TANGENCE
Number 137, 2025

ISSN: 1710-0305
PISSN: 1189-4563

CT-Guided Oxygen-Ozone Intradiscal Injection Therapy for Thoracolumbar Intervertebral Disc Herniations (Hansen Type II) in Dogs

Farkhondeh Fahim Dezhban[1], Majid Masoudifard[2*], Mohammad Mehdi Dehghan[3], Mohammad Molazem[4], Alireza Bahonar[5], Amir Ezzati[6], Morteza Ezati[7]

[1]DVM, Board-certified veterinary radiologist, Department of Surgery and Radiology, Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran.

[2]DVM, DVSc, Postal address: Department of Surgery and Radiology, Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran.

[3]DVM, DVSc, Postal address: Department of Surgery and Radiology, Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran.

[4]DVM, DVSc, Postal address: Department of Surgery and Radiology, Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran.

[5]DVM, PhD, Postal address: Department of Food Hygiene, Faculty of Veterinary Medicine, University of Tehran, Tehran, Iran.

[6]MD, Infectious diseases Specialist, General hospital, Tehran, Iran.

[7]DVM, Board-certified Small Animal Internist, Department of Internal Medicine, University of Tehran, Tehran, Iran.

Corresponding author: mmfard@ut.ac.ir, Postal address: Department of Surgery and Radiology, Faculty of Veterinary Medicine, University of Tehran, Azadi Avenue, Enqelab Square, Tehran, Iran.

Ethical approval: All applicable international, national and/or instutitional guidelines for the care and use of animals were followed.

ABSTRACT

Objective*:* To study the treatment of thoracolumbar herniated intervertebral discs (IVD) using Oxygen-Ozone (O_2-O_3) intradiscal injection.

Animals*:* 3 client-owned dogs with 13 herniated intervertebral discs

Procedure*:* These intervertebral discs were treated using 0.5 ml percutaneous injection of O_2-O_3 gas mixture, with an O_3 intradiscal concentration of 32 µg/µl under CT guidance, following MRI diagnosis. Five weeks after injection, a second MRI was performed, and six indices (introduced in the paper) were evaluated in all treated IVDs.

Results*:* All indices were improved after the treatment. In addition, three were statistically significant at $P<0.5$. All 3 dogs returned to their normal life after this study.

Conclusion: Intradiscal O_2-O_3 injection reduces the size of the herniated disc via disc shrinkage and decreasing the pressure of herniated part on the spinal cord.

KEYWORDS

Herniated intervertebral disc, Ozone-oxygen, intradiscal injection, dog

Introduction:

Intervertebral discs are located between all vertebral bodies of the vertebral column, except for the first and second cervical vertebrae (C1 and C2), and sacrum. Intervertebral discs possess a complex

ISSN: 1710-0305
PISSN: 1189-4563

structure: a thicker fibrous cartilage outer ring (the annulus fibrosus), and a more gelatinous core (the nucleus pulposus). They function as shock absorbers between each vertebra in the vertebral column, allow slight movement of vertebrae, function as ligaments to maintain the vertebrae, and absorb shock to the spine. Dehydration of the nucleus pulposus occurs when it loses water (1). However, the dehydrated IVDs will no longer be able to function as proper shock absorbers. Intervertebral disc disease (IVDD) usually pushes the nucleus pulposus against the spinal canal and compresses the spinal cord. Effects of IVDD range from bulging to protrusion or extrusion of IVD (2). A slow aging process in non-chondrodystrophic dogs mainly results in IVD degeneration (3). If the nucleus pulposus extrudes into the spinal canal and hits the spinal cord (concussion), it will require surgical intervention. More often, it slowly pushes its way into the spinal canal, resulting in pressure over the spinal cord (compression) (2).

IVDD is diagnosed using a combination of physical and neurological examinations, along with radiographs such as X-rays. In more advanced methods, imaging techniques such as MRI are obtained from the spine (3).

