is it possible that dogs are more resistant to developing clinically important urinary tract complications than humans? It also raises the issue of the difficulty of determining a clinically relevant infection in this population of dogs. In humans, the most common signs of impending urinary tract infection are fever, increased spasticity and pain, signs that the patient can report. To help detecting the occurrence of urinary tract infection, a questionnaire has been developed by Prof. Olby in an attempt to gather the information needed to discern clinically important infection that should be treated (**Appendix 1**) from those that would not require medical attention. This questionnaire was designed for the owner of the dog.

Finally, it would also be interesting to test the effect of urine acidifiers (ammonium chloride) or antiseptics (in particular nitrofurantoin or methanamine) to prevent the development of urinary tract infection in the chronic phase of spinal cord injury in paraplegic dogs.

Pharmacological Interventions for Bladder Management

In the chronic phase of supra-sacral spinal cord injury, the bladder tone might increase independently from brain control, leading to involuntary detrusor contractions, although this type of dysfunction does not seem to be a common referral request or complaint from owners. Perhaps it facilitates bladder emptying in some cases. In that instance, muscarinic receptor antagonists (e.g., tarafenacin, oxybutynin, imidafenacin, propioverine) can be used. In particular, the bioavailability of imidafenacin in dogs is known (65) making it a potential drug candidate to test. But there is no report of their use in pet dogs and these molecules can come with marked adverse effects in humans (dry mouth, bradycardia followed by tachycardia, arrythmias, constipation, blurred vision etc.). Therefore, more targeted detrusor relaxants causing fewer side effects have been researched. In particular, β3 adreno-receptor agonists have emerged as cleaner molecules in humans and are known to be safe in dogs, such as mirabegron (66); solabegron can relax the bladder while increasing the micturition reflex in experimental dogs (67). These drugs might benefit pet dogs in the chronic phase of spinal cord injury but have not been used in the clinic so far.

Local delivery of drugs constitutes another option. Botulinum-A toxin injection into the detrusor muscle via cystoscopy was first described in 2000 in humans (68) and is an effective way to inhibit acetylcholine release from parasympathetic nerves resulting in a decrease in bladder overactivity and increase in bladder capacity, altogether greatly reducing leaking episodes (69). This technique applies best to overactive "upper motor neuron" bladders and is in theory feasible in the clinic in dogs with chronic paraplegia and frequent emission of spurts of urine due to involuntary detrusor contractions. It has only been tested once in female dogs with non-neurogenic incontinence with good results (70).

Surgical Interventions

Surgical interventions to help improve the clinical signs and management of urinary incontinence can be separated in those that focus on the effector organs, the bladder and/or sphincters, and those that exploit viable peripheral nervous structures below the lesion.

In the sub-acute to chronic phase of spinal cord injury, placement of drainage systems such as low-profile cystotomy tubes might allow comfortable drainage of the urine for the dog and the owner in the few weeks after injury while waiting for potential recovery. Although this requires surgery and can lead to bladder infection, it remains a simple and relatively non-invasive procedure that can ensure complete bladder emptying and seems well rated by owners (71). Aside from this option, there are no reports of surgeries on the bladder itself in companion dogs with spinal cord injury, whereas bladder augmentation is commonly used in people (72) to improve bladder capacity. This might reflect a low demand from owners of affected dogs for advanced treatment of the incontinence, often content with their management or reluctant to put their dog through more surgical interventions. Our lack of experience in that field means that we are currently unable to describe to owners the potential risks or benefits of bladder surgeries aimed at improving incontinence. Some of these, such as vesicostomy are certainly feasible as described in experimental dogs (42). Vesicostomy limits the development of detrusor hypertrophy, which could lead in the long term to total lack of urine storage but seems to come with a number of issues such as bladder infection, surgical failure or skin irritation.

For sacral lesions, the challenge to restore continence is greater than with supra-sacral injuries because of the loss of neurons or peripheral nerves leading to weak sphincters and constant leakage. To circumvent this, two avenues have been researched: re-innervating or re-constructing the urethral sphincter. Re-innervation can be done with peripheral nerve transfer: femoral to pudendal nerve transfer (73), obturator to pelvic and sciatic to pudendal nerve transfers (74–77) have been done in experimental dogs with promising results but have not reached the clinic. These procedures seem to have merit and are probably worth exploring in the future. Re-construction of the bladder neck and urethra in dogs appears in a textbook (5) suggesting that this can be successful but no data are published. There might also be a place for implantation of hydraulic occluders (i.e., artificial urethral sphincter) that are able to restore urethral pressure (78). A good wealth of experience has been gained with this method over recent years for the treatment of urethral sphincter mechanism incompetence (79) which could potentially be exploited in the field of canine spinal cord injury.

Functional Electrical Stimulation

Dogs have been employed for a long time in neuroscience to study the pathophysiology of the bladder after spinal cord injury and ways to restore micturition. From the seventies mainly, spinal cord transection models (80) seemed to re-create in dogs the state of bladder denervation observed in humans (we are providing some references here in footnotes because these are not easily accessible to us but might be to other scientists—see

Footnote[1]), although the data was published in Russian, Japanese or French and is not readily accessible. These transection models, possibly ethically debatable, are still often used in China and Japan and a vast amount of data are available (81).

Various approaches have been used to improve bladder function and restore normal continence, and functional electrical stimulation has been one of the major lines of research followed (81, 82). Experiments in dogs have included stimulation at different levels including transcutaneous bladder stimulation (83), pelvic nerve stimulation (84) or pudendal nerve stimulation (43, 85) outside the spinal canal or intraspinal sacral roots stimulation (25, 86).

These techniques are often quite convincing [e.g., pudendal nerve stimulation delays bladder fibrosis and improves continence in dogs after spinal cord injury (43)] but have rarely converted into clinical applications, perhaps due to a perceived invasiveness, cost or possibly because the reported data do not reach the right audience. On the contrary, it seems that the data gathered in dogs has successfully been implemented in the clinic to humans with spinal cord injury (81) stressing the need to revisit the applicability of these studies to companion dogs.

One striking example of the knowledge gained from canine experiments is sacral nerve stimulation, described in the 19,000 and later in several studies (83, 87, 88). This technique was very successful and translated from bench to bed in <15 years following work from Brindley who developed a human sacral anterior root stimulator (89). This system consists of placing implants along the pairs of sacral nerves via a lumbo-sacral laminectomy. The implants are connected to a sub-cutaneous receiver that the patient can activate with a transducer. The transducer needs to be positioned above the skin under which the receiver is located, and a remote control delivers the electrical current to the transducer to activate the system. This provides nearly complete bladder emptying, solving the problem of lack of voiding. It also restores continence when the sacral sensory nerve roots are rhizotomised in surgery during implantation thereby preventing reflex incontinence (90). The Brindley's neuroprosthesis exploits the fact that the bladder is a smooth muscle, therefore relaxing slowly, and that the external urethral sphincter is striated, therefore relaxing quickly and functions by delivering short burst of stimulation intercalated by periods with no stimulation. Hence, during sacral nerve stimulation, both the smooth detrusor muscle and external urethral sphincter contract, leading to increase in bladder pressure but no emission of urine—this can be seen as a built-up pressure against a closed sphincter. Once the first short burst of stimulation finishes, the external urethral sphincter muscle relaxes quickly but the detrusor, being a smooth muscle relaxes more slowly, leaving a period of time during which the pressure in the bladder exceeds that of the sphincters leading to emission of urine. A series of on and off stimulations are then delivered until this "artificial dyssynergia" empties the bladder fully. The Brindley device has been translated to dogs successfully, a good example of "reverse" translation and described in 9 dogs in 2013 (91). The voiding efficiency of the system in dogs was >90%. Feces are frequently passed during bladder stimulation as well in implanted dogs, hence limiting loss of feces during the day, as is also observed in humans (92). The best candidates for this treatment are dogs with T3-L3 lesions and an upper motor neuron bladder that have not regained deep pain perception and locomotion. These animals can be implanted from a few weeks to years after their injury. Dogs receive the neuroprosthesis most commonly on the second pair of sacral nerves via a routine lumbo-sacral laminectomy (**Figure 1**). The first author of this review has now treated 25 dogs with this system. Finally, it does not appear needed in dogs to perform the sacral nerve sensory rhizotomy, although this is possible (93), and the efficiency of the system is such that if used regularly (e.g., four times in a day), the bladder remains small enough to prevent leakage of urine.

Finally, there is a view that stimulation of lower centers/motor neurons might re-train the bladder. Firing induced by electrical stimulation can be expected to alter circuitry strength and promote specific patterns of connectivity through Hebbian mechanisms. Indeed, recent experiments in rodents from the Edgerton laboratory confirmed that repeated electrical stimulation of the spinal cord, at frequencies of 40 Hz, changed neural networks controlling bladder function (94). Similarly, Grégoire Courtine and Daniel Eberli's laboratories found that multisystem neuroprosthesis developed to train locomotor function in rats also had an effect on bladder function (80). Data from humans and studies in rats showed that locomotor training exercises after spinal cord injury improved bladder function (95); however, the use of various orthoses as support systems rather than active forms of training neither improve the abilities to stand and walk nor improve the physiological health of humans with spinal cord injury, suggesting that active methods of exercise may have more physiological benefits for spinal cord injury subjects (96).

MANAGEMENT OF FECAL INCONTINENCE

Guidelines for the management of fecal incontinence in the acute and chronic phases are proposed in **Tables 1**, **3**, along with suggestions to manage the dog's environment and feeding.

The primary concern expressed by owners of dogs with fecal incontinence is management of the "mess" produced by inadvertent defecation, with secondary concerns of skin damage due to contamination with fecal material and contamination of the vulva causing urinary tract infections. Careful questioning of the owner is needed to determine when the incontinence is occurring and the nature and volume of the stool. The most practical and effective management technique is to use a low residue diet (e.g., gastro-intestinal/hypoallergenic diets) that reduces stool volume sometimes dramatically, and usually

[1]Mokhort V. Findings on the restoration of bladder function in experimental spinal cord injury. *Vopr Neirokhir.* (1963) 27:38–41.
Kondo A. The contraction of the ureter. II. Observations of the ureter of cord injured patients and cord transected dogs. *Nagoya J Med Sci.* (1970) 32:395–406.
Vishnevskii A. Restoration of bladder function following injury to the spinal cord. (1971) 47:34–40.
Sarramon JP, Lazorthes Y, Buffet J, Arbus L. Spinal cord stimulation and neurologic bladder. Experimental study and first clinical applications. *J Urol Nephrol (Paris)* (1973) 79:571–80.

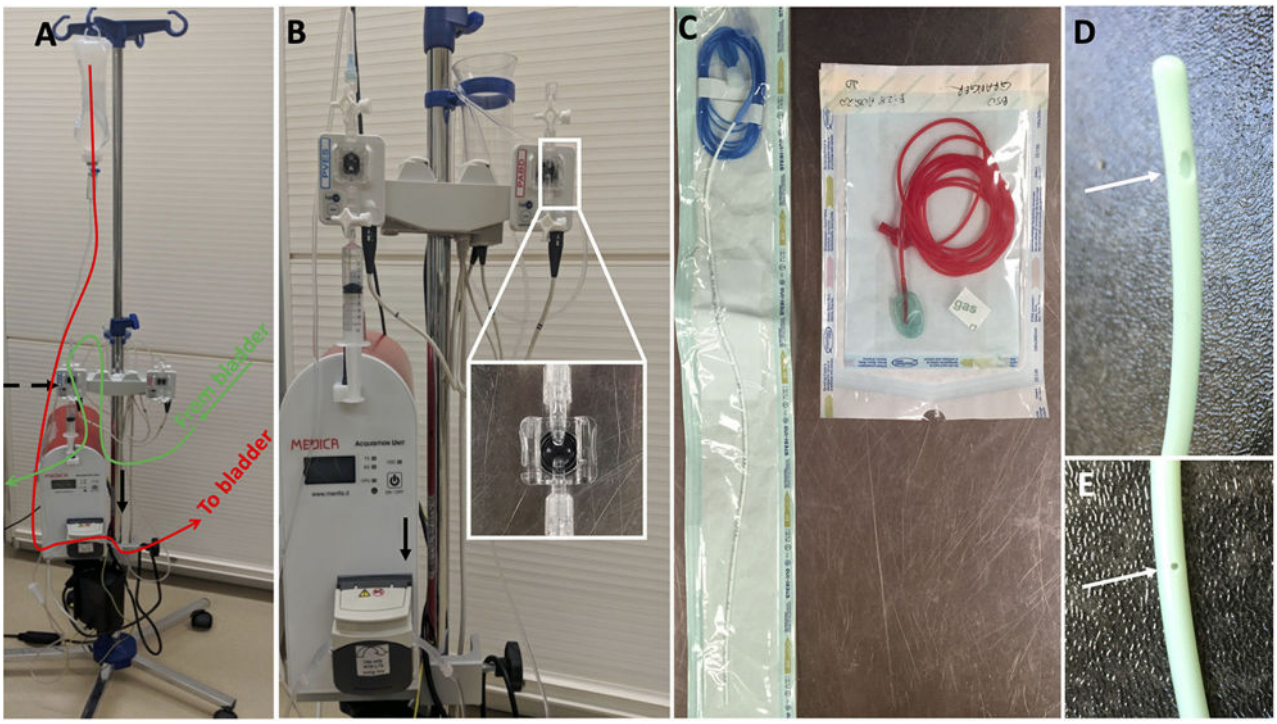

FIGURE 1 | Urodynamic equipment to perform cystometry in dogs; **(A)** it is composed of a pump (black arrow) infusing sterile fluid into the bladder through a dual lumen catheter placed in the bladder (follow red line); the dual lumen catheter measures water pressure in the bladder which is recorded at the level of a pressure transducer (black dashed arrow) connected to a computer software; **(B)** shows the pump more closely and the two pressure transducers typically used to measure bladder pressure and rectal pressure; **(C)** on the left a dual lumen catheter with one lumen used to infused sterile saline in the bladder (transparent port) and one lumen used to measure pressure in the bladder (blue port with blue line extension); on the right a rectal catheter used to measure indirectly abdominal pressure (see **Figure 3** for related pressure curves); **(D,E)** show the two ports of the dual lumen catheter, one large at the tip allowing sterile saline infusion and one 5 cm caudal, smaller and measuring fluid pressure.

resolves loose stool or diarrhea. All dietary indiscretions should be avoided because the consequences can be challenging.

Once stool volume and consistency have been addressed, further management techniques can be used to reduce accidents. First, a bathroom routine should be established providing ample opportunities to defecate. Owners are advised to start by taking the dog out first thing in the morning for bladder expression. Typically, they will defecate at that time, but if not, some owners find light perineal stimulation (either digital wearing a disposable glove, or with a cotton-tipped applicator) can trigger defecation, as well as manual bladder emptying. In some dogs, a meal or exercise is necessary to trigger defecation and owners rapidly learn the best routine for their dog. The question of diapers becomes important for some. For example, in dogs that routinely defecate in their sleep, if they produce only small volumes of hard stool, the problem is easy to deal with in the morning. However, some owners find use of diapers overnight can reduce contamination of bedding. This is effective but skin health should also be considered, and an alternative is use of plastic backed "pee" pads (i.e., absorbent bed pads such as those used for puppy toilet training). Overall, the key is to reduce the volume and moisture of stool, resulting in a reduced frequency of defecation and producing stool that is easy to clean up.

