

Histomorphometric Evaluation of Musculocutaneous, Median, and Ulnar Nerves: Relationship Between Gross Diameter and Fascicular Architecture and its clinical significance

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Abstract

Purpose:

The musculocutaneous, median, and ulnar nerves are particularly important because they carry motor, sensory, and autonomic fibers to functionally critical regions of the arm, forearm, and hand. The nerve is composed of multiple fascicles, each surrounded by perineurium and embedded within endoneurial and epineurial connective tissue. The number, size, distribution, and rearrangement of fascicles may vary along the course of the nerve. The current study aimed to investigate the histomorphometry of MCN, MN, and UN and to establish the relationship between gross diameter and fascicular architecture.

Methods:

A descriptive histomorphometric study was conducted on 62 upper-limb specimens at a tertiary care hospital. The gross diameters of musculocutaneous nerve, medial root of median nerve, lateral root of median nerve, median nerve, and ulnar nerve were measured. Total cross-sectional area, total mean feret diameter, fascicular cross-sectional area, and fascicular total mean feret diameter were assessed for the musculocutaneous, median, and ulnar nerves.

Results:

Among the gross measurements, the median nerve showed the greatest mean diameter, followed by the lateral root of the median nerve and the ulnar nerve. The musculocutaneous nerve showed the smallest mean gross diameter. Histologically, the ulnar nerve showed the highest total mean cross-sectional area, while the median nerve showed the highest mean fascicular cross-sectional area and mean fascicular Feret diameter.

Conclusion:

The current study has an integrated approach of gross and microscopic assessment of upper-limb nerves. Histomorphometric evaluation may improve anatomical understanding relevant to nerve repair, grafting, entrapment neuropathy, and regional anaesthetic procedures.

Introduction

The peripheral nerves of the upper limb show a high degree of anatomical and functional specialization. The musculocutaneous, median, and ulnar nerves are particularly important because they carry motor, sensory, and autonomic fibers to functionally critical regions of the arm, forearm, and hand. The musculocutaneous nerve (MCN) is a branch of lateral cord and it supplies the anterior compartment of the arm. The median nerve (MN) arises from two roots, one root from the lateral cord named as lateral

root of median nerve (LRMN) and other from the medial cord named as medial root of median nerve (MRMN). Sometimes MN arises from three or more roots.[1] MN provides major motor supply to the pronator compartment of the forearm and thenar muscles. The ulnar nerve arises from the medial cord and it supplies intrinsic muscles of the hand and contributes to fine motor control. Because these nerves are long, relatively superficial in several regions, and vascular structures, they are vulnerable to trauma, compression, iatrogenic injury, and entrapment.[2]

The nerve is composed of multiple fascicles, each surrounded by perineurium and embedded within endoneurial and epineurial connective tissue. The number, size, distribution, and rearrangement of fascicles may vary along the course of the nerve. Sunderland et.al found that peripheral nerves possess complex internal architecture, including fascicular redistribution, variable proportions of connective tissue and fascicular tissue.[2] Stewart et.al observed that fascicular architecture has clinical significance. The author emphasized on a point that any injury to particular nerve fascicles may lead to selective motor or sensory deficits rather than complete nerve palsy.[3]

Histomorphometric assessment of the total nerve cross-sectional area, fascicular area, fascicular diameter, and connective tissue arrangement, provides detailed information of the nerve that cannot be obtained by gross diameters alone. Previous studies reported on the internal topography of major nerves of the brachial plexus. The studies detailed on the fascicular arrangement that are sufficiently organized for guiding the nerve repair, but also variable to demand caution during operative procedures. [4, 5] Quantitative studies shown that fascicular dimensions, fascicle number, and cross-sectional area may vary according to nerve and the level of the nerve (Forearm, arm and hand), the morphometric data of this is critical for surgical planning and intervention.[6]

Communications between the musculocutaneous and median nerves are among the most frequently described variations in the infraclavicular part of the brachial plexus. Their presence can modify the expected pattern of weakness or sensory deficit after the nerve injury. Ballesteros et.al reported that musculocutaneous–median nerve communications in cadaveric specimens and emphasized their clinical relevance in diagnosis and surgical management.[7] A systematic review and meta-analysis by Sirico et.al reported that musculocutaneous nerve variations are not rare and that their classification should also taken into consideration for the presence and position of communicating branches.[8] Similarly, the median–ulnar communications such as Martin–Gruber, Marinacci, Riché–Cannieu, and Berrettini anastomoses may alter electrodiagnostic findings and operative anatomy. [9, 10] Such communicating branches may carry selected fascicles between nerves and therefore, their diameter and internal architecture may influence the clinical expression of nerve injury.

