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The Leakage Pathway and Effect of Needle Gauge on Degree of Disc Injury Post Anular Puncture

A Comparative Study Using Aged Human and Adolescent Porcine Discs

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Study Design. An *in vitro* biomechanical study using aged human and adolescent porcine discs.

Objectives. To find the leakage pathway and effect of needle gauge on the degree of disc injury post anular puncture.

Summary of Background Data. Spinal needles are widely used for minimal invasive disc surgeries and disc degeneration/regeneration research. Applications of anular puncture require different diameters of spinal needles. However, the effect of needle diameters on the disc injury has not been systematically studied yet.

Methods. Four groups of experiments were conducted: 1) porcine thoracic disc, 2) human thoracic disc, 3) porcine thoracic disc with 200 N external loading, and 4) porcine lumbar discs. The disc was punctured consecutively with needles from smaller diameter to larger diameter. After each anular puncture, the quantitative discomanometry technique was conducted to quantify the disc rupture pressure and volume. The association between needle gauge and rupture pressure and volume was analyzed.

Results. The degree of disc injury increased with the diameter of needle. For an aged human thoracic disc, the anulus fibrosus cannot hold pressure more than 2 MPa after a 21-gauge-needle-anular-puncture. The leakage pathway of injected saline was through the anular fissure but was through the endplate when the disc was next to an osteoporotic vertebrae. The pressure holding power of porcine disc is stronger than of human disc. The rupture pressure of porcine lumbar disc is higher than of porcine thoracic disc. The axial compressive external loading increased the disc rupture pressure. The rupture volumes were not affected by the dimension of injury fissure. The rupture volume was at level of 0.3 mL without external loading and at 0.2 mL with external loading.

Conclusion. A spinal needle of ≤22 gauge and injection volume of ≤0.2 mL are recommended to prevent postsurgery leakage.

Key words: disc integrity, spinal needle, quantitative discomanometry, endplate leakage. **Spine 2007;32:1809–1815**

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Intervertebral disc disorders, such as the herniation of nucleus pulposus and disc degeneration, are among the major musculoskeletal diseases. Conventional treatments include medication, steroid injection, physical therapy, and surgery. When nonsurgical treatments fail, surgical intervention, such as nucleotomy[1] or microdiscectomy,[2] may be required. Minimal invasive spine procedures,[3] such as the intradiscal electrothermal therapy, radiofrequency ablation, percutaneous laser disc decompression, chemonucleolysis, and percutaneous lumbar discectomy require 17-gauge to 22-gauge spinal needles to deliver the treatment into the disc (Table 1).[4–12] Spinal needles are also used to deliver materials, such as growth factor, plasmids, and cells into the nucleus for disc regeneration study.[13] It is possible that the anular puncture might create disc injury and the injected materials may leak through the anular fissure due to the anular puncture. Knowledge about the leakage pathway of injected materials and the degree of injury induced by anular puncture is helpful for designing surgery protocols.

In addition to delivering treatments or materials into the nucleus, the anular puncture technique was also used to create stab injury for disc degeneration research (Table 2).[14–30] Needles with diameters ranging from 16 gauge to 27 gauge were used for anular puncture in the rabbit model. The results showed that anular puncture with 16-gauge and 18-gauge needles produced a significant MRI degradation. However, this is not the case with the 21-gauge anular puncture.[16] A very fine needle anular puncture (27 gauge) was proved not to produce the disc degeneration.[22] Extrapolating the "disc injury–needle gauge" association from rabbit disc to human disc may be inaccurate due to geometric dissimilarity. The porcine disc is analogous to the human disc because of the morphologic and geometric similarity.[31] Therefore, it was used for disc degeneration study through the stab endplate injury[29,30] or the stab disc injury model.[28] Information about the effect of needle gauge on disc injury in the porcine disc will be helpful for future *in vivo* porcine disc degeneration/regeneration research.

The purpose of this study is to find the leakage pathway and degree of disc injury induced by different needle gauges in aged human and adolescent porcine discs. Based on the results, advice about needle usage in human or porcine discs in clinical and research applications can be provided.