Similar to dogs, management of gastrointestinal dysfunction in humans after spinal cord injury is often limited. While evidence for these interventions is lacking in dogs, they nevertheless can give some useful ideas for management of dogs with chronic spinal cord injury. Treatments for upper gastrointestinal dyspepsia include acid-suppression with proton-pump inhibitors and dysmotility along the entire gastrointestinal tract can be targeted with prokinetics. Pharmacological interventions targeting bladder overactivity with anticholinergics has become commonplace but carries the potential for unintended side effects of diminished gastrointestinal motility, as mentioned above. In addition to prokinetics, neurogenic bowel in humans is treated by increasingly invasive procedures. If diet and fluid management combined with manual evacuation are unsuccessful, individuals may resort to chemical stimulants containing glycerine or bisacodyl. Fluid management is problematic as individuals balance the need to bladder catheterize (lower fluid intake to reduce need for catheterisation) with demands of a bowel program (increase fluid intake to facilitate elimination). Extreme cases of colonic dysmotility may lead individuals to consider surgical procedures such as antegrade continence enema and colostomy, which are not reported in dogs in the context of

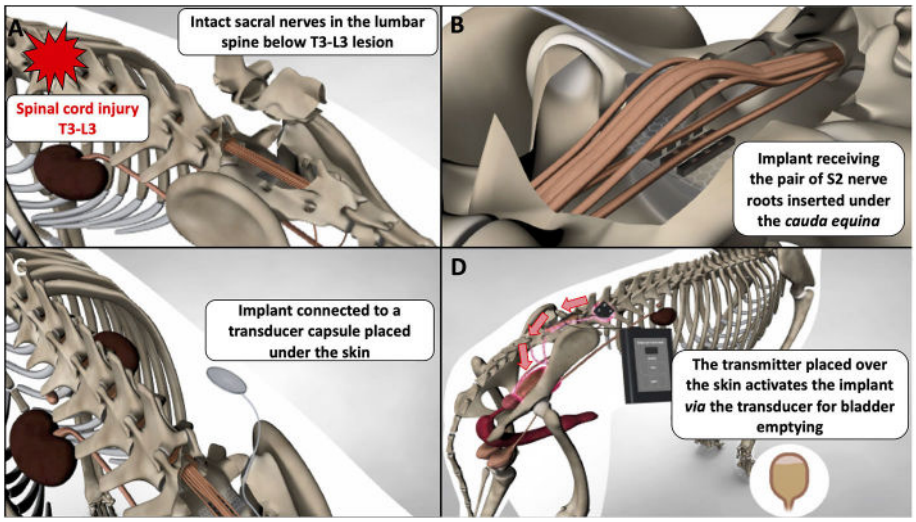

FIGURE 2 | Schematic demonstrating placement and function of a canine sacral nerve stimulator for bladder emptying in chronically paraplegic dogs; **(A)** in dogs with T3-L3 spinal cord lesions, the sacral nerves below the lesion remain intact and can be accessed via lumbo-sacral laminectomy; **(B)** a "book" electrode containing two gutters can receive a pair of sacral nerves (e.g., the S2 pair) when the implant is slotted underneath the dural cone and *cauda equina*; **(C)** the implant is connected via a cable (named a Cooper cable) to a sub-cutaneous transducer that can be palpated by the clinician and the owner; **(D)** the transducer is activated with a remote system brought close to the skin and the transducer; this generates an electrical current that flows to the implant, stimulate the sacral nerves, and leads to efficient bladder emptying.

spinal cord injury. Other people will benefit from sacral nerve stimulation via implanted devices (97). This is also possible in dogs with the sacral root stimulator described above (91): the system in dogs does allow stimulation of defecation (either using bladder voiding parameters or by setting parameters for bowel emptying), often leading to a routine of twice a day elimination of feces at the time the sacral stimulation is done for bladder emptying.

Another aspect that has been studied in humans and animals after spinal cord injury is a change in the gut microbiome which has been shown to be greatly altered after stroke (98) and traumatic brain injury (99). Only a few studies have investigated the spinal cord injury microbiome in both humans and rodent models and two treatment options have shown evidence of improving recovery after spinal cord injury (100, 101). Both probiotics and melatonin have shown to restore some of the microbiota disturbances triggered by spinal cord injury; however, future research should focus on the temporal changes of the spinal cord injury-induced dysbiosis and consider that microbial shifts can also potentiate inflammation which is known to occur following spinal cord injury and possibly add to the chronic inflammatory state of spinal cord injury individuals.

THE STUDY OF URINARY INCONTINENCE WITH THE CANINE TRANSLATIONAL MODEL OF SPINAL CORD INJURY

Recovery of urinary continence and pelvic floor muscle function in the chronic phase of spinal cord injury is highly valued by humans (1, 2). Dogs have been proposed as a large animal model for this purpose in particular those with T3-L3 spinal cord injury resulting in "upper motor neuron" bladder dysfunction (102, 103). The physiology of urination in dogs is more likely to be closer to humans compared to rodents because they are "house trained," and undergo larger fluctuations in bladder size pre-injury, having a similar number of micturitions per day to humans compared to rodents. Dogs are of larger size than rodents and the natural occurrence of spinal cord injury gives advantages to study bladder dysfunction over other animal models. It is possible to employ urodynamic techniques—such as cystometry or profilometry (**Figure 2**)—to study bladder physiology in dogs (104). This gives access to measures such as bladder compliance (i.e., the change of pressure for a given change of volume that describes the ability of the bladder to store urine), involuntary bladder contractions (**Figure 3**), pressure and volume at capacity, voiding efficiency etc. and these measures have been clearly defined by the International Continence Society (105, 106). As such, randomized control trials exploiting companion dogs to study the effect of promising interventions (druggable molecules or cell therapies) for chronic spinal cord injury repair have reported cystometry or continence outcomes. We are still at an early stage of describing the pathophysiological consequences of spinal cord injury on continence in dogs. Most studies of chronically paralyzed dogs did not document an improvement in continence (107–109), which may reflect the complexity of restoring long distance tracts responsible for brain controlled functions. An interesting study recently showed that sub-cutaneous injection of the broad-spectrum MMP inhibitor, GM6001, in the acute phase of the injury (<48 h) led to an increase in bladder compliance compared to controls at 6 weeks of follow-up (23). The pattern of voiding was however

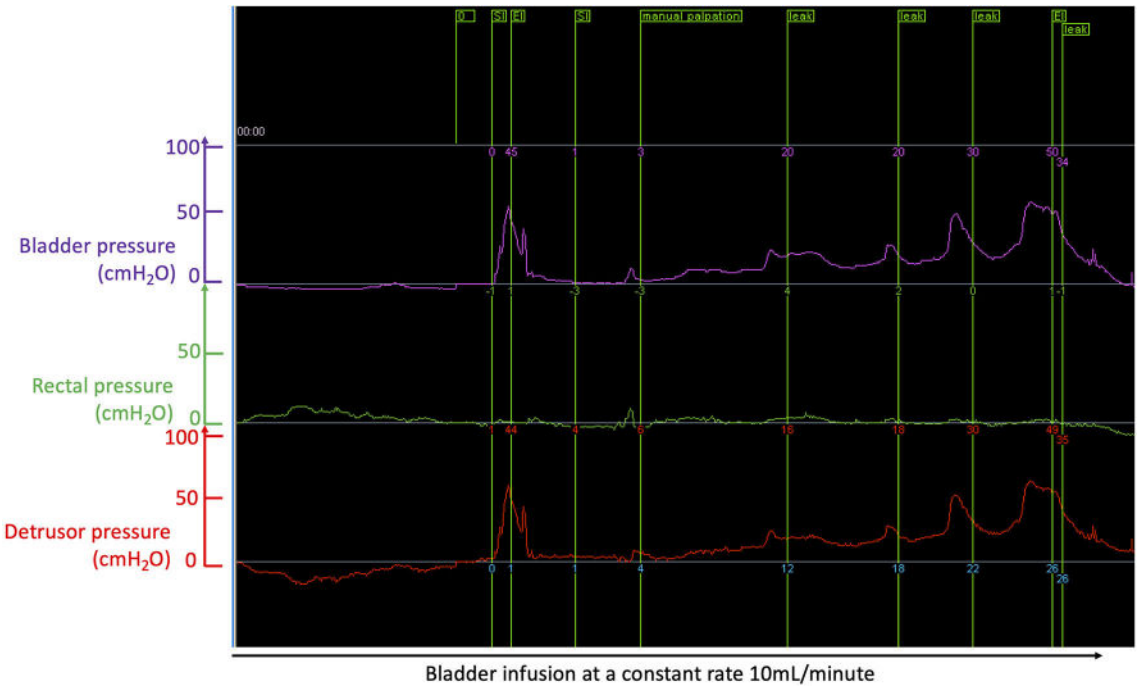

FIGURE 3 | Cystometry curves recorded during bladder filling at a constant rate of 10 mL/min with a dual lumen catheter placed in the bladder through the urethra; the purple trace shows bladder pressure; the green trace shows rectal pressure measured from a rectal balloon; the red trace is the "true" bladder pressure or "detrusor" pressure obtaining by subtracting the bladder pressure (purple curve) by the rectal pressure (green trace): this allows correction for increase bladder pressure peaks due to increase in abdominal pressure, e.g., when the dog moves or barks. In this example of a dog with chronic severe T3-L3 spinal cord injury (causing paraplegia and incontinence), one can see a peak of pressure corresponding to manual palpation by the clinician (green flag at the top "manual palpation" used as a test control); the first peak of pressure to the left of the recording is an artifact; further to the right, involuntary peaks of pressure are recorded and flagged (see green flags at the top "leak") and lead to involuntary emission of urine (i.e., incontinence); during filling, the detrusor pressure slowly rises (here to pressure >50 cmH_2O); however, full voluntary emptying should occur in normal animals when the detrusor pressure reaches ~20 mH_2O and this has not happened here.

unchanged, and these dogs had incomplete spinal cord injury and recovered from their injury, constituting a paradigm different from the problem of reversing incontinence in the chronic phase. Nevertheless, it raises the possibility of investigating this medication in dogs with complete lesions in order to study the long-term effects and sustainability of the changes on urinary continence in the chronic phase of injury.

CONCLUSION

Appropriate care of the lower urinary tract system in dogs after spinal cord injury is critical because this can affect quality and timing of recovery of function and the future life of the animal. Fortunately, most dogs with incomplete thoracolumbar spinal cord injury regain manageable continence, although mechanisms through which this occurs and methods by which this can be further improved upon remain unclear. Continued studies into the acute and also chronic changes of the lower urinary tract after spinal cord injury are important in order to better define the impact of incontinence on the animal and to understand changes over time that could be improved. Development of guidelines for treatment and careful recording of data on urinary tract function will benefit this population of dogs as well as provide much needed data for better understanding of this system after injury.

While evidence-based guidelines for the management of urinary incontinence in companion dogs are scarce, there is even less known about fecal incontinence in dogs. For example, pathological changes to the bladder or gut are largely unknown in companion dogs as are their consequences on function? Owners are frequently managing their dog's incontinence using "common sense" with little veterinary input and the incidence and importance of bacteriuria or urinary tract infections is unknown. While owners rapidly learn to cope with fecal dysmotility, it is very likely that our lack of understanding in the changes in autonomic function and microbiome result in a failure to recognize therapeutic opportunities that could improve the quality of life of animals.

Finally, it is possible that better characterization of autonomic dysfunction in companion dogs will reveal important knowledge that could be transferrable to humans with spinal cord injury. Here, companion dogs could play a pivotal role as a natural model of spinal cord injury and effectively complement other models of spinal cord injury.

AUTHOR CONTRIBUTIONS

NG, NO, and YN-L participated in manuscript conception, preparation, and editing with the first NG leading the conception

and writing and creating the figures and tables. The additional members of the Canine Spinal Cord Injury Consortium (CANSORT-SCI) consortium contributed to manuscript conception, editing, and review. All authors contributed to the article and approved the submitted version.

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REFERENCES

1. Liu CW, Attar KH, Gall A, Shah J, Craggs M. The relationship between bladder management and health-related quality of life in patients with spinal cord injury in the UK. *Spinal Cord.* (2010) 48:319–24. doi: 10.1038/sc.2009.132
2. Anderson KD. Targeting recovery: priorities of the spinal cord-injured population. *J Neurotrauma.* (2004) 21:1371–83. doi: 10.1089/neu.2004.21.1371
3. Freeman PM, Holmes MA, Jeffery ND, Granger N. Time requirement and effect on owners of home-based management of dogs with severe chronic spinal cord injury. *J Vet Behav Clin Appl Res.* (2013) 8:439–43. doi: 10.1016/j.jveb.2013.06.001
4. Olby N, Levine J, Harris T, Muñana K, Skeen T, Sharp N. Long-term functional outcome of dogs with severe injuries of the thoracolumbar spinal cord: 87 Cases (1996-2001). *J Am Vet Med Assoc.* (2003) 222:762–9. doi: 10.2460/javma.2003.222.762
5. Lorenz MD, Coates JR, Kent M. Disorders of micturition. In: Lorenz MD, Coates JR, Kent M. *Handbook of Veterinary Neurology.* Saint-Louis: Elsevier Saunders (2010) p. 58—74.
6. De Lahunta A, Glass EN. Control of defecation. In: de Lahunta A, Glass EN. *Veterinary Neuroanatomy Clinical Neurology*. Saint-Louis: Elsevier Saunders (2014) p. 187–8.
7. Hu HZ, Granger N, Jeffery ND. Pathophysiology, clinical importance, and management of neurogenic lower urinary tract dysfunction caused by suprasacral spinal cord injury. *J Vet Intern Med.* (2016) 30:1575–8. doi: 10.1111/jvim.14557
8. de Groat WC, Griffiths D, Yoshimura N. Neural control of the lower urinary tract. *Compr Physiol.* (2015) 5:327–96. doi: 10.1002/cphy.c130056
9. Häbler HJ, Jänig W, Koltzenburg M. Myelinated primary afferents of the sacral spinal cord responding to slow filling and distension of the cat urinary bladder. *J Physiol.* (1993) 463:449–60. doi: 10.1113/jphysiol.1993.sp019604
10. Birder LA. Nervous network for lower urinary tract function. *Int J Urol.* (2013) 20:4–12. doi: 10.1111/j.1442-2042.2012.03210.x
11. De Lahunta A, Glass EN. Control of urination. In: de Lahunta A, Glass EN. *Veterinary Neuroanatomy Clinical Neurology.* Saint-Louis: Elsevier Saunders. p.184–187.
12. Nishizawa O, Sugaya K, Shimoda N. Pontine and spinal modulation of the micturition reflex. *Scand J Urol Nephrol Suppl.* (1995) 82:177–80.
13. Holmes GM, Blanke EN. Gastrointestinal dysfunction after spinal cord injury. *Exp Neurol.* (2019) 320:113009. doi: 10.1016/j.expneurol.2019.113009
14. Nout YS, Leedy GM, Beattie MS, Bresnahan JC. Alterations in eliminative and sexual reflexes after spinal cord injury: defecatory function and development of spasticity in pelvic floor musculature. *Prog Brain Res.* (2006) 152:359–72. doi: 10.1016/S0079-6123(05)52024-7
15. Vanner S, Macnaughton WK. Submucosal secretomotor and vasodilator reflexes. *Neurogastroenterol Motil.* (2004) 16:39–43. doi: 10.1111/j.1743-3150.2004.00473.x
16. Gonella J, Bouvier M, Blanquet F. Extrinsic nervous control of motility of small and large intestines and related sphincters. *Physiol Rev.* (1987) 67:902–61. doi: 10.1152/physrev.1987.67.3.902
17. Shaw TA, De Risio L, Laws EJ, Rose JH, Harcourt-Brown TR, Granger N. Prognostic factors associated with recovery of ambulation and urinary continence in dogs with acute lumbosacral spinal cord injury. *J Vet Intern Med.* (2017) 31:825–31. doi: 10.1111/jvim.14702
18. Jonas U, Jones LW, Tanagho EA. Recovery of bladder function after spinal cord transection. *J Urol.* (1975) 113:626–8. doi: 10.1016/S0022-5347(17)59539-7
19. Tulloch AGS, Rossler AB. The autonomic nervous system and the bladder during spinal shock-an experimental study. *Paraplegia.* (1975) 13:42–8. doi: 10.1038/sc.1975.8
20. Hodshon AW, Thomas WB. Transient depression of pelvic limb reflexes in dogs with acute focal thoracolumbar myelopathy. *J Am Vet Med Assoc.* (2018) 253:1022–31. doi: 10.2460/javma.253.8.1022
21. Smith PM, Jeffery ND. Spinal shock - comparative aspects and clinical relevance. *J Vet Intern Med.* (2005) 19:788–93. doi: 10.1892/0891-6640(2005)19[788:SSAACR]2.0.CO;2