MRI scanners employ strong magnetic fields, magnetic field gradients, and radio waves to generate diagnostic images. It is the best diagnostic tool for the diagnosis of IVD degeneration prior to herniation or extrusion. With the invention of MRI, since it provided more detailed images from the spinal cord, myelography became obsolete (4).

O_2-O_3 injection for the treatment of disc herniation has been primarily proposed in human medicine. It is now frequently used as a minimally-invasive treatment for IVD herniation. Aside from being a cost-effective procedure, intradiscal injection of this gas demonstrated acceptable clinical results. Along with shrinking the herniated disc, O_2-O_3 gas possesses anti-inflammatory and analgesic effects on the compressed spinal cord (5). Many studies have confirmed the positive effects of ozone therapy in human medicine (6, 7, and 8). In veterinary medicine, HYUN-JUNG HAN *et al.* studied the impact of intradiscal O_2-O_3 injection on improving IVD herniation. In their study, CT was employed to measure the A-index, according to the following formula (5):

$$\text{A-index (\%)} = \frac{\text{Area of disc herniation x 100}}{\text{Area of the spinal canal}}$$

However, IVD is visible in a CT only if the disk is calcified. Therefore, using CT, IVD herniation, and spinal cord compression can be measured with very low accuracy. Therefore, using CT results increases the risk of false smaller measurements on disks that are not shrunk. Moreover, in CT, the difference between a disk and other tissues are not distinguishable (4).

To increase accuracy and introduce a diagnostic strategy that poses less radiation on the animal (2), we have conducted this study using MRI. We also measured multiple indices to evaluate the effect of O_2-O_3 therapy on IVD more accurately.

This study aims to investigate the effect of intradiscal O_2-O_3 gas therapy on herniated IVDD.

Materials and Methods:

Animals: Among the dogs admitted to the Small Animal Teaching from 25 March 2018 to 28 February 2020, 3 dogs with various types of neurologic abnormalities were included in this study based on their owner's request. These three dogs had clinical signs related to thoracolumbar IVDD and positive deep pain perception (DPP). In addition, dogs under 8 with no clinical signs relevant to thoracolumbar IVDD, along with samples with other major diseases such as cardiac or endocrinopathy, were excluded.

Samples 1 and 3 were female, while sample number 2 was male, and were 12, 10, and 9 years old, respectively. The patients demonstrated long-lasting periodic problems in gaining weight. Their body

ISSN: 1710-0305
PISSN: 1189-4563

weight ranged from 7 to 28 Kgs. The patients were gone through complete neurological examination by an experienced surgeon and were diagnosed with thoracolumbar intervertebral disc herniation (IVDH) on physical, neurological, diagnostic imaging views (radiography and MRI), complete blood count profile, and serum biochemical analysis. Different ranges of disc herniation were detected in thirteen intervertebral joint spaces. Deep pain perception (DPP) was positive in all patients, and they had no other systemic diseases. However, they had difficulty in jumping and running.

On the contrary, none had urinary or fecal incontinence. Sample 1 had delayed right hind limb paw replacement and hopping test, while samples 2 and 3 both had bilateral delayed paw replacement and hopping test before injection. None of the samples had a history of glucocorticoid therapy or surgery beforehand.

Premedication and Anesthesia: The dogs were given intravenous Pantoprazole (1 mg/kg) and subcutaneous Metoclopramide (0.5 mg/kg), four to six hours before anesthesia. They were sedated using intravenous injection of Ketamine (5 mg/kg) and Diazepam (0.2 mg/kg) and were maintained in sedation with intravenous Ketamine (4 mg/kg).

Diagnostic Imaging: The CT-scan device (Siemens, SOMATOM Spirit 2, 70KV, 100 mAs, Germany) was employed for 1 mm thick slices at 1.5 mm intervals under general anesthesia for intradiscal injections. MRI scan (General Electric Medical System, 1.5 Tesla) was performed before and five weeks after O_2-O_3 therapy. Index measurements were performed using Marco PACS software.