Despite the available literature on gross variations and the fascicular architecture, relatively fewer studies have attempted to integrate gross nerve diameter with histological architecture. External nerve diameter is easy to measure and is often used during dissection and surgery. However, a larger external diameter may reflect larger fascicles, more numerous fascicles, increased epineurial tissue, or a combination of these factors. Conversely, a nerve with a smaller external diameter may still possess

compact but functionally significant fascicles. This is clinically relevant because surgeons performing nerve repair, nerve grafting, neurolysis, fascicular dissection, or nerve transfer often depend on the internal topography of the nerve rather than only its external size.

The aim of the current study is to investigate the histomorphometry of MCN, LRMN, MRMN, MN, and UN and to establish the relationship between gross diameter and fascicular architecture. This integrated approach of the anatomical information will be beneficial to the clinical anatomists, peripheral nerve surgeons, and anaesthesiologists.

Materials and Methods

The present study included 62 adult upper limb specimens which were donated as cadaver to the Department of Anatomy at a tertiary care hospital. The limbs with deformities and damage involving the axillary region were excluded from the present study. The gender and side of the specimen were also noted.

Gross morphometry:

The dissection of specimens was done following the steps of Cunningham's manual of practical anatomy. Following the reflection of the skin and fascia, the tip of coracoid process providing attachments to coracobrachialis and short head of biceps were identified. Following the identification of axillary artery and axillary vein, the MCN entering deep surface of coracobrachialis was traced back to its origin from the lateral cord. The MN lying medial to coracobrachialis was traced back to its origin from the lateral and medial cords. The UN lying behind the axillary vein was traced back to its origin from the medial cord. The diameters were measured using the digital vernier caliper with accuracy of 0.02 mm (Aerospace) at the level of diversion of these nerves from their roots and cords (**Figure: 1**).

Histomorphometric analysis:

A 2 cm long segments of MCN, MN and UN were obtained from the level of diversion of nerves from the cords to analyse the histomorphometry and fixed in 10% neutral buffered formalin (NBF). The specimens were dehydrated in ascending grades of ethanol, cleared in xylene, embedded in paraffin wax. Serial sections of 7µm thickness were obtained using a rotary microtome. The sections were stained with luxol fast blue and eosin. The photomicrographs were taken using "Olympus CX 41 Microscope". The images were acquired at 10X, 20X and 40X magnification from six adjacent locations in five alternate sections per nerve and analyzed in Image J software. At 10X magnification the whole cross-section of nerve was selected by using the main menu option, polygon selection. The total cross sectional area and total ferret diameter of each nerve were measured using the "measure" option. After outlining the outer edge of perineurium of each fascicle using the polygon selection, its number, area and diameter were obtained by using the "measure" option.

All the statistical analyses were performed with IBM SPSS Statistics, version 19 (IBM Corp., Armonk, New York, USA). The difference in the proportion of these variations between the sides were analysed using Mann Whitney U test and student unpaired *t* test.

Results

Gross Morphometry:

Gross anatomical measurements were recorded across 62 adult upper limbs to determine the outer diameters of the major nerves arising from the lateral and medial cord of brachial plexus. (Fig. 2). The UN and musculocutaneous nerve MCN demonstrated mean diameters of 3.21 ± 0.50 mm and 2.76 ± 0.69 mm, respectively. The MN has the largest mean diameter at 4.53 ± 0.76 mm. The LRMN measured 3.23 ± 0.97 mm, while the MRMN measured 2.96 ± 0.62 mm. (Fig. 3: **a-d**)

According to the side of the upper limbs, (31 right limbs, 31 left limbs), no statistically significant differences were observed ($p > 0.05$). (Table 1)

Table 1
Gross diameters of the nerves based on the sides.

