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Regional differences within the human supraspinous and interspinous ligaments: a sheet plastination study

Received: 1 May 2001
Revised: 16 October 2001
Accepted: 1 December 2001
Published online: 2 February 2002
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Abstract The extent to which neighboring muscles and the fascia contribute to the formation of the supraspinous and interspinous ligaments is not clear from the literature. The purpose of this investigation is to examine the midline attachments of tendons and the posterior layer of thoracolumbar fascia in order to determine their respective contributions to the formation of these ligaments throughout the thoracolumbar spine. Study of the dense connective tissue organization in the posterior ligamentous system was carried out on two cadavers serially sectioned into thin (2.5-mm) epoxy resin plastinated slices. Additional observations were taken from a gross anatomical study of the midline anatomy in two adult cadavers. The results show that the spinal attachments of trapezius, rhomboideus major and splenius cervicis combine with the deep fascia to form the supraspinous ligament in

the upper thoracic spine. The posterior layer of the thoracolumbar fascia makes a major contribution to the supraspinous and interspinous ligaments in the lower thoracic spine. In addition to the posterior layer of thoracolumbar fascia, longissimus thoracis and multifidus combine to form the lumbar supraspinous and interspinous ligaments. Their spinal attachments produce a system of dense connective tissue with marked regional variation in fiber orientation and arrangement. The findings support the description of the supraspinous and interspinous ligaments as structures formed by both muscle tendons and aponeuroses along the length of the thoracic and lumbar spine, with regional differences in their connective tissue architecture.

Keywords Connective tissue · Fascia · Ligaments · Muscle attachments · Thoracolumbar spine

Introduction

The supraspinous ligament forms part of the posterior ligamentous system of the vertebral column. This extends from the C7 spinous process throughout the length of the thoracic spine to the level of L4 in 73% of adults [21]. Anteriorly, the supraspinous ligament merges with the interspinous ligaments – the sheets of fibrous tissue found between the spinous processes of adjacent vertebrae [2]. At the gross anatomical level, the true structural configuration of the supraspinous and interspinous ligaments in the lum-

bar spine is confounded by the manner in which they blend with neighboring musculature and fascial attachments [4, 5, 6, 7, 11, 13, 19, 21, 23]. For example, three parts of the lumbar supraspinous ligament are distinguished [21], but only the middle portion, formed by multifidus, longissimus thoracis and the thoracolumbar fascia, is well described [21]. Other investigators report that the supraspinous ligament is located superficial to [23], or between the spinal attachments of [11], the thoracolumbar fascia. Conflicting descriptions of the anatomical divisions of the interspinous ligaments [2, 19, 20, 21, 22] further attest to the difficulty in determining their structural formation accurately.

In our recent study on the ligamentum nuchae, the equivalent structure to the supraspinous ligament in the cervical spine, distinct regional differences were found in its connective tissue architecture [14]. Likewise, there are indications that anatomical differences exist within the supraspinous and interspinous ligaments of the thoracic and lumbar spine. Within the lumbar spine, the supraspinous ligament is most easily distinguishable at the L2-L3 level, and gradually diminishes below this level [21]. Changes in the interspinous ligament have been noted at the L5-S1 level due to the change in contribution from the aponeurosis of longissimus thoracis in the lower lumbar spine [21]. Compared with the lumbar spine, where the majority of studies have been carried out, the supraspinous and interspinous ligaments in the thoracic region are thin membranous structures, and only at the thoracolumbar junction do they become better defined [18]. The three-fold increase in the cross-sectional area of the supraspinous and interspinous ligaments (9 mm² at the T1/T2 level through to 30 mm² at the T8/T9 level) indicates that regional differences also exist between different levels within the thoracic spine [7]. Incremental increases in tensile strength along the thoracic supraspinous ligaments further support this view [16].

