# **Neurohistology of lumbar spine ligaments**

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A study was conducted to identify neural elements in the posterior ligaments of the lumbar spine by using a modified gold-chloride method. Three morphologic types of mechanoreceptors were identified: Ruffini corpuscles, Ruffini end organs, and pacinian corpuscles. Free nerve endings, which are thought to be responsible for pain production, were also demonstrated within the ligaments.

The nerve supply of the human lumbar spine has been reviewed by Bogduk (1983), who concluded that the posterior ligamentous structures were innervated by both the dorsal rami and the sinuvertebral nerve. However, it is not clear whether the nerve fibers end within the substance of the ligaments. Stilwell (1956) failed to find any nerve endings in the interspinous and flaval ligaments of monkey, but Hirsch et al. (1963) identified fine free fibers and complex unencapsulated endings in the same structures in man. The ligamenta flava have been described as containing no (Jackson et al. 1966) or only a small number of sensory endings (Hirsch et al. 1963). These histologic studies were performed mainly by using silver-nitrate and methyleneblue staining methods. However, as suggested by Jackson et al. (1966), these techniques do not stain neural tissue exclusively.

A modified gold-chloride technique has been introduced recently to investigate the nerve endings within the knee ligaments (O'Connor and Gonzales 1979, Schultz et al. 1984, Zimny et al. 1985, 1986, Schutte et al. 1987). This technique has been found to be specific and suitable for the impregnation of myelinated nerve fibers and specialized nerve endings.

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## Patients and methods

Biopsy specimens of the supraspinous ligament, interspinous ligament, and ligamentum flavum were obtained from 10 adult patients (aged 30–45 years) who underwent surgery for disc herniation and from 4 young patients (aged 13–19 years) with idiopathic scoliosis prior to Harrington rod instrumentation.

Fresh or thawed frozen tissue samples were stained in bulk using a modification of the gold-chloride method (Zimny et al. 1985). The samples were placed in small vials and covered with a solution of three parts filtered, freshly squeezed lemon juice and one part 88 percent formic acid at 4 °C. The vials were then placed in darkness at room temperature for 10 minutes. The samples were removed from the first solution and patted dry with a gauze sponge, replaced in their respective vials, and barely covered with a 1 percent solution of gold chloride at 4 °C. The vials were again placed in darkness at room temperature for 20 minutes. The gold chloride was decanted off, the tissues again patted dry, and once more covered with a 25 percent formic acid solution at room temperature. The tissues were kept in the dark overnight for 15-16 hours. They were washed in distilled water and then placed in isopentane for 30 seconds. Frozen sections were then cut on a slicing microtome at 100 µm and stored in water. The sections were floated on alcoholic gelatin, mounted on slides, and allowed to dry. The slides were immersed in 100 percent ethanol for 2 minutes, passed through xylene twice for 3 minutes each, and coverslips were affixed The serial sections were studied with a light microscope (Zeiss, Photomicroscope III).











Figure 4

Figure 1. Light micrograph showing the afferent myelinated axon (A) of a nerve entering the ligamentum flavum (L5–S1, subject 37 years old) from the surrounding connective tissue sheath and terminating in a receptor (R). Note bifurcation of parent axon (A). Length marker 100  $\mu$ m.

Figure 2. Light micrograph showing a group of two encapsulated Ruffini corpuscles (R) in the supraspinous ligament (L4–L5, subject 19 years old) close to a blood vessel (BV). Note the afferent axon (a) of one of the corpuscles. Length marker 25  $\mu$ m.

Figure 3. Light micrograph showing a Ruffini end organ (R) resembling a Golgi tendon organ within the interspinous ligament (T11-T12, subject 13 years old). Length marker 250 μm.

Figure 4. Light micrograph showing two fusiform mechanore-ceptors (P) of the Pacini type within the supraspinous ligament (L2–L3, subject 13 years old). Note the axis cylinder, the surrounding finely granular layer, and the layers of cells ensheathing the whole receptors. Length marker 10  $\mu$ m.



Figure 5. Light micrograph showing a free nerve ending (N) near a blood vessel (BV) within the interspinous ligament (T11–T12, subject 13 years old). Length marker 100 µm.

# Results

The posterior ligaments were found to have an extensive neural network. Neural fibers enter the ligaments by means of an axon from connective tissue and terminate in various receptors (Figure 1). At least four different types of nerve endings were observed in these ligaments. Consistent with earlier histologic studies, three of them could be readily characterized as Ruffini end organs, Ruffini corpuscles, and pacinian corpuscles (Boyd 1954, Stilwell 1956, 1957, Hromada and Polacek 1958, Jackson et al. 1966, O 'Connor and Gonzales 1979, Schultz et al. 1984, Zimny et al. 1986). The remaining type of endings consisted of very fine free fibers.

Ruffini corpuscles were found in all the three posterior ligaments investigated. They were composed of two to five encapsulated corpuscles, each consisting of an afferent axon and a globular capsule (Figure 2). The corpuscles are about  $80 \,\mu\text{m}$  in maximum diameter and  $35 \,\mu\text{m}$  in minimum diameter. They occurred in the vicinity of blood vessels and were arranged tridimensionally within the ligament substance. This type of mechanoreceptor was present at or near the periphery of all the regions of the posterior ligaments.

