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The Spine Journal 16 (2016) 889–895

THE SPINE JOURNAL

Basic Science

Closure of the annulus fibrosus of the intervertebral disc using a novel suture application device—in vivo porcine and ex vivo biomechanical evaluation

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Received 18 November 2015; revised 18 February 2016; accepted 4 March 2016

Abstract

BACKGROUND CONTEXT: Defects in the annulus fibrosus (AF) remain a challenge in the surgical treatment of lumbar disc herniations with persistent defects, allowing potential re herniation of nucleus pulposus (NP) tissue. A cervical porcine model was chosen to simulate human lumbar intervertebral disc (IVD).

PURPOSE: The aim of this study was to determine the technical feasibility of closure of the AF of the IVD using a novel minimally invasive Kerrison-shaped suture application device.

STUDY DESIGN: Ex vivo biomechanical and in vivo porcine device evaluations were performed.

METHODS: Ex vivo biomechanical evaluation: 15 porcine spinal units were explanted and subjected to mock discectomy. The annular defect was closed using 2-0 non-absorbable (ultra-high molecular-weight polyethylene, UHMWPE) suture and Dines knot. The knot was backed up with two, three, or four throws. The spinal unit was subject to 4000 cycles of flexion/extension with 1500 N of axial load, and assessed for knot slippage. In vivo porcine device evaluation: three pigs (53–57 kg) were anesthetized and underwent a ventral surgical approach to the cervical spine. The AF of two discs was incised, and simulated partial NP discectomy was performed. The defect was closed at one level using the AnchorKnot device to apply the suture with a Dines knot and four throws. The pigs were observed for 4 weeks before euthanasia, allowing 7T magnetic resonance imaging (MRI) and histological evaluation.

FDA device/drug status: Not approved for this indication (AnchorKnot Suture Passer, AnchorKnot Knot Pusher).

Author disclosures: ***AHB:*** Grant: Industry-matched grant from Ontario Centres of Excellence (E, Paid to the institution). ***CB:*** Grant: Industry-matched grant from Ontario Centres of Excellence (E, Paid to the institution). ***MKA:*** Grant: Industry-matched grant from Ontario Centres of Excellence (E, Paid to the institution). ***AHWC:*** Grant: Industry-matched grant from Ontario Centres of Excellence (E, Paid to the institution). ***RDH:*** Grant: Industry-matched grant from Ontario Centres of Excellence (E, Paid to the institution). ***WO:*** Grant: Industry-matched grant from Ontario Centres of Excellence (E, Paid to the institution). ***AJMY:*** Grant: Industry-matched grant from Ontario Centres of Excellence (E, Paid to the institution). ***SMM:*** Grant: Industry-matched grant from Ontario Centres of Excellence (E, Paid to the institution).

The disclosure key can be found on the Table of Contents and at www.TheSpineJournalOnline.com.

This project was supported by a research grant from Anchor Orthopaedics XT Inc. Rob Harrison is an employee of Anchor Orthopaedics XT Inc. Stuart McGill has received honoraria from Anchor Orthopaedics XT Inc. The remaining authors have no disclosures.

Animal Care Committee ethics approval for this in vivo animal study was obtained through Sunnybrook Research Institute.

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http://dx.doi.org/10.1016/j.spinee.2016.03.005

RESULTS: A Dines knot with four throws experienced no slippage after 4000 cycles. This configuration was tested in vivo. Clinically, the neurological examination in treated pigs was normal following surgery. Histological and MRI assessment confirmed sustained defect closure at 4 weeks. There was no reaction to the suture material and no NP extrusion at any of the sutured levels.
CONCLUSIONS: This study demonstrates that it is technically feasible to perform AF defect closure in a porcine model. This novel device achieved AF defect closure that was maintained through 4 weeks in vivo.