A more detailed analysis at the ultrastructural level is needed to better define the nature of regional differences within the supraspinous/interspinous complex and, in particular, the extent of the contribution from the neighboring muscles. Technically, this is problematic due to the close aggregation of muscle tendons and aponeuroses at the midline [6]. In a recent study of the ligamentum nuchae [14], thin (2.5-mm) epoxy resin slices obtained using the E12 sheet plastination technique [1, 24] allowed examination of its connective tissue structure at the macroscopic/microscopic levels [10]. One advantage these slices have over standard histological sections is their much increased size. This allows the *in situ* identification of structures attaching to the midline [15]. Accordingly, the same approach is used to examine features of the connective tissue architecture within the supraspinous/interspinous ligament complex in the thoracic and lumbosacral regions. The purpose of this investigation is to examine the midline attachments of tendons and the posterior layer of thoracolumbar fascia in order to determine their respective contributions to the formation of these ligaments throughout the thoracolumbar spine.

Materials and methods

Four human cadavers fixed in 10% formalin were used in this study. A detailed analysis of the connective tissue arrangement was carried out on serial thin (2.5-mm) slices from two cadavers processed according to the E12 sheet plastination technique [1, 24]. In brief, the cadavers were frozen at -85°C before slicing, dehydrating and degreasing. The body slices were subsequently impregnated with E12 epoxy resin. One cadaver (a female, aged 65 years) was sectioned in the horizontal plane. The serial slices

extending from T1 to the tip of the coccyx were selected for the study of the tissue. With the second cadaver (a female, aged 54 years), three sagittal slices were taken through the middle portion of the vertebral column. The sagittal slice extending from T11 to the tip of the coccyx was used in this study.

Detail of the connective tissue was examined by placing each slice over a radiographic light-box, making it possible to trace the origin, fiber orientation and destination of the dense connective tissue attaching to the midline. The connective tissue organization in the horizontal slices was also examined systematically under a stereomicroscope (Wild MZ8; Leica, Heerburgg, Switzerland) at magnifications ranging from $\times 0.63$ to $\times 3$.

Gross anatomical dissection was conducted on the posterior midline region of the spine in two embalmed cadavers (males, aged 76 and 84 years) to acquire an overall impression of the representative midline anatomy and confirm our findings in the plastinated slices. Isolated bundles of dense connective tissue at the midline were traced under an operative microscope with fine forceps to establish their origin and attachments relative to each other.

In this study, the supraspinous and interspinous ligaments are defined as the discrete bands of dense connective tissue fibers connecting the apices of the spinous processes, and adjoining spinous processes, respectively. The interspinous ligaments were considered absent where the tissue lacked ventral attachments. The results were grouped with reference to the observations made on the gross dissections and/or the plastinated slices (horizontal or sagittal) and also into upper thoracic (T1-T5), lower thoracic (T6-T12), lumbar (L1-L5) and sacral regions (S1-S5).

Results

Upper thoracic spine (T1-T5)

The dense connective tissue attaching to the spinous processes in the upper thoracic spine originates mainly from muscle tendons. At the gross anatomical level, the tendons of trapezius and splenius cervicis blend together at the midline, creating an impression of a fine ligament running longitudinally over the spinous process tips (Fig. 1A). Detail of the dense connective tissue fiber arrangement is confirmed in the plastinated slices. Obliquely orientated, dense connective tissue fibers arising from the middle portion of trapezius meet in the midline, decussating prior to attaching to the tips of the T1-T4 thoracic spinous processes, and joined by the tendons of rhomboideus major and splenius cervicis (Fig. 1B). Deep fascia overlying and connecting with splenius cervicis, longissimus thoracis and iliocostalis also attach to the spinous processes (Fig. 1B). The interspinous ligaments are absent throughout the upper thoracic spine. Instead, the interspinous compartment is occupied by a thin layer of loose connective tissue located between the bilateral multifidus muscles (Fig. 1C).