Intradiscal O_2-O_3 Injection: A 0.5 ml O_2-O_3 mixture (O_3 concentration of 32 µg/µl) was injected into the intradiscal area of thirteen IVDs obtained from three dogs under CT guidance and general anesthesia. Medical O_2-O_3 gas was produced by the 5th generation technology medical ozone generator (Gardina, MC80, Spain).

Measurements:

1) Pfirrmann Grading: Pfirrmann grading is used to evaluate the degenerative changes of Nucleus Polposus in T2W images, before and 5 weeks after intradiscal injection. This grading is normally used in spinal cord researches and has different classifications (grade I to V) (Image 1) (9, 10).

2) Schneiderman classification scheme: This scheme evaluates the degenerative changes in height and signal intensity of the intervertebral IVDH discs in T2W images, before and 5 weeks after intradiscal injection (table 1) (11).

3) MSU index: MSU (Michigan State University) index indicates the size of disc herniation and its location in the spinal canal. Moreover, it assesses the degree of disc herniation before and 5 weeks after intradiscal injection. It was introduced by the MSU classification system and is defined as a simple and reliable method to measure herniated IVD objectively. (Image 2) (12).

4) MSI: Evaluation and comparison of the Mean Signal Intensity (MSI) of the IVD were performed before and 5 weeks after intradiscal injection (13).

5) A-index: Measurement of the disc herniation area, in comparison with the spinal canal area, was performed before and 5 weeks after intradiscal injection, as proposed in (Image 5) (5).

6) B-index: Measurement and comparison of herniated intervertebral disc area, before and 5 weeks after intradiscal injection, was performed using the following equation:

$$\text{B-index (\%)} = \frac{\text{Intervertebral disc surface area before ozone-oxygen injection x 100}}{\text{Intervertebral disc surface area after 5 weeks after ozone-oxygen injection}}$$

ISSN: 1710-0305
PISSN: 1189-4563

The injection sites in the dorsal thoracolumbar area were first marked by the CT guide and were then surgically scrubbed (Image 3). The skin was perforated using a 2.5 inch 22 G spinal needle, and the needle was guided through epaxial muscles to the center of the herniated disc from the lateroventral side of the articular facet (Image 4). The operation was under constant monitoring with frequent CT images. Next, the stylet needle was extracted, and a 0.5 ml fresh ozone-oxygen gas mixture was injected. Finally, the stylet was extracted, and the injection site was compressed for about 1 minute. All the dogs were monitored for 24 hours afterward and were discharged the following day. Owners were instructed to limit the dog activity during the first 2 weeks and increase their activity gradually in the next 3 weeks. Clinical and MRI scans were performed for disc shrinkage, its size, and spinal cord compression.

Statistical Analysis: The data were assessed using SPSS. A paired T-test was used to identify the correlations between measurements before and after injection. $P<0.05$ was considered as the level of significance.

Results:

The quantitative indices for the 13 affected intervertebral discs were measured using Marco PACS software and the mean data values were presented in Table 2.

The patients went through comprehensive neurological examination, radiographic studies, and 1.5 Tesla MRI. A total of 13 IVDs were injected in 3 dogs (from bulging to protrusion). In specific, 4 IVDs were injected in dogs 1 and 3, while 5 IVDs were injected in dog 2.

After determining the definite location of the involved IVDs, the dogs were sedated for intradiscal O_2-O_3 injection under CT scan guidance. This intervention lasted approximately 20 to 25 minutes, and was determined according to the patient's time of entrance, the process for anesthesia, involved IVD detection, and final injection and removing the spinal needle (for each involved IVD). All patients recovered from the procedure normally. Following the injection, some gas contents were seen through surrounding musculature, and even in the spinal canal. In any case, such incidents generally had no side-effects on recovery or the study (Image 5).

After 5 weeks from O_2-O_3 injection, the clinical signs of all three patients were improved. Then, they were reevaluated by 1.5 Tesla MRI, which showed progressive shrinkage of involved IVD in all patients.