Keywords: Annular repair; Annulus fibrosus closure; Disc repair; Discectomy; Intervertebral disc; Minimally invasive; Porcine model; Re-herniation; Surgery; Suture application device

Introduction

Herniation of the nucleus pulposus (NP) through the annulus fibrosus (AF) of the intervertebral disc (IVD) is a recognized cause of low back and radicular leg pain [1]. The surgical treatment of this condition involves partial NP discectomy with good clinical outcomes reported [2]. The procedure involves surgical removal of herniated NP tissue including any sequestered or detached fragments of NP and endplate. It does not, however, address the tear or fissure in the AF, which is the path through which the herniation occurred. In some cases, a surgical annulotomy may be required to perform the partial NP discectomy, and this leaves a persistent defect in the AF immediately postprocedure. AF defects may be important to consider when recognizing the risk of reherniation following lumbar discectomy [3]. Carragee et al reported that larger AF defects postdiscectomy are a risk factor for recurrent disc herniation and poor outcome [3]. The same group reported recurrent IVD herniation requiring surgery in 10% of cases following NP discectomy. They also observed a 25% loss of disc height 2 years postprocedure and showed higher recurrent herniation rates associated with larger annular defects [4].

Defects in the AF of the IVD thus remain a surgical challenge, and efforts have been made to develop new techniques for their closure and repair. Various techniques have been developed to effect closure of AF defects and stimulate healing and regeneration. The IVD is a relatively avascular structure with low cell numbers available for a healing response to injury. It is therefore likely that achieving AF repair will involve combined mechanical and biological strategies [5].

With the advent of minimally invasive surgical (MIS) approaches for lumbar microdiscectomy, surgical access and anatomy limit the types of devices that permit AF repair. Devices are currently being developed to allow surgical closure of the AF through standard and minimally invasive approaches to the spine [6–9]. In this study, a novel Kerrison-shaped AF closure device (AnchorKnot Tissue Repair System, Anchor Orthopedics XT Inc.) was evaluated. This device delivers a 2-0 ultra-high molecular-weight polyethylene (UHMWPE) suture with Dines knot to the AF, facilitating closure through commonly used surgical approaches. The purpose of this study was to evaluate the feasibility of AF closure in an ex vivo biomechanical and in vivo preclinical model.

Materials and methods

The AnchorKnot suture-passing device is designed for use in the lumbar spine. A pig cervical spine model was chosen to model human lumbar IVDs based on the comparative size of the discs. Porcine cervical IVD is a well-recognized animal model for human lumbar spine comparison based on its geometry, anatomy [10], and function [11]. Due to anatomical similarities, the ventral surgical approach to the cervical spine of a pig is similar to the anterolateral (Smith-Robinson) surgical approach.

We performed bench-top evaluation of the suture-knot constructs, followed by ex vivo biomechanical testing of AF closures performed on explanted pig cervical spinal units. Based on biomechanical results, additional in vivo evaluation was performed to assess the feasibility of defect closure and determine knot integrity for 4 weeks postoperatively.

The suture used throughout the study was 2-0 UHMWPE.

A Dines knot (Fig. 1) was selected and used throughout the study based on biomechanical and engineering factors. It is commonly used in arthroscopic surgery and has shown superiority over other arthroscopic knots in several biomechanical studies [12–15].

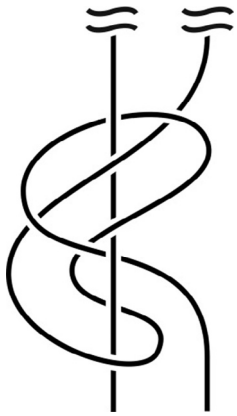

Fig. 1. A Dines knot.