Nerves	Diameter: Mean $\pm$ S.D (mm)			p-value
	Total(n = 62)	Right (n = 31)	Left (n = 31)	
MCN	2.76 ± 0.69	2.90 ± 0.67	2.63 ± 0.68	0.610
MRMN	2.96 ± 0.62	2.94 ± 0.62	2.99 ± 0.63	0.481
LRMN	3.23 ± 0.97	3.46 ± 0.93	3.00 ± 0.97	0.673
MN	4.53 ± 0.76	4.58 ± 0.82	4.49 ± 0.70	0.438
UN	3.21 ± 0.50	3.24 ± 0.51	3.18 ± 0.50	0.855
SD-Standard deviation, mm-millimeter ,MCN-Musculocutaneous nerve, MRMN- Medial root of median nerve, Lateral root of median nerve, MN-Median nerve, UN-Ulnar nerve				

According to gender differences (41 male and 21 female specimens) revealed distinct morphological variations. The gross diameter of the MN was significantly larger in male subjects (4.67 ± 0.85 mm) compared to female subjects (4.27 ± 0.44 mm), it was statistically significant ($p = 0.010$). The remaining neural structures, including the MCN, UN, and both median nerve roots, did not exhibit any significance. (Table 2)

Table 2
Gross diameters of the nerves based on the gender.

Nerves	Diameter: Mean $\pm$ S.D (mm)		p-value
	Males (n = 41)	Females (n = 21)	
MCN	2.85 $\pm$ 0.70	2.59 $\pm$ 0.62	0.859
MRMN	3.02 $\pm$ 0.66	2.86 $\pm$ 0.53	0.480
LRMN	3.32 $\pm$ 1.03	3.06 $\pm$ 0.83	0.240
MN	4.67 $\pm$ 0.85	4.27 $\pm$ 0.44	0.010*
UN	3.26 $\pm$ 0.49	3.12 $\pm$ 0.52	0.753
SD-Standard deviation, mm-millimeter ,MCN-Musculocutaneous nerve, MRMN- Medial root of median nerve, Lateral root of median nerve, MN-Median nerve, UN-Ulnar nerve. *Statistically significant			

Histomorphometric analysis:

Histomorphometric evaluation was conducted utilizing ImageJ software to definitively quantify the neural area and fascicular geometry of the MCN, MN, and UN. (Fig. 4: **a-c**). Analysis of the total cross-sectional nerve profiles demonstrated that the UN possessed the largest mean total cross-sectional area at $0.02891 \pm 0.00295 \text{ mm}^2$, with a total mean ferret diameter of. The MN exhibited a mean total cross-sectional area of $24.22 \pm 0.06 \text{ mm}^2$ and a total ferret diameter of $0.21202 \pm 0.00962 \text{ mm}$. The MCN yielded the smallest overall profile, presenting a mean cross-sectional area of $0.02071 \pm 0.00130 \text{ mm}^2$ alongside a total ferret diameter of $0.21655 \pm 0.00425 \text{ mm}$ (Table 3). The total mean cross sectional area of the nerves MCN,MN, and UN are statistically significant with $p = 0.0013$.

Table 3
Histomorphometric measurements of cross-sectional area and mean ferret diameter of the nerves

Nerve	Fascicular Cross-Sectional area (mm ²)	Mean fascicular Ferret diameter (mm)
	Mean $\pm$ SD	Mean $\pm$ SD
MCN	0.002257 $\pm$ 0.001954	0.06462 $\pm$ 0.03214
MN	0.003881 $\pm$ 0.001197	0.09548 $\pm$ 0.02068
UN	0.002349 $\pm$ 0.001330	0.07304 $\pm$ 0.02624
p-value	0.0018*	0.0018*
SD-Standard deviation, mm-millimeter ,MCN-Musculocutaneous nerve, MN-Median nerve, UN-Ulnar nerve. *Statistically significant		

Detailed assessment of the internal fascicular organization revealed further structural heterogeneity among the terminal branches. The MN displayed the largest mean fascicular cross-sectional area at

