phy,[5–7,9,13,14,20] which can lead to failed–back surgery syndrome.[21] To overcome these problems, various decompressive surgeries for LCS have been developed with the intention of preserving the posterior supporting structures of the lumbar spine.[16,17,19,22,26–29]

In 2002, we developed a new lumbar laminectomy technique for the treatment of LCS.[25] In this technique, which we named LSPSL (or the "split laminectomy"), the lamina is exposed by longitudinally splitting the spinous process into halves, with the muscular and ligamentous attachments to the spinous process left intact (Fig. 1). According to our retrospective study, a group of patients with persistent severe pain and progressive neural dysfunction caused by LCS benefited from LSPSL and their back muscles were significantly well preserved, compared with patients who underwent conventional laminectomy.[10,25] Based on our clinical experience with LSPSL, we assumed that the split laminectomy preserves the back muscles, leading to a reduction in acute wound pain after surgery.

The purpose of this prospective, randomized, controlled study was to evaluate acute postoperative wound pain after either LSPSL or conventional laminectomy.

Methods

This prospective, randomized study was approved by the medical ethics committee of Keio University Hospital.

Inclusion and Exclusion Criteria

To be eligible for the study, the following criteria had to be met: 1) presence of neurogenic claudication; 2) symptoms persistent for more than 6 months despite conservative therapy; 3) clinical symptoms and neurological signs in the lower limbs corresponding to the level of stenosis on MR imaging or myelography; and 4) 1- or 2-level decompression necessary.

Radiographic instability of the lumbar spine and degenerative spondylolisthesis were not regarded as exclusion criteria. However, the following exclusion criteria were adopted: 1) spinal canal stenosis due to congenital, spondylolytic, traumatic, and iatrogenic causes; 2) any previous operation in the lumbar area; 3) presence of other specific spinal disorders (such as ankylosing spondylitis, neoplasm, or metabolic diseases); 4) intermittent claudication resulting from peripheral arterial disease; 5) severe osteoarthrosis or arthritis in the lower limbs; 6) neurological disease causing impaired lower-limb function, including diabetic neuropathy; 7) psychiatric disorders; and 8) multilevel spinal canal stenosis requiring decompression at 3 or more levels.

Patient Population

Patients were recruited between December 2004 and December 2005. Before randomization, eligible patients were informed that they would be the candidates for a study

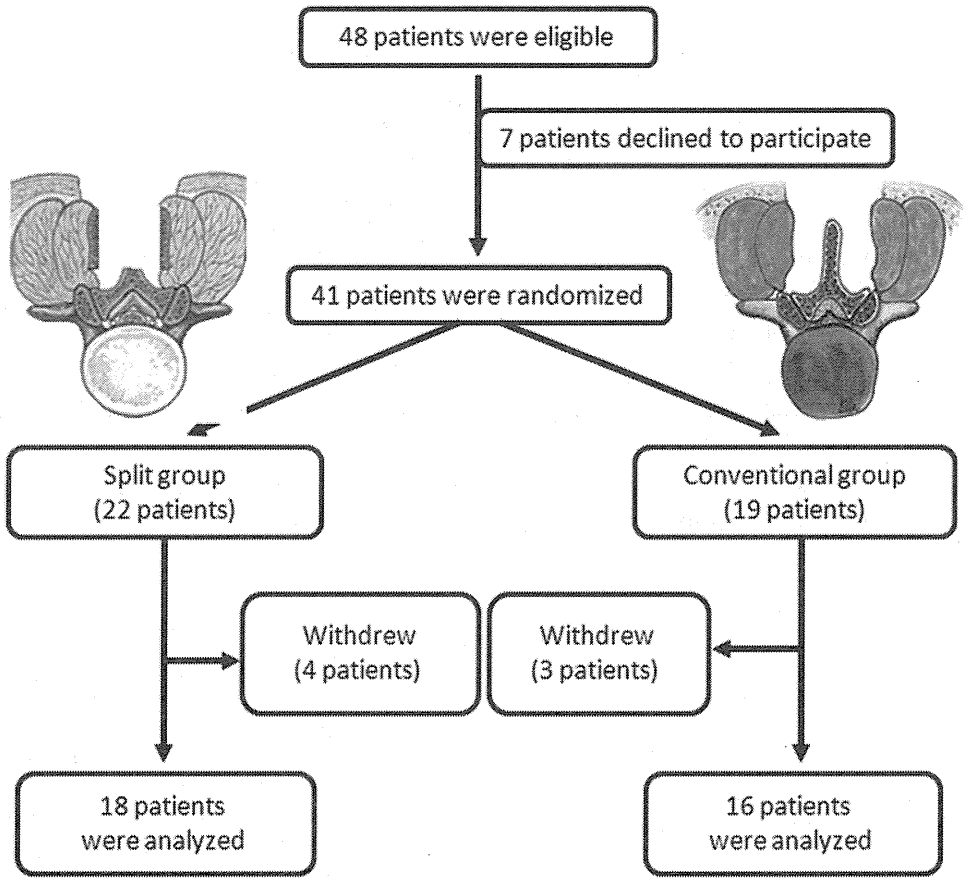

Fig. 1. Flow chart of the study and drawings representing the 2 laminectomies.

comparing 2 different procedures for the surgical treatment of LCS. Then, written consent was obtained from all participants. The baseline examinations consisted of plain radiography of the lumbar spine, myelography, CT, and MR imaging. Standard physical and neurological examinations were performed, and the JOA score for low-back pain[4] was calculated. The JOA Scale consists of 3 main parts: subjective symptoms, clinical signs, and ADL restrictions. The subjective symptoms section has 3 components: low-back pain, leg pain, and gait, with all 3 components having a possible score range of 0–3 points depending on severity. The clinical signs section also has 3 components: straight leg–raising test, sensory disturbance, and muscle disturbance, with all 3 components having a possible score range of 0–3 depending on the severity. The ADL section has 7 components based on the individual's ability to: turn over while lying down, stand, wash one's face, lean forward, sit for approximately 1 hour, lift or hold heavy objects, and ambulate. Each of these components is scored according to the severity of restriction (severe restriction, 0; moderate restriction, 1; no restriction, 2). After completion of the baseline examinations, identification data for the eligible patients who agreed to participate in the study were faxed to a central office, where they were randomized into either the LSPSL group or the conventional laminectomy group. The randomization was stratified according to sex, age (≤ 64 years or > 65 years), and the number of decompression levels (1 or 2). Only the attending surgeon of the patient was informed about which method to use for the specific patient. Overall, 5 spine surgeons participated in this study.

Forty-eight patients met the inclusion criteria for the study (Fig. 1), and 41 agreed to participate. These patients were assigned to either the LSPSL group (22 patients) or the conventional laminectomy group (19 patients). Four patients in the LSPSL group were subsequently withdrawn from the study because a conversion to posterior lumbar interbody fusion was necessary in 1 patient and the number of decompression levels was increased from 2 to 3 after randomization in 3 patients. Three patients in the conventional laminectomy group were also withdrawn from the study. In 1 of these 3 patients, a herniotomy was performed in addition to the laminectomy, and in the other 2 patients the number of decompression levels was increased from 2 to 3 levels after randomization.

Measurements

Questionnaires were administered on PODs 3 and 7. The questionnaire contained 3 questions regarding wound pain (intensity, depth, and duration) and questions regarding ADL. For the question concerning the intensity of the wound pain, a VAS consisting of a numerical rating scale from 0 (no pain) to 10 (worst possible pain) was used. For the question concerning depth of pain, a patient in whom pain felt "superficial" was assigned 1 point, "deep to the muscle" was assigned 2 points, and "deep to the bone" was assigned 3 points. For the question concerning duration of pain (that is, from the start of the pain to the relief of the pain), "a few seconds" was assigned 1 point, "a few minutes" was assigned 2 points, "a few hours" was assigned 3 points, and "all day" was assigned 4 points. For the evaluation of ADL, the ADL domain in the JOA Scale (JOA-ADL score) was used. One year postoperatively, standard physical and neurological examinations were performed, and the JOA score was calculated.