22. Atalan G, Parkinson TJ, Barr FJ, Innes JF, Holt PE. Urine volume estimations in dogs recovering from intervertebral disc prolapse surgery. *Berl Munch Tierarztl Wochenschr.* (2002) 115:303–305.
23. Levine JM, Cohen ND, Fandel TM, Levine GJ, Mankin J, Griffin JF, et al. Early blockade of matrix metalloproteinases in spinal-cord-injured dogs results in a long-term increase in bladder compliance. *J Neurotrauma.* (2017) 34:2656–67. doi: 10.1089/neu.2017.5001
24. Mari L, Behr S, Shea A, Dominguez E, Ricco C, Alcoverro E, et al. Predictors of urinary or fecal incontinence in dogs with thoracolumbar acute non-compressive nucleus pulposus extrusion. *J Vet Intern Med.* (2019) 33:2693–700. doi: 10.1111/jvim.15626
25. Abdel-Gawad M, Boyer S, Sawan M, Elhilali MM. Reduction of bladder outlet resistance by selective stimulation of the ventral sacral root using high frequency blockade: a chronic study in spinal cord transected dogs. *J Urol.* (2001) 166:728–33. doi: 10.1016/S0022-5347(05)66051-X
26. Hassouna M, LI JS, Sawan M, Duval F, Latt R, Elhilali MM. Effect of early bladder stimulation on spinal shock: experimental approach. *Urology.* (1992) 40:563–73. doi: 10.1016/0090-4295(92)90418-V
27. Aikawa T, Fujita H, Kanazono S, Shibata M, Yoshigae Y. Long-term neurologic outcome of hemilaminectomy and disk fenestration for treatment of dogs with thoracolumbar intervertebral disk herniation: 831 cases (2000-2007). *J Am Vet Med Assoc.* (2012) 241:1617–26. doi: 10.2460/javma.241.12.1617
28. Cerda-Gonzalez S, Olby NJ. Fecal incontinence associated with epidural spinal hematoma and intervertebral disk extrusion in a dog. *J Am Vet Med Assoc.* (2006) 228:230–5. doi: 10.2460/javma.228.2.230
29. Chen A V., Bagley RS, West CL, Gavin PR, Tucker RL. Fecal incontinence and spinal cord abnormalities in seven dogs. *J Am Vet Med Assoc.* (2005) 227:1945–51. doi: 10.2460/javma.2005.227.1945
30. Harris JE, Dhupa S. Lumbosacral intervertebral disk disease in six cats. *J Am Anim Hosp Assoc.* (2008) 44:109–15. doi: 10.5326/0440109
31. Holmes GM. Upper gastrointestinal dysmotility after spinal cord injury: is diminished vagal sensory processing one culprit? *Front Physiol.* (2012) 3:277. doi: 10.3389/fphys.2012.00277
32. Dowdle SM, Joubert KE, Lambrechts NE, Lobetti RG, Pardini AD. The prevalence of subclinical gastroduodenal ulceration in Dachshunds with intervertebral disc prolapse. *J S Afr Vet Assoc.* (2003) 74:77–81. doi: 10.4102/jsava.v74i3.515
33. Fenn J, Drees R, Volk HA, Decker S De. Comparison of clinical signs and outcomes between dogs with presumptive ischemic myelopathy and dogs with acute noncompressive nucleus pulposus extrusion. *J Am Vet Med Assoc.* (2016) 249:767–75. doi: 10.2460/javma.249.7.767
34. De Groat WC, Yoshimura N. Mechanisms underlying the recovery of lower urinary tract function following spinal cord injury. *Prog Brain Res.* (2006) 152:59–84. doi: 10.1016/S0079-6123(05)52005-3
35. Tyagi P, Kadekawa K, Kashyap M, Pore S, Yoshimura N. Spontaneous recovery of reflex voiding following spinal cord injury mediated by anti-inflammatory and neuroprotective factors. *Urology.* (2016) 88:57–65. doi: 10.1016/j.urology.2015.10.017
36. Sun X, Jones ZB, Chen X ming, Zhou L, So KF, Ren Y. Multiple organ dysfunction and systemic inflammation after spinal cord injury: a complex relationship. *J Neuroinflammation.* (2016) 13:260. doi: 10.1186/s12974-016-0736-y
37. Gallucci A, Dragone L, Menchetti M, Gagliardo T, Pietra M, Cardinali M, et al. Acquisition of involuntary spinal locomotion (Spinal Walking) in dogs with irreversible thoracolumbar spinal cord lesion: 81 dogs. *J Vet Intern Med.* (2q017) 31:492–7. doi: 10.1111/jvim.14651
38. Balducci F, Canal S, Contiero B, Bernardini M. Prevalence and risk factors for presumptive ascending/descending myelomalacia in dogs after thoracolumbar intervertebral disk herniation. *J Vet Intern Med.* (2017) 31:498–504. doi: 10.1111/jvim.14656
39. Castel A, Olby NJ, Mariani CL, Muñana KR, Early PJ. Clinical characteristics of dogs with progressive myelomalacia following acute intervertebral disc extrusion. *J Vet Intern Med.* (2017) 31:1782–9. doi: 10.1111/jvim.14829
40. Mari L, Behr S, Shea A, Dominguez E, Johnson PJ, Ekiri A, et al. Outcome comparison in dogs with a presumptive diagnosis of thoracolumbar fibrocartilaginous embolic myelopathy and acute non-compressive nucleus pulposus extrusion. *Vet Rec.* (2017) 181:293. doi: 10.1136/vr.104090
41. McKee WM, Downes CJ, Pink JJ, Gemmill TJ. Presumptive exercise-associated peracute thoracolumbar disc extrusion in 48 dogs. *Vet Rec.* (2010) 166:523–8. doi: 10.1136/vr.b4823
42. Agelan A, Braverman AS, Dean GE, Ruggieri MR. Refinement in the Management of the Denervated Canine Urinary Bladder Using an Abdominal Vesicostomy. *ILAR J.* (2008) 6:E9–14. doi: 10.1093/ilar.49.4.e8
43. Li P, Liao L, Chen G, Zhang F, Tian Y. Early low-frequency stimulation of the pudendal nerve can inhibit detrusor overactivity and delay progress of bladder fibrosis in dogs with spinal cord injuries. *Spinal Cord.* (2013) 51:668–72. doi: 10.1038/sc.2013.60
44. Caraty J, De Vreught L, Cachon T, Moissonnier P, Bongartz A, Viguier E, et al. Comparison of the different supports used in veterinary medicine for pressure sore prevention. *J Small Anim Pract.* (2019) 60:623–30. doi: 10.1111/jsap.13061
45. Iversen PO, Hjeltnes N, Holm B, Flatebo T, Strom-Gundersen I, Ronning W, et al. Depressed immunity and impaired proliferation of hematopoietic progenitor cells in patients with complete spinal cord injury. *Blood.* (2000) 96:2081–3. doi: 10.1182/blood.v96.6.2081.h8002081_2081_2083
46. Baigi SR, Vaden S, Olby NJ. The frequency and clinical implications of bacteriuria in chronically paralyzed dogs. *J Vet Intern Med.* (2017) 31:1790–1795. doi: 10.1111/jvim.14854
47. Carwardine DR, Rose JH, Harcourt-Brown TR, Granger N. Effectiveness of manual bladder expression in paraplegic dogs. *Am J Vet Res.* (2017) 78:107–12. doi: 10.2460/ajvr.78.1.107
48. Olby NJ, MacKillop E, Cerda-Gonzalez S, Moore S, Muñana KR, Grafinger M, et al. Prevalence of urinary tract infection in dogs after surgery for thoracolumbar intervertebral disc extrusion. *J Vet Intern Med.* (2010) 24:1106–11. doi: 10.1111/j.1939-1676.2010.0567.x
49. Cornejo-Dávila V, Durán-Ortiz S, Pacheco-Gahbler C. Incidence of urethral stricture in patients with spinal cord injury treated with clean intermittent self-catheterization. *Urology.* (2017) 99:260–4. doi: 10.1016/j.urology.2016.08.024
50. Stiffler KS, Stevenson MAMC, Sanchez S, Barsanti JA, Hofmeister E, Budsberg SC. Prevalence and characterization of urinary tract infections in dogs with surgically treated type 1 thoracolumbar intervertebral disc extrusion. *Vet Surg.* (2006) 35:330–6. doi: 10.1111/j.1532-950X.2006.00153.x
51. Bubenik LJ, Hosgood GL, Waldron DR, Snow LA. Frequency of urinary tract infection in catheterized dogs and comparison of bacterial culture and susceptibility testing results for catheterized and noncatheterized dogs with urinary tract infections. *J Am Vet Med Assoc.* (2007) 231:893–9. doi: 10.2460/javma.231.6.893
52. Levine JM, Levine GJ, Boozer L, Schatzberg SJ, Platt SR, Kent M, et al. Adverse effects and outcome associated with dexamethasone administration in dogs with acute thoracolumbar intervertebral disk herniation: 161 cases (2000-2006). *J Am Vet Med Assoc.* (2008) 232:411–7. doi: 10.2460/javma.232.3.411
53. Bubenik L, Hosgood G. Urinary tract infection in dogs with thoracolumbar intervertebral disc herniation and urinary bladder dysfunction managed by manual expression, indwelling catheterization or intermittent catheterization. *Vet Surg.* (2008) 37:791–800. doi: 10.1111/j.1532-950X.2008.00452.x
54. Olby NJ, Vaden SL, Williams K, Griffith EH, Harris T, Mariani CL, et al. Effect of cranberry extract on the frequency of bacteriuria in dogs with acute thoracolumbar disk herniation: a randomized controlled clinical trial. *J Vet Intern Med.* (2017) 31:60–8. doi: 10.1111/jvim.14613
55. Weese JS, Blondeau JM, Boothe D, Breitschwerdt EB, Guardabassi L, Hillier A, et al. Antimicrobial use guidelines for treatment of urinary tract disease in dogs and cats: antimicrobial guidelines working group of the international society for companion animal infectious diseases. *Vet Med Int.* (2011) 2011:263768. doi: 10.4061/2011/263768
56. Noël S, Massart L, Hamaide A. Urodynamic investigation by telemetry in Beagle dogs: Validation and effects of oral administration of current urological drugs: A pilot study. *BMC Vet Res.* (2013) 9:197. doi: 10.1186/1746-6148-9-197

57. Fischer JR, Lane IF, Cribb AE. Urethral pressure profile and hemodynamic effects of phenoxybenzamine and prazosin in non-sedated male beagle dogs. *Can J Vet Res.* (2003) 67:30–8.
58. Barnes KH, Aulakh KS, Liu C. Retrospective evaluation of prazosin and diazepam after thoracolumbar hemilaminectomy in dogs. *Vet J.* (2019) 253:105377. doi: 10.1016/j.tvjl.2019.105377
59. Bennett JK, Foote J, El-Leithy TR, Saleem MD, Green B, Archer CL, et al. Terazosin for vesicosphincter dyssynergia in spinal cord-injured male patients. *Mol Urol.* (2000) 4:415–420.
60. Haagsman AN, Kummeling A, Moes ME, Mesu SJ, Kirpensteijn J. Comparison of terazosin and prazosin for treatment of vesico-urethral reflex dyssynergia in dogs. *Vet Rec.* (2013) 173:41. doi: 10.1136/vr.101326
61. Muramatsu I, Taniguchi T, Okada K. Tamsulosin: α1-Adrenoceptor subtype-selectivity and comparison with terazosin. *Jpn J Pharmacol.* (1998) 78:331–335. doi: 10.1254/jjp.78.331
62. Bauer M, Glickman N, Glickman L, Toombs J, Golden S, Skowronek C. Follow-up study of owner attitudes toward home care of paraplegic dogs. *J Am Vet Med Assoc.* (1992) 15:1809–16.
63. Krause JS, Cao Y, DeVivo MJ, DiPiro ND. Risk and protective factors for cause-specific mortality after spinal cord injury. *Arch Phys Med Rehabil.* (2016) 97:1669–78. doi: 10.1016/j.apmr.2016.07.001
64. Garcia-Arguello LY, O'Horo JC, Farrell A, Blakney R, Sohail MR, Evans CT, et al. Infections in the spinal cord-injured population: a systematic review. *Spinal Cord.* (2017) 55:526–34. doi: 10.1038/sc.2016.173
65. Ohno T, Nakade S, Nakayama K, Kitagawa J, Ueda S, Miyabe H, et al. Absolute bioavailability of imidafenacin after oral administration to healthy subjects. *Br J Clin Pharmacol.* (2008) 65:197–202. doi: 10.1111/j.1365-2125.2007.02999.x
66. Schmid RD, Hovda LR. Mirabegron toxicosis in dogs: a retrospective study. *J Med Toxicol.* (2018) 14:160–7. doi: 10.1007/s13181-017-0644-2
67. Hicks A, McCafferty GP, Riedel E, Aiyar N, Pullen M, Evans C, et al. GW427353 (Solabegron), a novel, selective β3-adrenergic receptor agonist, evokes bladder relaxation and increases micturition reflex threshold in the dog. *J Pharmacol Exp Ther.* (2007) 323:202–9. doi: 10.1124/jpet.107.125757
68. Schurch B, Stöhrer M, Kramer G, Schmid DM, Gaul G, Hauri D. Botulinum-A toxin for treating detrusor hyperreflexia in spinal cord injured patients: a new alternative to anticholinergic drugs? *Preliminary results. J Urol.* (2000) 164:692–7. doi: 10.1016/S0022-5347(05)67283-7
69. Romo PGB, Smith CP, Cox A, Averbeck MA, Dowling C, Beckford C, et al. Non-surgical urologic management of neurogenic bladder after spinal cord injury. *World J Urol.* (2018) 36:1555–68. doi: 10.1007/s00345-018-2419-z
70. Lew S, Majewski M, Radziszewski P, Kuleta Z. Therapeutic efficacy of botulinum toxin in the treatment of urinary incontinence in female dogs. *Acta Vet Hung.* (2010) 58:157–65. doi: 10.1556/AVet.58.2010.2.2
71. Stiffler KS, McCrackin Stevenson MA, Cornell KK, Glerum LE, Smith JD, Miller NA, et al. Clinical use of low-profile cystostomy tubes in four dogs and a cat. *J Am Vet Med Assoc.* (2003) 223:325–9. doi: 10.2460/javma.2003.223.325
72. Gomez-Amaya SM, Barbe MF, De Groat WC, Brown JM, Tuite GF, Corcos J, et al. Neural reconstruction methods of restoring bladder function. *Nat Rev Urol.* (2015) 12:100–18. doi: 10.1038/nrurol.2015.4
73. Barbe MF, Gomez-Amaya S, Braverman AS, Brown JM, Lamarre NS, Massicotte VS, et al. Evidence of vagus nerve sprouting to innervate the urinary bladder and clitoris in a canine model of lower motoneuron lesioned bladder. *Neurourol Urodyn.* (2017) 36:91–7. doi: 10.1002/nau.22904
74. Ruggieri MR, Braverman AS, Bernal RM, Lamarre NS, Brown JM, Barbe MF. Reinnervation of urethral and anal sphincters with femoral motor nerve to pudendal nerve transfer. *Neurourol Urodyn.* (2011) 30:1695–704. doi: 10.1002/nau.21171
75. Tiwari E, Salvadeo DM, Braverman AS, Frara NA, Hobson L, Cruz G, et al. Nerve transfer for restoration of lower motor neuron–lesioned bladder and urethra function: establishment of a canine model and interim pilot study results. *J Neurosurg Spine.* (2019) 32:258–68. doi: 10.3171/2019.8.SPINE19265
76. Ruggieri MR, Braverman AS, D'Andrea L, Betz R, Barbe MF. Functional reinnervation of the canine bladder after spinal root transection and genitofemoral nerve transfer at one and three months after denervation. *J Neurotrauma.* (2008) 25:401–9. doi: 10.1089/neu.2007.0335
77. Gomez-Amaya SM, Barbe MF, Brown JM, Lamarre NS, Braverman AS, Massicotte VS, et al. Bladder reinnervation using a primarily motor donor nerve (femoral nerve branches) is functionally superior to using a primarily sensory donor nerve (genitofemoral nerve). *J Urol.* (2015) 193:1042–51. doi: 10.1016/j.juro.2014.07.095
78. Milodowski EJ, Friend EJ, Granger N, Doran ICP. Canine urethral sphincter pressure profile under incremental inflation of an artificial cuff: a cadaver study. *J Small Anim Pract.* (2019) 60:313–8. doi: 10.1111/jsap.12961
79. Gomes C, Doran I, Friend E, Tivers M, Chanoit G. Long-term outcome of female dogs treated with static hydraulic urethral sphincter for urethral sphincter mechanism incompetence. *J Am Anim Hosp Assoc.* (2018) 54:276–84. doi: 10.5326/JAAHA-MS-6709
80. Horst M, Heutschi J, Van Den Brand R, Andersson KE, Gobet R, Sulser T, et al. Multisystem neuroprosthetic training improves bladder function after severe spinal cord injury. *J Urol.* (2013) 189:747–53. doi: 10.1016/j.juro.2012.08.200
81. McGee MJ, Amundsen CL, Grill WM. Electrical stimulation for the treatment of lower urinary tract dysfunction after spinal cord injury. *J Spinal Cord Med.* (2015) 38:2135–46. doi: 10.1179/2045772314Y.0000000299
82. Ragnarsson KT. Functional electrical stimulation after spinal cord injury: current use, therapeutic effects and future directions. *Spinal Cord.* (2008) 46:255–74. doi: 10.1038/sj.sc.3102091
83. Sawan M, Duval F, Hassouna M, Li JS, Elhilali MM. A transcutaneous implantable bladder controller. *Neurourol Urodyn.* (1993) 12:281–4. doi: 10.1002/nau.1930120313
84. Holmquist B. Electromicturition by pelvic nerve stimulation in dogs. *Scand J Urol Nephrol.* (1968) 2:1–27.
85. Ju YH, Liao LM. Electrical stimulation of dog pudendal nerve regulates the excitatory pudendal-to-bladder reflex. *Neural Regen Res.* (2016) 11:676–81. doi: 10.4103/1673-5374.180757
86. Walter JS, Robinson CJ, Khan T, Wheeler JS, Wurster RD. Sacral epidural electrodes for voiding in the chronic spinal dog. *Stereotact Funct Neurosurg.* (1989) 53:113–24. doi: 10.1159/000099526
87. Wang S, Hou C, Chen A, Zhang S, Diao Y, Yin C, et al. Bladder controller in bladder function reconstruction of paraplegic dogs. *Zhonghua Wai Ke Za Zhi.* (2002) 40:780–2.
88. Hassouna M, Elmayergi N, Abdelhady M. Update on sacral neuromodulation: indications and outcomes. *Curr Urol Rep.* (2003) 4:391–8. doi: 10.1007/s11934-003-0014-2
89. Brindley GS, Polkey CE, Rushton DN, Cardozo L. Sacral anterior root stimulators for bladder control in paraplegia: the first 50 cases. *J Neurol Neurosurg Psychiatry.* (1986) 49:1104–14. doi: 10.1136/jnnp.49.10.1104
90. Chang S min, Hou C lin. Staggered rhizotomy of anterior and posterior sacral roots for bladder reservoir function in spinal cord injury: a canine experimental study and preliminary clinical report. *Orthop Surg.* (2009) 1:164–170. doi: 10.1111/j.1757-7861.2009.00025.x
91. Granger N, Chew D, Fairhurst P, Fawcett JW, Lacour SP, Craggs M, et al. Use of an implanted sacral nerve stimulator to restore urine voiding in chronically paraplegic dogs. *J Vet Intern Med.* (2013) 27:99–105. doi: 10.1111/jvim.12011
92. Varma JS, Binnie N, Smith AN, Creasey GH, Edmond P. Differential effects of sacral anterior root stimulation on anal sphincter and colorectal motility in spinally injured man. *Br J Surg.* (1986) 73:478–82. doi: 10.1002/bjs.1800730619
93. Haleem AS, Boehm F, Legatt AD, Kantrowitz A, Stone B, Melman A. Sacral root stimulation for controlled micturition: prevention of detrusor-external sphincter dyssynergia by intraoperative identification and selective section of sacral nerve branches. *J Urol.* (1993) 149:1607–12. doi: 10.1016/S0022-5347(17)36460-1
94. Gad PN, Roy RR, Zhong H, Lu DC, Gerasimenko YP, Edgerton VR. Initiation of bladder voiding with epidural stimulation in paralyzed, step trained rats. *PLoS One.* (2014) 9:e108184. doi: 10.1371/journal.pone.0108184
95. Hubscher CH, Montgomery LR, Fell JD, Armstrong JE, Poudyal P, Herrity AN, et al. Effects of exercise training on urinary tract function after spinal cord injury. *Am J Physiol - Ren Physiol.* (2016) 310:F1258–268. doi: 10.1152/ajprenal.00557.2015
96. Karimi M, Omar A, Fatoye F. Spinal cord injury rehabilitation: which way forward? *NeuroRehabilitation.* (2014) 35:325–40. doi: 10.3233/NRE-141124