Materials and Methods

Specimen Preparation. Fresh aged human thoracic spine and adolescent porcine thoracic and lumbar spine specimens were

Table 1. Spinal Needle Used for Clinical Application

Gauge No.	Application	Reference	Year
17G	Intradiscal electrothermal therapy (IDET)	Freeman *et al*[4]	2005
		Davis *et al*[5]	2004
		Saal and Saal[6]	2000
18G (lumbar) 22G (cervical)	Percutaneous laser disc decompression (PLDD)	Choy[7]	2004
18G	Chemonucleolysis (CNL)	Kim *et al*[8]	2002
		Burton *et al*[9]	2000
18G	Percutaneous lumbar discectomy (PLD)	Bonaldi[10]	2003
20G	Radiofrequency ablation (RFA)	Ercelen *et al*[11]	2003
		Barendse *et al*[12]	2001

Only recent representative research papers were listed.

used in this study. Fresh frozen human cadaveric thoracic discs from 3 spinal columns with an average age of 70 (range, 66–73 years) were used. The spine specimens were examined by MR (1.5T Excite HD, GE), CT (LightSpeed VCT, GE) and DEXA (Prodigy, GE) scanning. The human disc abnormality was examined by T2-weighted MR images. After the experiment, the human discs were cross-sectional dissected to find the grade of degeneration. The disc size was determined by the adjacent endplate geometry from CT stack images using an in-house imaging analysis tool. Bone qualities including bone mineral density (BMD) and T-score of adjacent vertebral body of tested discs were determined by DEXA scanning. Porcine thoracic and lumbar discs were harvested from 6-month-old swine. Six-month-old swine are approximately at the age of sexual maturity, but not musculoskeletal maturity. All specimens were carefully cleaned of muscle tissue, placed in double plastic bags, and frozen at −20°C. Before the experiments, the specimens were removed from the freezer and kept at room temperature for 6 hours. The specimens were wrapped with gauze and saline during thawing.

Quantitative Discomanometry (QD) Apparatus. The QD technique[32–35] was used to measure the injected fluid volume and developed hydrostatic pressure within the disc to quantify disc functional integrity.[34,35] A high pressure-QD apparatus was developed to differentiate the injured disc function from the intact one (Figure 1).[28] In brief, the QD apparatus was composed of a spinal needle, a high-pressure syringe, high-pressure 3-way stopcocks, a programmable X-table that produced horizontal thrust, a pressure transducer for pressure measurement, a linear potentiometer for volume measurement, and a computer for data acquisition. The maximum pressure of the system was 5 MPa. The linear range of the system was 4.5 MPa and the error of linearity was under 0.5%. The pressure and volume resolution of the system were 0.01 MPa and 0.002 mL, respectively. The in-

Table 2. Animal Models for Disc Degeneration Research

Animal	Gauge	DAI	Purpose of Needle Intervention	Reference	Year
Small size					
Rat	22G	II	NP exposure	Inoue *et al*[14]	2006
	27G	NA	CABC injection (CNL)	Norcross *et al*[15]	2003
Rabbit	#11 blade	II	NP exposure	Masuda *et al*[16]	2005
	16G	I	AF puncture	Sobajima *et al*[17]	2005
	16G	I	AF puncture	Masuda *et al*[16]	2005
	18G	I	AF puncture	Masuda *et al*[16]	2005
	18G	I	AF puncture	Kim *et al*[18]	2005
	21G	I	AF puncture	Masuda *et al*[16]	2005
	21G	I	NP aspiration and AF puncture	Kim *et al*[18]	2005
	23G	NA	Camptothecin injection	Kim *et al*[18]	2005
	25G	NA	CABC injection (CNL)	Toyomi *et al*[19]	1996
	26G	NA	CABC and chymopapain injection (CNL)	Sumida *et al*[20]	1999
	26G	NA	Chymopapain injection (CNL)	Lu *et al*[21]	2004
	27G	0	Osteogenic protein injection	An *et al*[22]	2005
Medium size					
Monkey	23G	NA	CABC and chymopapain injection (CNL)	Sugimura *et al*[23]	1996
Canine	23G	NA	CABC injection (CNL)	Takahashi *et al*[24]	1997
Large size					
Sheep	#11 blade	II	NP exposure	Lappalainen *et al*[25]	2002
	#20 blade	I	AF puncture	Lappalainen *et al*[25]	2002
	19G	I	IDET	Freeman *et al*[26]	2003
	21G	NA	CABC injection (CNL)	Sasaki *et al*[27]	2001
Mini-swine	18G	I	AF puncture	Wang *et al*[28]	2006
Swine	1.5-mm wire	0	Endplate injury	Cinotti *et al*[29]	2005
	3.5-mm drill	0	Endplate injury	Holm *et al*[30]	2004