Reduction of the involved IVD protrusions, after O_2-O_3 injection, was detected on MRI scans taken 5 weeks after the procedure. The following 6 indices were measured in our study:

1. Pfirrmann Grading: this index showed degeneration of IVD contents, except for 3 IVDs.
2. Schneiderman Score: this index was not significantly changed.
3. MSU Classification: this index did not experience considerable changes, which may be due to the large grading of this classification. Therefore, there were no significant changes in IVD protrusion status.
4. MSI: this index was changed considerably in all IVDs, up to one-third in some.
5. A-index: this index successfully changed, except in 2 IVDs, which experienced minor increases.
6. B-index: the variations of this index was interesting, except in 2 IVDs. In specific, there was a mild increase in one IVD and no change in only one IVD.

The results are shown in table 2. They were evaluated using SPSS software and Paired T-test.

ISSN: 1710-0305
PISSN: 1189-4563

Discussion:

The present study demonstrates the quantitative evaluation of IVD size, 5 weeks after intradiscal O_2-O_3 injection. Our data confirmed that the degeneration of IVDs occurs following intradiscal O_2-O_3 injection (up to 100% according to MSI index). However, A and B indices were enhanced in 85% of IVDs, which may need more time due to the chronicity of IVD shrinkage. Previous studies on human subjects demonstrated that IVD shrinkage following intradiscal O_2-O_3 injection is influenced by several factors, including the extension of the herniated IVD contents, degree of protrusion, and IVD classification (8). According to (5), comparing our data indicates that the volume of disc herniation is not related to the disc shrinkage.

As the results confirm, degenerative changes were delineated in 77% of IVDs in the first variant. On the other hand, the 2nd and the 3rd indices were not significantly changed, which can be due to the progressive time-consuming process of IVD degeneration.

Pfirrmann and Schneiderman scheme classifications did not experience significant changes. In our study, only one radiologist was responsible for the interpretation of the MR images, which justifies the possibility of error in the results (11). To overcome this shortcoming, factors should be evaluated by different radiologists to ensure inter-observer reliability (11). Another significant issue regarding these two factors is their detectable progressive variation. Hence, more time is needed to alter the MRI intensity of IVD.

MSU classification should be evaluated using MRI (12). As mentioned there, it is measured during a 5-year clinical follow-up on the cases. Therefore, more time is required to evaluate and determine the statistical meaningfulness of this index.

To determine and quantify the IVD degeneration, we evaluated the MSI (4th index). A significant change was detected before and after the data, which was statistically meaningful (0.0001, $P<0.05$). MRI enables assessment of the intensity of the IVD material before and after O_2-O_3 intradiscal injection.

Another valid index to measure the disc shrinkage and decompression of the spinal cord is the 3rd variant (A-index) (5). In our study, this index was statistically meaningful (0.006, $P<0.05$). This variant can be precisely estimated using 1.5 Tesla MRI because MRI delineates the accurate margin of the herniated material and compression of spinal cord parenchyma.

The last variant (B-index) is measured to evaluate the IVD shrinkage during the 5-week experiment period. The measured B-index value was also statistically valid, which was measured up to 0.003 ($P<0.05$). In addition, this variant was measured by MRI, which provides a gold standard of IVD diagnosis (due to the cartilaginous structure of the IVD).

According to A and B indices, we concluded that the IVD shrinkage occurred gradually in 5 weeks. On the other hand, according to (5), there is no relationship between disc shrinkage and the volume of disc herniation. However, more time is needed to evaluate other factors (due to Pfirrmann, Schneiderman, and MSU classification data in our study). Therefore, intradiscal ozone-oxygen injection yields progressive decompressive and anti-inflammatory effects on the spinal cord. Moreover, it has preventive influences on deterioration or recurrence of clinical signs. Nevertheless, more time is required to compare the results with laminectomy or fenestration (2).