97. Rasmussen MM, Kutzenberger J, Krogh K, Zepke F, Bodin C, Domurath B, et al. Sacral anterior root stimulation improves bowel function in subjects with spinal cord injury. *Spinal Cord.* (2015) 53:297–301. doi: 10.1038/sc.2015.2
98. Yin J, Liao SX, He Y, Wang S, Xia GH, Liu FT, et al. Dysbiosis of gut microbiota with reduced trimethylamine-n-oxide level in patients with large-artery atherosclerotic stroke or transient ischemic attack. *J Am Heart Assoc.* (2015) 4:e002699. doi: 10.1161/JAHA.115.002699
99. Treangen TJ, Wagner J, Burns MP, Villapol S. Traumatic brain injury in mice induces acute bacterial dysbiosis within the fecal microbiome. *Front Immunol.* (2018) 9:2757. doi: 10.3389/fimmu.2018.02757
100. Jing Y, Yang D, Bai F, Zhang C, Qin C, Li D, et al. Melatonin treatment alleviates spinal cord injury-induced gut dysbiosis in mice. *J Neurotrauma.* (2019) 36:2646–2664. doi: 10.1089/neu.2018.6012
101. Kigerl KA, Hall JCE, Wang L, Mo X, Yu Z, Popovich PG. Gut dysbiosis impairs recovery after spinal cord injury. *J Exp Med.* (2016) 213:2603–20. doi: 10.1084/jem.20151345
102. Jeffery ND, Granger N, Franklin RJM. Using naturally occuring spinal cord injury in domestic dogs to explore novel therapeutic options. In: Aldskogius H, editor. *Neuromethods 76: Animal Models of Spinal Cord Repair*. London: Humana Press, Springer protocols (2013). p.185–205.
103. Cook L, Byron J, Moore S. Urological sequelae to acute spinal cord injury in pet dogs: a natural disease model of neuropathic bladder dysfunction. *Top Spinal Cord Inj Rehabil.* (2019) 25:205–13. doi: 10.1310/sci2503-205
104. Goldstein RE, Westropp JL. Urodynamic testing in the diagnosis of small animal micturition disorders. *Clin Tech Small Anim Pract.* (2005) 20:65–72. doi: 10.1053/j.ctsap.2004.12.009
105. Abrams P, Cardozo L, Fall M, Griffiths D, Rosier P, Ulmsten U, et al. The standardization of terminology in lower urinary tract function: report from the standardisation sub-committee of the International Continence Society. *Urology.* (2003) 61:37–49. doi: 10.1016/S0090-4295(02)02243-4
106. Biering-Sørensen F, Craggs M, Kennelly M, Schick E, Wyndaele JJ. International lower urinary tract function basic spinal cord injury data set. *Spinal Cord.* (2008) 46:325–30. doi: 10.1038/sj.sc.3102145
107. Hu HZ, Granger N, Balakrishna Pai S, Bellamkonda R V., Jeffery ND. Therapeutic efficacy of microtube-embedded chondroitinase ABC in a canine clinical model of spinal cord injury. *Brain.* (2018) 141:1017–27. doi: 10.1093/brain/awy007
108. Granger N, Blamires H, Franklin RJM, Jeffery ND. Autologous olfactory mucosal cell transplants in clinical spinal cord injury: a randomized double-blinded trial in a canine translational model. *Brain.* (2012) 135:3227–37. doi: 10.1093/brain/aws268
109. Nishida H, Nakayama M, Tanaka H, Kitamura M, Hatoya S, Sugiura K, et al. Evaluation of transplantation of autologous bone marrow stromal cells into the cerebrospinal fluid for treatment of chronic spinal cord injury in dogs. *Am J Vet Res.* (2011) 72:1118–23. doi: 10.2460/ajvr.72.8.1118

Conflict of Interest: NG is employed by the company CVS Ltd as a clinical neurologist and the Royal Veterinary College, University of London, as Senior Research Fellow.

The remaining authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.

APPENDIX 1

NC State Veterinary Hospital
Date: _____
Urinary Tract Health Information in Paralyzed Dogs
History:

1. Has there been a recent change in the general mental attitude of your dog? ☐ Yes ☐ No. If yes, describe:_____
2. Has there been an increase in kicking, jerking, stiffness or spasms in the hind legs? ☐ Yes ☐ No. If yes circle the sign (s).
3. Has there been an increase in dribbling or inappropriate urination? ☐ Yes ☐ No
4. Is your dog licking its vulva or penis? ☐ Yes ☐ No
5. Is there a discharge from the vulva or penis? ☐ Yes ☐ No
6. Has the frequency of expressing/catheterization changed? ☐ Yes ☐ No If yes, provide details. _____
7. Has your dog been more painful recently? ☐ Yes ☐ No If yes, please explain and note location of pain:_____
8. Have you noticed any signs of pain or discomfort while expressing your dog's bladder? ☐ Yes ☐ No. If yes, please explain: _____
9. Has there been a change in the smell of urine? ☐ Yes ☐ No If yes, please describe: _____
10. Has there been a change in the appearance of urine? ☐ Yes ☐ No. If yes, please describe: _____

Physical exam:

1. Body temperature
2. UTI specific physical examination findings
   ☐ Changed mentation ☐ Lethargy and malaise ☐ Painful flank and bladder region ☐ Increased spasticity
   Additional comments:
3. Urine appearance and odor: ☐ Cloudy ☐ Dark ☐ Bloody ☐ Foul odor
4. Discoloration of skin around vulva or penis ☐ Yes ☐ No

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Prognostic Factors in Canine Acute Intervertebral Disc Disease

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Knowledge of the prognosis of acute spinal cord injury is critical to provide appropriate information for clients and make the best treatment choices. Acute intervertebral disc extrusions (IVDE) are a common cause of pain and paralysis in dogs with several types of IVDE occurring. Important prognostic considerations are recovery of ambulation, return of urinary and fecal continence, resolution of pain and, on the negative side, development of progressive myelomalacia. Initial injury severity affects prognosis as does type of IVDE, particularly when considering recovery of continence. Overall, loss of deep pain perception signals a worse outcome. When considering Hansen type 1 IVDE, the prognosis is altered by the choice of surgical vs. medical therapy. Concentration of structural proteins in the plasma, as well as inflammatory mediators, creatine kinase, and myelin basic protein in the cerebrospinal fluid (CSF) can provide additional prognostic information. Finally, cross-sectional area and length of T2 hyperintensity and loss of HASTE signal on MRI have been associated with outcome. Future developments in plasma and imaging biomarkers will assist in accurate prognostication and optimization of patient management.

Keywords: paraplegia, ambulation, dog, pain perception, spinal cord injury, acute intervertebral disc extrusion, acute non-compressive nucleus pulposus extrusion

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INTRODUCTION

Intervertebral disc disease (IVDD) is a common cause of acute spinal cord injury in dogs, due in large part to the high rates of early disc degeneration in chondrodystrophic breeds (1). Indeed, acute, severe thoracolumbar spinal cord injuries account for ∼4% of cases presenting to emergency rooms in North America, with 74% of these cases due to some form of IVDD (2) and ∼20,000 surgeries for the condition taking place annually (3). Paralysis is an extremely stressful clinical problem for pet owners, who have to process complex information on the underlying disease, the risks and costs of the procedures needed to diagnose and treat their pet, and the possibility that their pet might suffer permanent deficits or death. Thus, rapid and accurate patient assessment and a realistic portrayal of the clinical picture and prognosis is important for the owner at time of presentation to the veterinarian. Moreover, this information aids the veterinarian with appropriate patient triage and with setting realistic functional recovery goals that will allow early detection when a patient is deviating from an expected course. Indeed, the importance of prognostication to humans with traumatic spinal cord injury is such that multivariable clinical prediction models

have been developed for this purpose (4). In addition, as increasing numbers of clinical trials are performed in this clinically diverse population of dogs, identifying prognostic biomarkers that quantify injury severity more accurately than clinical assessment alone can refine patient inclusion criteria or serve as covariates in trials, increasing study power and efficiency.

The prognosis, or "before (from the Greek, pro) knowledge (from the Greek, gignoskein)" of a disease is a forecast of disease course following its onset, in this case, using standard treatment. It refers to the possible outcomes of a disease and the frequency with which these outcomes can be expected to occur. A prognostic factor is a measurement that is associated with clinical outcome in the absence of therapy or with the application of a standard therapy (5). A biomarker is "a defined characteristic that is measured as an indicator of normal biological processes, pathogenic processes, or biological responses to an exposure or intervention, including therapeutic interventions. Molecular, histologic, radiographic, or physiologic characteristics are types of biomarkers" (https://www.ncbi.nlm.nih.gov/books/NBK326791/). Moreover, a prognostic biomarker is "a biomarker used to identify likelihood of a clinical event, disease recurrence or progression in patients who have the disease or medical condition of interest." In the context of intervertebral disc extrusion (IVDE)-induced spinal cord injury, the clinical events being prognosticated include the *recovery of independent walking* (both speed and level of recovery) and *fecal and urinary continence*. Resolution of pain is also important, and usually considered for certain subsets of IVDH that cause extreme pain such as cervical IVDE. Finally, the chances of *development of progressive myelomalacia*, a complication that is usually fatal, is also prognosticated in Hansen type 1 IVDE specifically.

Over the last few decades, there have been numerous studies evaluating prognostic factors for recovery of ambulation and continence after IVDH-induced thoracolumbar spinal cord injury, fewer after cervical spinal cord injury. Many of these studies consider clinical parameters, but their findings can be contradictory. Quantification of lesion extent using imaging, originally using myelography and more recently, magnetic resonance imaging (MRI), has received a lot of attention. Finally, biomarkers measured within the blood and cerebrospinal fluid (CSF) have been investigated. In this article we will consider prognostic factors for dogs with acute spinal cord injury due to acute IVDH. The majority of published data reports outcomes in thoracolumbar IVDH, but data on cervical IVDH have also been included. We will focus on Hansen type 1 IVDE but have also included prognostic information on acute non-compressive nucleus pulposus extrusions (ANNPE) and FCEM (fibrocartilagenous embolic myelopathy) as well as hydrated nucleus pulposus extrusions (HNPE). Fibrocartilagenous embolism is included both because of its clinical similarity to ANNPE and because it represents a form of intervertebral disc-induced acute spinal cord injury.

When presenting data on prognosis to clients, it is extremely important to educate them on what that data represents for their individual dog and on how to use the data in the recovery period. Clients need accurate information in order to make the best decision initially and perhaps the most difficult concept for them is understanding that the data presented represents the behavior of a population; the precise prediction of outcome for an individual is not possible. Once beyond the initial decisions about treatment, providing clients with a timeline for certain thresholds to be crossed, while again explaining the variability across a population and factors that might influence their individual dog, can ensure that a dog that is not following an expected recovery curve, perhaps due to a comorbidity or a complication of the initial injury, is identified in a timely manner. This discussion can also help clients to understand that decisions can be made at many different stages of injury and recovery, and avoid a decision being made in a time of great stress that might be regretted at a later date.

PROGNOSTIC FACTORS

Clinical Presentation

Several different parameters have been evaluated for their prognostic utility including signalment, speed of onset and duration of signs (in particular duration of paralysis), and severity of neurological deficits. The most likely underlying condition is also taken into account because it will affect prognosis. This occurs because different forms of IVDD cause differing types and distribution of spinal cord pathology. The vast majority of these studies are retrospective in nature and variation in patient populations, diagnostic and therapeutic protocols and reporting and methods of follow up make it difficult to draw strong conclusions.

Severity of Neurological Deficits

The exception to the limitation of clinical parameters as prognostic factors is severity of neurological deficits, which is the most immediately accessible, simple and reliable prognostic indicator for animals with spinal cord injury. This is true regardless of neuroanatomic location and is equally true in human spinal cord injury (6). Given the poor regenerative capabilities of the adult CNS, it is not surprising that gauging the extent of permanent tissue loss is extremely important to establish a prognosis. It is common practice to assign animals with thoracolumbar spinal cord injury to one of six categories based on the severity of their clinical signs in a scale known widely as the Modified Frankel Scale (MFS) (7, 8). The frequency of signs within each category in this scale has been estimated (**Table 1**) (9). There are numerous slightly different variations on this scale in the veterinary literature, with numbers assigned in different directions and subcategories developed. This makes comparisons between studies confusing and so for the purposes of accurate reporting in this paper, clinical severity has been categorized using description of the signs (**Table 1**).

Evaluation of Pain Perception

Prognosis for recovery of independent ambulation is influenced by the *presence of pain perception* (10–12). Not surprisingly for such an important clinical variable, there is a range of

TABLE 1 | Definition of categories of neurologic injury commonly used clinically.

Description	MFS Scale 1 (7)	MFS Scale 2 (8)	Frequency in reported studies (9)	ASIA impairment scale (6)
Normal	0	6	NA	E
Painful, no neurological deficits	1	5	NA	
Ambulatory paraparetic/ataxic	2	4	30%	D
Non-ambulatory paraparetic	3	3	22%	C
Paraplegic intact DPP	4	2 (superficial and deep pain present)	30%	B
Might be subcategorized by:		1 (lacks superficial pain)		B
• *Presence of pain in both hind feet and tail vs. only one location*				
• *Presence of superficial pain*				
• *Presence of voluntary urination*				
Paraplegic NDPP	5	0	16%	A

Note numbers are assigned using opposite conventions by different authors. These categories correspond well to the human American Spinal Injury Association (ASIA) scale (6). In this article the description of each category is used to avoid confusion when interpreting a wide range of literature. MFS, Modified Frankel Scale; NA, not applicable—data derived from studies of dogs presenting for surgery; DPP, deep pain perception; NDPP, no deep pain perception.

different terminology used in the literature. Historically the term "deep pain perception" (DPP) has been used—referring to the response to an extremely noxious stimulus applied over the bone of a digit. More recently, the term deep has been omitted and authors use the terms pain perception and nociception. Sometimes "deep nociception" appears. In this article we use the term DPP when discussing prognostic indicators because it is familiar to most veterinarians and conveys the importance of applying a strong noxious stimulus when determining the presence of pain perception.