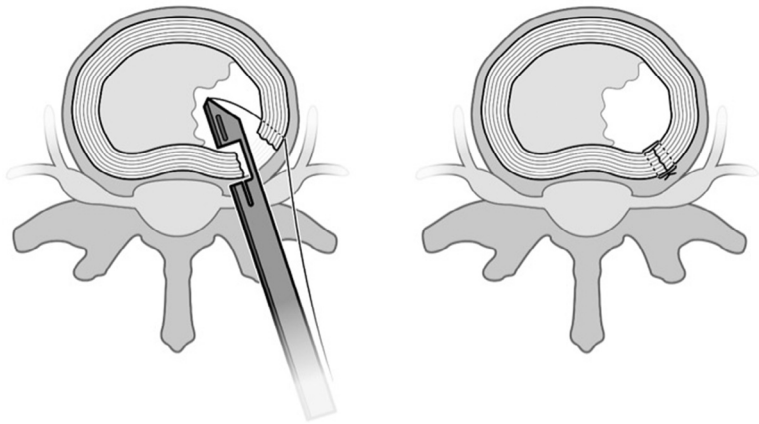

Fig. 2. Application of suture using the AnchorKnot device.

Bench-top knot evaluation

We tested the straight-pull strength of the Dines knot backed up with two, three, or four half-hitches (n=10 per group). Suture loops were tied around a 23 mm diameter metal rod, and each half-hitch was secured using a knot pusher.

A dynamic servohydraulic load testing system (Model 8511, Instron Canada, Burlington, Ontario, Canada) preloaded each suture loop with 5 N of tension and then displaced the loops from each other at a constant rate of 0.5 mm/s. The failure point where the force-displacement graph deviated from a linear force-displacement relationship indicated where the knot began to slip or the suture fail.

One-way ANOVA was used to determine any significant difference between the different half-hitch configurations with respect to the amount of load sustained prior to failure. A Bonferroni correction was applied to allow for multiple comparisons between groups.

Ex vivo biomechanical evaluation

Functional spinal units (C3–C4 or C5–C6) were harvested from 15 pig spines (age: 6 months; average weight: 80 kg). All specimens met Galante's criteria of a normal disc [16]. The specimens were secured using non-exothermic dental stone (Denstone, Miles, South Bend, IN, USA) and 18-gauge wire looped bilaterally around the lamina and anterior processes. Saline-soaked cloth and plastic wrap was used to prevent desiccation, and each specimen was preloaded with 300 N of axial compression for 15 minutes to counter any postmortem swelling that may have occurred [17,18]. Passive range of motion testing with 1500 N axial load established the linear region of axial displacement versus torque, similar to the neutral zone described by Panjabi et al [19].

A 15-mm hole was drilled through the laminae to provide access to the IVD of each specimen. A vertical incision was made in the AF. The suture-passing device (AnchorKnot Suture Passer, Anchor Orthopedics XT Inc., Mississauga, ON) was inserted into the disc through the incision and rotated such that a single pull of the trigger passed the suture through the tissue 3 mm lateral to the incision. The device was then rotated 180°, and another pull of the device's trigger retrieved the suture through the opposing margin (Fig. 2). In this way, the suture strand was looped around the "inside" of the defect, with two free limbs available on the "outside" for knot tying.

A preloaded Dines knot was advanced and tightened with a knot pusher (AnchorKnot Knot Pusher, Anchor Orthopedics XT Inc.). The Dines knot was backed up with two, three, or four half-hitches (n=5 per group) and again secured with the knot pusher. Using the same servohydraulic load testing system, each specimen was subjected to repetitive flexion and extension to the angles established during the passive range of motion tests. These were conducted with 1500 N of axial compression. Pilot testing demonstrated that if the suture was going to slip, it would do so early on, so measurements were made at 50, 100, 200, 300, 500, 1000, 2000, and 4000 cycles.

Durability testing was conducted on five further specimens prepared in an identical manner to previous specimens. AF closure was achieved using the four half-hitch knot configuration and specimens were tested to 85,000 cycles.

Statistical analysis was performed using the Mann-Whitney *U* test to compare the knot slippage data in AF tissue between the two, three, and four half-hitch groups.

In vivo AF repair

Animal Care Committee ethics approval for this in vivo animal study was obtained through Sunnybrook Research Institute.