In addition to the questionnaire, we also evaluated the pre- and postoperative serum levels of CRP and CPK, the amount of pain medication taken during the 3-day postoperative period, and the muscle atrophy rate as demonstrated on 1-month postoperative axial MR images. Pain medication usage was determined by counting the number of tablets dispensed by the nurses at each patient's request. A nonsteroidal antiinflammatory drug, Loxoprofen, which is a popular analgesic in Japan, was used in the present study.

To evaluate the magnitude of the surgical damage to the paraspinal muscles, the cross-sectional area of the paraspinal muscles (multifidus) was measured on preoperative and 1-month postoperative T2-weighted axial MR images using the NIH Image software (Imaging Research). The axial images were obtained at the decompressed intervertebral level. In cases of 2-level decompression, the caudal level was chosen for the evaluation of muscle atrophy because the influences of the injury to the medial branch of the posterior ramus of the spinal nerve would be more evident at the caudal level. The rate of muscle atrophy was calculated using the following formula: atrophy rate (%) = (1 − total postoperative area/total preoperative area) × 100.

Statistical Analysis

Data were analyzed using the SPSS 16.0 J statistical software package. A Mann-Whitney U-test was used to compare the mean scores or values between the groups. Statistical significance was defined as $p < 0.05$ for a 2-sided hypothesis. Mean data are presented ± SD.

Surgical Techniques

Split Laminectomy. For a 1-level (L4–5) decompression, a posterior midline skin incision is made between the L-3 and L-5 spinous processes to expose the tip of the L-4 spinous process (Fig. 1). The cortex of the tip of the L-4 spinous process is removed at the midline using a high-speed drill with a fine 2-mm diamond-tipped bur, and then, using an osteotome, the spinous process is divided to the base and detached from the L-4 lamina, leaving the bilateral paraspinal muscles attached to the lateral aspects of the split spinous process. The supra- and interspinous ligaments between L3–4 and L4–5 are also split longitudinally with a scalpel. An ample working space for the laminectomy is obtained by retracting the split halves of the spinous process bilaterally, together with its attached paraspinal muscles. The L-4 lamina is removed using a high-speed drill and Kerrison rongeurs, and the nerve tissue is decompressed in the standard fashion. After the decompression of the affected nerve roots and the thecal sac, the halves of the split L-4 spinous process are reapproximated using a strong nonabsorbable suture. The 1-level posterior L4–5 decompression, thus, can be accomplished by removing the L-4 caudal portion of the lamina, preserving the supra- and interspinous ligaments of L3–4 and L4–5 and the L-4 spinous process, with minimal damage to the paraspinal muscles.

Conventional Laminectomy. A posterior midline skin incision is made between the L-3 and L-5 spinous processes to expose the L4–5 interlaminar space (Fig. 1). Bilateral paraspinal muscles are detached from the L-4 and L-5 spinous processes, and then, using a chisel to expose the L4–5 interlaminar space, the L-4 and L-5 spinous processes are detached from the lamina. Last, the nerve tissue is decompressed in the same manner as for the LSPSL.

Postoperative Care

The patients in both groups were allowed to sit up and walk without lumbar support on the 1st POD.

Results

Data in 18 patients (10 men and 8 women) in the LSPSL (split-laminectomy) group and 16 patients (8 men and 8 women) in the conventional laminectomy group were included in the final analyses. We observed no significant intergroup differences in mean age at the time of surgery, mean number of decompressed levels, mean operative time, mean intraoperative blood loss, or number of surgeries performed by each surgeon (Table 1). The mean preoperative JOA score in the LSPSL group was 16.4 ± 4.7 and that in the conventional laminectomy group was 14.9 ± 4.0 (Table 2). The subjective symptoms portion of the JOA score indicated that lower-leg pain and gait disturbance were severe in both groups. No significant intergroup difference was recognized in 3 parts of subjective symptoms, clinical signs, and ADL restrictions. At 1 year postoperatively, the mean preoperative JOA score of 16.4 ± 4.7 was increased to 25.8 ± 3.4 (mean recovery rate 75% ± 21%) in the split-laminectomy group, whereas in the conventional laminectomy group the mean preoperative JOA score of 14.9 ± 4.0 was increased to 25.4 ± 2.9 (mean recovery rate 74% ± 17%). Significant intergroup differences were not recognized in these clinical parameters. No obvious perioperative complication was recognized. One patient in the split-laminectomy group underwent posterior intervertebral fusion 1 year later to treat symptoms that recurred 6 months after LSPSL. One patient in each group did not attend the 1-year follow-up examination because of general health problems.

TABLE 1: Demographics of the split-laminectomy group (LSPSL) and the conventional laminectomy group*

Parameter	Laminectomy Group (%)	
	Split (LSPSL)	Conventional
no. of patients	18	16
mean age (yrs)†	69 ± 10	71 ± 8
male/female ratio†	10:8	8:8
mean no. of decompressed levels†	1.4 ± 0.5	1.4 ± 0.5
mean intraoperative time (min)†	69 ± 29	82 ± 36
mean intraoperative blood loss (g)†	44 ± 75	55 ± 48
no. of ops performed by each surgeon‡		
K.I.	4 (22)	6 (38)
Y.O.	3 (17)	1 (6)
H.T.	4 (22)	2 (13)
M.N.	1 (6)	1 (6)
M.M.	6 (33)	6 (38)

* Mean values are presented ± the SD.
† No significant intergroup difference according to the Mann-Whitney U-test.
‡ No significant intergroup difference according to the chi-square test.

TABLE 2: Clinical results preoperatively and 1-year postoperatively*

Parameter	Possible Range	Laminectomy Group	
		Split (LSPSL)	Conventional
mean preop JOA score	0–29	16.4 ± 4.7	14.9 ± 4.0
subjective symptoms	0–9	3.5 ± 1.3	3.5 ± 1.3
low-back pain	0–3	1.7 ± 0.9	1.7 ± 0.8
lower-leg pain	0–3	0.9 ± 0.2	0.8 ± 0.4
gait ability	0–3	0.9 ± 0.9	1.0 ± 0.7
clinical symptoms	0–6	5.1 ± 1.0	5.0 ± 0.7
ADL restrictions	0–14	8.9 ± 2.7	7.5 ± 2.9
mean postop 1-yr JOA score	0–29	25.8 ± 3.4	25.4 ± 2.9
subjective symptoms	0–9	7.5 ± 1.4	7.3 ± 1.0
low-back pain	0–3	2.5 ± 0.5	2.3 ± 0.5
lower-leg pain	0–3	2.4 ± 0.8	2.3 ± 0.5
gait ability	0–3	2.7 ± 0.5	2.7 ± 0.5
clinical symptoms	0–6	5.7 ± 0.7	5.7 ± 0.6
ADL restrictions	0–14	12.8 ± 1.6	12.5 ± 1.9
recovery rate (%)†		75 ± 21	74 ± 17

* No significant intergroup difference according to the Mann-Whitney U-test for the JOA score or recovery rate. Mean values are presented ± the SD.
† Recovery rate: (postoperative points − preoperative points)/(29 − preoperative points).

The VAS Score for Wound Pain

On POD 3, the mean VAS score was 43 ± 27 mm in the LSPSL group and 44 ± 26 mm in the conventional laminectomy group. Although on POD 7 the mean score was reduced to 16 ± 17 mm in the LSPSL group and 34 ± 31 mm in the conventional group ($p = 0.002$), a significant decrease was recognized only in the former group. Additionally, on POD 7 a significant intergroup difference was recognized ($p = 0.04$) (Fig. 2).

Depth of Pain

On POD 3, the mean depth-of-pain score was 1.6 ± 0.7 in the LSPSL group and 1.6 ± 0.7 points in the conventional laminectomy group. On POD 7, although the score was significantly reduced to 0.9 ± 0.6 in the split-laminectomy group ($p = 0.021$), no change was recognized in the conventional group (depth-of-pain score 1.7 ± 0.8). On POD 7, a significant intergroup difference was recognized ($p = 0.013$) (Fig. 3).