Because of its clinical implications, assessment of pain perception should be made extremely carefully in any animal that lacks motor function. This is performed ideally in a calm animal, using an instrument with relatively wide jaws such as needle drivers or pliers (to avoid cutting the skin as pressure is applied). While placing an animal on its side to perform this test allows a clear view of the response, if they are fighting to get up, it can be difficult to interpret their behavior. If this occurs, the animal should be placed in whatever position allows clear access to the limb being tested with the animal resting quietly. Pressure is applied over the digit being tested and a gentle squeeze is applied to produce a withdrawal reflex (if present) and then pressure is increased until the patient demonstrates perception of the stimulus such as vocalization, looking around, or moving away (**Supplementary Video 1**). In animals with blunted perception, the response might be as subtle as an alteration in breathing pattern or dilation of pupils. Any repeatable behavioral indication that the animal can feel the stimulus is taken as DPP being present. Both medial and lateral digits should be tested in each foot and the base of the tail should be tested (using the handles of the forceps). Presence of pain perception in any one of these locations places the animal in the prognostic category of having DPP (10, 13).

Recovery in Animals With Intact DPP

The prognosis for recovery of independent ambulation and continence in animals that have intact DPP, even if apparently blunted, is good to excellent depending on the treatment pursued (**Table 2**) and the type of disc herniation that occurred (**Table 3**). The speed of that recovery is influenced by the severity of motor impairment at presentation, altering prognosis for walking at 2, 4–6, and 12 weeks (**Table 2**). These benchmarks are extremely useful to indicate when a dog might not be recovering as expected, triggering a timely re-evaluation by the veterinarian. Recovery of fecal and urinary continence in dogs with DPP due to Hansen Type 1 IVDE matches recovery of walking. However, persistent fecal and urinary incontinence have both been reported in animals with incomplete injuries due to ANNPE and FCEM in spite of recovery of ambulation (**Table 3**).

Recovery of independent ambulation and resolution of pain in dogs with cervical IVDE has also been reported with and without spinal cord decompression and in general is excellent with surgery (**Table 4**). However, potentially serious complications of hemorrhage, hypoventilation and bradycardia, vertebral subluxation and aspiration pneumonia have all been reported and the development of an adverse event of this manner does worsen prognosis (28, 42–45). The majority of dogs with hydrated nucleus pulposus extrusions (HNPE) present with cervical extrusions. These dogs have an excellent prognosis for recovery with or without surgery, even in the presence of respiratory compromise (**Table 4**).

Recovery in Animals With No DPP

The prognosis for animals that lack DPP is less certain, with recovery rates for independent walking in dogs with surgically managed thoracolumbar IVDE ranging from 30 to 75% in different studies (11–16). Overall, ∼60% of dogs with Hansen type 1 IVDE recover DPP and ambulation by 6 months after injury (**Table 2**). The timing of recovery of pain perception is important, because once it is present, the prognosis for recovery of ambulation is excellent. One study found that 62% of dogs

TABLE 2 | Summary of prognosis for acute TL-IVDE based on presenting grade of injury and treatment choice.

Grade	Overall recovery with conservative management (%)	Overall recovery with surgery (%)	Recovery at 2 w (%)	Recovery at 4–6 w (%)	Recovery at 3 m (%)	Development of PMM (%)
Ambulatory paraparetic	72.5 ($n = 116$)	98.4 ($n = 318$)	84	92	93.9	0
Non-ambulatory paraparetic	79.8 ($n = 74$)	93 ($n = 341$)	77.8	88.9	92.8	0.6
Paraplegic with DPP	56 ($n = 77$)	93 ($n = 548$)	70.8	78.2	83.2	2.7
Paraplegic NDPP	22.4% ($n = 48$)	61 ($n = 502$)	26.5	42.3	53.8	13.9

w, weeks; m, months; PMM, progressive myelomalacia; NDPP, no deep pain perception; DPP, deep pain perception (10, 12–21).

TABLE 3 | Outcomes of dogs with different types of acute thoracolumbar intervertebral disc disease.

Grade		Ambulatory paraparetic	Non-ambulatory paraparetic	Paraplegic with DPP	Paraplegic with NDPP
ANNPE (22, 23)	Amb	100% ($n = 84$)	100% ($n = 105$)	100% ($n = 40$)	Unknown
	UC	96.9% ($n = 65$) 100% ($n = 19$)	91.1% ($n = 90$) 100% ($n = 15$)	82.1% ($n = 28$) 92% ($n = 12$)	Unknown
	FC	92.3% ($n = 65$) 100% ($n = 19$)	75.7% ($n = 90$) 93% ($n = 15$)	46.4% ($n = 28$) 58% ($n = 12$)	Unknown
FCEM/ ischemic myelopathy (24–26)	Amb		87.5% ($n = 301$)		43% ($n = 7$)
	UC		99% ($n = 40$) 70.4% ($n = 51$)		Unknown
	FC		97% ($n = 40$) 59.3% ($n = 51$)		Unknown

ANNPE, acute non-compressive nucleus pulposus extrusion; FCEM, fibrocartilagenous embolic myelopathy; Amb, ambulatory; UC, urinary continence; FC, fecal continence; NDPP, no deep pain perception; DPP, deep pain perception.

that did recover DPP recovered it within 4 weeks, another 30% by 12 weeks and one dog (8%) recovered it at 36 weeks (16). The prognosis for recovery of fecal and urinary continence in these dogs is not quite the same as recovery of independent walking. In dogs with Hansen type 1 IVDE that do recover DPP and walking, ∼40% do not recover normal fecal continence and 30–53% do not recover normal urinary continence (13, 16). While in the majority of cases, owners find the level of continence acceptable, it is important to note it might not be normal and accidents will be more likely to happen than prior to injury.

The prognosis for recovery in dogs with ANNPE and FCEM that present with paraplegia without DPP is considered poor and the majority of these dogs are euthanized within a week of injury. As such, these cases are scarcely reported in the literature and it is extremely difficult to establish what their prognosis would be if managed long term. There is a recent report of three dogs with ANNPE that recovered walking, but none recovered fecal continence and 2/3 remained urinary incontinent (22). A review of the literature on FCEM revealed seven dogs in this category for which long term outcomes were available; three of the seven recovered ambulation but there is no information on their continence (24).

TABLE 4 | Outcomes of dogs with different types of acute cervical intervertebral disc disease.

	Conservative	Surgery
Hansen type 1 IVDE (27–31)	72.8% ($n = 197$)	96.2% ($n = 786$)
HNPE (32–37)	98.5% ($n = 45$)	95.5% ($n = 97$)
ANNPE (38)	100% ($n = 12$)	
FCEM (39–41)	82.7% ($n = 39$)	

A successful outcome is recovery of ambulation and resolution of cervical pain. All severities of injury are considered together because studies have shown no effect of injury severity on outcome. Persistent cervical pain is the most common reason for treatment failure for Hansen type 1 IVDE. ANNPE, acute non-compressive nucleus pulposus extrusion; FCEM, fibrocartilaginous embolic myelopathy.

The prognosis of dogs with cervical lesions that lack DPP is difficult to report because so few dogs present with this severity of injury (29). This reflects the high mortality rate due to

hypoventilation and brady-arrhythmias in dogs with functionally complete cervical spinal cord injury (46).

There are special considerations in dogs that lack DPP when discussing prognosis. The first is the development of progressive myelomalacia (PMM) and the second is the prognosis for recovery of ambulation if DPP is not recovered. Progressive myelomalacia is a very important consideration due to the gravity of this condition, and the most important risk factor for development of this condition is injury severity (47, 48) (**Table 2**). Indeed between 9 and 33% of DPP negative dogs with Hansen type 1 IVDE can develop this condition, with most studies reporting a rate between 9 and 17.5%, and one study reporting a rate of 33% in French bulldogs specifically (11, 14–16, 47, 48). It is extremely important to warn owners of the potential development of PMM in paraplegic DPP negative dogs and to monitor for it both pre and post-operatively (49).

The prognosis of dogs developing independent walking without recovery of DPP has also been investigated in dogs that have suffered Hansen type 1 IVDE. Two studies have reported this specifically and in total 27/88 (31%) dogs that did not regain DPP recovered the ability to walk. In both studies the median time to walking was 9 months with a range of 2–28 months (13, 16). It is important to note that while these dogs did regain some ability to urinate and defecate, none had normal continence. The prognosis for recovery of walking in this population of dogs has been reported to be improved to a recovery rate of 59% with intensive physical rehabilitation (50).

Spinal Shock and Schiff Sherrington Posture

The presence of spinal shock and Schiff Sherrington posture at presentation have been described in dogs (9, 51). Spinal shock occurs much more frequently with FCEM and ANNPE due to the peracute onset of signs in these conditions (52). The prognostic significance of spinal shock has been evaluated and its presence is associated with the development of fecal incontinence in ANNPE but does not appear to affect recovery of ambulation (22, 52). The prognostic significance of the Schiff Sherrington posture has not been evaluated. The assumption of its importance is likely because it is easily recognized in severe thoracolumbar SCI cases, but the presence or lack thereof of DPP should be used as the indicator of prognosis in these cases, as previously discussed.

TABLE 5 | Summary of studies evaluating the relationship between signalment and prognosis.

	Study	Population of dogs	Conclusion
Breed	Ruddle et al. (12) retrospective	*N* = 250 all non-ambulatory with and without pain perception	No effect of breed on recovery speed or final outcome
	Castel et al. (47) retrospective	*N* = 197, all deep pain negative	No effect of breed on final outcome. Recovery speed not examined
Age	Ruddle et al. (12) retrospective	*N* = 250 all non-ambulatory with and without pain perception	No effect of age on recovery speed, increasing age worsens outcome
	Olby et al. (16) retrospective	*N* = 64, all deep pain negative	Increased age slows speed of recovery but not final outcome
	Davis and Brown (54) retrospective	*N* = 112, all non-ambulatory with pain perception	No effect of age on recovery speed or final outcome
	Jeffery et al. (15) prospective	*N* = 78, all deep pain negative	Age has no effect on final outcome. Recovery speed not examined
	Castel et al. (47) retrospective	*N* = 197, all deep pain negative	Age has no effect on final outcome. Recovery speed not examined
Weight	Ruddle et al. (12) retrospective	*N* = 250 all non-ambulatory with and without pain perception	Weight has no effect on speed of recovery or final outcome
	Olby et al. (16) retrospective	*N* = 64, all deep pain negative	Increased weight slows speed of recovery but not final outcome
	Davis and Brown (54) retrospective	*N* = 112, all non-ambulatory with pain perception	Weight has no effect on speed of recovery or final outcome
	Macias et al. (55). Retrospective, dogs >20 kg	*N* = 63, all severities of injury	Weight has no effect on final outcome when compared to other studies
	Bull et al. (56) retrospective	*N* = 238, all severities of injury, cervical and TL	Dogs >20, g had a worse outcome than dogs <20 kg
	Shaw et al. (57) retrospective	*N* = 121, split T3-L3 and L4-S3. ANNPE, HNPE, FCE and Hansen type 1 IVDE	Dogs weighing >15 kg had a worse outcome than dogs <15 kg
	Hillman et al. (30) retrospective	*N* = 32, cervical IVDE, all non-ambulatory	Dogs weighing <15 kg are 6× more likely to recover completely than dogs weighing >15 kg
	Cherrone et al. (29) retrospective	*N* = 190, cervical IVDE, small vs. large breed, all severities of injury	Large breed dogs more likely to have a recurrence. No effect on speed of recovery or final outcome

Unless noted specifically, all studies evaluated dogs with TL-IVDE treated with decompressive surgery.

Signalment

Breed and age affect the likelihood of a particular type, location and severity of IVDD. For example, younger dogs with acute TL IVDE present with more severe neurological signs (53) and acute TL IVDE occurs at a younger age, a more caudal site and a greater severity of neurological deficits in French bulldogs when compared with Dachshunds. As a result of the severity and tendency for a more caudal, lumbar location of their spinal cord injury, French bulldogs are more likely to develop PMM with rates as high as 33% in deep pain negative dogs (11). However, in these examples, the prognostic factors at presentation are severity of signs and location of disc extrusion.

Breed has been evaluated as a prognostic factor in several studies, but analysis is somewhat hampered by the overwhelming prevalence of Dachshunds (**Table 5**). Regardless, no study has found an effect of breed on prognosis. Similarly, sex does not alter prognosis (12, 15, 47). Several studies have evaluated the effect of age on prognosis and results have been somewhat contradictory, with one study showing increased age slows the speed of recovery in acute TL IVDE, but does not alter the final outcome, another suggesting it reduces the final recovery level, while others show no effect on final recovery (12, 16). A clear conclusion on the role of age as a prognostic factor cannot be drawn. Body weight has been investigated and results are similarly conflicting. One study on TL IVDE found increased body weight slowed speed of recovery and another found that dogs that weighed >20 kg had a worse outcome. Other studies found no effect of body weight on final outcome (12, 16, 47, 54–56). By contrast, when evaluating a population of non-ambulatory tetraparetic dogs, small breed dogs are six times more likely to have a successful recovery than large breeds (30).

Onset and Duration of Signs

Various studies have evaluated the speed of onset and the duration of signs, in particular the duration from onset of non-ambulatory status to surgical decompression (**Table 6**).

TABLE 6 | Summary of studies evaluating the relationship between speed of onset and duration of non-ambulatory status.

Parameter	Study	Population of dogs	Conclusion
Speed of onset	Scott et al. (14) retrospective	*N* = 34, all deep pain negative	Peracute onset (<1 h) has a negative effect on outcome. Speed of recovery not examined
	Ferreira et al. (58) retrospective	*N* = 71, all paraplegic deep pain positive	Peracute onset (<2 h) has a negative effect on outcome but not speed of recovery
	Olby et al. (16) retrospective	*N* = 64, all deep pain negative	No effect on final outcome. Speed of recovery not examined
	Jeffery et al. (15) prospective	*N* = 78 all without pain perception	No effect on final outcome. Speed of recovery not examined
	Castel et al. (47) retrospective	*N* = 197, all deep pain negative	No effect on final outcome. Speed of recovery not examined
Duration of non-ambulatory status	Scott et al. (14) retrospective	*N* = 34, all deep pain negative	No effect on final outcome or speed of recovery
	Ferreira et al. (58) retrospective	*N* = 71, all paraplegic deep pain positive	No effect on final outcome but duration of paralysis >6 days slows speed of recovery
	Davis and Brown (54) retrospective	*N* = 112, all non-ambulatory with pain perception	Increased duration of paralysis increased speed of recovery, no effect on final outcome
	Olby et al. (16) retrospective	*N* = 64, all deep pain negative	No effect on final outcome. Speed of recovery not examined
	Jeffery et al. (15) prospective	*N* = 78, all deep pain negative	No effect on final outcome. Speed of recovery not examined
	Castel et al. (47) retrospective	*N* = 197, all deep pain negative	No effect on final outcome. Speed of recovery not examined. >12 h duration, increased risk of PMM

TABLE 7 | Prognostic factors associated with location of herniated intervertebral disc.

Parameter	Study	Population of dogs	Conclusion
Location	Ruddle et al. (12) retrospective	*N* = 250, all non-ambulatory TL IVDE with and without pain perception	Location of disc herniation has no effect on outcome
	Cardy et al. (60) retrospective	*N* = 162, split between dachshunds and Cocker spaniels, all severities.	Caudal lumbar discs associated with less severe signs and therefore better outcome
	Shaw et al. (57) retrospective	*N* = 121, split T3–L3 and L4–S3. ANNPE, HNPE, FCE and Hansen type 1 IVDE	L4–S3 associated with worse outcome in terms of continence
	Castel et al. (47) retrospective	*N* = 197, all deep pain negative split T3–L3 and L4–S3.	No effect on recovery of ambulation

TABLE 8 | Summary of studies evaluating the relationship between biomarkers and outcome.