We also introduced intradiscal O_2-O_3 injection as an alternative therapy for herniated IVDD with low invasion, long-lasting effects. Moreover, compared with glucocorticoid therapeutics and surgery, this method is a cost-effective solution. However, despite being useful in chronic patients, intradiscal injection of O_2-O_3 is not a desirable therapy in patients that require rapid decompression. In specific, it is beneficial in patients who demonstrate clinical improvements by anti-inflammatory therapies and have frequent relapses. All the dogs included in this study had chronic problems and did not require emergency surgery. Samples with no deep pain perception in the clinical examination were excluded since they need emergency operation to decompress the spine.

ISSN: 1710-0305
PISSN: 1189-4563

The density of the involved disc is decreased after O_2-O_3 intradiscal injection. This intradiscal shrinkage creates free space for the compressed spine to move regardless of herniated disc materials in the spinal cord. This confirmed by MRI data and clinical signs. Disc shrinkage also guaranteed that the treated IVD would not remain herniated or extruded, and thus, the possibility of recurrence of pain and clinical signs were reduced considerably (5). Therefore, the O_2-O_3 therapy offers a replacement for previous conservative methods, which may prevent further extrusion or recurrence of herniation of IVD (14).

There are other advantages in using the O_2-O_3 mixture as a non-harmful gas with analgesic and anti-inflammatory effects, even when this gas is in direct contact with the spinal cord. The anti-inflammatory feature of the ozone-oxygen mixture functions via inhibiting inflammatory inducers and pain-producing mediators such as prostaglandins (15). Moreover, due to its strong bactericidal activity, it has fewer complications, which considerably decreases the risk of abscess formation from skin contamination (16, 17).

The other noticeable this method is its simplicity and short anesthesia duration, which plays an essential role in these patients. In addition, it requires minimal hospitalization (approximately 24 hours) for post-surgical checks of the side-effects. In laminectomy and other surgical methods, hospitalization time is considerably more. Furthermore, the cost of this procedure is significantly lower than surgery or other alternatives, specifically in the long-term, as this method prevents further problems with the injected IVD.

In this study, a recognizable disc shrinkage was confirmed in 1.5 Tesla MRI. However, the degree of this shrinkage requires more time and is related to the extent of herniated disc material degeneration. Nevertheless, no specific complications were detected in this study. In conclusion, it is observed that intradiscal O_2-O_3 mixture injection can be helpful, and minimally invasive, in the decompression of disc herniations.

Acknowledgements:

This work was performed at Faculty of Veterinary Medicine of University of Tehran.

The Veterinary medical Teaching Hospital of University of Tehran supports Ms. Fahim Dezhban for using radiology and CT scan equipments.

Tehran Medical Imaging Center cooperates Ms. Fahim Dezhban in using MRI system.

The authors declare that there were no conflicts of interests

References:

1. Andreula C. Ozone therapy. Neuroradiology. 2011 Sep 1;53(1):207.

2. Berent AC, Tobias KM. Hepatic vascular anomalies. Veterinary surgery small animal. 2012;1:1624-58.

3. Brisson BA. Intervertebral disc disease in dogs. Veterinary Clinics of North America, Small Animal Practice. 2010;40(5):829-58.

4. Robertson IA, Thrall DE. Imaging dogs with suspected disc herniation: pros and cons of myelography, computed tomography, and magnetic resonance. Veterinary radiology & ultrasound. 2011 Mar;52:S81-4.

5. Han HJ, Kim JY, Jang HY, Lee B, Yoon JH, Jang SK, Choi SH, Jeong SW. Fluoroscopic-guided intradiscal oxygen-ozone injection therapy for thoracolumbar intervertebral disc herniations in dogs. in vivo. 2007 Jul 1;21(4):609-13.

ISSN: 1710-0305
PISSN: 1189-4563

6. Buric J, Rigobello L, Hooper D. Five and ten year follow-up on intradiscal ozone injection for disc herniation. International journal of spine surgery. 2014 Jan 1;8.