Duration of Pain

On POD 3, the mean duration-of-pain score was 2.5 ±

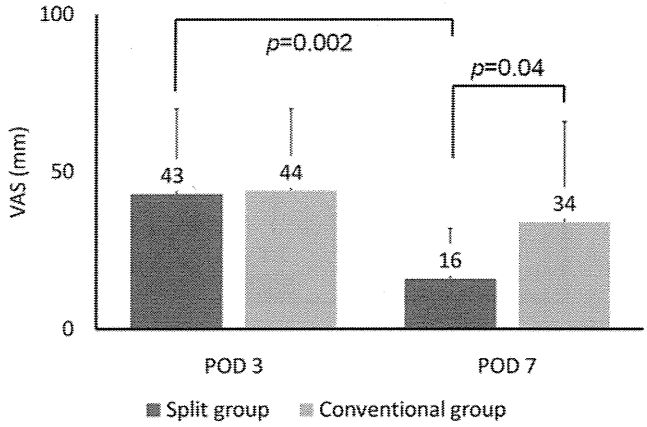

Fig. 2. Bar graph identifying VAS scores of acute postoperative wound pain on PODs 3 and 7. The mean VAS score of 43 ± 27 mm in the split-laminectomy group on POD 3 was significantly reduced to 16 ± 17 mm on POD 7 ($p = 0.002$). A significant difference was recognized between the 2 groups on POD 7 ($p = 0.04$).

1.4 in the LSPSL group and 2.9 ± 1.3 in the conventional group. On POD 7, the score was significantly reduced to 1.5 ± 1.5 in the split-laminectomy group ($p = 0.045$) and 2.5 ± 1.6 in the conventional laminectomy group. Although the mean scores were lower in the LSPSL group, no significant differences were recognized between the 2 groups (Fig. 4).

The JOA-ADL Scores

The preoperative mean JOA-ADL score was 8.8 ± 2.7 in the LSPSL group and 7.5 ± 2.7 in the conventional group. The scores decreased to 6.3 ± 3.0 in the former and 4.9 ± 4.5 in the latter group POD 3. The scores increased to 7.9 ± 2.6 and 7.8 ± 4.2, respectively, on POD 7. No significant intergroup differences were recognized (Fig. 5).

Serum CRP and CPK Levels

The mean preoperative serum CPK level was 99 ± 64 U/L in the split-laminectomy group and 115 ± 86 U/L in the conventional laminectomy group. The value increased to 126 ± 93 U/L on POD 3 and then decreased to 71 ± 50 U/L on POD 7 in the split-laminectomy group. In the conventional laminectomy group, the value increased to 207 ± 150 U/L on POD 3 and then decreased to 106 ± 86 U/L on POD 7. A significant difference was recognized between the 2 groups only on POD 3 ($p = 0.02$) (Fig. 6).

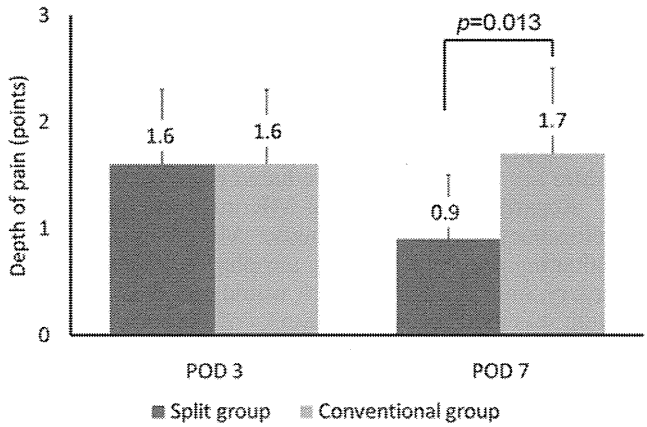

Fig. 3. Depth-of-pain scores reflecting acute postoperative wound pain on PODs 3 and 7. The mean score of 1.6 ± 0.7 in the split-laminectomy group on POD 3 was significantly reduced to 0.9 ± 0.6 ($p = 0.021$). A significant difference was recognized between the 2 groups on POD 7 ($p = 0.013$).

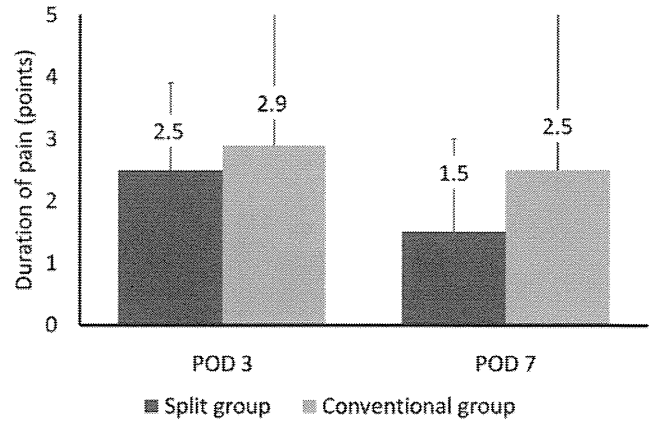

Fig. 4. Duration-of-pain scores on PODs 3 and 7. The mean score of 2.5 ± 1.4 in the split-laminectomy group on POD 3 was significantly reduced to 1.5 ± 1.5 ($p = 0.045$). No significant differences were recognized between the groups.

The mean preoperative serum CRP level was 0.3 ± 0.3 mg/dl in the LSPSL group and 0.2 ± 0.3 mg/dl in the conventional laminectomy group. In the LSPSL group, the value increased to 5.3 ± 3.7 mg/dl on POD 3 and then decreased to 1.1 ± 0.6 mg/dl on POD 7. In the conventional laminectomy group, the value increased to 5.5 ± 3.2 mg/dl on POD 3 and then decreased to 1.9 ± 1.5 mg/dl on POD 7. A significant intergroup difference was recognized between the 2 groups on POD 7 ($p = 0.04$) (Fig. 6).

Analgesics Taken During the 3-Day Postoperative Period

Although the mean number of analgesic medications taken during the 3-day postoperative period was smaller in the LSPSL group than the conventional group (1.7 ± 1.3 tablets vs 2.3 ± 2.4 tablets, $p = 0.22$), no significant difference was recognized.

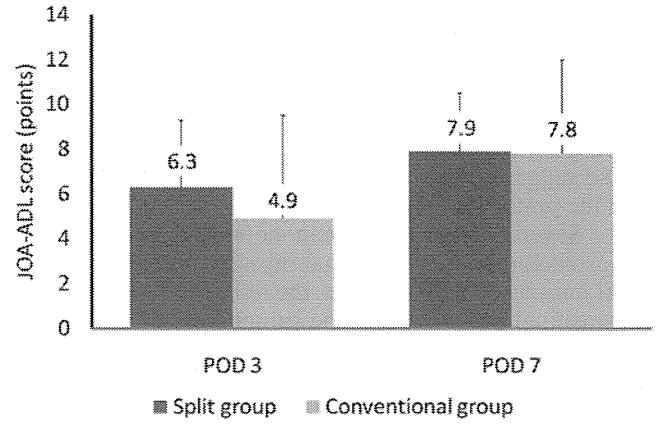

Fig. 5. Bar graph identifying JOA-ADL scores on PODs 3 and 7. The mean JOA-ADL score of 6.3 ± 3.0 in the split-laminectomy group and 4.9 ± 4.5 in the conventional laminectomy group on POD 3 increased to 7.9 ± 2.6 and 7.8 ± 4.2, respectively, on POD 7. No significant intergroup differences were recognized.