Lower thoracic spine (T6-T12)

A marked transition in the midline connective tissue organization coincides with the spinal attachments of the posterior layer of the thoracolumbar fascia becoming evident

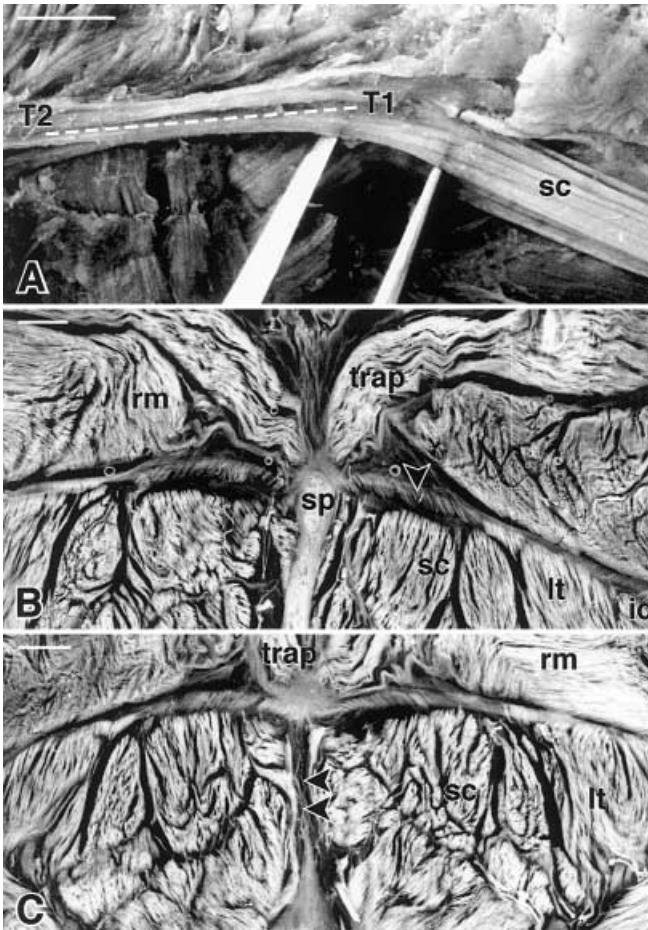


Fig. 1 **A** Tendinous fibers from splenius cervicis (*sc*) between T1 and T2 (*dotted line*), forming part of the supraspinous ligament in the upper thoracic spine. **B** Horizontal plastinated slice (T2 level) showing the supraspinous ligament formation by decussating tendons of trapezius (*trap*), rhomboideus major (*rm*) and adjoining deep fascia (*single arrowhead*). The deep fascia overlies splenius cervicis (*sc*), longissimus thoracis (*lt*) and iliocostalis (*ic*). **C** Horizontal plastinated slice (T2–T3 level). View of the fine membranous tissue (*double arrowheads*) occupying the compartment between neighboring muscles

at the T6 level. At the gross anatomical level, the fibers of the thoracolumbar fascia pass in an oblique direction towards the midline, deep to the muscular attachments of trapezius (Fig. 2A). In the horizontal plastinated slices, the decussating fibers of trapezius can be identified. They are joined by fibers from the posterior layer of the thoracolumbar fascia, which attach directly to the lateral aspect of the T6–T9 spinous processes as a single layer of connective tissue. At the T9 level, the lower portion of trapezius gives rise to a tendinous aponeurosis, which spans the lower thoracic and upper lumbar spinous processes. The T9 level also marks the commencement of fiber decussation of the thoracolumbar fascia in the horizontal plastinated slices. Microscopically, these fibers are seen to form small fibrous compartments around individual fibers run-