0.003881 ± 0.001197mm², corresponding to a total fascicular ferret diameter of 0.09548 ± 0.02068mm. In contrast, the UN and MCN demonstrated much finer internal architectures. The UN was characterized by a mean fascicular area of 0.002349 ± 0.001330 mm² and a total mean fascicular ferret diameter of 0.07304 ± 0.02624 mm. Similarly, the MCN exhibited a mean fascicular cross-sectional area of 0.002257 ± 0.001954 mm² with a concurrent ferret diameter of 0.06462 ± 0.03214 mm. (Table 4) The total mean fascicular cross sectional area and mean fascicular ferret diameter of the nerves MCN,MN, and UN are statistically significant with p = 0.0018.

Table 4
Histomorphometric measurements of the fascicular organization of the nerves

Nerves	Total cross-sectional area (mm ²), Mean ± SD	Mean Ferret diameter (mm), Mean ± SD
MCN	0.02071 ± 0.00130	0.21655 ± 0.00425
MN	0.02422 ± 0.00324	0.20605 ± 0.00968
UN	0.02891 ± 0.00295	0.21202 ± 0.00962
p-value	0.0013*	0.1729
SD-Standard deviation, mm-millimeter ,MCN-Musculocutaneous nerve, MN-Median nerve, UN-Ulnar nerve. *Statistically significant		

Relationship between gross diameter and histological architecture

On measuring the gross diameter, the median nerve showed the largest mean external diameter. Histologically, the ulnar nerve showed the highest total cross-sectional area, while the median nerve showed the highest mean fascicular cross-sectional area and mean fascicular Feret diameter. These results suggested that the relationship between gross diameters and histomorphometric analysis is not strictly linear. External diameter may reflect the overall caliber of the nerve, but the internal composition depends on the relative contribution of fascicular tissue and connective tissue. Thus, gross diameter alone should not be considered as a complete indicator of functional or histological complexity.

Discussion

This current study provides an integrated approach of gross and histomorphometric analysis of the MCN, LRMN, MRMN, MN and UN. Earlier studies revealed that the nerve variations are significant because they show that external nerve diameter is the only source to depict any injury, but that does not fully represent the internal fascicular organization of a peripheral nerve, which is anatomically and clinically significant.

The MCN showed the smallest gross diameter among the other nerves. This is because of the supply of the nerve is more restricted muscular territory in the anterior compartment of the arm and continues as a cutaneous nerve to the lateral forearm. However, its clinical relevance should not be underestimated. Chiarapattanakom et.al studied the internal topography of the MCN and their fascicular groups.[11] This is clinically significant for nerve transfer procedures, particularly in brachial plexus injury, where fascicular selection and donor–recipient matching influence functional recovery.

MN arises from both lateral and medial cords of the brachial plexus and carries contributions from C5 to T1 spinal segments. Its calibre is large, reflects the broad distribution of the nerve. In the present study, the MN had a mean diameter of 4.53 ± 0.76 mm, which was greater than the MCN and UN. Brill et.al reported that the functional importance of the MN indicating substantial fascicular complexity within upper limb nerves. [6] The MN showed the highest fascicular cross-sectional area and fascicular Feret diameter in the present study. This observation supports the concept that the median nerve contains relatively prominent fascicular components. Earlier studies of median nerve fascicular anatomy have shown that its internal organization is clinically important for selective neurolysis, nerve repair, and neuroprosthetic applications. Planitzer et.al studied median nerve fascicular anatomy as a basis for distal neural prostheses and emphasized that knowledge of fascicular topography may improve selective stimulation and surgical reconstruction.[12] Franco et.al studied the relevance of intraneural median nerve anatomy in treating mixed median nerve injuries in the hand.[13] The current study details on the larger fascicular profile in the median nerve is therefore consistent with the functional and surgically significant.