AF indicates anulus fibrosus; NP, nucleus pulposus; CABC, chondroitinase ABC; IDET, intradiscal electrothermal therapy; CNL, chemonucleolysis; DAI, degree of anular injury; NA, not available; 0, no immediate injury; I, minor injury that disc degenerates gradually; II, major injury that the nucleus protrudes immediately.

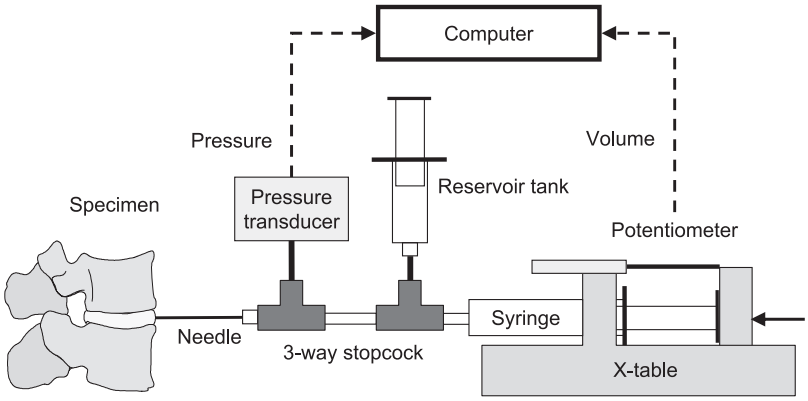

Figure 1. Apparatus of quantitative discomanometry (QD). An industrial X-table was used to drive the syringe at a constant flow rate. The potentiometer was used to record the volume of saline injected into the disc. The pressure transducer was used to record the developed pressure. The system was able to produce and measure the pressure up to 5 MPa.

jection volume was 0.5 mL, and the injection rate was fixed at 0.5 mL/min.

Protocol of QD Test. Four groups of experiments were conducted. The first group, which served as the baseline for comparison, was the test of porcine thoracic disc without external loading. The second group of the experiment was to test the difference of aged human specimen from adolescent porcine specimen. The third group of the experiment was to test the effect of external loading. The fourth group of the experiment was to find the difference between porcine thoracic and lumbar discs. Numbers of specimens of each group are shown in Table 3.

The approach of QD test was through one site of anterolateral region of disc and the anular puncture was through the other site of anterolateral region of disc. A pilot study showed that the results of QD test was not affected by the approach of anular puncture. The needle tip of QD apparatus was carefully placed into the center of the nucleus with the help of a plastic stopper/indicator. The disc was first anular punctured with a fine spinal needle (22 gauge for thoracic discs and 18 gauge for lumbar discs), and then the QD test was conducted. During the QD test, the site where the injected saline leaked was searched. If the saline leaked through the vertebrae due to the failure of the endplate, no more anular punch and QD tests were done. If the leakage was through the anular fissure induced by anular puncture, the disc was then punctured by the spinal needle with a smaller diameter (21 gauge for thoracic discs and 17 gauge for lumbar discs) and gradually increased to that of 14 gauge. The subsequent punctures by increasingly larger needles were done through the exactly already existed fissured path. The gradually increased injury protocol was adopted from a verified protocol, the "incremental trauma."[37] The incremental trauma protocol allows for the investigation of injury progression from mild injury to severe injury. The cumulative damage of incremental trauma protocol was proved to be minimal.[38]

Table 3. No. of Specimens in Each Group

Group	Specimen	External Loading	No. of Total Specimens	No. of Sample That Leaked Through Endplate
G1	Porcine thoracic disc	No	8	0
G2	Human thoracic disc	No	15	8
G3	Porcine thoracic disc	200 N	8	0
G4	Porcine lumbar disc	No	17	8

Data are mean (SD).