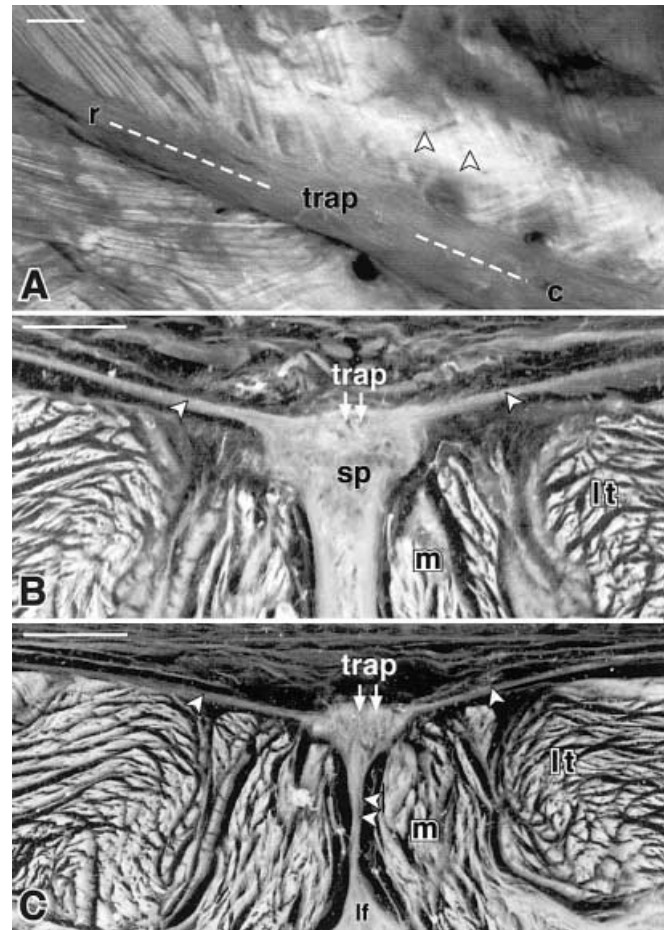


Fig. 2 **A** The supraspinous ligament (*dotted line*) formed by the tendon of trapezius (*trap*) and the posterior layer of the thoracolumbar fascia (*single arrowheads*) running in a rostral (*r*) to caudal (*c*) direction along the midline of the lower thoracic spine. **B** Horizontal plastinated slice (T10 level). Individual fibers belonging to trapezius (*trap* – *double arrows*) between the decussating fibers of the posterior layer of the thoracolumbar fascia. Contribution to the supraspinous ligament at the T10 spinous process (*sp*) level from multifidus (*m*) and longissimus thoracis (*lt*). **C** Horizontal plastinated slice (T10–T11 level). Formation of the interspinous ligament (*double arrowheads*) by the anterior reflection of the posterior layer of the thoracolumbar fascia and its continuity with the ligamentum flavum (*lf*). (*Bar scales* = 4 mm)

ning longitudinally within the lower tendinous portion of trapezius (Fig. 2B).

The interspinous ligament commences at the T6 level as an anterior extension of the thoracolumbar fascia forming a single sheet of dense connective tissue bridging between borders of the spinous processes of adjacent vertebrae (Fig. 2C). The posterior layer of the thoracolumbar fascia becomes progressively thicker below the T10 level and, correspondingly, the interspinous tissue becomes better defined and bilaminar in form.