Three pigs (53–57 kg) were selected for this study. Initial assessment confirmed good health and the absence of any clinically apparent neurological deficit. The pigs were fasted for 12 hours before surgery and a non-steroidal anti-inflammatory drug (Metacam; 0.4 mg/kg intramuscular (I.M.)) was administered. Anesthetic induction was performed using Ketamine (10–15 mg/kg) and Atropine (0.04–0.05 mg/kg) I.M. The pigs were placed in a dorsally recumbent position, and the ventral aspect of the neck was prepared for surgery using an aqueous 10% povidone-iodine solution. Anesthesia was maintained

using inhalation anesthesia (2 % isoflurane in oxygen 2 L/min). A ventral surgical approach to the cervical spine was performed through a 20-cm incision parallel to the trachea. The C3 to C5 vertebrae were exposed including the associated IVDs. A vertical incision was made in the AF of two discs (C3–C4; C4–C5) using a scalpel. Simulated partial NP discectomy was performed using a pituitary rongeur. Three passes of the rongeur were used in each case to standardize the amount of NP removed. One defect was closed at a randomly selected level using the Anchorknot annular closure system described above. The second defect served as unrepaired control. The wound was sutured and closed in layers.

Following surgery, the antibiotic Duplocillin was administered at 2.5 mL/50 kg I.M. for 5 days. The pigs were monitored twice daily for the first 48 hours postoperatively including neurological assessments. Once analgesia was no longer required, the animals were monitored once daily. Four weeks after surgery, the pigs were euthanized under general anesthesia using a pentobarbital overdose (120 mg/kg, I.V.). The pig cervical spines were harvested for 7T magnetic resonance imaging (MRI) and histological evaluation. To be able to identify the suture path in the histology slides and to distinguish it from processing artifacts, the suture was stained with tissue dye. Unoperated control discs in each animal were also evaluated for baseline comparison.

After decalcification of the endplates, 5-mm transverse histology sections were stained with hematoxylin and eosin as well as Safranin-O to stain the IVDs. Histological specimens were reviewed for continuity of the AF closure, evidence of NP extrusion, and evidence of an inflammatory reaction to the suture material. Observation of any inflammatory cells adjacent to the suture material was taken as evidence of a possible inflammatory response.

MR images were acquired using a 7T Bruker Biospin (Bruker BioSpin MRI GmbH, Ettlingen, Germany). T2-weighted rapid acquisition with relaxation enhancement (RARE) images were acquired with a echo time (TE) of 54 ms, repetition time (TR) of 5 s, 8 averages, 0.2×0.2–mm/pixel, and 0.5-mm slice thickness.

IVD material was manually segmented on the axial MR images to eliminate bright signal from surrounding tissue. Each voxel within the segmented region was multiplied by the normalized signal intensity to reduce partial volume effects, especially in the damaged discs. These weighted values were summed and multiplied by the voxel volume to produce an estimate of the disc volume for control, sham, and sutured discs. One-way ANOVA was used to look for any statistical differences in volume between controls, sham, and sutured discs.

Results

Bench-top knot evaluation

Data for the straight-pull testing are presented in Table 1. The four half-hitch knots tolerated significantly more load before failure compared to the three half-hitch (p≤.001) or two half-hitch (p≤.001) knots. The predominant mode of failure for the four half-hitch knots was breakage, rather than slippage.

Table 1
Mean failure load for the suture loops tested

	Mean failure load (N)	Standard deviation (N)
Two half-hitches	46.7*	11.8
Three half-hitches	74.7†	17.3
Four half-hitches	105.0	17.1

* Indicates statistical significance when compared with the 4 half-hitch configuration (p≤.001).

† Indicates statistical significance when compared with the 4 half-hitch configuration (p≤.001).

Ex vivo biomechanical testing

The mean values for knot slippage after 4000 cycles of flexion/extension are provided in Table 2. Under the testing conditions described, the four half-hitch knot showed no knot slippage compared with 2.8 mm for three half-hitches (p=.032) and 8.8 mm for two half-hitches (p=.008). The suture in the four half-hitch specimens was qualitatively observed to be tight after testing and maintained closure of the annular defect. After further testing and exposure to 85,000 cycles, no knot slippage was evident in 3 out of 5 four-half-hitch specimens, while two specimens each showed 2 mm of knot slippage.