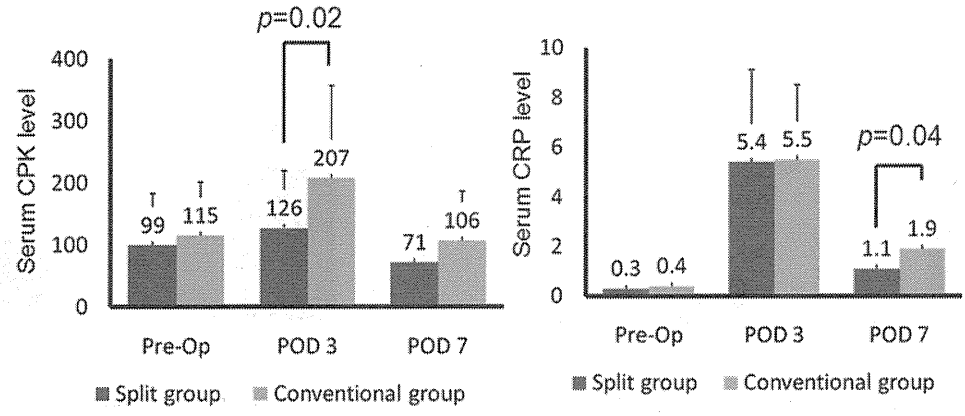

Fig. 6. Serum CPK and CRP levels before surgery (Pre-Op) and on PODs 3 and 7. The mean preoperative serum CPK level of 99 ± 64 U/L in the split-laminectomy group increased to 126 ± 93 U/L on POD 3, and then decreased to 71 ± 50 U/L on POD 7. A significant difference was recognized between the groups only on POD 3 ($p = 0.02$). The mean preoperative serum CRP level of 0.3 ± 0.3 mg/dl in the split-laminectomy group increased to 5.4 ± 3.7 mg/dl on POD 3, and then decreased to 1.1 ± 0.6 mg/dl on POD 7. A significant difference was recognized between the groups only on POD 7 ($p = 0.04$).

Atrophy Rate of Paraspinal Muscles at the 1-Month Postoperative Examination

The mean atrophy rate in the split-laminectomy group was significantly lower than that in the conventional group (24% ± 15% and 43% ± 22%, respectively; $p = 0.004$).

Discussion

The results of the present study indicated that, after posterior decompression surgery for LCS, acute postoperative wound pain was milder in the LSPSL (split-laminectomy) group than in the conventional laminectomy group. All mean scores regarding wound pain were lower in the split-laminectomy group. In particular, significant differences were observed between the groups in the VAS scores on POD 7 and the depth of pain on POD 7. The reduced postoperative wound pain in the LSPSL group may be due to the reduction of damage to the paraspinal muscles. Because serum CPK and CRP levels are the indicators of muscle damage,[8,11,12,23] the results (mean serum CPK levels on POD 3 and CRP level at POD 7 being significantly lower in the split-laminectomy group) indicated that the extent of the surgical invasiveness to the paraspinal muscles was lesser in the split-laminectomy group. Additionally, the muscle atrophy rate at 1 month being significantly lower in the split-laminectomy group also suggested that the procedure is less invasive in terms of the paraspinal muscles.

Several assumptions can be made concerning the reasons for the more minimal invasiveness to the paraspinal muscles in LSPSL. First, the reduced dissection of the paraspinal muscles from the spinous process—the result of splitting the spinous process—might also reduce the invasiveness of the procedure to the paraspinal muscles. Second, in conventional laminectomy, because the midline structures disturb one's access to the lateral recesses, paraspinal muscle dissection would extend to the medial side or sometimes to the lateral side of the facet joints. This would be associated with an increased chance of damaging the medial branch of posterior ramus. The posterior ramus emerges from the intervertebral foramen and passes dorsolateral to the superior articular process, and it splits into their terminal medial and lateral branches. The medial branch then enters the multifidus.[1–3,18] Because the medial branch is tethered to the periosteum, lateral to the facet joints, by fibers of the intertransverse ligaments,[1] the dissection of the muscles lateral to the facet joints may increase the risk of injuring the medial branch of the posterior ramus, which will finally result in denervation of the multifidus.[1] Furthermore, the medial branch of the posterior ramus is at risk of being injured, even by lateral retraction of the paraspinal muscles,[1,15] whereas in the split laminectomy a wide visualization of the central canal and the lateral recess is achieved by retracting the split spinous processes bilaterally, enabling minimal dissection of the paraspinal muscles and minimal detachment of the muscles from the facet joints. Kawaguchi et al.[5–7] reported that muscle injury is closely related to retraction pressure in their experimental and clinical trials. In a split laminectomy, the broad visualization of the central canal and the lateral recess may diminish the retraction pressure on paraspinal muscles. Additionally, interposition of the split spinous processes between the paraspinal muscles and the retractors may acts as a mechanical buffer to reduce the retraction pressure against the paraspinal muscles. All of these factors may explain why the paraspinal muscles in the split-laminectomy procedure are associated with reduced invasiveness.

Regarding the timing of MR imaging, based on our preliminary data of consecutive MR images taken for evaluating paraspinal muscle atrophy after decompressive surgery, muscle edema, which would cause the muscles to swell, was evident within 2 weeks of surgery. Sometimes, the muscle area after conventional laminectomy was larger than that of preoperative areas and that of the split laminectomy because of muscle edema. Thus, we decided that the measurement of muscle area as a parameter of invasiveness within 2 weeks would be inappropriate. Because the muscle edema would decrease to an almost normal level by 1 month postoperatively, we chose to obtain MR images 1 month after surgery to evaluate muscle atrophy.

Limitations of this study were as follows. The surgical technique was not identical among the patients because the surgeries were not performed by a single surgeon. Overall, 5 spine surgeons participated in this study. However, before starting the study, the procedures were standardized among the 5 surgeons to minimize the technical differences. Furthermore, the proportions of surgeries performed by each surgeon were almost identical between the 2 treatment groups. The other limitation was the withdrawal of the 7 patients (17%) from the study. The reasons for the withdrawal were the extension of decompression levels or the conversion of the procedure from decompression to fusion after randomization. However, we do not think that these withdrawals had a major impact on the results. Another limitation was that the measurements used in this study for evaluating the wound pain were not well validated except for the VAS. Because we believed that evaluation of the wound pain by only the VAS was insufficient, we added our nonvalidated original measurements to provide supplemental data, knowing that the measurements were less reliable. However, despite these limitations, we believe the results of the present study indicated that split laminectomy was less invasive, and reduced postoperative wound pain compared with conventional laminectomy.

Conclusions

The present prospective, randomized, controlled study confirmed that LSPSL for LCS reduces acute postoperative wound pain compared with conventional laminectomy, possibly because of minimized damage to the paraspinal muscles.

Disclosure

The authors report no conflict of interest concerning the materials or methods used in this study or the findings specified in this paper.

Author contributions to the study and manuscript preparation include the following. Conception and design: Matsumoto, Watanabe, Toyama, Chiba. Acquisition of data: Watanabe, Ikegami, Tsuji, Ishii, Ogawa, Takaishi, Nakamura. Analysis and interpretation of data: Watanabe, Nishiwaki. Drafting the article: Watanabe. Critically revising the article: Watanabe. Reviewed final version of the manuscript and approved it for submission: all authors. Statistical analysis: Ikegami, Nishiwaki. Study supervision: Matsumoto, Toyama, Chiba.

Acknowledgments

The authors thank the nurses who work in the 6-3 and 10N wards of Keio University Hospital for their assistance and cooperation during this study.

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Manuscript submitted November 19, 2009.
Accepted September 22, 2010.
Please include this information when citing this paper: published online December 10, 2010; DOI: 10.3171/2010.9.SPINE09933.
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Japanese Orthopaedic Association Back Pain Evaluation Questionnaire (JOABPEQ) : An Association Study in Patients with Lumbar Disc Herniation and Lumbar Spinal Canal Stenosis

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Key words : low back pain, lumbar disc herniation, lumbar spinal canal stenosis, questionnaire

Introduction

In 1997, the JOA revised the JOA score for low back pain and developed a new scientific outcome measure called JOABPEQ. The basic concept for developing JOABPEQ was to evaluate patients with low back pain from various specific perspectives, such as dysfunction, disability, handicap, and psychological problems. Furthermore, such measures should be patient-oriented, and its reliability and validity should be confirmed by statistical analysis. A paper has been published confirming the availability of JOABPEQ evaluating the short-term effects of surgical treatment for patients with lumbar disc herniation (LDH) or lumbar spinal stenosis (LSS)[5]. The aim of this study is to confirm the availability of JOABPEQ again using longer follow-up surveys.