Our validation study also showed that the results of QD test remained unchanged even after 5 repetitive QD tests. Two types of testing environment were applied during QD test, one is discs without external load, and another one is the discs with 200 N axial compressive loading through out the QD test.

After each anular puncture, the QD technique was used to quantify the disc rupture pressure and volume. The rupture pressure is the maximal pressure during the injection history (Figure 2). The higher rupture pressure means better fluid sealing capability of the disc. The effect of needle diameter on the rupture pressure and volume was analyzed. The internal and external diameters of the needles are showed in Table 4.

Statistical Analysis. One-tailed independent *t* test was performed to find the difference of rupture pressure for Group 1 *versus* Group 2, Group 1 *versus* Group 3, and Group 1 *versus* Group 4 under the same anular puncture condition. The one-tailed independent *t* test was also performed to examine the difference in BMD and T-score between the endplate leakage and anular leakage of human specimens. The one-tailed dependent *t* test was conducted to find the difference in rupture pressure for discs punctured with different needle gauges. All tests were considered to be significant at the α level of 0.05.

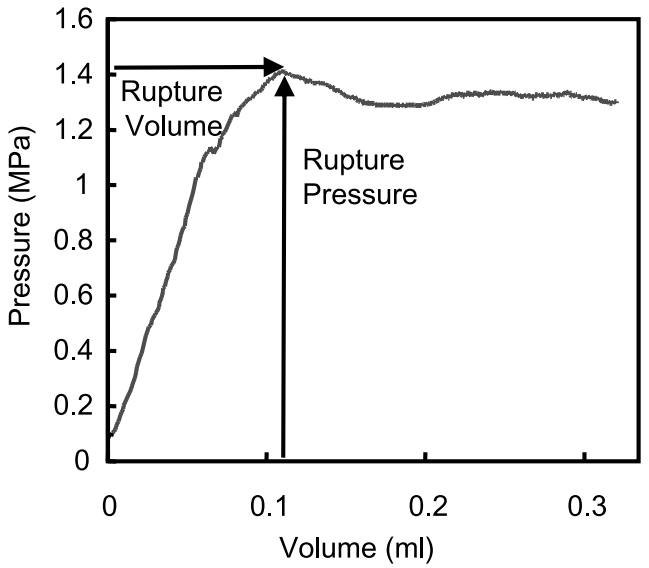

Figure 2. Schematics of a pressure-volume curve obtained using QD apparatus. The rupture pressure is the maximal pressure during the injection history.

Table 4. Diameters (mm) of Needle Gauge

	Gauge No.													
	14G	15G	16G	17G	18G	19G	20G	21G	22G	23G	24G	25G	26G	27G
Inside diameter	1.6	1.4	1.37	1.17	0.96	0.77	0.69	0.52	0.42	0.33	0.31	0.26	0.21	0.16
Outside diameter	2.0	1.8	1.67	1.47	1.26	1.07	0.90	0.82	0.72	0.63	0.55	0.50	0.45	0.40

■ Results

All porcine discs used were rated Grade 1 according to the criteria by Galante.[39] The T2-weighted MR image of human discs showed no sign of disc bulging, protrusion, or herniation. The cross-sectional dissection of human disc showed Grade 2 degeneration according to Galante's criteria.[39] The sizes of the tested human and porcine discs are similar to those of human discs documented in literature (Table 5).[40–43]

The rupture pressures of injured discs significantly decreased as the diameter of the needle gauge increased ($P < 0.05$) (Figures 3, 4). The adolescent porcine disc punctured with 20-gauge and 16-gauge spinal needles cannot hold a disc pressure of more than 2 MPa and 1 MPa. The pressure holding capability of human thoracic disc was weaker than of porcine thoracic disc under the same stab injury condition ($P < 0.05$ for needles of 18 gauge and larger diameters, comparison between Group 1 and Group 2) (Figure 3). The aged human thoracic discs punctured with 21-gauge and 18-gauge spinal needles cannot hold a disc pressure of more than 2 MPa and 1 MPa. On average, the external loading increased the disc pressure holding capability by 0.91 MPa (SD, 0.25 MPa) in 21-gauge and 20-gauge-needle-injured discs ($P < 0.05$), and by 0.35 MPa (SD, 0.07 MPa) in 19-gauge to 14-gauge-needle-injured discs ($P = 0.021$–0.151, comparison between Group 1 and Group 3) (Figure 3). The pressure holding capability of porcine lumbar discs was higher than the one of porcine thoracic discs ($P < 0.05$, comparison between Group 1 and Group 4) (Figure 4). The porcine lumbar disc punctured with 17-gauge and 14-gauge spinal needles cannot hold a disc pressure of more than 2 MPa and 1 MPa, respectively.