In the gross morphometry of UN, the mean diameter was smaller than that of the median nerve. Histologically, however, it showed the highest total cross-sectional area among the three nerves. At the same time, its fascicular cross-sectional area was lower than that of the median nerve. This suggests that a larger total microscopic profile may not necessarily mean larger fascicular tissue. The difference needs to be explained in detail by variations in epineurial or perineurial connective tissue, fascicle number, fascicle spacing, or tissue processing effects. Peripheral nerves contain varying proportions of connective tissue, and the connective tissue component has important mechanical and protective functions. It allows the nerve to tolerate stretching, compression, and movement across joints, but it may also influence the appearance of cross-sectional size. Franco et.al described the connective tissues associated with peripheral nerves as clinically important in relation to regional anesthesia and interpretation of the nerve sheath environment.[14]

The absence of statistically significant right–left differences is notable. Although the right-sided mean values were numerically higher for MCN, LRMN, MN, and UN, the differences were not statistically significant. This suggests that side-related asymmetry may not be a major determinant of nerve diameter. However, this should be interpreted cautiously.

The nerve communications are important and have a lot of clinical and functional significance. Median–ulnar communications may also complicate clinical interpretation. Martin–Gruber anastomosis,

Marinacci anastomosis, Riché–Cannieu anastomosis, and Berrettini anastomosis represent different patterns of crossover between median and ulnar nerve territories. Ballesteros et.al found musculocutaneous–median nerve communications in 19.8% of upper limbs in their cadaveric series and emphasized their relevance in diagnosis and management of upper-limb nerve lesions.[7] Sirico et.al in a systematic review discussed about MCN variations are sufficiently common to warrant careful anatomical consideration .[8] Roy et.al, observed that the nerve anastomoses have variable incidence and its clinical implications for electrophysiological interpretation.[9] Smith et.al summarized how these communications may alter the motor or sensory innervation patterns in the forearm and hand.[10]

In traumatic lacerations and nerve repair aims to align the corresponding fascicular groups as accurately as possible. When internal topography of the nerve is complex or when communicating branches alter the expected fiber distribution, functional recovery may be incomplete despite technically adequate epineurial repair. Jabaley et.al described the clinical application of internal topographic knowledge in repair, lysis, and grafting of major forearm nerves.[4] Some studies emphasized that knowledge of internal anatomy is important for nerve repair in the forearm and hand.[5] The present study adds to this concept by showing that external nerve size alone cannot be relied upon to infer internal fascicular pattern.

The perineurium and epineurium influence diffusion, mechanical protection, and response to local anesthetic or compression. [14] A nerve with greater connective tissue may respond differently to compression than a nerve with densely packed fascicles. Thus, histomorphometric data may contribute to understanding why similar degrees of compression can produce different clinical patterns. Regional anesthesia is another relevant application. Peripheral nerve blocks depend on accurate localization of nerves and predictable spread of anesthetic around fascicles. Intrafascicular injection is a recognized mechanism of nerve injury, and local anesthetic exposure may damage the axons present in the nerve, particularly when injected within fascicles.[15] Therefore, the relationship between gross diameter and histological architecture was interpreted descriptively rather than by correlation analysis

Conclusion

The present study demonstrates that the musculocutaneous, median, and ulnar nerves show measurable differences in gross diameter and histomorphometric architecture. The median nerve had the greatest gross diameter and the most prominent fascicular dimensions, while the ulnar nerve showed the highest total microscopic cross-sectional area. These findings indicate that gross nerve diameter and internal histological structure are related but not equivalent. Gross diameter alone should not be used as a complete indicator of fascicular complexity. Combined gross and microscopic assessment provides a more accurate understanding of upper-limb nerve morphology and may be useful in anatomical teaching, peripheral nerve repair, nerve grafting, interpretation of neuropathies, and regional anesthesia.

Abbreviations

Abbreviation

Definition

MCN

Musculocutaneous nerve

MN

Median Nerve

MRMN

Medial root of Median Nerve

LRMN

Lateral root of Median Nerve

UN

Ulnar Nerve

Declarations

Competing interests

The authors declare no potential conflicts of interest relevant to this article.

Ethics approval

The study was conducted on cadaveric specimens obtained through the voluntary body donation program of the Department of Anatomy, Jawaharlal Institute of Postgraduate Medical Education and Research (JIPMER), Puducherry. Informed written consent for academic, educational, and research purposes—including scientific publication—was obtained from the donors' next of kin in accordance with institutional guidelines. Formal Institutional Ethics Committee (IEC) review was waived pursuant to institutional policy governing research on donated cadaveric material.