In vivo results

Clinical examination of pigs included a neurological assessment and was normal preoperatively, postoperatively, and at the prescribed observation points for 4 weeks following surgery. No anesthetic or surgical complications were observed in any of the three pigs.

Histological results showed maintained AF closure (Fig. 3) at 4 weeks in all three pigs and no significant soft tissue or inflammatory type reaction. Qualitatively on histological review, there were no significant differences comparing sutured discs with unrepaired discs.

MRI volumetric assessment showed reduced volume in both unrepaired (p=.03) and sutured (p=.04) discs compared with unoperated control discs. Although we did not have preop-

Table 2
Mean knot slippage data for the various knot configurations used. This was obtained with 4000 cycles of flexion/extension with load

	Mean knot slippage (mm)	Standard deviation (mm)
Two half-hitches	8.8*	5.1
Three half-hitches	2.8†	2.3
Four half-hitches	0	0

* Indicates statistical significance when compared with the four half-hitch configuration (p=.008).

† Indicates statistical significance when compared with the four half-hitch configuration (p=.032).

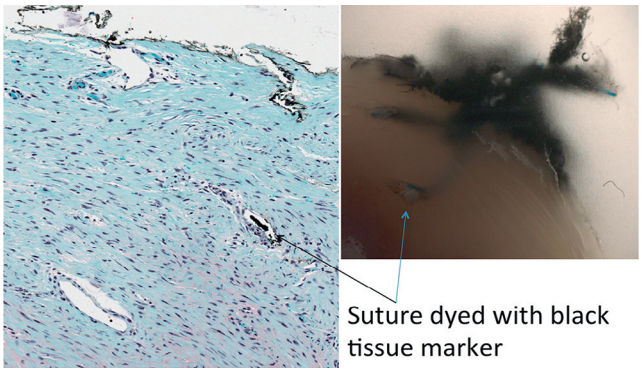

Fig. 3. Transverse histological specimen and corresponding paraffin block sample from sutured disc. Suture material was dyed postmortem with black tissue dye.

erative and immediate postoperative MRI to more accurately quantify the amount of NP removed, we estimated based on follow-up MRI scan and histology that approximately 25% of the NP was removed. There was no significant difference observed between sutured and unsutured discs (p=.86). The volumes are presented in Table 3.

Discussion

There are over 300,000 lumbar discectomy procedures performed in the United States annually [2,5,20]. The incidence of symptomatic reherniation requiring revision surgery is not insignificant, and revision spine surgery has worse outcomes when compared with index procedures [4,21]. Disc height loss after discectomy may also be associated with ongoing back pain symptoms and disability [22–24]. There are theoretical advantages of AF repair that include containment of remaining NP tissue to minimize the risk of recurrent disc herniation and maintain pressurization of the gel-like NP. There are a variety of potential strategies for repair of AF defects. These include suturing strategies as well as biologic strategies such as cell, protein, or gene delivery with or without synthetic or naturally occurring scaffolds [5]. Our AF closure method has several advantages relative to other available methods. The suture device was designed to be operated in a manner intuitive for spinal surgeons: the hand-held device is similar in size and in shape to a spinal Cloward/Kerrison rongeur and engages disc tissue through a familiar hand-grip closure maneuver. A strong suture-knot configuration has been chosen, extrapolated from our experience in orthopedic arthroscopy, which to our knowledge has not been previously applied to the spine. Finally, the device was designed to be usable for both open and MIS spine surgical approaches.