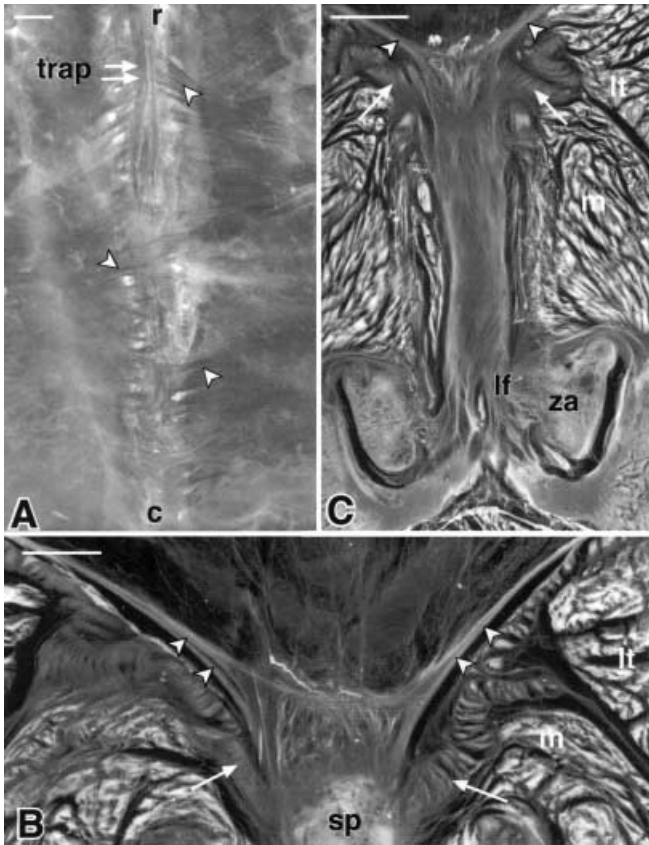


Fig. 3 **A** Formation of the lumbar supraspinous ligament by trapezius (*trap* – double arrows) and the posterior layer of thoracolumbar fascia (*single arrowheads*) orientated in the rostral (*r*) to caudal (*c*) direction. **B** Horizontal plastinated slice (L3 level). Formation of the supraspinous ligament by the posterior layer of the thoracolumbar fascia. Longissimus thoracis (*lt*) and multifidus (*m*) attach to the spinous process (*sp*) laterally. **C** Horizontal plastinated slice (L1–L2 level). The contribution of the posterior layer of the thoracolumbar fascia, multifidus and longissimus thoracis to the interspinous ligament. The interspinous ligament merges with the ligamentum flavum (*lf*) and capsule of the zygoapophyseal joints (*za*). (Bar scales = 4 mm)

Lumbar spine (L1–L5)

The midline attachments of the posterior layer of the thoracolumbar fascia, longissimus thoracis and multifidus form the principal connective tissue component of the supraspinous ligament in the lumbar spine. At the gross anatomical level in the upper lumbar spine, the tendinous aponeurosis from trapezius intertwines with the midline attachments of the thoracolumbar fascia (Fig. 3A). In the horizontal plastinated slices, distinctive bands of dense connective tissue fibers from the thoracolumbar fascia cross the midline to merge with its contralateral fibers to contribute to the structure of both the supraspinous (Fig. 3B, Fig. 4A) and interspinous ligaments (Fig. 3C). Fibers originating from the posterior layer of the thoracolumbar fascia split and pass anteriorly obliquely or