Consent to participate

All specimens utilized in this study were obtained through the institutional voluntary body donation program. Informed written consent was provided prior to or at the time of donation, explicitly permitting the use of the donors' bodies for anatomical research and medical education.

Consent to publish

Not applicable. No personally identifiable information or distinguishing anatomical features are disclosed in this manuscript or its accompanying figures, ensuring donor confidentiality is strictly maintained.

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Author Contribution

Protocol development: Dr Raveendranath Veeramani

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Data analysis: Dr Vani PC and Dr Nikilesh Sankaran

Manuscript writing/editing: Dr Nikilesh Sankaran Supervision: Dr Raveendranath Veeramani

All authors read and approved the final manuscript.

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Data availability statement

The histomorphometric datasets generated and analyzed during the current study are not publicly deposited due to institutional protocols, but are available from the corresponding author upon reasonable request.

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Figures



Figure 1

Digital Vernier caliper

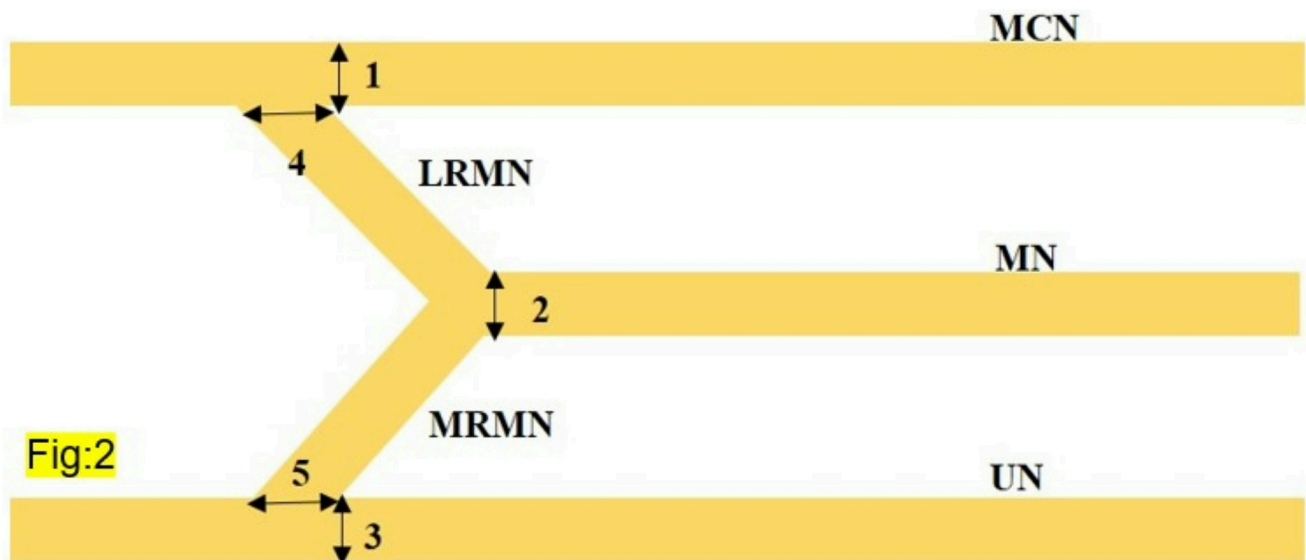


Figure 2

Schematic representation of gross diameter measurement of nerves. 1-Musculocutaneous nerve (MCN), 2-Median nerve (MN),3-Ulnar nerve (UN),4- Lateral root of median nerve (LRMN), 5-Medial root of median nerve (MRMN).