Table 3
Estimated disc volumes in milliliters (based on weighted axial MRI images taken immediately postsacrifice)

Pig	Unoperated (mL)	Unrepaired (mL)	Sutured (mL)
1	0.57	0.29	0.31
2	0.43	0.12	0.09
3	0.86	0.09	0.15
Mean	0.62	0.17	0.18
1 Standard deviation	0.22	0.11	0.11

MRI, magnetic resonance imaging. One-way ANOVA was used. Sutured vs unrepaired (p=.86); unoperated vs sutured (p=.038); unoperated vs unsutured (p=.033).

Our study demonstrates the feasibility of suture repair of the AF using a novel Kerrison-shaped AF closure device with sustained closure through 4 weeks in vivo. Ex vivo biomechanical testing demonstrated that a Dines knot with four half-hitches provided the optimal strength configuration under described testing conditions. Surgical access to the AF during discectomy may be limited in common with other MIS approaches such as those used for shoulder rotator cuff repair. The arthroscopic literature was therefore reviewed, and previous studies have determined that one of the best arthroscopic knots is the Dines knot [12–15]. This knot showed the least knot slippage and can be easily applied through a limited approach.

The biomechanical testing protocol simulated the approximate in vivo distraction forces across the AF defect experienced in a 2-week period [25]. 1500 N of axial load approximates the forces experienced in the human lumbar spine during lifting a light load [26]. Despite these efforts to simulate likely forces through the AF, it is not practical to fully simulate the in vivo forces through the AF in an ex vivo model. After initial testing showed early knot slippage in the two and three half-hitch groups, further endurance testing focused on the four half-hitch configuration. In vivo, we would expect a gristling process that could conceivably stiffen the knot and reduce slippage. This was absent in our cadaveric model, and so endurance testing was limited to 85,000 cycles. Future studies evaluating effects of combined flexion-torsion will be important to consider in clinical translation of this research. Under prolonged fatigue testing, without any biological healing, the distance between the two vertebral bodies of the motion segment was reduced. This reduction was due to high levels of annular delamination [11] and bulging [27] beyond that seen in the 4000 cycle tests. The delamination and bulging occur radially, potentially stretching out the suture construct. This mechanism could have resulted in the late slippage observed in the endurance tests.

Our in vivo study validated sustained AF closure through 4 weeks postoperatively. A novel and attractive element of this suture closure device is its Kerrison rongeur-like design, which holds tactile familiarity among spine surgeons performing discectomy. The addition of half-hitches following initial application of the Dines knot requires a knot pusher to hand tighten the hitches to the initial knot. AF closure instruments have been designed for ease of insertion through both minimally invasive tubular systems as well as conventional microdiscectomy surgical approaches.

MRI assessment of the in vivo IVD showed higher volumes for sutured and unsutured discs in pig #1 compared with the

other two because these data were acquired in fixed rather than fresh tissue. We found much greater T2 contrast between the disc and the surrounding tissue when imaging fresh tissue, so the volumes in pig #1 may be overestimated. Additionally, the disc volume appears to vary from disc to disc, so it is possible that a difference in volume between sutured and unsutured discs would be seen in a larger study. In our study, we did not perform preoperative MRI scans of the pig spines to compare potential differences in disc volume between adjacent disc levels a priori. Each MRI scan performed requires general anesthesia in our animal model. We did, however, at the time of surgery randomly assign selection of each disc for suture repair or no suture repair in an effort to minimize potential bias. The endpoint of this study was primarily to test the feasibility of repair using the novel suture device and to assess maintenance of closure of the disc repair site at 4 weeks.

There is evolving literature on the use of AF suture repair strategies following lumbar discectomy. In one 2-year multicenter matched-cohort prospective study of 76 patients, the authors reported greater maintenance of disc height and improved 1-year leg pain, back pain, and low-back disability in their annular repair (Barracaid) cohort when compared with controls. By 2-year follow-up, symptomatic recurrent same-level disc herniation was observed in 6.5% of patients in the control cohort compared with 0% in the annular closure cohort. In their follow-up report, potentially significant costs savings with annular closure were estimated to be $222,573 per 100 primary discectomy procedures [28]. In another randomized control trial of the Xclose Tissue Repair System (Anulex Technologies, Minnetonka, MN), the authors observed a significant reduction in the rate of reherniation at 3 and 6 months postoperatively in patients with leg-dominant pain receiving annular repair after discectomy [29]. Non-significant trends favoring repair were observed through 2 years. Annular repair did not lead to increased risks for patients.