Materials and Methods

1. Patients

Since April 2008, nineteen institutions were asked to recruit patients with LDH or LSS, who were admitted to their hospital for surgery, with the intention of collecting at least 200 patients with each disorder. Patients with typical LDH and LSS symptoms who underwent surgery at these institutions were included. Exclusion criteria were patients with previous surgery, other musculoskeletal disease requiring medical treatment, mobility aids or psychiatric disease that could potentially lead to inappropriate answers.

2. Applying the Questionnaire

The recruited patients were asked to complete the self-estimated questionnaire (JOABPEQ) and VAS scores (low back pain and leg pain) preoperatively, six and twelve months postoperatively. Additionally, patients were asked to judge the five self-rated

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Table 1 Comparison of the JOABPEQ scores in the LSS and LDH groups

		LBP	Lumbar ability	Walking ability	Social life ability	Mental health
LSS group	Pre-ope	43.0	58.0	21.0	36.5	48.5
	6 M	100**	83.0	79.0**	65.0	64.5
	12 M	100**	83.0	71.0**	65.0	66.0
LDH group	Pre-ope	29.0	50.0	29.0	32.0	46.5
	6 M	100**	100**	100**	78.0**	67.5
	12 M	100**	100**	100**	86.0**	59.0

Wilcoxon signed-ranks test **$p<0.01$

improvement grades regarding each of the five categories of JOABPEQ using a four-step rating scale (improved, slightly improved, no change and aggravated). The attending doctor was asked to record the JOA score, and the overall grades of improvement (improved, slightly improved, no change and aggravated).

3. Statistical Analysis

SPSS Statistics (Japanese version 17.0) was used to perform statistical analyse. Mann-Whitney's U test and t-test were used to evaluate the effect of surgical treatment demonstrated by JOABPEQ, JOA score and VAS. Spearman's rank correlation coefficient was calculated to determine the relationship between the total points of the JOA score and each factor of the JOABPEQ score, and the relationship between the changes of postoperative JOABPEQ scores and the grade of improvement assessed by patients or attending doctors. p values less than 0.05 were considered significant.

Results

In this study, three hundred and forty-three patients (LDH group : 75 patients, LSS group : 268 patients) were surveyed between April 2008 and February 2010. Mean patient ages in the LSS group and LDH group were 68.0 years and 46.0 years, respectively, showing a significant difference between the two groups.

1. JOA score

Mean JOA scores of the LSS group were 15.4, 22.8 and 23.0 points preoperatively, 6 and 12 months postoperatively, and mean JOA scores of the LDH group were 14.6, 25.3 and 25.8 preoperatively, 6 and 12 months postoperatively, respectively.

2. JOABPEQ

The median preoperative and postoperative JOABPEQ scores are shown in Table 1. In the LSS group good recovery was obtained only in two functional domains (low back pain and walking ability). In the LDH group, good recovery was obtained in four functional domains (low back pain, lumbar function, walking ability and social life function).

3. VAS

The average VAS scores for low back in LSS/LDH group was 5.7/4.7 preoperatively and 2.3/1.7 and 2.6/1.9 at 6 and 12 months postoperatively, respectively. The average VAS scores for leg pain was 6.3/6.4 preoperatively and 2.6/1.5 and 2.8/2.0 at each postoperative time point. Each score was apparently decreased after surgery in both groups.

4. The Correlation between JOA score and JOABPEQ

We assessed the correlation between JOA score and JOABPEQ preoperatively and postoperatively (Table 2). In the LSS group, there was a correlation between JOA score and each functional category of JOABPEQ both preoperatively and postoperatively. This suggest-

Table 2 The correlation between JOA score and JOABPEQ

		Preoperatively	After 6 months	After 12 months
LSS group	Low back pain	0.35**	0.42**	0.47**
	Lumbar function	0.45**	0.53**	0.56**
	Walking ability	0.34**	0.57**	0.74**
	Social life function	0.43**	0.57**	0.73**
	Mental health	0.31**	0.50**	0.61**
LDH group	Low back pain	0.42**	0.41**	0.24
	Lumbar function	0.45**	0.41**	0.54**
	Walking ability	0.46**	0.42**	0.42**
	Social life function	0.40**	0.42**	0.51**
	Mental health	0.33**	0.30*	0.24

*$p<0.05$, **$p<0.01$

Table 3 The correlation between JOABPEQ and self-assessments by doctors and patients

		Assessment by doctors		Assessment by patients	
		6 M ($n=210$)	12 M ($n=135$)	6 M ($n=235$)	12 M ($n=150$)
LSS group	Low back pain	0.27**	0.37**	0.33**	0.46**
	Lumbar function	0.20**	0.27**	0.24**	0.32**
	Walking ability	0.47**	0.50**	0.48**	0.61**
	Social life function	0.35**	0.45**	0.40**	0.48**
	Mental health	0.34**	0.33**	0.46**	0.40**
		6 M ($n=65$)	12 M ($n=36$)	6 M ($n=65$)	12 M ($n=36$)
LDH group	Low back pain	0.38**	0.26	0.36**	0.13
	Lumbar function	0.18	0.03	0.23	0.03
	Walking ability	0.26*	0.14	0.22	0.19
	Social life function	0.25*	0.38*	0.31*	0.45**
	Mental health	0.31**	0.39*	0.33**	0.26

*$p<0.05$, **$p<0.01$

ed that these measure would show a stronger correlation at a later postoperative time. In the LDH group, there was a correlation between the JOA score and every functional category of JOABPEQ except for the factor of low back pain and mental health 12 months preoperatively. And the tendency toward a stronger correlation at a later postoperative time was not recognized in this group.

5. Correlation between the Changes of Postoperative JOABPEQ scores and Assessments by Doctors and Patients

Table 3 shows the correlation between the postoperative changes in the points for each factor of JOABPEQ and changes in the grades assessed by attending doctors and patients themselves. In the LSS group, there was a significant correlation in every functional category. These findings suggested that JOABPEQ could properly reflect the assessments by attending doctors and patients themselves in the LSS group. In the LDH group, there were not as many correlations demonstrated between JOABPEQ and assessments by doctors or patients, and this showed a tendency toward fewer correlations 12 months postoperatively than there were 6 months postoperatively.

Discussion

The new outcome measure for low back pain, JOABPEQ published in April 2007, has gradually

become more widely used[6,7]. Its remarkable features are follows :

The First, JOABPEQ is a patient-oriented outcome measure that is preferable to exclude physician's bias, while the JOA score is a disease specific and physician-oriented outcome measure that mainly assesses the neurological status of the patient and enables surgeons to evaluate the results of surgical treatment. The Second, JOABPEQ includes 25 questions that are divided into five domains designated as : (1) low back pain, (2) lumbar function, (3) walking ability, (4) social life function and (5) mental health, making it a comprehensive outcome measure. The Third, JOABPEQ contains original equations that yield scores for the five factors. Then the equations to calculate the score for each domain are assembled in order to intuitively indicate the status of patients in five different functional domains.

The reliability and validity of JOABPEQ have been confirmed by statistical analysis[1~4]. Investigations have been performed in order to verify the sensitivity of the functional scores for treatment results in patients with low back pain, although another investigation is required to verify its sensitivity for each lumbar spinal disorder ; such as lumbar disc hernia or lumbar spinal canal stenosis.

This study was performed to confirm the validity of JOABPEQ for evaluating the effects of surgical treatment in patients with LDH or LSCS, and the first report has been published evaluating the short-term results[5]. We reported results analyzing 6 and 12 months postoperative data from 343 patients (LSCS : 268 patients, LDH : 75 patients).

These results indicated that JOABPEQ is highly sensitive for assessing treatment results in both LSS and LDH patients. Furthermore, it was shown that there was a difference in the assessment of postoperative results between the LSS and LDH groups.