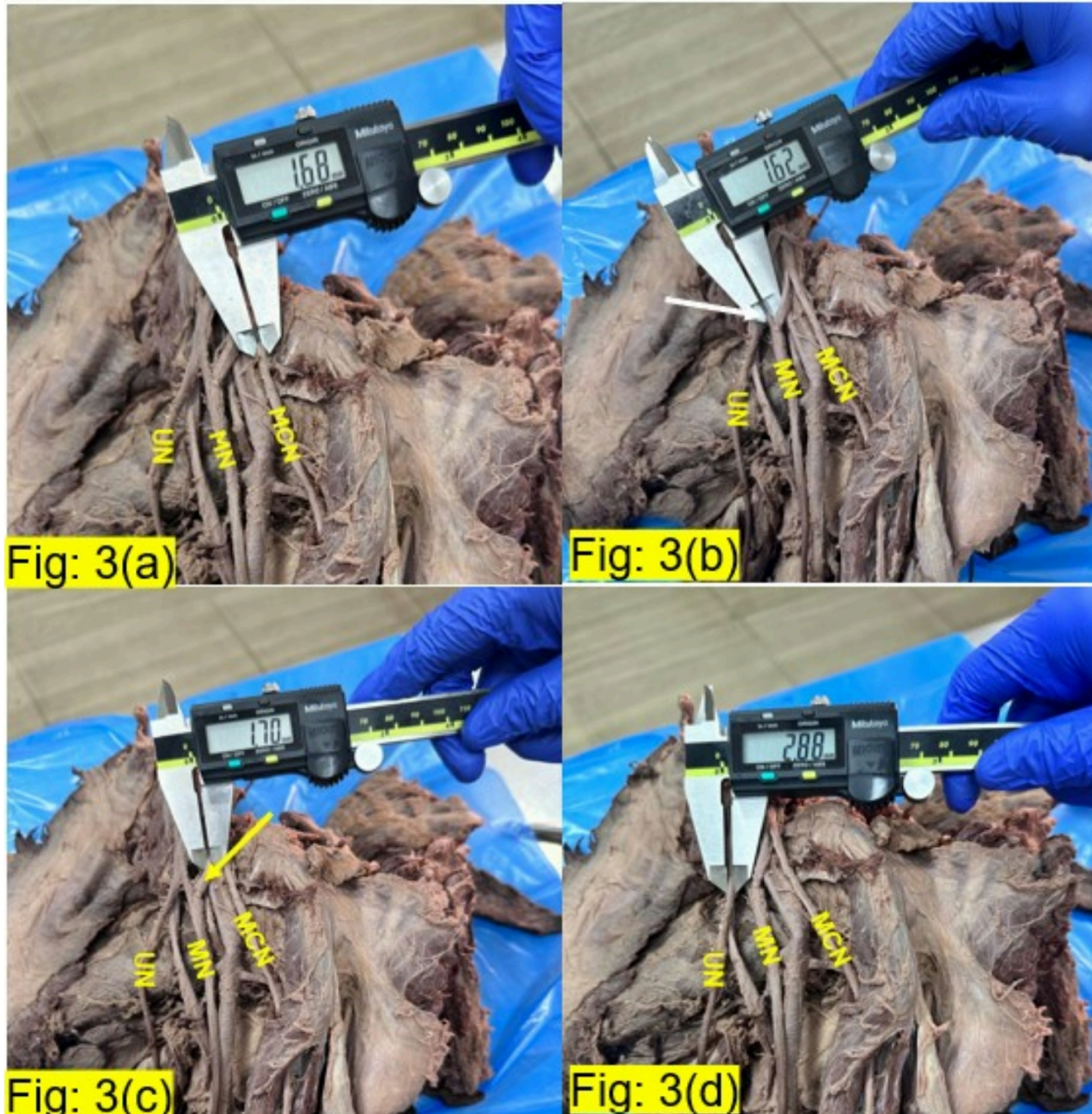


Figure 3

Gross diameter measurement of nerves using vernier caliper. a) Measurement of the Musculocutaneous nerve. b) Measurement of the medial root of the median nerve. c) Measurement of the lateral root of the median nerve. d) Measurement of ulnar nerve. MCN-musculocutaneous nerve, MN-median nerve,

MRMN-medial root of median nerve, LRMN-lateral root of median nerve, and UN-ulnar nerve. White arrow- Medial root of median nerve. Yellow arrow- lateral root median nerve.