Our study has several limitations. A porcine cervical spine was chosen to model the human lumbar spine for which the device is optimized. The porcine cervical spine is a suitable analog for human lumbar spine comparison based on its geometry, anatomy [10], and function [11]. Despite the similarities, it is important to recognize that there are also differences including increased hydration of pig NP as compared with human NP. This difference would in theory increase the likelihood of NP extrusion in our young healthy pig model. Our in vivo study involved a simulated open approach to the cervical disc, and we did not evaluate the use of the device through a tubular system. We also need to recognize the heterogeneity in the morphology of human disc herniations. There may be a subset of patients in whom AF suture repair strategies would be applicable. For example, AF tissues need to be able to be reapproximated and held together by suture closure. Large uncontained AF defects pose a challenge and may require alternate biologic or combined strategies.

Conclusions

In summary, this study shows promise in the use of a novel suture closure device for AF repair following surgical discectomy. A Dines knot with four half-hitches provided the optimal configuration, and the in vivo study validated the feasibility of AF closure sustained through 4 weeks. This motivates a planned clinical study to further evaluate the feasibility, safety, and effectiveness of this device in patients requiring surgery for lumbar disc herniation.

Acknowledgments

This work has been supported through financing provided by the Health Technology Commercialization Program through htx.ca—The Health Technology Exchange (HTX) and funding support from Ontario Centers of Excellence (OCE). This program has also been made available through the support of the Ontario Ministry of Research and Innovation (MRI).

References

[1] Mixter WJ, Barr JS. Rupture of the intervertebral disc with involvement of the spinal cord. N Engl J Med 1934;211:210–14.
[2] Weinstein JN, Lurie JD, Tosteson TD, et al. Surgical vs nonoperative treatment for lumbar disk herniation: the Spine Patient Outcomes Research Trial (SPORT) observational cohort. JAMA 2006;296:2451–9.
[3] Carragee EJ, Han MY, Suen PW, Kim D. Clinical outcomes after lumbar discectomy for sciatica: the effects of fragment type and anular competence. J Bone Joint Surg Am 2003;85-A:102–8.
[4] McGirt MJ, Eustacchio S, Varga P, et al. A prospective cohort study of close interval computed tomography and magnetic resonance imaging after primary lumbar discectomy: factors associated with recurrent disc herniation and disc height loss. Spine 2009;34:2044–51.
[5] Guterl CC, See EY, Blanquer SB, et al. Challenges and strategies in the repair of ruptured annulus fibrosus. Eur Cell Mater 2013;25:1–21.
[6] Blanquer SB, Sharifi S, Grijpma DW. Development of poly(trimethylene carbonate) network implants for annulus fibrosus tissue engineering. J Appl Biomater Funct Mater 2012;10:177–84.
[7] Bron JL, van der Veen AJ, Helder MN, van Royen BJ, Smit TH. Biomechanical and in vivo evaluation of experimental closure devices of the annulus fibrosus designed for a goat nucleus replacement model. Eur Spine J 2010;19:1347–55.
[8] Pirvu T, Blanquer SB, Benneker LM, et al. A combined biomaterial and cellular approach for annulus fibrosus rupture repair. Biomaterials 2015;42:11–19.
[9] Sharifi S, van Kooten TG, Kranenburg HJ, et al. An annulus fibrosus closure device based on a biodegradable shape-memory polymer network. Biomaterials 2013;34:8105–13.
[10] Yingling VR, Callaghan JP, McGill SM. The porcine cervical spine as a model of the human lumbar spine: an anatomical, geometric, and functional comparison. J Spinal Disord 1999;12:415–23.
[11] Tampier C, Drake JD, Callaghan JP, McGill SM. Progressive disc herniation: an investigation of the mechanism using radiologic, histochemical, and microscopic dissection techniques on a porcine model. Spine 2007;32:2869–74.
[12] Dines JS, Moynihan D, Uggen C, Dines DM. The DMD knot: a new locking, flip knot. J Surg Orthop Adv 2008;17:115–18.
[13] Hassinger SM, Wongworawat MD, Hechanova JW. Biomechanical characteristics of 10 arthroscopic knots. Arthroscopy 2006;22:827–32.