Biomarker	Study	Population studied	Findings
CMC CSF MBP and CK	Levine et al. (61)	$N = 54$, all non-ambulatory	CSF MBP < 3 ng/mL and CK < 38 U/L highly predictive for recovery
	Witsberger et al. (62)	$N = 54$, all non-ambulatory	
CMC CSF cytology	Srugo et al. (63)	$N = 54$, all non-ambulatory	% macrophages and macrophage/mononuclear ratio have high sensitivity and specificity for recovery
	Witsberger et al. (62)	$N = 54$, all non-ambulatory	Cytology has no relationship to outcome
CMC CSF tau	Roerig et al. (64)	$N = 51$, TL and Ce	CSF concentration has high specificity and sensitivity for recovery
CMC CSF inflammatory mediators	Taylor et al. (65)	$N = 39$, all non-ambulatory	CSF concentration of MCP-1 is negatively associated with outcome
Serum pNfH	Nishida et al. (66)	Paraplegic with ($n = 22$) and without deep pain ($n = 38$)	Serum concentration has high specificity but low sensitivity for recovery. Elevated in dogs with PMM
	Olby et al. (67)	$N = 31$, paraplegic without deep pain	Serum concentrations at time of presentation were not associated with recovery
Serum GFAP	Sato et al. (68) Olby et al. (67)	$N = 51$, non-ambulatory $N = 31$, paraplegic without deep pain	Presence has high sensitivity and specificity for recovery and for PMM
Serum S100beta	Olby et al. (67)	$N = 31$, paraplegic without deep pain	Serum concentrations at time of presentation were not associated with recovery

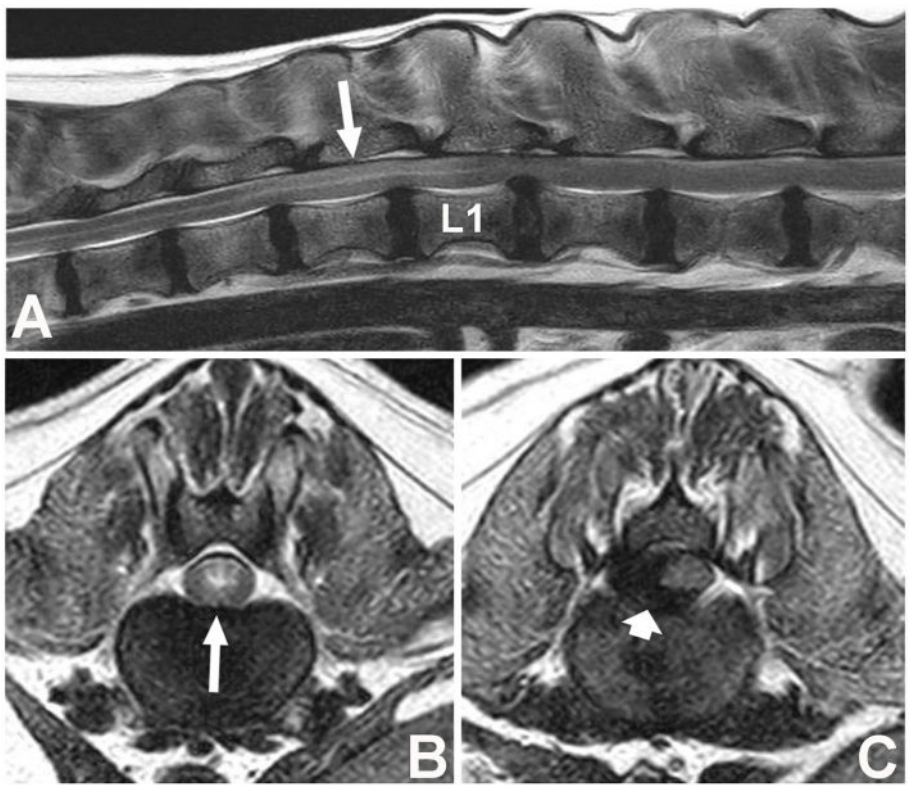

FIGURE 1 | Images of a female spayed, 6-year-old, mixed breed dog with an acute onset of non-ambulatory paraparesis and spinal pain **(A)**. Sagittal T2W image showing moderate ventral spinal cord compression secondary to intervertebral disc extrusion at L1–L2, with associated spinal cord hyperintensity spanning over T13 to L1 (long arrow) **(B)**. Transverse T2W image at T13–L1 showing spinal cord hyperintensity (long arrow) cranial to the compressive lesion **(C)**. Transverse T2 image showing lateralized spinal cord compression caused by a hypointense material between L1–2 (short arrow) found to be extruded disc material at surgery. L1 vertebral body is labeled (L1).

These studies are necessarily hampered by reliance on owner observations and periods during which pet dogs are not observed. In addition, the definition of the times can differ between studies with varying definition of onset (onset of ataxia, vs. pain for example), time to presentation or time to surgery as well as different populations of dog being examined. Large case cohorts are presented in **Table 6**. Overall, there is no consensus on an effect of speed of onset of signs or duration of signs on overall outcome, but there is some evidence that duration of signs might influence the speed of recovery. There is also some evidence that an interval of >12 h between onset of non-ambulatory status and surgical decompression increases the risk of PMM (47). Finally, there is evidence that delaying surgery until the day following presentation increases the risk of clinical deterioration from which the dog might not recover (59).

Location of Intervertebral Disc Extrusion

Several studies have compared outcome in dogs with Hansen type 1 IVDE and found no difference in dogs with T3–L3 vs. L4–S3 localization (**Table 7**). One study found a worse prognosis in dogs with lower motor neuron (LMN) signs of incontinence (57). Perhaps the most important prognostic detail in this category is the increased risk of development of PMM with discs located in the caudal lumbar vertebrae in dogs that are paraplegic with no pain perception (47, 48). When considering cervical IVDE, adverse events are more likely to occur with disc herniations at C7/T1 (28).

Blood and Cerebrospinal Fluid Biomarkers

While the prognosis for recovery can be established quite well in dogs with DPP prior to diagnostics and treatment, those that lack DPP pose a greater challenge. These dogs dichotomize into a group that shows a recovery comparable to those with incomplete lesions and a group in which there is no or limited recovery. These data show us that there is a profound floor effect when evaluating paraplegic DPP negative dogs and that this group includes dogs that have permanent interruption of conduction and those in which that interruption was temporary, perhaps representing conduction block due to edema, energy failure, etc. As a result, there have been many attempts to identify biomarkers that will allow differentiation of these dogs. Such attempts are plagued by the challenges of the influence of duration of injury and the ability to measure them at the time of presentation, to establish meaningful prognoses for owners. None have yet reached the point of clinical utility.

A plasma or serum biomarker would be ideal because it would allow prognostication prior to embarking on expensive advanced diagnostics, but they are somewhat removed from the central nervous system (CNS) compartment and so are likely to require very sensitive measuring techniques. Biomarkers that

TABLE 9 | Association of spinal cord hyperintensity detected on T2 weighted MRI in dogs with intervertebral disc extrusion and outcome.

Study	Design and population	Magnet strength	MR imaging changes	Conclusion	Comments
Ito et al. (79)	Retrospective 77 dogs Dogs paraplegic Blinded study Morphologic and morphometric	0.3 T	Only dogs with T2W SC hyperintensity equal to L2 vertebra	Hyperintensity better predictor of outcome than absence of DPP	Large variation between onset signs and MRI—median 7d
Levine et al. (53)	Retrospective 129 dogs with follow-up Ambulatory and non-ambulatory dogs Blinded study Morphologic and morphometric	1.0 T for the majority of cases	Any degree of T2W SC hyperintensity	Direct association between the length ratio of hyperintensity with long-term functional outcome	The large variation in injury severity makes comparison with other studies challenging
Boekhoff et al. (80)	Retrospective 63 dogs All dogs paraplegic Blinded study Morphologic and morphometric	1.0 T	SC T2W hyperintensities ranging from half of L2 vertebra to >2 times L2	Association between extent of T2 hyperintensity with delayed ambulation, not statistically significant	Large variation between onset signs and MRI—62% between 2 and 7 days
Wang-Leandro et al. (81)	Prospective 35 dogs All dogs paraplegic Blinded study Morphologic and morphometric	3.0 T	SC T2W hyperintensities assessed in sagittal plane using L2 vertebra as reference	Length of SC T2 hyper-intensity had no association with motor functional recovery	Only 2 dogs were enrolled with >7 days of onset of signs
Otamendi et al. (82)	Retrospective 47 dogs	3.0 T	SC T2W hyperintensities assessed in sagittal plane using L2 vertebra as reference	No association between T2 hyperintensity and recovery of motor function or PMM	Abstract only

SC, spinal cord; PMM, progressive myelomalacia.

show the most promise are CNS structural proteins, glial fibrillary acidic protein (GFAP) and phosphorylated neurofilament heavy chain (pNfH) (**Table 8**) (66–68). Both can be measured in the plasma and serum using ELISA and provide some insight into the severity of CNS injury. Of the two, GFAP is the most discriminating at time of injury, predicting both recovery of ambulation and development of PMM with an accuracy of >80%. Serum pNfH concentrations are more variable at the time of injury and S100β was found to be less useful. Unfortunately, at this time there is no rapid point of care test available, but they have proven useful as a covariate of injury severity in clinical trials (69).

Evaluation of CSF biomarkers takes the clinician closer to the CNS, but as a result such tests are more invasive and require general anesthesia. Markers evaluated include cytology, myelin basic protein (MBP) with or without creatine kinase (CK), tau, glutamate, matrix metalloproteinase-9 (MMP-9) and inflammatory cytokines (61–65). Some studies have conflicting results, but all are summarized in **Table 8** and MBP and CK CSF concentrations in particular are both highly predictive of return of ambulatory function.

Imaging and Prognosis IVDD

Myelography

The first imaging modality used to prognosticate cases of IVDE was myelography. It was based on the extension of an intramedullary pattern, which was interpreted as indirect evidence of the severity and extent of spinal cord swelling (70). Spinal cord swelling, calculated as the ratio of the length of the loss of the dorsal and ventral contrast columns to the second lumbar vertebra (spinal cord swelling: L2 ratio), was correlated with a poor prognosis when it was found to be five or more vertebral bodies. However, a subsequent study could not confirm these findings (14). In the latter study, the ratio for dogs with a successful outcome was 1.7, compared to 2.0 for those with an unsuccessful outcome. Only two dogs had intramedullary pattern longer than five bodies and both dogs recovered.

Myelographic studies of dogs with acute non-compressive nucleus pulposus extrusion (ANNPE) demonstrated an intramedullary pattern and an additional extradural pattern was seen in approximately half of the dogs. The degree of spinal cord swelling was not associated with severity of clinical signs or outcome (71).

An extensive intramedullary pattern with evidence of contrast medium infiltration into the spinal cord has been reported as an indication of progressive myelomalacia (PMM) (72). Infiltration of contrast within the spinal cord parenchyma, however, is not pathognomonic for PMM since it can also be iatrogenic or represent other intramedullary lesions such as syringohydromyelia (72, 73).

Magnetic Resonance Imaging

The utility of MRI for diagnostic purposes has been very well-defined and characterized. Its ability to serve as a biomarker is not as clear, although several studies have proposed imaging markers as prognostic indicators. As MRI is routinely acquired as part of diagnostic work-up of IVDE cases, the identification of reliable imaging markers identified on MRI would be invaluable for clinicians and owners.

TABLE 10 | Association of MRI abnormalities associated with progressive myelomalacia (PMM) in dogs with intervertebral disc extrusion.

Study	Design and population	Magnet strength	MR imaging changes	Conclusion	Comments
Okada et al. (83)	Retrospective 12 dogs; five confirmed, seven presumptive Time between onset signs and MRI: median 3.7 d	Low field (0.4 and 0.5 T) for 11/12 dogs 1.5 T for one dog	Length of T2W SC hyperintensity equal L2 vertebra ranged from 6 to 20 times L2	Hyperintensity longer than six times body of L2 characteristic of PMM	Small sample size and only five dogs confirmed
Gilmour et al. (84)	Retrospective five dogs with PMM—necropsy confirmed Time between onset signs and MRI–median 2 ds	1.5 T	T2W length SC hyperintensity/L2: 2.3 and 1.2 (mean, median) Loss of CSF signal ≥ L2 (ratio CSF:L2) on HASTE equal to 10.7 and 8.9 (mean and median)	A ratio of 7.4 of loss of CSF signal ≥ L2 on HASTE had a sensitivity of 100% and specificity of 75%.	Small sample size
Castel et al. (85)	Retrospective 20 dogs with PMM and MRI Number necropsy confirmed not reported Time frame between onset signs and MRI not reported	1.5T	T2W SC hyperintensity longer than six times L2 seen in 45% dogs Loss of CSF signal ≥ 7.4× L2 (ratio CSF:L2) on HASTE seen in 85% dogs	Loss of CSF signal equal of longer 7.4× L2 more reliable than T2 hyperintensity	The three dogs with a ratio CSF: L2 HASTE <7.4 were imaged within 12–24 h following onset paraplegia
Balducci et al. (48)	Retrospective 13 dogs with MRI–none necropsy confirmed	0.2 T	-T2W SC hyperintensity longer than 4.57 times L2 seen in 84.6% dogs	Dogs with hyperintensity > 4.57 times L2 were 17.2 times more likely to develop PMM	Dogs may not show T2 SC hyperintensity when imaged <24 h after onset of paraplegia and still develop PMM

Spinal cord hyperintensity on T2W images has been the most widely investigated parameter (**Figure 1**). This spinal cord (SC) hyperintensity identified on T2W images has been associated with necrosis, myelomalacia, intramedullary hemorrhage, inflammation, and edema (74–76). Without differentiating the pathologic process more specifically, T2 hyperintensity has been shown to correlate well with the severity of neurologic signs at presentation in dogs with IVDE (53, 77, 78). Its utility as a prognostic indicator is less clear. Even though the first report of the utility of spinal cord hyperintensity indicated that extension of the area of T2W spinal cord hyperintensity on low-field MRI was a reliable predictor of outcome, even more reliable than the absence of deep pain perception (79), these findings could not be reproduced in other studies, primarily those using high-field MRI (**Table 9**). Use of high-field MRI leads to increase in signal-to noise ratio and consequently to a change in image resolution; therefore, mild intramedullary hyperintensities in sagittal T2W sequences may be more frequently evident using high field magnetic fields compared to low field ones.

Identification of MRI features suggestive of progressive myelomalacia is crucial for prognostic purposes since its identification indicates an abysmal prognosis. The length of spinal cord T2W hyperintensity and the length of intramedullary pattern reflected as loss of CSF signal on HASTE/MR myelography sequences have been used and are presented on **Table 10**. A recent 3T MRI study proposed that intramedullary hypointensity on T2W images was associated with PMM (86).

High field MRI changes have been associated with outcome in dogs with ANNPE (22, 38) and FCEM (39). A larger lesion on transverse images, quantified as a greater percentage cross sectional area of the spinal cord, has been considered the most useful MRI variable to predict the short- and long-term outcome of dogs with ANNPE and FCEM (22, 38, 39). In ANNPE, dogs with a smaller lesion had a shorter interval to unassisted ambulation. In contrast, a percentage cross sectional area that equals or exceeds 90% of the spinal cord had a 92% chance of having an unsuccessful long-term outcome (38), and a lesion exceeding 40% of the transverse area has been associated with an increased likelihood of long-term urinary and fecal incontinence (22). However, a low-field MRI study of 21 dogs with ANNPE found no association between any MRI parameter with outcome (71).

Advanced MRI techniques have been proposed to increase reliability of MRI as biomarker. A recent study proposed a semi-automated assessment of SC signal changes aiming to minimize interobserver variability (86). Diffusion tensor imaging (DTI) has been used in dogs with naturally occurring spinal cord injury secondary to IVDE (81, 87–89). The spinal cord, primarily white matter, microstructural changes are captured through quantification using DTI techniques. As such DTI is able to detect abnormal SC areas that appear macroscopically normal on T2W sequences. Specific DTI parameters, including tractography, have the potential to serve as prognostic biomarkers, although no specific parameter has been identified (81, 87–89). More information regarding DTI in IVDE can be found in a companion article entitled Diagnostic imaging in intervertebral disc disease.

Electrophysiological Testing

Attempts have been made to use electrophysiological testing to quantify injury severity and predict prognosis, both at the time of injury and in chronically paralyzed dogs. The majority of this work has been completed in dogs with thoracolumbar IVDE. Two different approaches have been used. The first is to evaluate the descending pathways using magnetic stimulation of the motor cortex (90) and the second is to evaluate the ascending pathways within the spinal cord and projecting to the brain using somatosensory evoked potentials (SSEP) (91).

Motor Evoked Potentials

Transcranial magnetic motor evoked potentials (TMMEP) can be elicited reliably in dogs under sedation (92, 93). However, they are extremely sensitive to spinal cord injury and are lost completely in dogs that are paraplegic (94, 95). With less severe injuries, latency increases and amplitude decreases, but these values do not discriminate initial severity as well as clinical assessment and evaluation of MEPs at time of presentation does not provide prognostic information (94, 95). There has been interest in the utility of repeated TMMEP evaluation in dogs that were paraplegic at presentation. Dogs that show recovery of pain perception and motor function recover TMMEPs, leading to the suggestion that this tool can be used to complement assessment of recovery (96). Two groups have evaluated the presence and latency of TMMEPs in dogs that do not regain deep pain perception, and reached different conclusions with one group finding an association between TMMEP presence and recovery or walking, and the other failing to find this association (97, 98). At this time, there is no evidence that evaluation of TMMEP at time of injury can provide prognostic information in acute spinal cord injury due to IVDE.