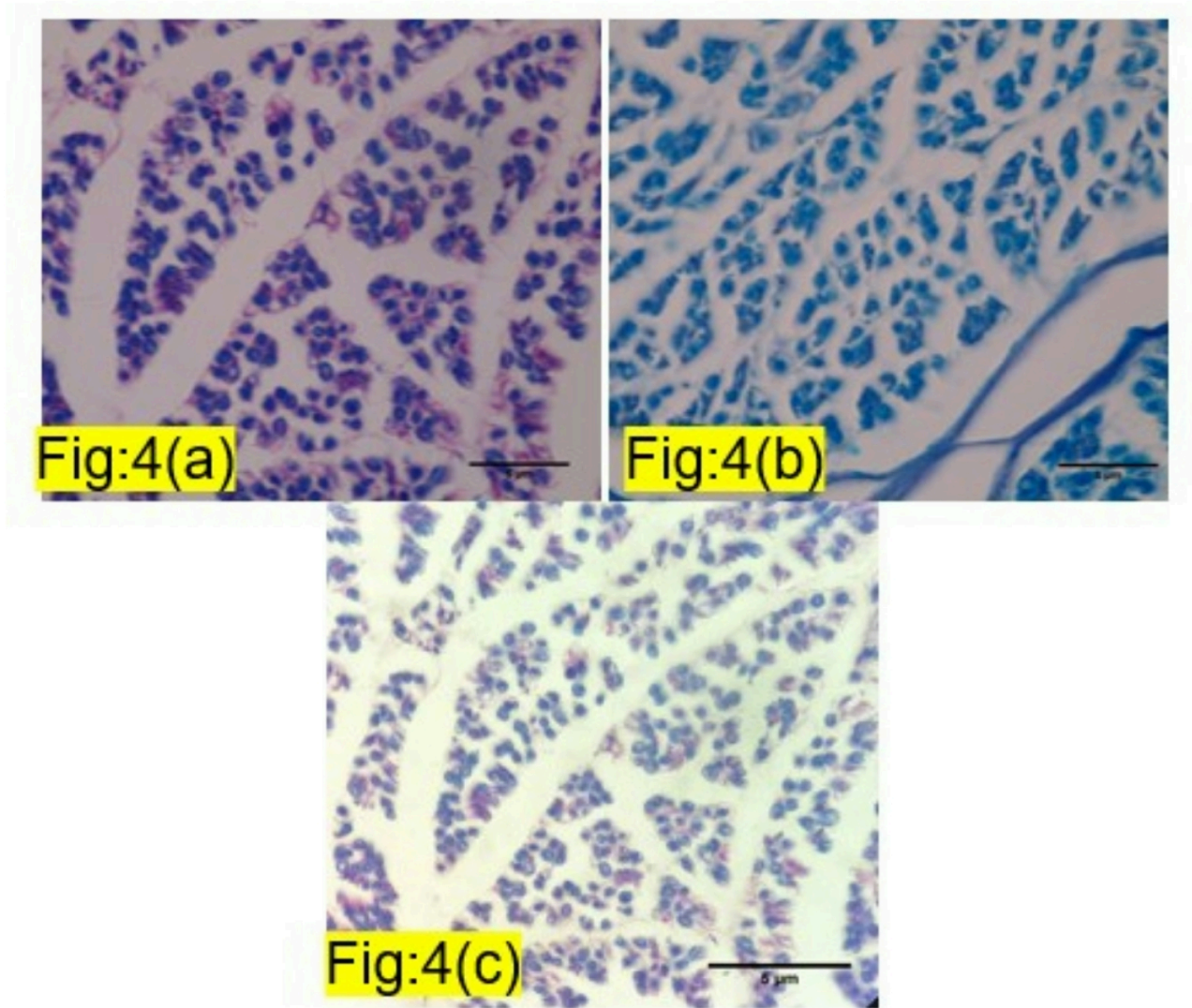


Figure 4

Histomorphometric assessment of nerves using Image J software. a) Musculocutaneous nerve, stain: Eosin, Magnification: 40X. b) Median nerve, stain: Luxol fast blue, Magnification: 40X c) Ulnar nerve, stain: Eosin, Magnification: 40X. MCN-musculocutaneous nerve, MN-median nerve, MRMN-medial root of median nerve, LRMN-lateral root of median nerve, and UN-ulnar nerve.