

Size and separability of the calcaneal and the medial and lateral plantar nerves in the distal tibial nerve

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Received: 25 November 2008 / Accepted: 15 April 2009 / Published online: 16 May 2009
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Abstract The tibial nerve (TN) has three main terminal branches: the medial and lateral plantar nerves and the calcaneal branch (CB), which innervates the foot sole. The design and implantation of nerve cuff electrodes with separate channels for each of these three terminal branches would provide significant sensory information, which can be used in functional electrical stimulation systems to assist standing or to correct foot drop. Detailed quantitative anatomical data about fascicular size and separability of the terminal branches of TN are needed for the design and implantation of such cuff electrodes. Therefore, the branching pattern, the fascicular separability and the fascicular size of the TN posterior to the medial malleolar-calcaneal axis were examined in this study, using ten human TN specimens. The TN branching patterns were highly dispersed. For the CBs, multiple branches were identified in five (50%) of the specimens. For the TN, the bifurcation point was located within the tarsal tunnel in eight (80%) of the cases. The distance proximal to the medial malleolar-calcaneal axis for which the TN could be split ranged from 0 to 41 mm. Quantitative and qualitative data were obtained for the fascicular size and separability

of the TN. Only the CB of the TN proved separable for a sufficient length for nerve cuff electrode implantation. The results suggest the use of a two-channel cuff with one common channel for the lateral and medial plantar nerves, having multiple electrodes for selective recording, and one channel for the CB.

Keywords Tibial bifurcation and fascicular distributions · Calcaneal branch pattern · Fascicular size and separability of the medial and lateral plantar nerves and the calcaneal branches · Cuff electrodes

Abbreviations

CB	Calcaneal branch
FES	Functional electrical stimulation
LPN	Lateral plantar nerve
MMCA	Medial malleolar-calcaneal axis
MPN	Medial plantar nerve
SNR	Signal-to-noise ratio
TN	Tibial nerve
TT	Tarsal tunnel

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Introduction

The distal part of the tibial nerve (TN) branches into the calcaneal branch (CB), the medial plantar nerve (MPN) and the lateral plantar nerve (LPN). These branches innervate the calcaneal, the medial plantar and the lateral plantar areas of the foot, respectively (See Fig. 1), and carry sensory information from those areas.

Sensory information from the skin and from muscles and tendons as well can be used as a feedback signal for a functional electrical stimulation (FES) system used to

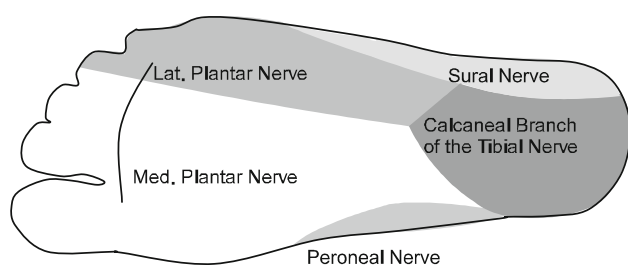


Fig. 1 Schematic drawing representing the areas innervated by the three main sensory nerves of the foot sole. Adapted from Gray (1995)

stimulate paralysed muscles for the restoration of movement in spinal cord-injured individuals (Haugland et al. 1997; Havel et al. 1988; Navarro et al. 2005). One modality to obtain such information is the use of a cuff electrode as a neural interface, enabling the recording of compound nerve signals, which includes the sensory information carried by the nerve (Andreasen and Struijk 2002, 2006; Andreasen et al. 2000; Haugland et al. 1997; Havel et al. 1988; Hoffer 1990; Stein et al. 1975).

The distal TN with its branches is of special interest for FES applications such as the correction of foot drop (Haugland and Sinkjær 1995) and FES for standing and walking of spinal cord-injured people (Andreasen and Struijk 2003a). In the case of a foot drop FES system, the CB will convey information about the heel strike and the heel lift, whereas the MPN and the LPN carry information about the floor contact of their respective innervation areas, which may be used to detect inversion or eversion of the foot during walking. The heel-contact signal is essential for timing of the peroneal nerve stimulation in a foot drop system, whereas inversion or eversion may call for a change of the stimulation parameters. In the case of FES for standing and walking, in addition to the information mentioned above, the nerve signals from the three innervation areas give information about the centre of pressure (Andreasen and Struijk 2003a), which is needed for the control of standing.

Ideally, a nerve cuff electrode should be placed on each of the three terminal branches of the TN to obtain maximally selective signals from the three main innervation areas (see Fig. 1) and to obtain a maximal signal-to-noise ratio (SNR). Alternatively, a multichannel cuff with three separate channels, one for each of the three branches of the TN, could be used. The SNR is optimised by the choice of a cuff electrode with the lowest possible diameter (Struijk 1997). However, a tightly fitting cuff may induce nerve damage (Hoffer 1990). These requirements for the cuff design and implantation pose two questions: (1) can the branches of the TN be safely separated from each other to allow for a multichannel-cuff implant, and (2) what are the dimensions of the three nerves?