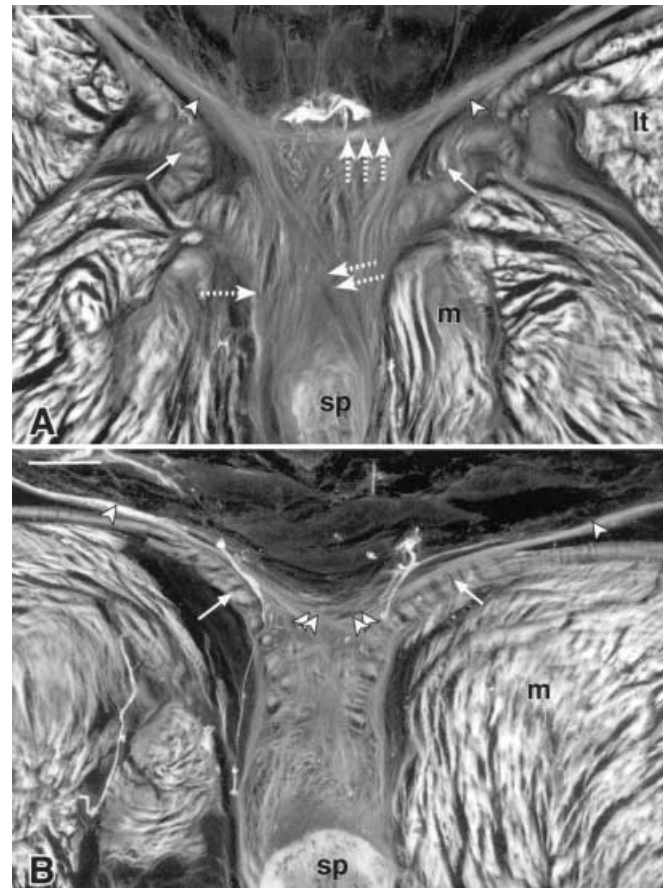


Fig. 4 **A** Horizontal plastinated slice (L3 level). Connective tissue fiber orientation within the supraspinous ligament and attachments to the spinous process (*sp*). Anteriorly (*single dashed arrow*), obliquely (*double dashed arrows*) and horizontally (*triple dashed arrows*) directed fibers are evident. The posterior layer of the thoracolumbar fascia (*single arrowheads*) and shared attachment (*single arrow*) from longissimus thoracis (*lt*) and multifidus (*m*). **B** Horizontal plastinated slice (L5 level). Decussation of connective tissue fibers (*double arrowheads*) from the posterior layer of thoracolumbar fascia is visible superficial to the erector spinae aponeuroses. Multifidus merges with the interspinous ligament to attach onto the spinous process. (Bar scales = 4 mm)

transversely before attaching to the spinous processes (Fig. 3B). In the mid and lower lumbar spine, the attachments of longissimus thoracis and multifidus also contribute to the supraspinous ligament formation (Fig. 4A). At L5, the dense connective tissue becomes further modified to create a horizontal T-bar formation as the posterior layer of the thoracolumbar fascia joins with the common erector spinae aponeurosis to attach onto the L5 spinous process (Fig. 4B).

In the horizontal plastinated slices, the interspinous ligament merges anteriorly with the posterior capsule of the zygoapophyseal joints (Fig. 3C). The combined contribution of the posterior layer of the thoracolumbar fascia, the aponeuroses of longissimus thoracis and tendons of multi-

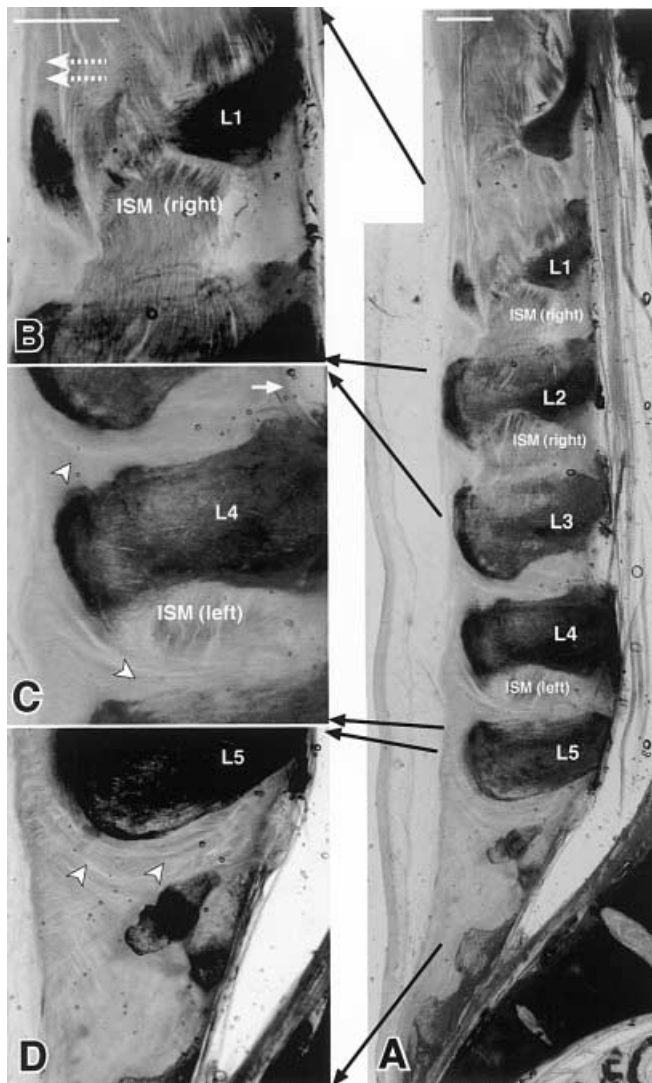


Fig. 5 A Sagittal plastinated slice through the lumbosacral spine region. The interspiniales muscles: on the right – *ISM (right)* – attaching between L1 and L2, and L2 and L3, and on the left – *ISM (left)* – visible between L4 and L5. A complete interspinous ligament is located between L3 and L4. **B** Expanded view: L1 level. Longitudinal fibers (*double dashed arrows*) of the supraspinous ligament terminating at L1 spinous process. **C** Expanded view: L4 level. Dense connective tissue fibers (*single arrowheads*) curve over the spinous processes and merge with the ligamentum flavum (*single arrow*) (above L4), or attach onto the upper border of the spinous process below (below L4). **D** Expanded view: L5-S1 level. Decussating connective tissue fibers (*single arrowheads*) sweep in an anterior direction between the L5/S1 junction. (Bar scales = 4 mm)

fidus to the interspinous tissue is further confirmed in the sagittal plane plastinated slice (Fig. 5A–D). Dense connective tissue fibers pass caudally over the tips of each of the lumbar spinous processes (L1–L5) and attach to the superior border of the bony lamina below forming the posterior part of the interspinous ligament (Fig. 5D). The

interspinous tissue merges anteriorly with the ligamentum flavum (Fig. 5C). In the lower lumbar spine, evidence of fiber decussation is seen in the sagittal plane (Fig. 5D).

Sacrum (S1–S5)

The tendinous origins of multifidus and the erector spinae aponeurosis contribute to the midline dense connective tissue arrangement at this level. Caudal to S3, there is no contribution from the surrounding musculature and the fascia gradually diminishes at the level of the coccyx.

Discussion

The key contribution of this study is the description of the in situ connective tissue structure of the posterior ligamentous system throughout the entire length of the thoracolumbar spine. Undoubtedly, a limitation of the study is that key results were taken from the horizontal E12 epoxy resin plastinated slices using a single cadaver. Adding more specimens would not be a practical option, because the costs would have been prohibitive. However, the data obtained from these slices were supported by further information obtained from anatomical dissections on two cadavers and the single sagittal slice from another cadaver. This latter slice was also processed with E12 epoxy resin. While it can be argued that the findings in the study represent an anatomical variation, and are therefore not a legitimate description, they are consistent with results of previous gross anatomical studies in the lumbar spine [11, 13, 19, 21, 22]. For example, connections between the interspinous ligament and the zygoapophyseal joint (Fig. 3C) have been previously reported [2, 20]. In order to understand the role of the posterior ligamentous system, the detail of configuration, shape, attachments and their topographical relationships to contiguous spinal structures is needed [13]. The results obtained from the plastinated slices in this study begin to fill this gap.

However, additional factors leading to variability in the structural arrangement within the supraspinous and interspinous ligaments between individuals, such as age and sex, cannot be discounted. For example, it is conceivable that alterations in their fiber orientation may accompany the increases in vertebral body height that are seen in the growth period. Fatty infiltration and fibrocartilaginous metaplasia of the supraspinous ligament have also been reported to occur with age [21]. Likewise, variation in the thickness in the spinal attachments of the posterior layer of the thoracolumbar fascia in the thoracic region of the spine [3] may be associated with sex, or possibly occupational influences.

The results in this study describe the changes in connective tissue structure of the supraspinous and interspinous ligaments at different levels along the length of

the thoracic and lumbosacral spine. In the upper thoracic spine, the supraspinous ligament is a formation of tendons and adjoining fascia (Fig. 1A,B), with no interspinous ligament to be found (Fig. 1C). It is only in the lower thoracic and lumbar spine where the posterior layer of thoracolumbar fascia joins with the other tendinous insertions that the interspinous ligaments become recognizable anatomical entities (Fig. 2C, Fig. 3C). The plastinated slices confirm that the lumbar supraspinous ligament is formed principally by the posterior layer of the thoracolumbar fascia, with contributions from longissimus thoracis and multifidus [21] (Fig. 4A). However, contrary to the view that the posterior component of the lumbar interspinous ligaments belong to the aponeurosis from longissimus [21], our results indicate that this is formed by the fibers of the thoracolumbar fascia in both the lower thoracic and lumbar spines (Fig. 2C, Fig. 3C). Hence, in this respect, the interspinous ligaments in the lumbar spine can be considered to be a deep component of the thoracolumbar fascia [19]. The interspinous-supraspinous-thoracolumbar ligamentous tissue complex has a major role in anchoring the major fascial planes of the back to the spinous processes [13, 25]. Demonstration of the attachments of the posterior layer of the thoracolumbar fascia further endorses its spinal stabilizing role via the substance of the midline ligaments [4, 22].

It is apparent that there are regional differences in the structural make-up of the posterior ligamentous system along the whole length of the spine. In this investigation, the architectural arrangement of the dense connective tissue appears to be related directly to spinal level, with fiber orientation varying according to the level under consideration. This appeared to be directly attributable to the arrangement of tendons and aponeuroses overlying each of the spinous processes. In the upper thoracic spine, an oblique and longitudinal fiber arrangement was seen in the supraspinous ligament due to the combined attachments of rhomboideus major, splenius cervicis and, more superficially, trapezius (Fig. 1B).

Our results indicate that there is a wide variation in fiber direction within the connective tissue architecture of the posterior ligamentous tissue, within both the supraspinous (Fig. 4A) and interspinous (Fig. 5A–D) ligaments. This may explain some of the complex loading patterns that have been found within this tissue [8, 9, 12, 17]. One such example is seen with the high proportion of stiffness retained by the lumbar interspinous ligaments on disrup-

tion [8]. It has also been suggested that their collagen fibers behave as a fiber-reinforced composite in order to withstand force [8]. While this point is still debatable, the multiple directions of connective tissue fibers within the supraspinous and interspinous ligaments indicates that they are capable of transmitting loads in more than one direction.

Recognition of regional differences within the posterior ligamentous system has several biomechanical implications. The validity of spinal models can only be achieved by the incorporation of precise anatomical information regarding geometric, physical and material representation of its structural components [13]. For instance, inclusion of the anatomical differences within the thoracolumbar spine and the multiple components contributing to its structure may provide a more realistic appraisal of biomechanical responses of the spine than extrapolating models of the ligaments based on a single axial cable system. Variations in mechanical properties found throughout the length of the supraspinous and interspinous ligaments [16] are also explained by the differences in structural morphology throughout the thoracic and lumbosacral spine. However, the clear demonstration that structurally they are essentially an assembly of tendinous attachments indicates that their function cannot be evaluated independently from their muscles of origin.

Conclusions

The results in this study support a description of the supraspinous and interspinous ligaments as structures formed by a combination of muscle tendons and aponeuroses along the length of the thoracic and lumbar spine, with regional differences in their connective tissue architecture. The interspinous ligaments are absent in the upper thoracic spine and are at least partially formed by the posterior layer of the thoracolumbar fascia in the lower thoracic spine. Incorporation of regional differences of the supraspinous/interspinous ligament complex into biomechanical models will give a more accurate representation of their anatomy.

Acknowledgements The generosity of the Department of Anatomy, Auckland University, for providing access to the collection of plastinated slices and financial assistance from the Deans Fund (Otago Medical School) are acknowledged. Brynley Crosado and Russell Barnett, Department of Anatomy & Structural Biology, University of Otago, are thanked for their technical assistance.

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