Somatosensory Evoked Potentials

Somatosensory evoked potentials can be elicited by stimulation of a peripheral nerve in a pelvic or thoracic limb (99). Needles are introduced percutaneously to the level of the interarcuate ligament to record from different levels of the spinal cord and subcutaneously to record over the sensory cortex. Various parameters can be recorded including presence or absence of a waveform, latency, amplitude and duration of the potential at the level of the sensory cortex, the conduction velocity of ascending volleys along the spinal cord, particularly across a lesion, the presence and location of conduction block, and the presence and amplitude of an injury potential (100–103). Early work suggested that lack of recordable cortical evoked potential was associated with failure to recover ambulation in dogs with TL IVDE (100). Another study evaluating conduction velocity and amplitude of spinal evoked potentials recorded at T10/11 found that a ratio of conduction velocity to amplitude was predictive of outcome (101). Later studies did not come to the same conclusions, and combined various parameters to discriminate initial injury severity to the level of clinical evaluation (102). The location of conduction block can be evaluated using the evoked injury

potential and the distance between conduction block and site of compression might contribute useful information but this has not been investigated further (103). SSEPs have also been evaluated in chronically deep pain negative dogs, but results are conflicting and are not of prognostic utility (97, 98). Currently, there is no clear evidence that prognosis can be established in acute IVDE using SSEPs.

CONCLUSIONS

Understanding the prognosis of spinal cord injury secondary to IVDE is important for client education, optimal patient management and clinical trial design and execution. The prognosis varies with type of IVDE and is influenced by treatment choices. Severity of initial clinical presenting signs is the most useful guide to prognosis at the time of presentation. Biomarkers within the blood, CSF and on imaging can also help to predict outcome. Understanding time to recovery and differentiating between motor and autonomic recovery (continence) can provide invaluable information for veterinarians as they manage dogs that have suffered a spinal cord injury. The manner in which these data are presented and discussed with the owner is a critical part of patient care and it is vital both that accurate information is provided and that owners understand the nature of that information as it relates to their dog. As we move into an era with increased availability of bedside tests of injury severity, and improved accuracy of imaging prognostication, it is likely that patient stratification will improve our ability to perform well-designed clinical trials and to optimize patient care on an individual basis.

AUTHOR CONTRIBUTIONS

NO, RdC, JL, VS, and CANSORT SCI contributed to conception of the study. NO, RdC, JL, and VS designed the study. NO wrote the first draft. RdC wrote a subsection. All authors reviewed, revised, and approved the submitted version.

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SUPPLEMENTARY MATERIAL

The Supplementary Material for this article can be found online at: https://www.frontiersin.org/articles/10.3389/fvets.2020.596059/full#supplementary-material

Supplementary Video 1 | Assessment of deep pain perception in an ∼3-year-old male castrated mix breed dog. He is paraplegic at time of testing. Pressure is applied over the lateral and medial digits of the left hind and there is a withdrawal reflex but no behavioral acknowledgment of the stimulus. However, when pressure is applied to digits of the right foot the dog withdraws the limb and rapidly sits up and looks around, indicating that he has sensation in this foot.

REFERENCES

1. Bergknut N, Auriemma E, Wijsman S, Voorhout G, Hagman R, Lagerstedt A-S, et al. Evaluation of intervertebral disk degeneration in chondrodystrophic and nonchondrodystrophic dogs by use of Pfirrmann grading of images obtained with low-field magnetic resonance imaging. *Am J Vet Res*. (2011) 72:893–8. doi: 10.2460/ajvr.72.7.893
2. Rossi G, Stachel AF, Lynch A, Olby NJ. Intervertenral disc disease and aortic thromboembolism are the most common causes of acute paralysis in DOgs and Cats presenting to an emergency clinic. *Vet Rec*. (2020). doi: 10.1136/vr.105844. [Epub ahead of print].
3. Moore SA, Early PJ, Hettlich BF. Practice patterns in the management of acute intervertebral disc herniation in dogs. *J Sm Anim Pract*. (2016) 57:409–15. doi: 10.1111/jsap.12496

4. DeVries Z, Hoda M, Rivers CS, Maher A, Wai E, Moravek D, et al. Development of an unsupervised machine learning algorithm for the prognostication of walking ability in spinal cord injury patients. *Spine J*. (2020) 20:213–24. doi: 10.1016/j.spinee.2019.09.007
5. Clark GM. Prognostic factors versus predictive factors: examples from a clinical trial of erlotinib. *Mol Onc*. (2007) 1:406–12. doi: 10.1016/j.molonc.2007.12.001
6. Fawcett JW, Curt A, Steeves JD, Coleman WP, Tuszynski MH, Lammertse D, et al. Guidelines for the conduct of clinical trials for spinal cord injury as developed by the ICCP panel: spontaneous recovery after spinal cord injury and statistical power needed for therapeutic clinical trials. *Spinal Cord*. (2007) 45:190–205. doi: 10.1038/sj.sc.3102007
7. Sharp N, Wheeler SJ. *Thoracolumbar Disc Disease. Small Animal Spinal Disorders*. 2nd ed. London: Mosby Ltd (2005). doi: 10.1016/B978-0-7234-3209-8.50012-1
8. Levine JM, Ruaux CG, Bergman RL, Coates JR, Steiner JM, Williams DA. Matrix metalloproteinase-9 activity in the cerebrospinal fluid and serum of dogs with acute spinal cord trauma from intervertebral disk disease. *Am J Vet Res*. (2006) 67:283–7. doi: 10.2460/ajvr.67.2.283
9. Granger N, Carwardine D. Acute spinal cord injury: tetraplegia and paraplegia in small animals. *Vet Clin North Am Small Anim Pract*. (2014) 44:1131–56. doi: 10.1016/j.cvsm.2014.07.013
10. Olby N, Harris T, Burr J, Muñana K, Sharp N, Keene B. Recovery of pelvic limb function in dogs following acute intervertebral disc herniations. *J Neurotrauma*. (2004) 21:49–59. doi: 10.1089/089771504772695940
11. Aikawa T, Shibata M, Asano M, Hara Y, Tagawa M, Orima H. A comparison of thoracolumbar intervertebral disc extrusion in French bulldogs and dachshunds and association with congenital vertebral anomalies. *Vet Surg*. (2014) 43:301–7. doi: 10.1111/j.1532-950X.2014.12102.x
12. Ruddle TL, Allen DA, Schertel ER, Barnhart MD, Wilson ER, Lineberger JA, et al. Outcome and prognostic factors in non-ambulatory hansen type I intervertebral disc extrusions: 308 cases. *Vet Comp Ortho Trauma*. (2006) 19:29–34. doi: 10.1055/s-0038-1632970
13. Aikawa T, Fujita H, Kanazono S, Shibata M, Yoshigae Y. Long-term neurologic outcome of hemilaminectomy and disk fenestration for treatment of dogs with thoracolumbar intervertebral disk herniation: 831 cases (2000–2007). *J Am Vet Med Assoc*. (2012) 241:1617–26. doi: 10.2460/javma.241.12.1617
14. Scott HW, McKee WM. Laminectomy for 34 dogs with thoracolumbar intervertebral disc disease and loss of deep pain perception. *J Small Anim Pract*. (1999) 40:417–22. doi: 10.1111/j.1748-5827.1999.tb03114.x
15. Jeffery ND, Barker AK, Hu HZ, Alcott CJ, Kraus KH, Scanlin EM, et al. Factors associated with recovery from paraplegia in dogs with loss of pain perception in the pelvic limbs following intervertebral disk herniation. *J Am Vet Med Assoc*. (2016) 248:386–94. doi: 10.2460/javma.248.4.386
16. Olby N, Levine J, Harris T, Muñana K, Skeen T, Sharp N. Long-term functional outcome of dogs with severe injuries of the thoracolumbar spinal cord: 87 cases (1996-2001). *J Am Vet Med Assoc*. (2003) 222:762–9. doi: 10.2460/javma.2003.222.762
17. Langerhuus L, Miles J. Proportion recovery and times to ambulation for non-ambulatory dogs with thoracolumbar disc extrusions treated with hemilaminectomy or conservative treatment: a systematic review and meta-analysis of case-series studies. *Vet J*. (2017) 220:7–16. doi: 10.1016/j.tvjl.2016.12.008
18. Davies JV, Sharp NJH. A comparison of conservative treatment and fenestration for thoracolumbar intervertebral disc disease in the dog. *J Sm Anim Pract*. (1983) 24:721–9. doi: 10.1111/j.1748-5827.1983.tb00360.x
19. Levine JM, Levine GJ, Johnson SI, Kerwin SC, Hettlich BF, Fosgate GT. Evaluation of the success of medical management for presumptive thoracolumbar intervertebral disk herniation in dogs. *Vet Surg*. (2007) 36:482–91.
20. Baumhardt R, Ripplinger A, Aiello G, Schwab ML, Ferrarin DA, Wrzesinski MR, et al. Clinical management of dogs with presumptive diagnosis of thoracolumbar intervertebral disc disease: 164 cases (2006–2017). *Pesq Vet Bras*. (2020) 40:55–60. doi: 10.1590/1678-5150-pvb-6067
21. Mann FA, Wagner-Mann CC, Dunphy ED, Ruben DS, Rochat MC, Bartels KE. Recurrence rate of presumed thoracolumbar intervertebral disc disease in ambulatory dogs with spinal hyperpathia treated with anti-inflammatory drugs: 78 cases (1997–2000). *J Vet Emer Crit*. (2007) 17:53–60. doi: 10.1111/j.1476-4431.2006.00195.x
22. Mari L, Behr S, Shea A, Dominguez E, Ricco C, Alcoverro E, et al. Predictors of urinary or fecal incontinence in dogs with thoracolumbar acute non-compressive nucleus pulposus extrusion. *J Vet Intern Med*. (2019) 33:2693–700. doi: 10.1111/jvim.15626
23. McKee WM, Downes CJ, Pink JJ, Gemmill TJ. Presumptive exercise-associated peracute thoracolumbar disc extrusion in 48 dogs. *Vet Rec*. (2010) 166:523–8. doi: 10.1136/vr.b4823
24. Bartholomew KA, Stover KE, Olby NJ, Moore SA. Clinical characteristics of canine fibrocartilaginous embolic myelopathy (FCE): a systematic review of 393 cases (1973–2013). *Vet Record*. (2016) 179:650–656. doi: 10.1136/vr.103863
25. Mari L, Behr S, Shea A, Dominguez E, Johnson PJ, Ekiri A, et al. Outcome comparison in dogs with a presumptive diagnosis of thoracolumbar fibrocartilaginous embolic myelopathy and acute non-compressive nucleus pulposus extrusion. *Vet Rec*. (2017) 181:293–3. doi: 10.1136/vr.104090
26. Fenn J, Drees R, Volk HA, De Decker S. Comparison of clinical signs and outcomes between dogs with presumptive ischemic myelopathy and dogs with acute noncompressive nucleus pulposus extrusion. *J Am Vet Med Assoc*. (2016) 249:767–75. doi: 10.2460/javma.249.7.767
27. Levine JM, Levine GL, Johnson SI, Kdermin SC, Hettlich BF, Fosgate GT. Evaluation of the success of medical management for presumptive cervical intervertebral disk herniation in dogs. *Vet Surg*. (2007) 36:492–9. doi: 10.1111/j.1532-950X.2007.00296.x
28. Rossmeisl JH, White C, Pancotto TE, Bays A, Henao-Guerrero PN. Acute adverse events associated with ventral slot decompression in 546 dogs with cervical intervertebral disc disease. *Vet Surg*. (2013) 42:795–806. doi: 10.1111/j.1532-950X.2013.12039.x
29. Cherrone KL, Dewey CW, Coates JR, Bergman RL. A retrospective comparison of cervical intervertebral disk disease in nonchondrodystrophic large dogs versus small dogs. *J Am Anim Hosp Assoc*. (2004) 40:316–20. doi: 10.5326/0400316
30. Hillman RB, Kengeri SS, Waters DJ. Reevaluation of predictive factors for complete recovery in dogs with nonambulatory tetraparesis secondary to cervical disk herniation. *J Am Anim Hosp Assoc*. (2009) 45:155–63. doi: 10.5326/0450155
31. Russell SW, Griffiths RC. Recurrence of cervical disc syndrome in surgically and conservatively treated dogs. *J Am Vet Med Assoc*. (1968) 153:1412–7.
32. Neßler J, Flieshardt C, Tünsmeyer J, Dening R, Tipold A. Comparison of surgical and conservative treatment of hydrated nucleus pulposus extrusion in dogs. *J Vet Intern Med*. (2018) 32:1989–95. doi: 10.1111/jvim.15304
33. Borlace T, Gutierrez-Quintana R, Taylor-Brown FE, De Decker S. Comparison of medical and surgical treatment for acute cervical compressive hydrated nucleus pulposus extrusion in dogs. *Vet Rec*. (2017) 181:625–5. doi: 10.1136/vr.104528
34. Falzone C. Canine acute cervical myelopathy: hydrated nucleus pulposus extrusion or intraspinal discal cysts? *Vet Surg*. (2017) 46:376–80. doi: 10.1111/vsu.12631
35. Manunta ML, Evangelisti MA, Bergknut N, Grinwis GCM, Ballocco I, Meij BP. Hydrated nucleus pulposus herniation in seven dogs. *Vet J*. (2015) 203:342–4. doi: 10.1016/j.tvjl.2014.12.027
36. Beltran E, Dennis R, Doyle V, De Stefani A, Holloway A, De Risio L. Clinical and magnetic resonance imaging features of canine compressive cervical myelopathy with suspected hydrated nucleus pulposus extrusion. *J Sm Anim Pract*. (2012) 53:101–7. doi: 10.1111/j.1748-5827.2011.01166.x
37. Dolera M, Malfassi L, Marcarini S, Mazza G, Sala M, Carrara N, et al. Hydrated nucleus pulposus extrusion in dogs: correlation of magnetic resonance imaging and microsurgical findings. *Acta Vet Scand*. (2015) 57:58. doi: 10.1186/s13028-015-0151-x
38. De Risio L, Adams V, Dennis R, McConnell FJ. Association of clinical and magnetic resonance imaging findings with outcome in dogs with presumptive acute noncompressive nucleus pulposus extrusion: 42 cases (2000-2007). *J Am Vet Med Assoc*. (2009) 234:495–504. doi: 10.2460/javma.234.4.495
39. De Risio L, Adams V, Dennis R, McConnell FJ, Platt SR. Association of clinical and magnetic resonance imaging findings with outcome in dogs