Qualitative descriptions of the TN and its branches are readily available in the literature, but detailed quantitative data are not given anywhere. At the level of the ankle, the TN bifurcates into the MPN and LPN (see Fig. 2). This bifurcation point varies only a little among specimens, whereas a great dispersion exists in the bifurcation level of the CB. Most studies indicate that the tibial bifurcation is located in the tarsal tunnel in the great majority of cases (Table 1). Davis and Schon (1995) found that the TN divides within the tarsal tunnel, within 2 cm of the medial-malleolar-calcaneal axis (MMCA, see Fig. 2) in 16 out of 18 feet. In the remaining two feet the bifurcation took place 5 and 9 cm proximal to the MMCA. However, Bareither et al. (1990) identified the bifurcation more proximally in up to 31% of the feet studied. A single case in which the TN completely failed to divide into terminal branches has been reported as well (Sammarco and Conti 1994), but this must be considered to be extremely seldom.

In order to place a cuff electrode on the TN, in which the fascicles of the CB, LPN and MPN run inside separate channels, the branches have to be separated proximal to the tarsal tunnel. In this work we examine the dimensions and the separability of the CB, LPN and MPN, and the location of the bifurcation point of the TN and the branching point of the CB. The examination is based on cross-sections of the studied nerves allowing for the study of the fascicular branching pattern at the perineural level, the importance of which was recently emphasised by Kudoh and Sakai (2007).

Materials and methods

Anatomy

The medial plantar nerve

The MPN is the largest and most anterior of the terminal branches of the TN. It has muscular, cutaneous, articular and vascular branches and supplies the medial part of the foot sole, anterior to the heel (see Fig. 1). As soon as the nerve enters the sole of the foot, its cutaneous branches arise and course downward in the interval between the abductor of the big toe and the flexor digitorum brevis to supply the inner aspect of the foot sole. The cutaneous branches anastomose with terminal branches of the medial calcaneal nerve (Sarrafian 1983).

The lateral plantar nerve

The LPN has muscular, cutaneous, articular and vascular branches and supplies the lateral part of the foot sole, anterior to the heel (see Fig. 1). After the bifurcation of the

Fig. 2 The tibial nerve bifurcates into its terminal branches below the flexor retinaculum, at the medial malleolus-calcaneal axis. The *right side* of the figure shows cross-sections of nerve N9 in this study. The cross-section at the *bottom* shows clearly separable fascicles of the LPN, MPN and the CB. In the cross-section shown in the middle, the *line* indicates where the fascicles can be separated. The *top figure* of the cross-sections shows the same nerve 1 mm more proximally. The perineurium of the MPN and LPN can no longer clearly be distinguished, and the fascicles have merged

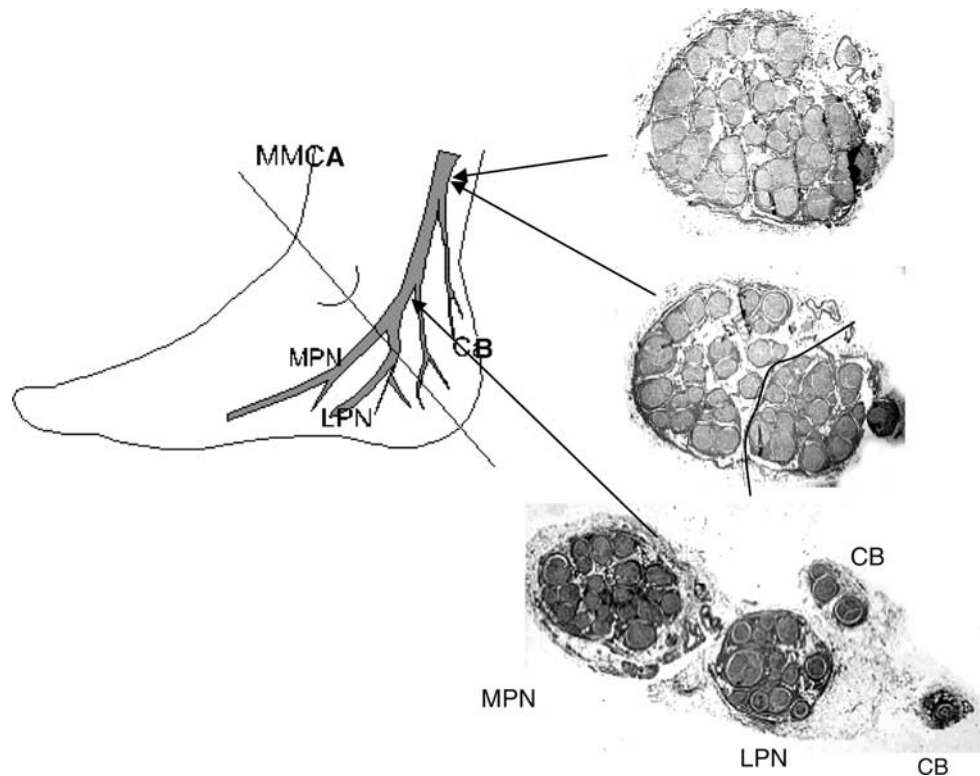


Table 1 Bifurcation patterns of the tibial nerve modified from Davis and Schon (1995)