7. Zhang Y, Ma Y, Jiang J, Ding T, Wang J. Treatment of the lumbar disc herniation with intradiscal and intraforaminal injection of oxygen-ozone. Journal of back and musculoskeletal rehabilitation. 2013 Jan 1;26(3):317-22.

8. Lehnert T, Naguib NN, Wutzler S, Nour-Eldin NE, Bauer RW, Kerl JM, Vogl TJ, Balzer JO. Analysis of disk volume before and after CT-guided intradiscal and periganglionic ozone–oxygen injection for the treatment of lumbar disk herniation. Journal of Vascular and Interventional Radiology. 2012 Nov 1;23(11):1430-6.

9. Mwale F, Iatridis JC, Antoniou J. Quantitative MRI as a diagnostic tool of intervertebral disc matrix composition and integrity. European Spine Journal. 2008 Dec 1;17(4):432.

10. Grunert P, Hudson KD, Macielak MR, Aronowitz E, Borde BH, Alimi M, Njoku I, Ballon D, Tsiouris AJ, Bonassar LJ, Härtl R. Assessment of intervertebral disc degeneration based on quantitative MRI analysis: an in vivo study. Spine. 2014 Mar 15;39(6):E369.

11. Nagy SA, Juhasz I, Komaromy H, Pozsar K, Zsigmond I, Perlaki G, Orsi G, Schwarcz A, Walter N, Doczi T, Bogner P. A statistical model for intervertebral disc degeneration: determination of the optimal t2 cut-off values. Clinical neuroradiology. 2014 Dec 1;24(4):355-63.

12. Mysliwiec LW, Cholewicki J, Winkelpleck MD, Eis GP. MSU classification for herniated lumbar discs on MRI: toward developing objective criteria for surgical selection. European spine journal. 2010 Jul 1;19(7):1087-93.

13. Niemeläinen R, Videman T, Dhillon SS, Battié MC. Quantitative measurement of intervertebral disc signal using MRI. Clinical radiology. 2008 Mar 1;63(3):252-5.

14. Andreula CF, Simonetti L, De Santis F, Agati R, Ricci R, Leonardi M. Minimally invasive oxygen-ozone therapy for lumbar disk herniation. American Journal of Neuroradiology. 2003 May 1;24(5):996-1000.

15. Bonetti M, Fontana A, Cotticelli B, Dalla Volta G, Guindani M, Leonardi M. Intraforaminal O2-O3 versus periradicular steroidal infiltrations in lower back pain: randomized controlled study. American journal of neuroradiology. 2005 May 1;26(5):996-1000.

16. Rilling S, Viebahn R. The use of ozone in medicine. KF Haug Publishers; Portland, OR: US distributor, Medicina Biologica; 1987.

17. Viebahn-Haensler, R., & Lee, A. (2002). The use of ozone in medicine (pp. 4-21). Heidelberg: AUDREY publishers.

Table 1. Schneiderman classification scheme (11)

Grade	0	1	2	3
Appearance	Normal height and signal intensity	Speckled pattern or heterogeneous decreased intensity	Diffuse loss of signal	Signal void

ISSN: 1710-0305
PISSN: 1189-4563

Table 2. Mean data values before and after intradiscal O_2-O_3 injection in 13 IVDs

Stage	PG	SS	MSU	MSI	A-Index	B-Index
Before:						
Mean	2.2308	1.1538	1.1538	398.6154	12.5708	271.5385
Std. Error of Mean	0.36080	0.22206	0.10415	39.83726	1.80314	25.03449
Std. Deviation	1.30089	0.80064	0.37553	143.63527	6.50132	90.26315
After:						
Mean	2.6923	1.3077	1.0000	206.6154	6.3554	253.4615
Std. Error of Mean	0.28610	0.13323	0.16013	18.95154	1.44976	23.57057
Std. Deviation	1.03155	0.48038	0.57735	68.33074	5.22717	84.98492
Total:						
Mean	2.4615	1.2308	1.0769	406.5621	9.4631	262.5000
Std. Error of Mean	0.23026	0.12779	0.09484	2.261107	1.29269	16.94164
Std. Deviation	1.17408	0.65163	0.48358	8.152538	6.59147	86.38576