The leakage of injected saline in porcine thoracic discs was always through anular fissure. However, leakage of injected fluid in aged human thoracic discs and adolescent porcine lumbar discs were through either the endplate or the anular fissure. The rupture pressure of the human thoracic disc was 0.79 MPa (SD, 0.41 MPa) in the observance of endplate leakage. The average BMD of the human specimens that leaked through endplate was 0.652 g/cm^2 (T-score = −3.3), and the one that leaked through anulus fissure was 0.781 g/cm^2 (T-score = −2.0). The rupture pressure of the porcine lumbar disc was 3.84 MPa (SD, 0.43 MPa) in the observance of endplate leakage (Table 6).

The rupture volumes were not affected by the dimension of injury fissure. The average rupture volume of porcine thoracic discs was 0.29 mL (SD, 0.06 mL), whereas the one of human thoracic discs was 0.34 mL (SD, 0.02 mL). The external loading decreased the rupture volume to 0.21 mL (SD, 0.05 mL). The rupture volume of porcine lumbar disc (0.27 mL [SD, 0.06 mL]), however, was not larger than the one of thoracic discs.

■ Discussion

The QD test is a reliable biomechanical technique for measuring disc functional integrity. The degree of disc injuries can also be evaluated by medical imaging or biomechanical function. The quantitative indexes of medical imaging include disc height[22,44–46] and MRI signal intensity. The quantitative indexes of biomechanical function include spinal stability,[47,48] disc compressive stiffness,[49–52] and anulus fibrosus hydraulic permeability.[50] The advantage of the QD test is that it measures the "confine fluid" capability of disc, *i.e.*, an essential biomechanical function related to the disc degeneration pathway.[53] A disc with a higher degree of functional integrity is expected to reach a higher degree of spinal

Table 5. Dimensions (mm) of Human and Porcine Thoracic and Lumbar Discs

	Porcine Thoracic disc in Current Study	Human Thoracic Disc in Current Study	Human Thoracic Disc in Literature	Porcine Lumbar Disc in Current Study	Human Lumbar Disc in Literature
Disc height	5.2 (0.3)	4.5 (0.6)	6.6 (1.4)[40]	7.0 (0.8)	8.9 (0.11)[41] 10.7 (0.5)[40]
Disc width	32.0 (2.6)	EPDu = 35.9 (5.1) EPDl = 36.3 (4.2)	EPWu* = 28.6 (4.7)[42] EPWl* = 31.0 (5.3)[42]	36.5 (1.8)	EPWu* = 44.4 (2.6)[43] EPWl* = 47.1 (2.7)[43]
Disc depth	24.1 (0.5)	EPWu = 33.6 (5.6) EPWl = 33.5 (5.8)	EPDu* = 26.2 (4.6)[42] EPDl* = 27.3 (4.4)[42]	24.9 (1.0)	EPDu* = 34.8 (0.5)[43] EPDl* = 34.4 (0.9)[43]

Data are mean (SD).
*Data are based on the measurement of endplate.
EPWu indicates upper endplate width; EPWl, lower endplate width; EPDu, upper endplate depth; EPDl, lower endplate depth.

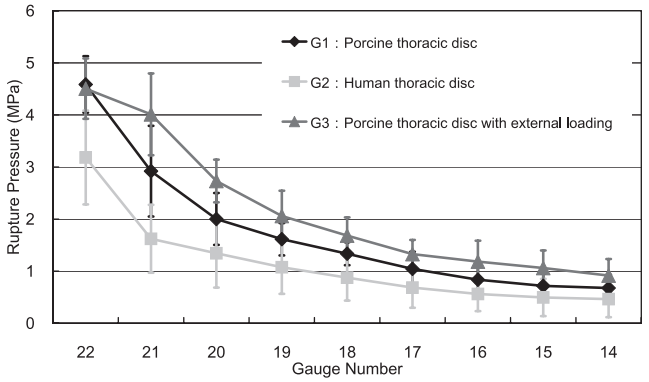

Figure 3. The effect of needle gauge on rupture pressures of G1 (porcine thoracic discs), G2 (human thoracic disc), and G3 (porcine thoracic discs with 200 N external loading). Error bars represent ± SD.

stability, disc stiffness, and lower anulus fibrosus permeability.

The porcine disc is analogous to the human disc in terms of anatomic, geometric and functional characteristics.[31,54] The loading axis of the quadruped spine, like the human spine, was mainly along its long axis.[55] These factors make the swine a valuable animal for disc research. In this study, we compare the results between adolescent porcine and aged human discs. The disparity of rupture pressure between adolescent porcine and aged human discs can result from the difference in the morphology or physical condition (healthy *vs.* degenerated) of the discs. It is difficult to obtain degenerated porcine discs as well as healthy young human discs to make a fair comparison. The "disc injury-needle gauge" curves of aged human and adolescent porcine discs showed in Figure 3 and 4 may be the range of the curve for healthy young human discs.

The bone quality of adjacent vertebral body affected the pattern of fluid leakage. The injected saline leaked through either the endplate or anular fissure in aged human discs (which are adjacent to osteoporotic vertebral bodies), but leaked only through the anular fissure in adolescent porcine thoracic discs (which are adjacent to intact healthy vertebral bodies). The bone quality (BMD,

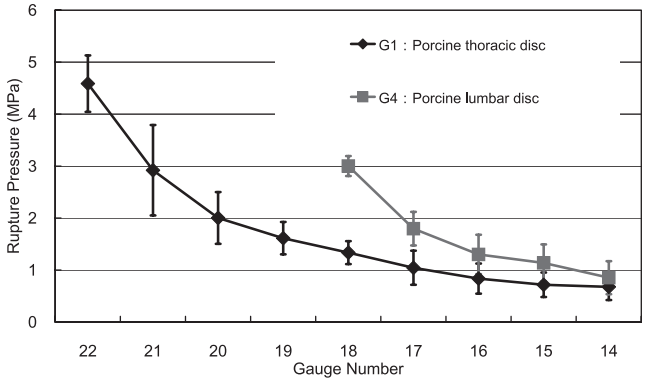

Figure 4. The effect of needle gauge on rupture pressures of G1 (porcine thoracic discs) and G4 (porcine lumbar disc). Error bars represent ± SD.

$P = 0.080$ and T-score, $P = 0.113$) of the endplate leakage group tended to be lower than that of the anular leakage group. The rupture pressure of the endplate is connected with the bone quality of the adjacent vertebral body. It is predicted that, for an intact disc of severely osteoporotic spinal column, the pressurized fluid may protrude through the endplate instead of the anulus fibrosus. The disc rupture pressure of the endplate leakage group is virtually the rupture pressure of the endplate.

The rupture pressure of the intact healthy human disc has not been reported. Our previous study found that the rupture pressure of an intact porcine disc was above 5 MPa (*i.e.*, the limitation of the machine). The rupture pressure[28] of an intact bovine disc was 18 MPa.[56] However, discomanometry tests in the literature showed that the rupture pressure of human discs was only 0.96 MPa (SD, 0.13 MPa),[57] and the rupture pressure of the degenerated human disc was <0.5 MPa.[34] Our study showed that the rupture pressure of the endplate leakage group was at the level of 0.8 MPa. It is speculated that the reported disc rupture pressure was mainly due to the fracture of the endplate, rather than the failure of the anulus fibrosus. The *in vivo* measurements[58,59] showed that the intradiscal pressure was up to 2.3 MPa when lifting 20 kg with round flexed back. The rupture pressure of an intact healthy disc should be above 3 MPa for maintaining normal daily loading.

The rupture pressure of human lumbar discs may be speculated with results of porcine lumbar disc test. The rupture pressure of the porcine lumbar endplate (3.84 MPa [SD, 0.43 MPa]) is lower than that of thoracic endplate (above machine's limit, *i.e.*, 5 MPa). However, the pressure holding capability of porcine lumbar anulus fibrosus is higher than of thoracic anulus fibrosus under the same stab injury condition. If this association existed in human spine, it can be extrapolated that the human lumbar disc may have higher tolerance of using larger diameter of spinal needle. However, for a lumbar disc next to an osteoporotic vertebrae, the rupture pressure of lumbar endplate may be even lower than of thoracic endplate (0.8 MPa). The fluid/materials injected into the lumbar disc may easily leak to the adjacent osteoporotic vertebrae.