[14] Dahl KA, Patton DJ, Dai Q, Wongworawat MD. Biomechanical characteristics of 9 arthroscopic knots. Arthroscopy 2010;26:813–18.
[15] Baums MH, Sachs C, Kostuj T, Schmidt-Horlohe K, Schultz W, Klinger HM. Mechanical testing of different knot types using high-performance suture material. Knee Surg Sports Traumatol Arthrosc 2013;23:1351–8.
[16] Galante JO. Tensile properties of the human lumbar annulus fibrosus. Acta Orthop Scand 1967;(Suppl. 100):101–91.
[17] Callaghan JP, McGill SM. Intervertebral disc herniation: studies on a porcine model exposed to highly repetitive flexion/extension motion with compressive force. Clin Biomech (Bristol, Avon) 2001;16:28–37.
[18] Adams MA, McNally DS, Dolan P. "Stress" distributions inside intervertebral discs. The effects of age and degeneration. J Bone Joint Surg Br 1996;78:965–72.
[19] Panjabi MM, Duranceau JS, Oxland TR, Bowen CE. Multidirectional instabilities of traumatic cervical spine injuries in a porcine model. Spine 1989;14:1111–15.
[20] Atlas SJ, Deyo RA, Patrick DL, Convery K, Keller RB, Singer DE. The Quebec Task Force classification for spinal disorders and the severity, treatment, and outcomes of sciatica and lumbar spinal stenosis. Spine 1996;21:2885–92.
[21] Lebow RL, Adogwa O, Parker SL, Sharma A, Cheng J, McGirt MJ. Asymptomatic same-site recurrent disc herniation after lumbar discectomy: results of a prospective longitudinal study with 2-year serial imaging. Spine 2011;36:2147–51.
[22] Yorimitsu E, Chiba K, Toyama Y, Hirabayashi K. Long-term outcomes of standard discectomy for lumbar disc herniation: a follow-up study of more than 10 years. Spine 2001;26:652–7.
[23] Loupasis GA, Stamos K, Katonis PG, Sapkas G, Korres DS, Hartofilakidis G. Seven- to 20-year outcome of lumbar discectomy. Spine 1999;24:2313–17.
[24] Barth M, Diepers M, Weiss C, Thome C. Two-year outcome after lumbar microdiscectomy versus microscopic sequestrectomy: part 2: radiographic evaluation and correlation with clinical outcome. Spine 2008;33:273–9.
[25] Brinckmann P, Johannleweling N, Hilweg D, Biggemann M. Fatigue fracture of human lumbar vertebrae. Clin Biomech (Bristol, Avon) 1987;2:94–6.
[26] Nachemson AL. Disc pressure measurements. Spine 1981;6:93–7.
[27] Brinckmann P, Grootenboer H. Change of disc height, radial disc bulge, and intradiscal pressure from discectomy. An in vitro investigation on human lumbar discs. Spine 1991;16:641–6.
[28] Parker SL, Grahovac G, Vukas D, Ledic D, Vilendecic M, McGirt MJ. Cost savings associated with prevention of recurrent lumbar disc herniation with a novel annular closure device: a multicenter prospective cohort study. J Neurol Surg A Cent Eur Neurosurg 2013;74:285–9.
[29] Bailey A, Araghi A, Blumenthal S, Huffmon GV, Anular Repair Clinical Study Group. Prospective, multicenter, randomized, controlled study of anular repair in lumbar discectomy: two-year follow-up. Spine 2013;38:1161–9.