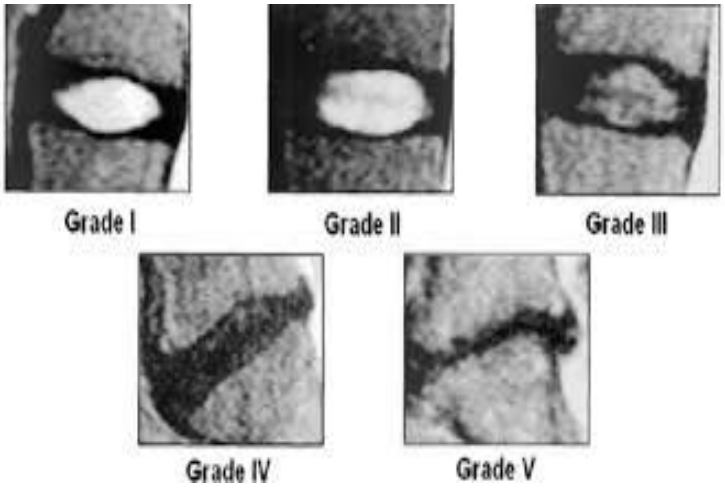

Image 1. Pfirrmann Grading

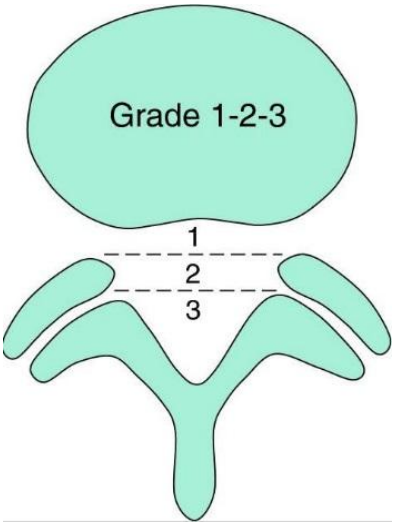

Image 2. MSU quantitative assessment

ISSN: 1710-0305
PISSN: 1189-4563

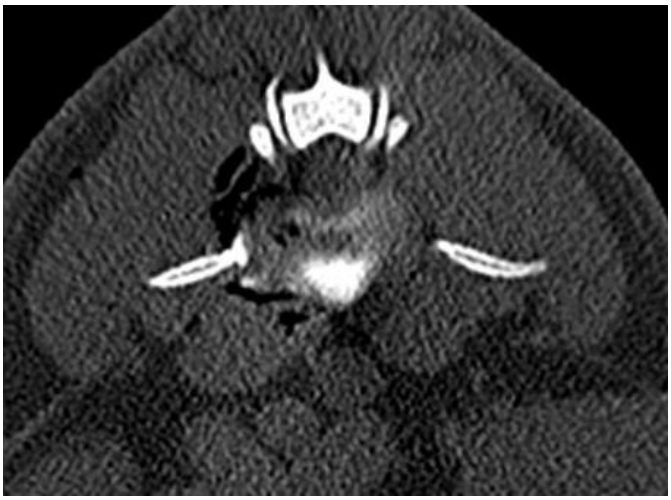

Image 3. Preparation of the patient and injection of the spinal needle through the IVD