The loss of disc rupture pressure due to anular puncture is a critical issue for long-term disc healing. An early study indicated that the exposure of nucleus pulposus would initiate disc degeneration due to the "loss of confined fluid mechanics."[60] Osti *et al*[61] showed that the disc degeneration can be induced by acute anular injury without immediate herniation using a sheep model. The loss of the disc integrity may not be the only reason for disc degeneration, but in the long-term, the degenerated disc cannot hold the disc integrity.[17] For example, the rupture pressure of the burst fractured human disc was 0.3 MPa,[35] and the rupture pressure of the degenerated human disc was below 0.5 MPa.[34] In this study, we found a disc with an injury fissure larger than an 18-gauge needle cannot hold the intradiscal pressure under

Table 6. No. of Specimens, Rupture Pressure, BMD, and T-Score of Human Thoracic Discs (G2) and Porcine Lumbar Discs (G4) That Leaked Through Endplate and Anular Fissure

Group	Leakage Site	No. of Specimens	Rupture Pressure (MPa)	BMD (g/cm^2)	T-Score
G2	Endplate	8	0.79 (0.41)	0.652 (0.140)	−3.3 (1.5)
	Anular fissure	7	Depends on needle gauge	0.781 (0.037)	−2.0 (0.5)
				$P = 0.080$	$P = 0.113$
G4	Endplate	8	3.84 (0.43)	—	—
	Anular fissure	9	Depends on needle gauge	—	—

Data are mean (SD).

normal loading and may create a vicious cycle for disc healing hence initiating the disc degeneration process. Our previous *in vivo* porcine study confirmed that an 18-gauge needle punctured disc cannot recover after a 2-month healing period.[28] The results of the present study may represent the *in vivo* acute stage as well as the midterm (2 month) consequence of the stab injury of the disc.

The *in vivo* needle selection may be advised with the results of current study. The intradiscal pressure was reported to be in the range of 0.5 MPa[62] to 1 MPa[63] during upright standing, of 0.6 MPa[62] to 1 MPa[63] during upright sitting, and of 0.1 MPa[62] to 0.3 MPa[63] during lying in the prone position. It can be inferred that 2 MPa is the range of intradiscal pressure during loaded activity and 1 MPa is the range during normal posture. This study found external loading increased the rupture pressure of injured discs due to the compressed anulus fibrosus and deceased size of injury. The *in vivo* external loading increases the rupture pressure of injured disc. However, the magnitude of *in vivo* external loading varies a lot, thus the quantitative protection due to external loading may not be easily defined. The human "rupture pressure–needle gauge" curve in Figure 3 can represent the worst scenario. If this is the case, when the postsurgery leakage is a concern, the patient/subject of an anular puncture using a 21-gauge needle or larger should minimize lifting or load-carrying activities, and in the case of an anular puncture using an 18-gauge needle or larger, lifting or load-carrying activities should be avoided.

■ Conclusion

When anular puncture is used for delivering material into the nucleus, a spinal needle of ≤22 gauge and injection volume of ≤0.2 mL is recommended to prevent postsurgery leakage.

■ Key Points

- The degree of disc stab injury increases with the diameter of the spinal needle.
- For an aged human thoracic disc, the anulus fibrosus cannot hold pressure more than 2 MPa after a 21-gauge needle anular puncture. When the disc is next to an osteoporotic vertebra, the injected fluid leaks through endplate, whose rupture pressure is 0.8 MPa.
- The pressure holding power of adolescent porcine disc is stronger than of aged human disc under the same stab injury condition.
- The rupture pressure of porcine lumbar disc is higher than of porcine thoracic disc.
- The axial compressive external loading increases the disc rupture pressure.
- When anular puncture is used for delivering material into the nucleus, a spinal needle of ≤22 gauge and injection volume of ≤0.2 mL is recommended to prevent postsurgery leakage.

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