Ruffini end organs resembling the Golgi tendon organs were observed mainly in the supraspinous and interspinous ligaments. They were larger than the Ruffini corpuscles and displayed individual variations in morphology (Figure 3). Their corpuscles were curved or coiled in a spiral around a blood vessel. They were oriented parallel to the direction of the collagen fascicles of the ligament.

The third type of nerve ending in the posterior ligaments was represented by simple encapsulated corpuscles with an inner core (pacinian corpuscles). They were found mainly in the dorsal part of the supraspinous ligament near blood vessels. Pacinian corpuscles were much less numerous in the ligamentum flavum and in the interspinous ligament. The corpuscles lie singly or in groups of up to five (Figure 4). The encapsulated corpuscles with an inner bulb were fusiform, being about 20–40  $\mu$ m wide and 100–300  $\mu$ m long. As with the Ruffini end organs, a capillary blood vessel was related to each pacinian corpuscle.

The last variety of nerve ending found in the spinal ligaments consisted of free nerve endings (Figure 5). Their diameter was often less than 1  $\mu$ m. They branched out between the collagen and elastin fibers of the ligament from parent axons whose diameter was  $1-2 \mu$ m. In the interspinous and supraspinous ligaments, the free nerve endings were located near the attachment to the spinous processes. In the ligamenta flava, they were found in the outermost layer of the dorsal surface, but never in the deeper substance.

# Discussion

Our observations are similar to those reported in the knee ligaments (Zimny et al. 1986), and they disagree with the conclusions of Jackson et al. (1986) that there are no nerve endings in the supraspinous and interspinous ligaments. It is known that the ligamentum flavum and the interspinous ligaments are innervated by the medial branches of the dorsal rami and that the recurrent sinuvertebral nerve sends a direct dorsal branch to the ligamentum flavum and the anterior facet joint (Bogduk 1983). Then, there are at least two potential sources innervating the posterior ligamentous system, that is, the lumbar dorsal rami and the sinuvertebral nerve. Stilwell (1956, 1957) found the Ruffini receptors only at the connection between the spinalis dorsi and the interspinous ligament and the pacinian corpuscles only in the lumbodorsal fascia. We have now found them also in the supraspinous ligaments and the ligamenta flava.

The function of these mechanoreceptors has been described by several authors (Boyd 1954, Schultz et al. 1984, Zimny et al. 1986, Grigg et al. 1982). They monitor proprioceptive information and signal potentially injurious deformations of the ligaments and joints (Schultz et al. 1984). In the posterior ligaments of the lumbar spine, they could be involved in the stress-monitoring system postulated by Gracovetsky and Farfan (1981).

Ruffini corpuscles that are slowly adapting mechanoreceptors (Freeman and Wyke 1967, Zimny et al. 1986) could be involved in the monitoring of compressive stresses during hyperextension of the spine. These receptors could act to prevent contact between spinous processes, because they are sensitive to compression in a plane perpendicular to the plane of the corpuscle (Grigg et al. 1982).

Ruffini end organs are also slow-adapting mechanoreceptors, but uniquely sensitive to in-plane axial stresses (Boyd 1954). They could monitor the axial tension developed in the interspinous and supraspinous ligaments during flexion. They may be responsible for the control of the reflex inhibitory mechanism suggested by Floyd and Silver (1955) to explain the relaxation of spinal muscles occurring during trunk flexion when the tension in the posterior ligaments is sufficient to sustain gravitational moment.

Pacinian corpuscles are known to give rise to rapidly adapting discharges (Freeman and Wyke 1967). They have a very low threshold and work as dynamic receptors at the beginning and end of a movement (Halata 1977). Their presence in the supraspinous ligaments confirms the existence of a system with a fast response time to monitor and control the stress (Gracovetsky and Farfan 1986). They could assess a sudden increased stress during the lifting of a weight and fire the erectores spinae muscles to prevent the rupture of the posterior ligamentous structures.

The free nerve terminals that we have observed conform with the generally accepted morphology of pain receptors (Freeman and Wyke 1967). Most of them ran at some distance from the blood vessels. It is believed that they constitute the pain-receptor system of the posterior ligamentous structures. In the supraspinous and interspinous ligaments, the free nerve terminals are found near the attachment of the spinous processes, whereas in the ligamenta flava they are located in the dorsal surface (Yahia et al. 1987). Using a specific immunohistochemical method, Korkala et al. (1985) failed to find any nociceptive nerve terminals in the ligamentum flavum. This may be due to regional variations, because these authors used only small samples of the tissue. We found free nerve endings only in the posterior layer of the ligamentum flavum. Recently, Giles and Taylor (1987) demonstrated similar free nerve endings in the synovial folds of the lumbar zygapophyseal joint. Thus, their demonstration in the interspinous and supraspinous ligaments has potential clinical significance in relation to low-back pain; the majority of interspinous ligament ruptures are located in the two lowest spaces of the lumbar spine (Rissanen 1960).

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