In the LSS group, there was a significant correlation between each functional category on JOABPEQ and the assessments by attending doctors and patients themselves. In the LDH group, there were poor correlations between JOABPEQ and assessments by doctors or patients, and there was a tendency toward fewer correlations at 12 months than at 6 months after surgery, even though there was a significant correlation in each functional category at 3 months after surgery[5].

As for the poor correlations shown by LDH patients 12 months after surgery, it might be associated with the fact that more than half of the LDH patients acquired the full mark at both 6 and 12 months after surgery in three functional domains (low back pain, lumbar function and walking ability). The limitations of this study are the small number of the LDH patients and the short follow-up duration. Further studies with a 2-year postoperative follow-up are needed.

Acknowledgments

We thank Kuniyoshi Abumi, M. D. (Hokkaido University), Shinichi Konno, M. D. (Fukushima Medical University), Kensei Nagata M. D. (Kurume University), Osamu Shirado, M. D. (Saitama Medical University) and Kazuhisa Takahashi, M. D. (Chiba University) for providing cases for the database.

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J Orthop Sci (2011) 16:785–790
DOI 10.1007/s00776-011-0135-2

ORIGINAL ARTICLE

Matrix metalloproteinase 13 in the ligamentum flavum from lumbar spinal canal stenosis patients with and without diabetes mellitus

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Received: 2 March 2011 / Accepted: 12 July 2011 / Published online: 10 August 2011
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Abstract
Background Lumbar spinal canal stenosis (LSCS) is one of the most common spinal disorders in the elderly, and ligamentum flavum (LF) hypertrophy is an important cause of LSCS. Matrix metalloproteinase 13 (MMP13) can degrade fibrillar collagens and elastic microfibrils, and is involved in inflammation and fibrosis. The purpose of this study was to compare the expression of MMP13 in the LF from LSCS patients with diabetes mellitus [DM (+)] with that in the LF from patients without DM [DM (−)] and to analyze the relationship among DM, MMP13 expression, and LF hypertrophy.
Methods LFs from 11 DM (+) and 24 DM (−) LSCS patients were analyzed in this study. Histology analysis using hematoxylin and eosin and Masson's trichrome stain was performed for each LF. The expression of MMP13 was analyzed by quantitative real-time PCR. The thickness of LF was measured by CT.
Results In the LF from DM (+) LSCS patients, the elastic fibers were more disorganized and had lower volumes than in the LF from DM (−) LSCS patients, while more fibrotic tissue was observed in the LF from DM (+) than from DM (−) LSCS patients. MMP13 expression was significantly higher in the LF from DM (+) LSCS patients (0.46 ± 0.61 vs. 0.05 ± 0.09, $P = 0.002$). The LF from the DM (+) LSCS patients was significantly thicker than that from the DM (−) LSCS patients (5.0 ± 0.9 vs. 3.1 ± 0.8 mm, $P < 0.01$), and the thickness was correlated with the expression of MMP13 (correlation coefficient = 0.43, $P = 0.01$, Pearson's correlation test).
Conclusion DM-related MMP13 expression can be one of the factors contributing to fibrosis and hypertrophy of the LF. Further research on the mechanism of this process may lead to new therapies for LF hypertrophy.

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Introduction

Lumbar spinal canal stenosis (LSCS) is one of the most common spinal disorders in the elderly. The causes of LSCS include ligamentum flavum (LF) hypertrophy, hypertrophy of the facet joints, bulging of the intervertebral discs, and vertebral endplate osteophytosis. Among them, LF hypertrophy plays a dominant role in the narrowing of the lumbar spinal canal [1]. In the lower lumbar spine, the LF is composed of thick elastic fibers that are densely arranged with interspersed collagen fibers [2]. Histological changes in the hypertrophied LF from LSCS patients include fibrosis, degradation of elastic fibers with an increase in collagen fibers, granulation tissue proliferation, chondroid metaplasia, and calcification [3–6]. Fibrosis is considered to be the main cause of LF hypertrophy, and transforming growth factor (TGF)-β released by endothelial cells may stimulate the fibrosis, especially during the early phase of hypertrophy [3]. However, the pathomechanism of LF hypertrophy remains unclear.

The matrix metalloproteinases (MMPs) include over 20 zinc-dependent enzymes that degrade or modify

extracellular matrix molecules, such as elastin, collagen, and proteoglycans [7, 8]. Several recent studies have demonstrated a role of MMP3 [9] or MMP inhibitors in the pathology of LF [10].

Of the MMPS, MMP13 can degrade fibrillar collagens, including type I, II, and III collagens, into gelatin. It can also degrade the elastic microfibrils that are involved in extracellular matrix remodeling [11]. In addition, MMP13 plays a role in inflammatory and fibrotic processes [12–14]. MMP13 was reported to be more highly expressed in the LF of LSCS patients than in that of disc herniation patients, and is expressed in LF fibroblasts [15]. Moreover, high plasma glucose increases the expression of MMP13 in vessels [16] and the cornea [17].

We hypothesize that diabetes mellitus (DM) can cause an increased expression of MMP13 in the LF, which may lead to fibrosis and extracellular matrix remodeling and finally cause LF hypertrophy. The purpose of this study was to compare the expression of MMP13 in the LF of LSCS patients with DM [DM (+)] with that in the LF of patients without DM [DM (−)] and to analyze the relationship among DM, MMP13 expression, and LF hypertrophy.

Materials and methods

Thirty-five LF samples were obtained from 35 LSCS patients who underwent decompressive laminectomy for neurogenic claudication. The demographic data of the patients are shown in Table 1. All patients gave informed consent to participate in this study, and the study was approved by the Institutional Review Board of our institute. The mean age was 76.7 ± 7.6 years in the DM (+) group and 72.6 ± 7.4 years in the DM (−) group. There was no difference in age or gender between the two groups ($P > 0.05$). CT images of the lumbar spine of 1 mm thickness were made for all the patients before surgery.

Histologic analysis with hematoxylin-eosin staining and Masson's trichrome staining

During surgery, the LF was removed en-bloc, and the epidural fat was detached. Half of each sample was immediately stored in a −80°C freezer for subsequent quantitative real-time PCR analysis; the other half was fixed in 4% neutral formalin and decalcified in 20% ethylenediaminetetraacetic acid (EDTA) for 4 weeks, then embedded in paraffin for histologic analyses. Two consecutive sections (4 μm thick) were cut on a microtome and subjected to hematoxylin-eosin (H&E) and Masson's trichrome staining, respectively. H&E staining was used to analyze the degradation of the elastic fibers, and Masson's trichrome staining was used to determine the degree of fibrosis [18]. Masson's trichrome staining exhibited elastic fibers as pink and collagen fibers as blue in color.

Paraffin sections of yellow ligament were stained with rabbit anti-MMP13 antibody (Abcam, no. ab39012) followed by Alexa488-conjugated goat anti-rabbit antibody (molecular probe) and TOTO3 (molecular probe) as a nuclear stain, and then observed under a confocal microscope (FV1000, Olympus).

Table 1 Demographic data of the patients

	DM (+)	DM (−)
Age (years)	76.7 ± 7.6 (range 67–84)	72.6 ± 7.4 (range 60–88)
Gender	Female 5, male 6	Female 12, male 12
Level	L2/3: 1, L3/4: 2, L4/5: 7, L5/S1: 1	L3/4: 3, L4/5: 20, L5/S1: 1

Quantitative real-time PCR

The total RNAs were isolated from LF samples by TRIzol reagent (Invitrogen Corp.), and the concentration and quality were determined with an ND-1000 spectrophotometer (NanoDrop). The first-strand cDNAs were synthesized using an Advantage RT-for-PCR kit (Clontech Laboratories Inc.), then subjected to real-time PCR analysis using SYBR Premix ExTaq II (Takara Bio Inc.) according to the manufacturer's instructions. The MMP13 mRNA was normalized to the β-actin mRNA in each sample. PCR amplification was carried out on a Thermal Cycler Dice Real-Time System (Takara Bio Inc.), and gene expression was quantified using the delta-delta Ct method [19]. Nucleotide sequences of the primers were as follows: MMP13 forward: 5′-GCCAGAACTTCCCAACCAT-3′, MMP13 reverse: 5′-GGGCCCAGAATTTTCTCC-3′, β-actin forward: 5′-TGAGCGCGGCTACAGCTT-3′, and β-actin reverse: 5′-TCCTTAATGTCACGCACGATTT-3′.