suspected to have ischemic myelopathy: 50 cases (2000–2006). *J Am Vet Med Assoc*. (2008) 233:129–35. doi: 10.2460/javma.233.1.129
40. Gandini G, Cizinauskas S, Lang J, Fatzer R, Jaggy A. Fibrocartilaginous embolism in 75 dogs: clinical findings and factors influencing the recovery rate. *J Sm Anim Pract*. (2003) 44:76–80. doi: 10.1111/j.1748-5827.2003.tb00124.x
41. Hawthorne JC, Wallace LJ, Fenner WR, Waters DJ. Fibrocartilaginous embolic myelopathy in miniature schnauzers. *Am Anim Hosp Assoc*. (2001) 37:374–83. doi: 10.5326/15473317-37-4-374
42. Kube S, Owen T, Hanson S. Severe respiratory compromise secondary to cervical disk herniation in two dogs. *J Am Anim Hosp Assoc*. (2003) 39:513–7. doi: 10.5326/0390513
43. Posner LP, Mariani CL, Swanson C, Asakawa M, Campbell N, King AS. Perianesthetic morbidity and mortality in dogs undergoing cervical and thoracolumbar spinal surgery. *Vet Anaesth Analg*. (2016) 41:137–44. doi: 10.1111/vaa.12127
44. Beal MW, Paglia DT, Griffin GM, Hughes D, King LG. Ventilatory failure, ventilator management, and outcome in dogs with cervical spinal disorders: 14 cases (1991–1999). *J Am Vet Med Assoc*. (2001) 218:1598–602. doi: 10.2460/javma.2001.218.1598
45. Stauffer JL, Gleed RD, Short CE, Erb HN, Schukken YH. Cardiac dysrhythmias during anesthesia for cervical decompression in the dog. *Am J Vet Res*. (1988) 49:1143–6.
46. Clark DM. An analysis of intraoperative and early postoperative mortality associated with cervical spinal decompressive surgery in the dog. *J Am Anim Hosp Assoc*. (1986) 22:739–44.
47. Castel A, Olby NJ, Ru H, Mariani CL, Muñana KR, Early PJ. Risk factors associated with progressive myelomalacia in dogs with complete sensorimotor loss following intervertebral disc extrusion: a retrospective case-control study. *BMC Vet Res*. (2019) 15:1–9. doi: 10.1186/s12917-019-2186-0
48. Balducci F, Canal S, Contiero B, Bernardini M. Prevalence and risk factors for presumptive ascending/descending myelomalacia in dogs after thoracolumbar intervertebral disk herniation. *J Vet Intern Med*. (2017) 353:63–7. doi: 10.1111/jvim.14656
49. Muguet-Chanoit AC, Olby NJ, Lim JH, Gallagher R, Niman Z, Dillard S, et al. The cutaneous trunci muscle reflex: a predictor of recovery in dogs with acute thoracolumbar myelopathies caused by intervertebral disc extrusions. *Vet Surg*. (2012) 41:200–6. doi: 10.1111/j.1532-950X.2011.00921.x
50. Gallucci A, Dragone L, Menchetti M, Gagliardo T, Pietra M, Cardinali M, et al. Acquisition of involuntary spinal locomotion (spinal walking) in dogs with irreversible thoracolumbar spinal cord lesion: 81 dogs. *J Vet Intern Med*. (2017) 222:762–6. doi: 10.1111/jvim.14651
51. Smith PM, Jeffery ND. Spinal shock–comparative aspects and clinical relevance. *J Vet Intern Med*. (2005) 19:788–93. doi: 10.1111/j.1939-1676.2005.tb02766.x
52. Full AM, Heller HLB, Mercier M. Prevalence, clinical presentation, prognosis, and outcome of 17 dogs with spinal shock and acute thoracolumbar spinal cord disease. *J Vet Emerg Crit*. (2015) 26:412–8. doi: 10.1111/vec.12438
53. Levine JM, Fosgate GT, Chen AV, Rushing R, Nghiem PP, Platt SR, et al. Magnetic resonance imaging in dogs with neurologic impairment due to acute thoracic and lumbar intervertebral disk herniation. *J Vet Intern Med*. (2009) 23:1220–6. doi: 10.1111/j.1939-1676.2009.0393.x
54. Davis GJ, Brown DC. Prognostic indicators for time to ambulation after surgical decompression in nonambulatory dogs with acute thoracolumbar disk extrusions: 112 cases. *Vet Surg*. (2002) 31:513–8. doi: 10.1053/jvet.2002.36015
55. Macias C, McKee WM, May C, Innes JF. Thoracolumbar disc disease in large dogs: a study of 99 cases. *J Sm Anim Pract*. (2002) 43:439–46. doi: 10.1111/j.1748-5827.2002.tb00010.x
56. Bull C, Fehr M, Tipold A. Bandscheibenvorfälle beim hund: retrospektive studie über den klinischen verlauf von 238 hunden (2003–2004). *Berl Munch Tierarztl Wochenschr*. (2008) 121:10–21. doi: 10.2376/0005-9366-121-159
57. Shaw TA, De Risio L, Laws EJ, Rose JH, Harcourt-Brown TR, Granger N. Prognostic factors associated with recovery of ambulation and urinary continence in dogs with acute lumbosacral spinal cord injury. *J Vet Intern Med*. (2017) 31:825–31. doi: 10.1111/jvim.14702
58. Ferreira AJA, Correia JHD, Jaggy A. Thoracolumbar disc disease in 71 paraplegic dogs: influence of rate of onset and duration of clinical signs on treatment results. *J Sm Anim Pract*. (2002) 43:158–63. doi: 10.1111/j.1748-5827.2002.tb00049.x
59. Martin S, Liebel FX, Fadda A, Lazzerini K, Harcourt-Brown T. Same-day surgery may reduce the risk of losing pain perception in dogs with thoracolumbar disc extrusion. *J Sm Anim Pract*. (2020) 51:598–7. doi: 10.1111/jsap.13147
60. Cardy TJA, Tzounos CE, Volk HA, De Decker S. Clinical characterization of thoracolumbar and lumbar intervertebral disk extrusions in English cocker spaniels. *J Am Vet Med Assoc*. (2016) 248:405–12. doi: 10.2460/javma.248.4.405
61. Levine GJ, Levine JM, Witsberger TH, Kerwin SC, Russell KE, Suchodolski J, et al. Cerebrospinal fluid myelin basic protein as a prognostic biomarker in dogs with thoracolumbar intervertebral disk herniation. *J Vet Intern Med*. (2010) 24:890–6. doi: 10.1111/j.1939-1676.2010.0531.x
62. Witsberger TH, Levine JM, Fosgate GT, Slater MR, Kerwin SC, Russell KE, et al. Associations between cerebrospinal fluid biomarkers and long-term neurologic outcome in dogs with acute intervertebral disk herniation. *J Am Vet Med Assoc*. (2012) 240:555–62. doi: 10.2460/javma.240.5.555
63. Srugo I, Aroch I, Christopher MM, Chai O, Goralnik L, Bdolah-Abram T, et al. Association of cerebrospinal fluid analysis findings with clinical signs and outcome in acute nonambulatory thoracolumbar disc disease in dogs. *J Vet Intern Med*. (2011) 25:846–55. doi: 10.1111/j.1939-1676.2011.0739.x
64. Roerig A, Carlson R, Tipold A, Stein VM. Cerebrospinal fluid tau protein as a biomarker for severity of spinal cord injury in dogs with intervertebral disc herniation. *Vet J*. (2013) 197:253–8. doi: 10.1016/j.tvjl.2013.02.005
65. Taylor AR, Welsh CJ, Young C, Spoor E, Kerwin SC, Griffin JF, et al. Cerebrospinal fluid inflammatory cytokines and chemokines in naturally occurring canine spinal cord injury. *J Neurotrauma*. (2014) 31:1561–9. doi: 10.1089/neu.2014.3405
66. Nishida H, Nakayama M, Tanaka H, Kamishina H, Izawa T, Hatoya S, et al. Evaluation of serum phosphorylated neurofilament subunit nf-h as a prognostic biomarker in dogs with thoracolumbar intervertebral disc herniation. *Vet Surg*. (2014) 43:289–93. doi: 10.1111/j.1532-950X.2014.12144.x
67. Olby NJ, Lim JH, Wagner N, Zidan N, Early PJ, Mariani CL, et al. Time course and prognostic value of serum GFAP, pNFH, and S100β concentrations in dogs with complete spinal cord injury because of intervertebral disc extrusion. *J Vet Intern Med*. (2019) 33:726–34. doi: 10.1111/jvim.15439
68. Sato Y, Shimamura S, Mashita T, Kobayashi S, Okamura Y, Katayama M, et al. Serum glial fibrillary acidic protein as a diagnostic biomarker in dogs with progressive myelomalacia. *J Vet Med Sci*. (2013) 75:949–53. doi: 10.1292/jvms.12-0483
69. Zidan N, Fenn J, Griffith E, Early PJ, Mariani CL, Muñana KR, et al. The effect of electromagnetic fields on post-operative pain and locomotor recovery in dogs with acute, severe thoracolumbar intervertebral disc extrusion: a randomized placebo-controlled, prospective clinical trial. *J Neurotrauma*. (2018) 35:1726–36. doi: 10.1089/neu.2017.5485
70. Duval J, Dewey C, Roberts R, Aron D. Spinal cord swelling as a myelographic indicator of prognosis: a retrospective study in dogs with intervertebral disc disease and loss of deep pain perception. *Vet Surg*. (1996) 25:6–12. doi: 10.1111/j.1532-950X.1996.tb01371.x
71. Ros C, de la Fuente C, Ródenas S, Novellas R, Viu J, Aã Or S. Myelographic and low-field magnetic resonance imaging findings in dogs with presumptive acute hydrated non-compressive nucleus pulposus extrusion. *Vet Rec*. (2017) 181:594–9. doi: 10.1136/vr.104201
72. Lu D, Lamb CR, Targett MP. Results of myelography in seven dogs with myelomalacia. *Vet Rad US*. (2002) 43:326–30. doi: 10.1111/j.1740-8261.2002.tb01012.x
73. Kirberger RM, Wrigley RH. Myelography in the dog: review of patients with contrast medium in the central canal. *Vet Rad US*. (1993) 34:253–8. doi: 10.1111/j.1740-8261.1993.tb01573.x
74. Schouman-Claeys E, Frija G, Cuenod CA, Begon D, Paraire F, Martin V. MR imaging of acute spinal cord injury: results of an experimental study in dogs. *AJNR Am J Neuroradiol*. (1990) 11:959–65.

75. Flanders AE, Spettell CM, Friedman DP, Marino RJ, Herbison GJ. The relationship between the functional abilities of patients with cervical spinal cord injury and the severity of damage revealed by MR imaging. *Am J Neuroradiol*. (1999) 20:926–34.
76. Falconer JC, Narayana PA, Bhattacharjee MB, Liu S-J. Quantitative MRI of spinal cord injury in a rat model. *Magn Reson Med*. (1994) 32:484–91. doi: 10.1002/mrm.1910320410
77. Besalti O, Pekcan Z, Sirin YS, Erbas G. Magnetic resonance imaging findings in dogs with thoracolumbar intervertebral disk disease: 69 cases (1997–2005). *J Am Vet Med Assoc*. (2006) 228:902–8. doi: 10.2460/javma.228.6.902
78. Boekhoff TMA, Ensinger E-M, Carlson R, Bock P, Baumgärtner W, Rohn K, et al. Microglial contribution to secondary injury evaluated in a large animal model of human spinal cord trauma. *J Neurotrauma*. (2012) 29:1000–11. doi: 10.1089/neu.2011.1821
79. Ito D, Matsunaga S, Jeffery ND, Sasaki N, Nishimura R, Mochizuki M, et al. Prognostic value of magnetic resonance imaging in dogs with paraplegia caused by thoracolumbar intervertebral disk extrusion: 77 cases (2000-2003). *J Am Vet Med Assoc*. (2005) 227:1454–60. doi: 10.2460/javma.2005.227.1454
80. Boekhoff TM, Flieshardt C, Ensinger E-M, Fork M, Kramer S, Tipold A. Quantitative magnetic resonance imaging characteristics: evaluation of prognostic value in the dog as a translational model for spinal cord injury. *J Spinal Disord Tech*. (2011) 25:E81–7. doi: 10.1097/BSD.0b013e31823f2f55
81. Wang-Leandro A, Siedenburg JS, Hobert MK, Dziallas P, Rohn K, Stein VM, et al. Comparison of preoperative quantitative magnetic resonance imaging and clinical assessment of deep pain perception as prognostic tools for early recovery of motor function in paraplegic dogs with intervertebral disk herniations. *J Vet Intern Med*. (2017) 31:842–8. doi: 10.1111/jvim.14715
82. Otamendi AJ, Boudreau B, Gilmour LJ, Griffin J, Levine J, Jeffery ND. Imaging findings at 3T and clinical outcome in deep-pain negative dogs with intervertebral disc herniation. *J Vet Intern Med*. (2017) 31:1262.
83. Okada M, Kitagawa M, Ito D, Itou T, Kanayama K, Sakai T. Magnetic resonance imaging features and clinical signs associated with presumptive and confirmed progressive myelomalacia in dogs: 12 cases (1997–2008). *J Am Vet Med Assoc*. (2010) 237:1160–5. doi: 10.2460/javma.237.10.1160
84. Gilmour LJ, Jeffery ND, Miles K, Riedesel E. Single-shot turbo spin echo pulse sequence findings in dogs with and without progressive myelomalacia. *Vet Rad US*. (2016) 13:425–9. doi: 10.1111/vru.12463
85. Castel A, Olby NJ, Mariani CL, Muñana KR, Early PJ. Clinical characteristics of dogs with progressive myelomalacia following acute intervertebral disc extrusion. *J Vet Intern Med*. (2017) 31:1782–9. doi: 10.1111/jvim.14829
86. Boudreau B, Jeffery ND, Gilmour LJ. Relationship between automated identification of intramedullary T2 hypointensity and clinical outcome in dogs with severe spinal cord injury. In: *Proceedings American College of Veterinary Radiology Annual Meeting*. Baltimore, MD (2019). doi: 10.1089/neu.2020.7188
87. Wang-Leandro A, Hobert MK, Kramer S, Rohn K, Stein VM, Tipold A. The role of diffusion tensor imaging as an objective tool for the assessment of motor function recovery after paraplegia in a naturally-occurring large animal model of spinal cord injury. *J Trans Med*. (2018) 16:1–10. doi: 10.1186/s12967-018-1630-4
88. Lewis MJ, Yap P-T, Mccullough S, Olby NJ. The relationship between lesion severity characterized by diffusion tensor imaging and motor function in chronic canine spinal cord injury. *J Neurotrauma*. (2018) 35:500–7. doi: 10.1089/neu.2017.5255
89. Lewis MJ, Olby NJ, Early PJ, Mariani CL, Munana KR.6 Influence of duration of injury on diffusion tensor imaging in acute canine spinal cord injury. *J Neutrotrauma*. (2020). doi: 10.1089/neu.2019.678. [Epub ahead of print].
90. Strain GM, Prescott-Mathews JS, Tedford BL. Motor potentials evoked by transcranial stimulation of the canine motor cortex. *Prog Vet Neurol*. (1990) 1:321–9.
91. Holliday TA, Weldon NE, Ealand BG. Percutaneous recording of evoked spinal cord potentials of dogs. *Am J Vet Res*. (1979) 40:326–33.
92. Sylvestre AM, Brooke JD, Cockshutt JR, Parent JM. Transcranial magnetic motor evoked potentials in the hind limbs of normal dogs sedated with oxymorphone, midazolam, and acepromazine. *Prog Vet Neurol*. (1992) 3:72–6.
93. Amendt HL, Siedenburg JS, Steffensen N, Söbbeler FJ, Schütter A, Tünsmeyer J, et al. Transcranial magnetic stimulation with acepromazine or dexmedetomidine in combination with levomethadone/fenpipramide in healthy beagle dogs. *Vet J*. (2016) 217:40–2. doi: 10.1016/j.tvjl.2016.06.006
94. Sylvestre AM, Cockshutt JR, Parent JM, Brooke JD, Holmberg DL, Partlow GD. Magnetic motor evoked potentials for assessing spinal cord integrity in dogs with intervertebral disc disease. *Vet Surg*. (1993) 22:5–10. doi: 10.1111/j.1532-950X.1993.tb00360.x
95. Amendt HL, Siedenburg JS, Steffensen N, Kordass U, Rohn K, Tipold A, et al. Correlation between severity of clinical signs and transcranial magnetic motor evoked potentials in dogs with intervertebral disc herniation. *Vet J*. (2017) 221:48–53. doi: 10.1016/j.tvjl.2017.01.015
96. Siedenburg JS, Wang-Leandro A, Amendt H-L, Rohn K, Tipold A, Stein VM. Transcranial magnetic motor evoked potentials and magnetic resonance imaging findings in paraplegic dogs with recovery of motor function. *J Vet Intern Med*. (2018) 32:1116–25. doi: 10.1111/jvim.15058
97. Lewis MJ, Howard JF, Jr, Olby NJ. The relationship between trans-lesional conduction, motor neuron pool excitability, and motor function in dogs with incomplete recovery from severe spinal cord injury. *J Neurotrauma*. (2017) 34:2994–3002. doi: 10.1089/neu.2017.5012
98. Hu HZ, Jeffery ND, Granger N. Somatosensory and motor evoked potentials in dogs with chronic severe thoracolumbar spinal cord injury. *Vet J*. (2018) 237:49–54. doi: 10.1016/j.tvjl.2018.05.007
99. Holliday TA. Electrodiagnostic examination: somatosensory evoked potentials and electromyography. *Vet Clin North Am Small Anim Pract*. (1992) 22:833–57. doi: 10.1016/S0195-5616(92)50079-1
100. Bright RM, Breazile JE, Bojrab MJ. Prognostic application of cortical evoked responses in dogs with spinal cord injury. *Vet Surg*. (1977) 6:55–9. doi: 10.1111/j.1532-950X.1977.tb00562.x
101. Shores A, Redding RW, Knecht CD. Spinal-evoked potentials in dogs with acute compressive thoracolumbar spinal cord disease. *Am J Vet Res*. (1987) 48:1525–30.
102. Poncelet L, Michaux C, Balligand M. Somatosensory potentials in dogs with naturally acquired thoracolumbar spinal cord disease. *Am J Vet Res*. (1993) 54:1935–41.
103. Poncelet L, Michaux C, Balligand M. Study of spinal cord evoked injury potential by use of computer modeling and in dogs with naturally acquired thoracolumbar spinal cord compression. *Am J Vet Res*. (1998)59:300–6.

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