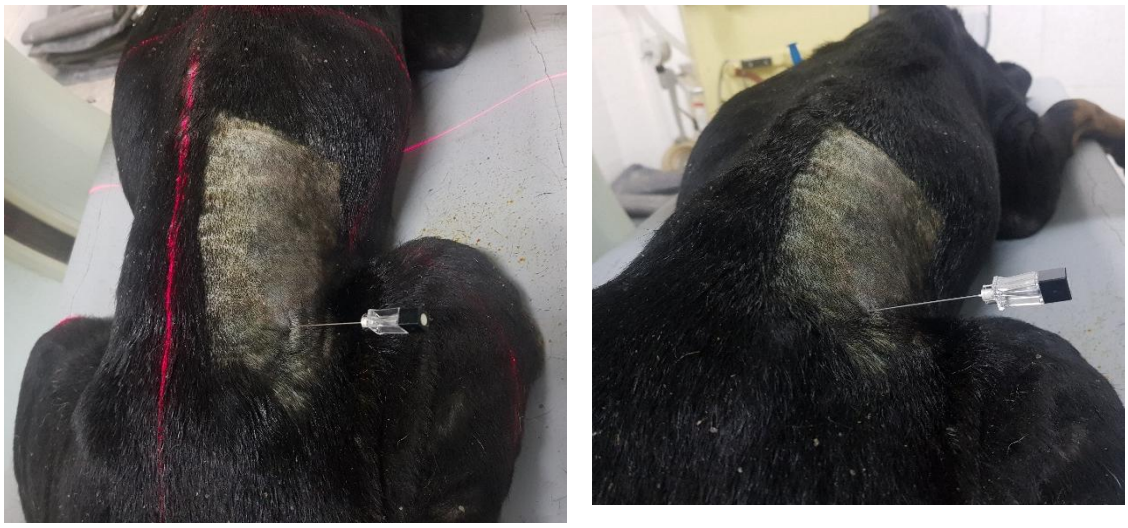

Image 4. O_2-O_3 intradiscal injection under CT guidance

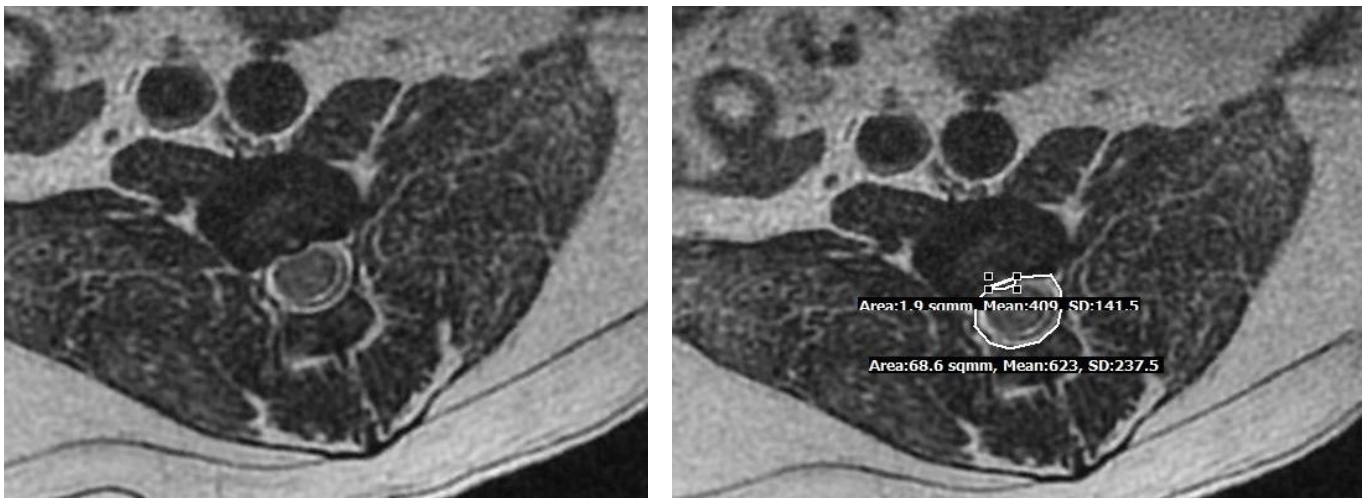

Image 5. T2W image at L3-L4 level from Dog 1 before O_2-O_3 intradiscal injection, with and without A-index measurement: