



Distinct fascicular and epineurial architecture of the obturator nerve: a comparative analysis with clinical and surgical relevance

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Abstract: Peripheral nerves of the lumbar plexus share a common origin but exhibit distinct anatomical and clinical features. Although the gross anatomy of these nerves has been described, differences in cross-sectional structure remain unclear. This study compared the histological architecture of the obturator nerve (ON) with that of the femoral nerve (FN) and lateral femoral cutaneous nerve (LFCN). Eighteen nerves (ON, FN, and LFCN) from six cadavers were examined using Masson's trichrome-stained cross-sections. Morphometric analysis included the short-to-long axis ratio and fascicular area ratio. Epineurium conspicuity was assessed using a semi-quantitative 3-point scale by three blinded observers. Inter-observer agreement was evaluated using weighted kappa statistics. Non-parametric tests were used for comparisons. The ON had a significantly higher short-to-long axis ratio (0.590 ± 0.10) than the FN (0.393 ± 0.08) and LFCN (0.359 ± 0.09) ($P=0.031$). The fascicular area ratio was also greater in the ON (0.470 ± 0.09) than in the FN (0.262 ± 0.09) and LFCN (0.226 ± 0.07) ($P=0.031$). Epineurium scores were higher in the ON (median 2, IQR 2–2) than in the FN and LFCN (median 1, IQR 0–1) ($P<0.05$), with substantial inter-observer agreement ($\kappa=0.71$). The ON exhibits a distinct fascicular tissue-rich architecture with a consistently well-developed epineurium, which may contribute to its characteristic macroscopic appearance and aid anatomical identification.

Key words: Anatomy, Cadaver, Peripheral nerve, Histology, Pelvis

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Introduction

Peripheral nerves of the lumbar plexus, including the obturator nerve (ON), femoral nerve (FN), and lateral femoral cutaneous nerve (LFCN), share a common segmental origin from the L2–L4 nerve roots, but play distinct functional roles in the lower extremity. The ON descends posterior to the psoas major and travels through the obturator foramen

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to innervate the thigh adductor muscles and provides limited sensory innervation to the medial thigh, whereas the FN primarily contributes to knee extension and hip flexion and also provides sensory innervation to the anterior and medial thigh [1]. In contrast, the LFCN is a purely sensory nerve supplying cutaneous sensation to the anterolateral thigh [2].

Despite their common origin, these nerves differ considerably in their anatomical course, branching patterns, and clinical vulnerability [3, 4]. The ON courses along the lateral pelvic wall and divides into anterior and posterior branches around the obturator foramen [5, 6], while the FN passes between the psoas major and iliacus muscles, courses beneath the inguinal ligament, and divides into multiple motor and sensory branches. The LFCN follows a more superficial course near the anterior superior iliac spine, making it particularly susceptible to entrapment syndromes [7].

Clinically, these anatomical differences are reflected in distinct patterns of neuropathy. Injury to the ON results in adductor weakness and medial thigh sensory disturbance, often associated with pelvic surgery [8]. Injury to the FN leads to quadriceps weakness and gait disturbance, commonly following retroperitoneal or orthopedic procedures [2]. The LFCN is frequently implicated in meralgia paresthetica due to its superficial course [7].

Although the macroscopic anatomy and clinical features of these nerves have been well described, subtle differences in their gross appearance are often noted during dissection. In particular, the ON is frequently perceived as having a relatively brighter or more distinct appearance compared with adjacent nerves. This observation raises the possibility that intrinsic structural differences, particularly in connective tissue composition, may underlie these macroscopic features.

However, comparative studies focusing on the cross-sectional morphology of these nerves remain limited. Detailed analysis of their cross-sectional architecture may provide insight into the structural basis of these observed differences. Such differences may not only contribute to their macroscopic appearance but also serve as useful anatomical indicators for identifying the ON within the pelvis. Furthermore, variations in connective tissue composition may influence nerve biomechanics, potentially affecting nerve course, mobility, and susceptibility to injury.

Therefore, the aim of this study was to compare the cross-sectional morphology of the ON, FN, and LFCN using cadaveric and histological analyses, and to clarify structural differences that may underlie their distinct anatomical characteristics.

Materials and Methods

Specimens and morphometric analysis

Eighteen nerves from six formalin-fixed anatomical donors (four female and two male) were dissected. The age at death ranged from 42 to 98 years, with a mean of 81.2 ± 20.3 years. The study was conducted in accordance with all relevant local and international ethical guidelines regarding the use of human anatomical donors in research [9].

Sample harvesting

The ON, FN, and LFCN on the same side were harvested at the L4–5 level from each anatomical donor.

Histological preparation

Specimens were processed using standard histological techniques and stained with Masson's trichrome to enhance visualization of collagen-rich connective tissue components, including the epineurium. All specimens were stained using the same protocol to ensure consistency across samples.

Morphometric analysis

Cross-sectional histological images of the FN, ON, and LFCN were obtained from the same specimens ($n=6$), allowing paired comparisons within each individual.

For each nerve, the total cross-sectional area was measured by manually tracing the outer boundary of the nerve along the epineurium using ImageJ (National Institutes of Health), with thresholding applied to assist in boundary identification (Fig. 1). In addition, the long and short axes of each nerve were measured, and the short-to-long axis ratio was calculated to assess cross-sectional shape, with values closer to 1.0 indicating a more rounded morphology.

Fascicles were identified based on the presence of a distinct perineurial boundary. Each fascicle was individually traced, and the total fascicular area was calculated as the sum of all fascicular areas within a given nerve.

The fascicular area ratio was calculated as:

$$\text{fascicular area ratio} = \text{fascicular area} / \text{total nerve area}$$

All morphometric measurements were performed by a single observer.

Semi-quantitative evaluation of the epineurium

The conspicuity of the epineurium was assessed semi-

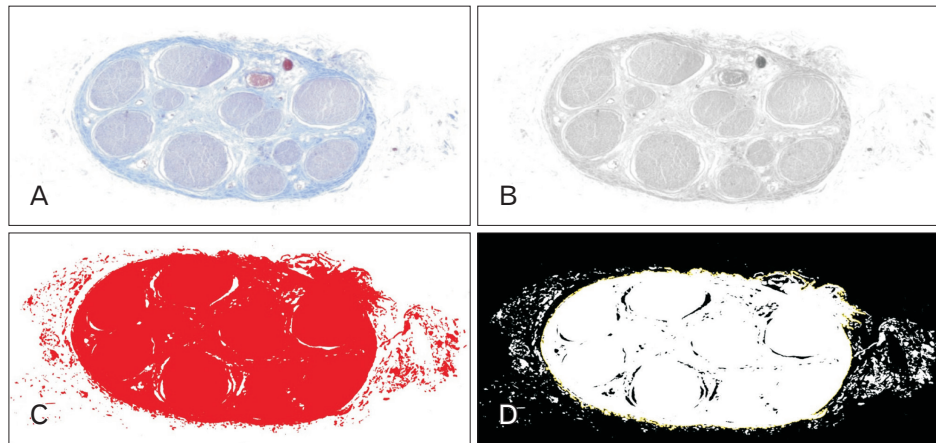


Fig. 1. Image processing workflow for morphometric analysis of peripheral nerve cross-sections. Representative histological image and processing steps used for morphometric measurements in ImageJ (National Institutes of Health). (A) Original histological section. (B) Conversion to 8-bit grayscale. (C) Thresholding applied to enhance contrast and facilitate identification of nerve boundaries. (D) Final manual tracing of the outer boundary of the nerve (yellow line) along the epineurium. Thresholding was used to assist boundary detection; however, all measurements were finalized by manual tracing.

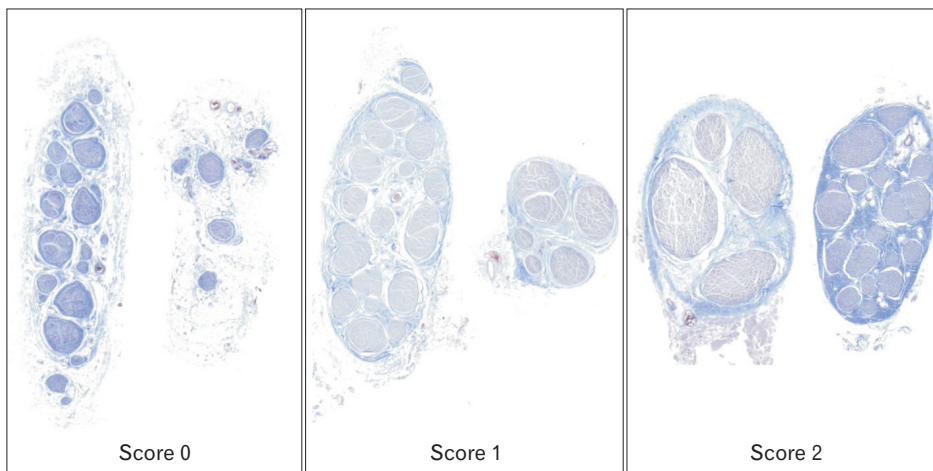


Fig. 2. Representative examples of each score (0–2) to reach consensus on the scoring criteria.

quantitatively using a 3-point ordinal scale:

- 0: not identifiable
- 1: partially identifiable or indistinct
- 2: clearly identifiable and well developed, with a continuously traceable outer boundary.

Prior to scoring, a calibration session was conducted using representative histological images not included in the study dataset. During this session, the scoring criteria were explained in detail, including continuity, visibility, and the ability to trace the outer boundary of the nerve. Observers reviewed representative examples of each score (0–2) and reached consensus on the scoring criteria (Fig. 2).

To reduce comparative and expectation bias, all images were presented individually in randomized order, and both

nerve type and specimen identity were blinded to the observers. In addition, histological images of other peripheral nerves, processed using the same staining protocol, were interspersed among the study images and excluded from the final analysis.

Each image was independently evaluated by three observers. For each nerve in each specimen, the final epineurium score was determined using the median value of the three ratings.

Inter-observer agreement

Inter-observer agreement was assessed using weighted kappa (κ) statistics with squared weights. Pairwise agreement between observers was calculated, and the mean κ

value was reported. Agreement was interpreted according to the criteria of Landis and Koch [10].

Statistical analysis

Because all measurements were obtained from matched nerves within the same specimens and the sample size was small ($n=6$), non-parametric statistical tests were used. Differences among the three nerves (FN, ON, and LFCN) were evaluated using the Friedman test. When significant differences were identified, *post hoc* pairwise comparisons were performed using the Wilcoxon signed-rank test. A P -value <0.05 was considered statistically significant.

The authors state that every effort was made to follow all local and international ethical guidelines and laws that pertain to the use of human anatomical donors and their images in anatomical research [11-13]. According to institutional policy, Institutional Review Board (IRB) approval was not required for cadaveric studies.

Results

Short-to-long axis ratio

The short-to-long axis ratios of the FN, ON, and LFCN were 0.393 ± 0.08 , 0.590 ± 0.10 , and 0.359 ± 0.09 , respectively. A significant difference was observed among the three nerves (Friedman test, $P < 0.01$). *Post hoc* pairwise comparisons revealed that the ON had a significantly higher short-to-long axis ratio than both the FN and LFCN (Wilcoxon signed-rank test, $P = 0.031$ for both comparisons), whereas no significant difference was observed between the FN and LFCN

($P > 0.05$).

Importantly, the short-to-long axis ratio of the ON exceeded that of the FN and LFCN in all specimens (Fig. 3).

Fascicular area ratio

The fascicular area ratios of the FN, ON, and LFCN were 0.262 ± 0.09 , 0.470 ± 0.09 , and 0.226 ± 0.07 , respectively. A significant difference was observed among the three nerves (Friedman test, $P < 0.01$). *Post hoc* analysis demonstrated that the ON had a significantly higher fascicular area ratio than both the FN and LFCN (Wilcoxon signed-rank test, $P = 0.031$ for both comparisons), whereas no significant difference was observed between the FN and LFCN.

Importantly, the fascicular area ratio of the ON exceeded that of the FN and LFCN in all specimens.

Epineurium score

Inter-observer agreement for epineurium scoring was substantial, with a mean weighted kappa value of 0.71 across observer pairs (Table 1).

The epineurium score differed significantly among the three nerves (Friedman test, $P < 0.01$). The ON consistently demonstrated higher scores than the FN and LFCN. *Post hoc* analysis revealed that the ON had significantly higher epineurium scores than both the FN and LFCN (Wilcoxon signed-rank test, $P < 0.05$), whereas no significant difference was observed between the FN and LFCN.

The epineurium score was highest in the ON (median 2, IQR 2–2), whereas both the FN and LFCN demonstrated

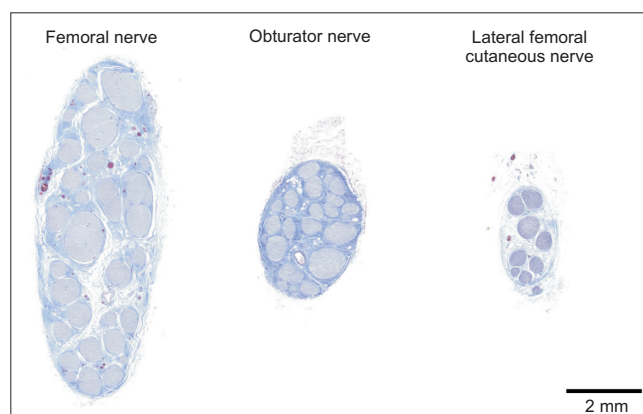


Fig. 3. Representative cross-sectional images of each nerve. The obturator nerve demonstrates a more rounded shape, less prominent interfascicular connective tissue with a well-defined epineurium compared with the femoral and lateral femoral cutaneous nerves.

Table 1. Inter-observer agreement for semi-quantitative epineurium scoring

Observer pair	Weighted κ	95% CI
Observer 1 vs. 2	0.74	0.49–0.90
Observer 1 vs. 3	0.68	0.41–0.87
Observer 2 vs. 3	0.71	0.46–0.89
Mean	0.71	-

Weighted kappa (κ) values were calculated for each pair of observers using squared weights. Agreement was substantial across all observer pairs. CI, confidence interval.

Table 2. Semi-quantitative epineurium scores

Nerve	Median	IQR
Femoral nerve	1	0–1
Obturator nerve	2	2–2
Lateral femoral cutaneous nerve	1	0–1

Values are presented as medians and interquartile ranges (IQRs). Scores were determined using the median of three independent observers for each specimen.

lower scores (median 1, IQR 0–1) (Table 2).

These findings indicate that the ON possesses a consistently well-developed epineurium.

Integrated structural characteristics

Taken together, the ON exhibited:

- a more rounded cross-sectional morphology,
 - a higher proportion of fascicular components, and
 - a consistently well-developed epineurium,
- compared with the FN and LFCN.

These findings indicate that the ON exhibits a distinct structural profile compared with the FN and LFCN (Fig. 3).

Discussion

The present study demonstrates that the ON exhibits a distinct structural profile compared with the FN and LFCN, characterized by a higher proportion of fascicular components, a more rounded cross-sectional geometry, and a consistently well-developed epineurium. These findings were consistent across specimens and supported by both quantitative and semi-quantitative analyses.

Peripheral nerves are composed of fascicular and non-fascicular components, including the epineurium, perineurium, and interfascicular connective tissue, each contributing to mechanical protection and functional integrity [14, 15]. The significantly higher fascicular area ratio observed in the ON suggests that the relative proportion of these components varies among peripheral nerves.

The epineurium, as the outermost connective tissue layer, plays an essential role in protecting nerve fibers from external mechanical stress and facilitating nerve mobility within surrounding tissues [14, 15]. The higher epineurium scores observed in the ON indicate a more developed external connective tissue envelope compared with the FN and LFCN.

Experimental studies have shown that connective tissue organization influences the mechanical behavior of peripheral nerves, including resistance to compression and deformation [16, 17]. A well-developed epineurium may provide more uniform external support, resulting in a more circular morphology.

The use of Masson's trichrome staining in the present study further supports this interpretation. This staining method allows clear visualization of collagen-rich connective tissue components [18], enabling reliable identification of the epineurium. The consistently higher epineurium scores in

the ON suggest that the outer connective tissue layer is more readily identifiable in the ON than in the comparison nerves. This finding is broadly consistent with previous anatomical descriptions indicating that funiculi occupy approximately 52% of the ON cross-sectional area at the sacroiliac joint, implying that nearly half of the nerve consists of fascicular tissue [14].

Interestingly, these structural differences may correspond to subtle macroscopic features observed during dissection. Empirically, the ON is often perceived as having a relatively brighter or more distinct appearance compared with adjacent nerves.

Although such observations have not been systematically evaluated, the present findings raise the possibility that the characteristic appearance of the ON may be related to its distinct structural profile. However, the present study did not directly assess macroscopic appearance, surface characteristics, or optical properties, and this interpretation should therefore be regarded as speculative. Future studies incorporating quantitative assessment of gross appearance or imaging characteristics may help clarify this relationship. Whether the distinct connective tissue architecture observed in the ON influences ultrasonographic visibility remains unknown. Future studies combining histological and ultrasonographic analyses may help clarify the relationship between nerve structure and ultrasound appearance [19, 20].

From an anatomical and surgical perspective, this feature may have practical implications. Identification of the ON within the pelvis can be challenging due to its deep location and proximity to surrounding structures. The presence of a relatively well-defined and reflective outer layer may serve as a useful supplementary visual cue during dissection or surgical exposure, particularly in complex pelvic procedures [21, 22]. The anatomical course of the ON along the pelvic wall also suggests potential exposure to mechanical compression, such as increased intra-pelvic pressure. The well-developed epineurium observed in this study may represent an adaptive feature that enhances mechanical protection.

An additional consideration is that the ON arises from the anterior divisions of the lumbar plexus, whereas the FN and LFCN originate from the posterior divisions. Therefore, it remains unclear whether the structural characteristics observed in the ON represent a unique feature of this nerve or reflect broader architectural differences between anterior- and posterior-division nerves. Developmental differences between these divisions may potentially influence connec-

tive tissue organization and fascicular arrangement. Future studies including other anterior division nerves, such as the accessory ON, may help clarify this issue.

Differences in connective tissue composition may further contribute to functional variation. In the ON, the well-developed epineurium, rather than an increased internal connective tissue content, may influence how the nerve responds to compression, traction, or surgical manipulation [15, 17]. A relatively fascicle-dense architecture with a well-developed epineurium, as observed in the ON. These structural differences may partly explain variations in clinical presentation, vulnerability to injury, and recovery patterns among different peripheral nerves [23]. In addition, the relatively well-developed epineurium and interfascicular connective tissues of the ON may facilitate surgical handling during procedures such as nerve grafting and suturing, as the nerve may be less susceptible to mechanical damage [24]. The consistently well-developed epineurium and higher fascicular proportion in the ON may also influence tactile feedback during needle advancement, as well as perineural injectate spread and resistance to injection.

Comparative morphometric studies of specific peripheral nerves remain limited. While detailed descriptions of nerve structure are well established [14, 15, 22], direct comparisons among anatomically related nerves are scarce. The present study, therefore, provides novel insight into regional variation in peripheral nerve architecture.

The consistency of the findings across all specimens is notable. In every case, the ON demonstrated higher fascicular area ratios and short-to-long axis ratios than the FN and LFCN, suggesting that these differences reflect intrinsic structural characteristics rather than incidental variation.

The reliability of the semi-quantitative scoring was supported by substantial inter-observer agreement (mean weighted $\kappa=0.71$). The kappa statistic is widely used to assess agreement beyond chance in categorical data [25], and the observed values indicate robust reproducibility.

Taken together, the findings suggest that the ON represents a structurally distinct entity among peripheral nerves, characterized by a fascicular tissue-rich architecture, consistently conspicuous epineurium and a more rounded cross-sectional geometry.

Limitations

Several limitations of this study should be acknowledged. First, the sample size was relatively small ($n=6$), which may

limit the generalizability of the findings. However, the paired study design and the consistent pattern observed across all specimens partially mitigate this limitation.

Second, the epineurium evaluation was semi-quantitative and based on visual assessment. Although inter-observer agreement was substantial, quantitative histochemical or imaging-based approaches may provide additional precision in future studies.

Third, the analysis was based on two-dimensional histological sections. Because peripheral nerves are three-dimensional structures, cross-sectional analysis may not fully capture spatial variations along their course.

Fourth, the LFCN and FN are posterior division nerves and ON is an anterior division nerve. Future studies could evaluate additional anterior division nerves such as the accessory ON.

Finally, the study population consisted of older cadaveric specimens, and age-related changes in connective tissue composition cannot be excluded. Future studies including a broader age range may further clarify the generalizability of these findings.

The ON exhibits a distinct structural profile compared with the FN and LFCN, characterized by a higher fascicular proportion, a more rounded cross-sectional geometry, and a consistently well-developed epineurium. These features collectively indicate a fascicular tissue-rich architecture with a consistently well-developed epineurium. The consistent differences observed across specimens suggest that the ON represents a structurally distinct entity among the branches of the lumbar plexus.

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Conceptualization: SI, MSH, RST, JI. Data acquisition: SI, MSH, JI. Data analysis or interpretation: SI, MSH, RST, JI. Supervision: RST, JI. Drafting of the manuscript: SI, NK, YT.

Critical revision of the manuscript: MSH, RST, JI. Approval of the final version of the manuscript: all authors.

Conflicts of Interest

No potential conflict of interest relevant to this article was reported.

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