Measurement of LF thickness

The LF thickness was measured using the preoperative axial CT images. The thickness of the LF on both sides was measured at its midpoints at the level of the decompressed intervertebral disc, following the method proposed by Fukuyama et al. [20], using image-analysis software (Real INTAGE, Tokyo, Japan) that allowed digital measurements with a precision of 0.1 mm.

The measurements were repeated five times by the first author (G.C.), and the results were averaged. To assess the intra- and inter-observer reliability of measurements, CT images of LF from 20 patients were chosen in a random fashion and were measured again by the first author

2 weeks after his first measurement and by the second author (K.W.). The intra- and inter-observer reliability were statistically tested using an intraclass correlation coefficient. The data presented in the results section are based on the measurements by the first author.

Real-time PCR for *mmp13*

To evaluate the influence of glucose on mmp13 expression in fibroblasts, which are the main component of the LF, NIH3T3 fibroblastic cells from mice were cultured in serum-free Dulbecco's Modified Eagle's Medium (Sigma-Aldrich) for 24 h. Then cells were treated with or without 3 mg/ml glucose for 19 h. Subsequently, total RNA was isolated from NIH3T3 cells using an RNeasy mini kit (Qiagen, Hilden, Germany). Single-stranded complementary DNAs (cDNAs) were synthesized with reverse transcriptase (Clontech Laboratories, Palo Alto, CA). Real-time PCR was performed using SYBR Premix ExTaq II (Takara Bio Inc., Otsu, Shiga, Japan) with a DICE Thermal Cycler (Takara Bio Inc.), according to the manufacturer's instructions. *β-Actin* expression served as an internal control. Primer sequences were as follows:

mmp13-forward:	5′-AACCTGGACAAGCAGTTCCAAAG-3′
mmp13-reverse:	5′-GAAATGGCTTTTGCCAGTGTAGG-3′
β-actin-forward:	5′-TGAGAGGGAAATCGTGCGTGAC-3′
β-actin-reverse:	5′-AAGAAGGAAGGCTGGAAAAGAG-3′

Statistical analysis

All values were reported as mean ± standard deviation. All data were analyzed with the SPSS system (version 13.0). Comparisons between the two groups were made by the Mann-Whitney *U* test. The relationship between MMP13 expression and LF thickness was analyzed by Pearson's test. $P < 0.05$ indicated statistical significance.

Results

Hematoxylin-eosin staining

In the LF from DM (+) LSCS patients, the elastic fibers were fragmented, disorganized and focally lost, accompanied by a proliferation of collagen fibers (Fig. 1a, b), while, in the LF from DM (−) LSCS patients, rich elastic fibers were arrayed in parallel order (Fig. 1c, d).

Masson's trichrome staining

In the Masson's trichrome staining of the LF from DM (+) patients, a large area was stained blue, indicating the presence of massive fibrosis (Fig. 1e, f), whereas in the LF from DM (−) patients, a large area was stained pink and showed a regular arrangement, indicating a normal, non-fibrotic condition (Fig. 1g, h).

The immunostaining of MMP13 using the rabbit anti-MMP13 antibody exhibited enhanced expression of MMP13 in the LF from DM (+) patients compared with DM (−) patients (Fig. 2).

MMP13 expression measured by quantitative real-time PCR

The ratio of MMP13 to β-actin mRNA was 0.46 ± 0.61 (range 0.0009–1.5801) for the DM (+) group and 0.05 ± 0.09 (range 0.000001–0.403321) for the DM (−) group (Fig. 3). The MMP13 mRNA expression was significantly higher in the LF from the DM (+) patients than in that from the DM (−) patients ($P = 0.002$).

Thickness of the LF

The mean thickness of the LF measured on CT images was 5.0 ± 0.9 mm (range 3.5–6.7 mm) in the DM (+) group and 3.1 ± 0.8 (range 1.9–4.7 mm) in the DM (−) group (Fig. 4). The LF from DM (+) patients was significantly thicker than that from DM (−) patients ($P < 0.01$). For intra-observer reliability of measurements, the intra-class correlation coefficient was 0.991 (95% confidence interval 0.984–0.996). For the inter-observer reliability, it was 0.970 (0.944–984). Thus, both intra- and inter-observer reliability were acceptably high.

Correlation between MMP13 expression and LF thickness

A positive correlation was observed between the MMP13 expression and thickness of the LF both in DM (+) LSCS patients (correlation coefficient = 0.646, $P = 0.032$, Pearson's correlation test) and in DM (−) LSCS patients (correlation coefficient = 0.542, $P = 0.006$, Pearson's correlation test). Thus, the correlation coefficient was higher in DM (+) patients than in DM (−) patients.

mmp13 expression in NIH3T3 cells

mmp13 expression in NIH3T3 cells analyzed by real-time PCR was 1.50 ± 0.06 times higher in the cells cultured with glucose than in those cultured without ($P < 0.001$) (Fig. 5).

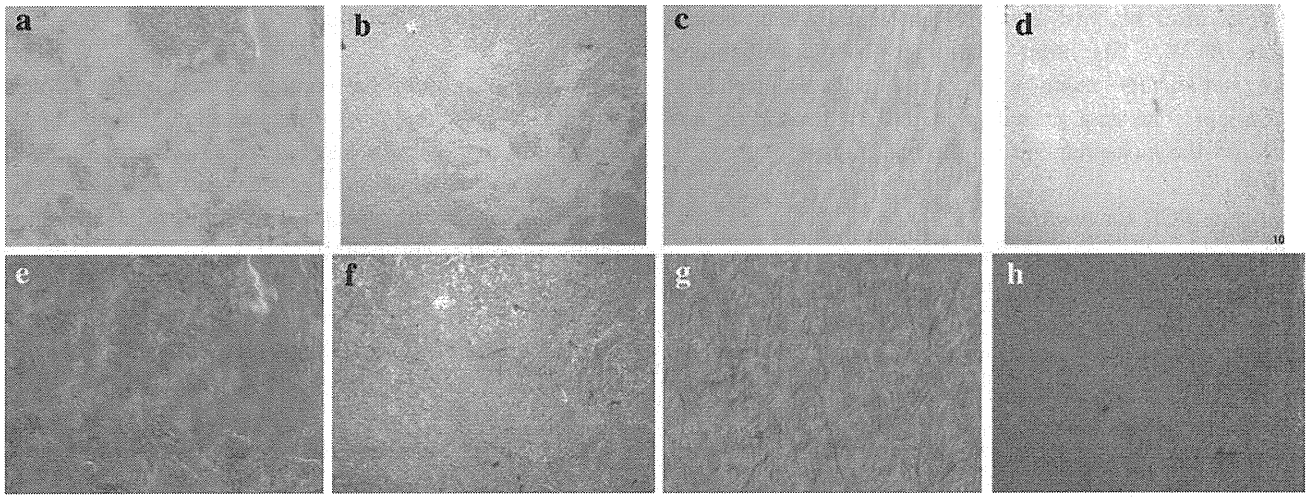

Fig. 1 **a** In the LF from DM (+) LSCS patients, elastic fibers were disorganized and focally lost, accompanied by a proliferation of collagen fibers. The elastic fibers also had low volumes and uneven diameters. H&E staining ×200. **b** LF from DM (+) LSCS patients. H&E staining ×40. **c** In the LF from DM (−) LSCS patients, a large area was stained pink with a regular arrangement, indicating a normal, non-fibrotic condition. H&E staining ×200. **d** LF from DM (−) LSCS patients. H&E staining ×40. **e** In the LF from DM (+) LSCS patients, a large area was stained blue, indicating the presence of massive fibrosis. Masson's trichrome staining ×200. **f** LF from DM (+) LSCS patients. Masson's trichrome staining ×40. **g** In the LF from DM (−) LSCS patients, rich elastic fibers were regularly arrayed, and the diameters of the elastic fibers varied only slightly. Masson's trichrome staining ×200. **h** LF from DM (−) LSCS patients. Masson's trichrome staining ×40

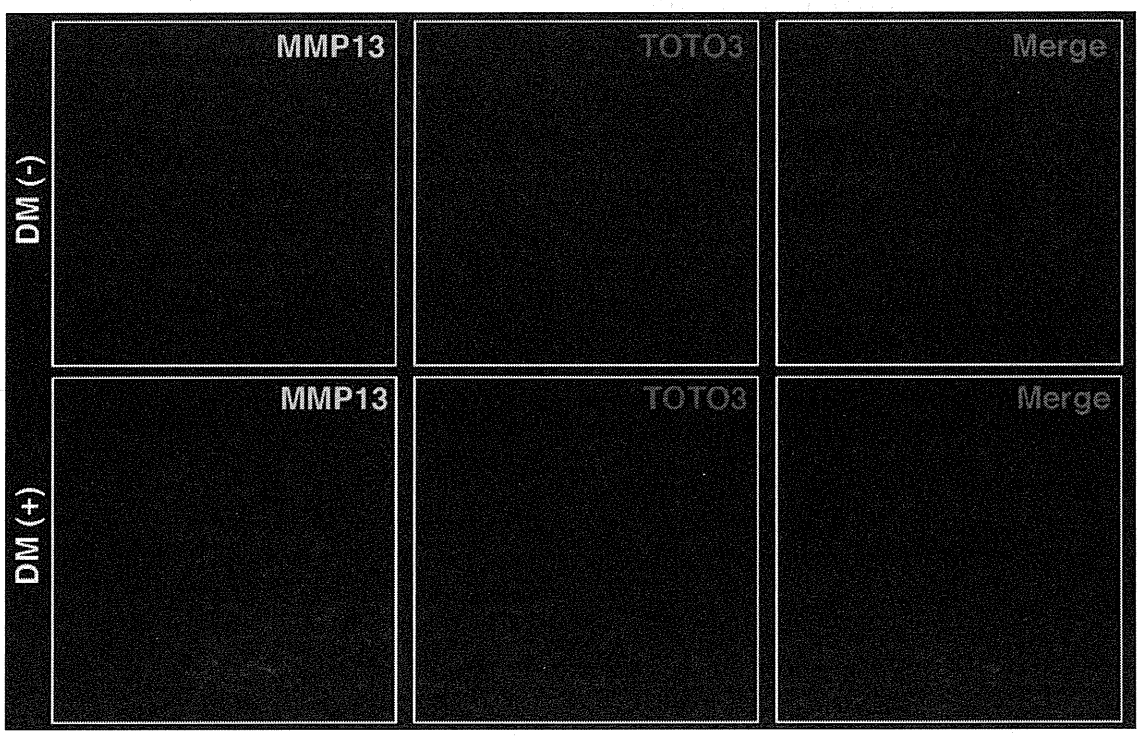

Fig. 2 Paraffin specimens of yellow ligament from individuals with DM [DM (+)] or non-DM [DM (−)] were stained with rabbit anti-MMP13 antibody followed by Alexa488-conjugated goat anti-rabbit antibody and observed under a confocal microscope. TOTO3 served as a nuclear stain

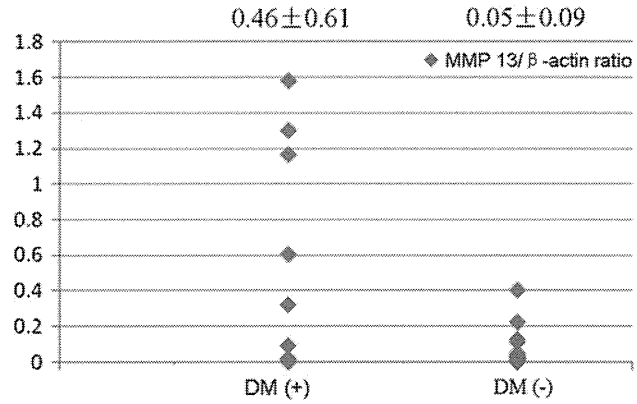

Fig. 3 Expression of MMP13 in the LF from DM (+) LSCS patients and DM (−) LSCS patients

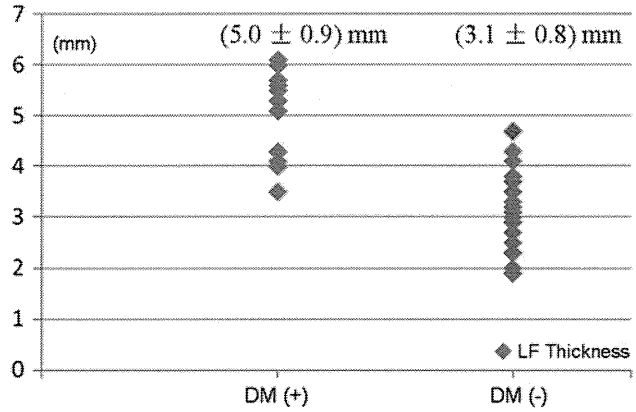

Fig. 4 Thickness of the LF from DM (+) and DM (−) LSCS patients

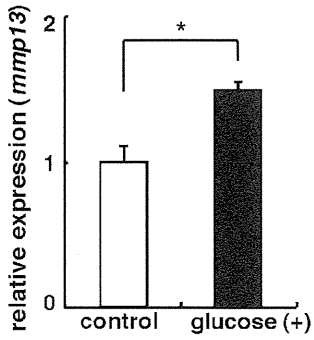

Fig. 5 NIH3T3 cells were cultured in the presence or absence of glucose (3 mg/ml) for 19 h, and *mmp13* expression was analyzed by real-time PCR. Data are shown as mean ± SD of *mmp13/β-actin* in cultured cells with glucose relative to that of without glucose. $*P < 0.001$

Discussion

In this study, we found an increased expression of MMP13 in the LF from DM (+) LSCS patients compared with the LF from DM (−) LSCS patients. In addition, elastin degradation and fibrosis of the LF were more severe in the DM (+) LSCS patients than in the DM (−) patients, and the LF of the DM (+) LSCS patients was significantly thicker. Our results suggest that an increased expression of MMP13, which may be related to DM, can be one of the factors contributing to fibrosis and hypertrophy of the LF, resulting in the progression of stenosis of the lumbar spinal canal.

Previous studies showed that the LF in the lumbar region is rich in elastic fibers, whose principal components are elastin and fibrillin [2]. Fibrillin can be degraded by MMP13, which is an important process in connective tissue remodeling [9]. MMP13 has been described as a trigger for the activation of a positive MMP2 and MMP9 feedback loop in asbestos-induced pulmonary fibrosis in mice [12]. The importance of MMP13 in fibrosis has also been shown in reports on bleomycin-induced pulmonary fibrosis in rats [21] and systemic sclerosis in humans [22]. In vessels and cornea, high plasma glucose increases the expression of MMP13 [14, 15]. In this study, the expression of MMP13 in the LF was higher in DM (+) than in DM (−) LSCS patients, and the expression of MMP13 was correlated with the thickness of the LF. Expression of mmp13 was upregulated by the presence of glucose in mice fibroblastic-like cells. MMP13 degrades both collagen fibers and elastic fibers, and is highly involved in extracellular remodeling [7, 9]. Thus, high plasma glucose levels may increase the expression of MMP13 in the LF and cause fibrosis. However, the exact mechanisms of the upregulation of MMP13 in DM patients should be elucidated by further studies.

In conclusion, we found a higher expression of MMP13 in the LF from DM (+) LSCS patients than in the LF from DM (−) LSCS patients. In addition, the elastin degradation and fibrosis of the LF was more severe in DM (+) patients than in DM (−) patients. These results suggest that the increased expression of MMP13 associated with DM can be one of the factors contributing to LF fibrosis and hypertrophy.

Conflict of interest This study was partly supported by a grant from Ono Pharmaceutical Co., Tokyo, Japan. All authors had no other financial relation regarding the study.

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