Author (year)	Specimens (N)	Bifurcation in tarsal tunnel ^a (%)	Bifurcation proximal to the tarsal tunnel ^a (%)	Bifurcation distal to the tarsal tunnel ^a (%)
Horwich (1938)	100	96	4	0
Dellon and Mackinnon (1984)	31	94	6	0
Havel et al. (1988)	68	93	7	0
Nagoaka (1990)	62	85	15	0
Bareither et al. (1990)	126	69	31	0
Davis and Schon (1995)	20	90	10	0
Current study, Andreassen Struijk et al. (2009)	10	80	10	10

^a Data are expressed as ± 2 cm from the MMC axis

TN, the LPN is located more posteriorly than the MPN. Two centimetres within its origin (Davis and Schon 1995), the LPN gives off a rather large muscular branch to the abductor digiti quinti, which courses almost transversely laterally anterior to the calcaneal tuberosities. This branch has often been referred to as a CB (Davis and Schon 1995).

The calcaneal branch

Newer studies of the CB patterns show a high variability among specimens in both number and origin of the CB (Davis and Schon 1995; Dellon and MacKinnon 1984; Havel et al. 1988), and some variability also exists between the feet of the same individual. The medial CB often

branches from the TN in the distal third of the leg and divides into two branches. The posterior branch covers the skin at the medial part of the Achilles and the posterior part of the heel. The anterior branch is a plantar branch, covering the posterior part of the foot sole. It courses along with the inner border of the foot, and some of its terminal branches anastomose with branches of the neighbouring nerves: the sural nerve, the CBs of the saphenous nerve and cutaneous branches of the LPN and MPN (Sarrafian 1983). Besides the cutaneous branches, the proximal CB, instead of the terminal portion of the TN, provided small articular branches and vascular branches to the posterior tibial artery in the cases studied by Davis and Schon (1995).

The diameter of the CB is rather small as compared to the ones of the MPN and LPN.

Material

The study was performed on ten TNs (three from left legs and seven from right legs) dissected from ten different human cadavers donated to the Anatomical Institute at the University of Aarhus, Denmark. None of the dissected cadavers showed any evidence of nerve disease or damage. The cadavers were routinely fixed by perfusion with formalin within 24 h postmortem.

Nerve processing

The TNs were dissected from a point at least 6 cm above the MMCA to a point well beyond the branching point into the LPN and MPN. During dissection all branches were marked with coloured threads and carefully described on a drawing of the nerve. The dissected nerve segment was gently removed, mounted on a cork board and further fixed in 2.5% glutaraldehyde in Sørensen's phosphate buffer, pH = 7.2–7.4 (Hospital Pharmacy, Aarhus County Hospital, Aarhus, Denmark) for a time period ranging from a few days to 5 months.

Prior to the embedding in paraffin, the nerves were cut transverse to the nerve axis at the level where the MMCA crossed the centre of the nerve. The length of the TN, the location of its branch points and the diameter of the branches at different locations along the TN posterior to the MMCA were measured using a microscope and a slide gauge. The nerves were dehydrated overnight and embedded in paraffin using a standard laboratory machine (LEICA tissue processor). To prepare the nerve for microtome slicing, the paraffin-embedded nerve was cut in 3-mm-long pieces and mounted vertically in blocks filled with paraffin to be sliced perpendicularly to the nerve axis. For each 0.5 mm of the length of the nerve, three 4- μ m-thick slices were cut using a sliding microtome. The slices were straightened in the water bath, and one of the three slices was stained using Masson's trichrome, after which all the slices were mounted on glass plates.

The glass plates were mounted on a microscope and photographed. Preliminary experiments on a single nerve showed that it was sufficient to examine sections representing every 1 mm length of the nerve in order to be able to study the fascicular separability of the nerve. The total magnification was 11.5–17.8 times, as determined from photographs of a scale.

Data analysis

The distance for which the TN could be separated into the LPN, MPN and CB was determined from the photographs. The fascicles were considered to be separable if each of the

fascicle groups had clear separate perineuriums (see Fig. 2) and a separate vascular supply (the fascicles were considered to have a separate vascular supply if the vascular supply was located within the fascicle groups or in the epineurium at either of the fascicles. If the main vascular supply was located between the fascicles, it was considered to be common, since the fascicles could not be split without injuring the supply). When this was no longer the case, it was concluded that the fascicles had merged and could no longer be safely separated.

The height and width of the cross-section of the fascicle groups belonging to the MPN, LPN and the CB, as well as the total cross-sectional height and width of the TN, were measured at the MMCA and at the merge points. The measurements were performed on the photographs, and the metric data were calculated using the known magnification. Measurements of fascicle group sizes with and without connective tissue were performed at the widest part of each group of fascicles and in a direction perpendicular to that line (Fig. 3).

Results

Bifurcation of the tibial nerve

In eight of the ten nerves, the bifurcation was located in the tarsal tunnel (TT: within ± 2 cm of the MMCA). Four of these (50%) divided distal to the MMCA. The remaining two nerves, which did not bifurcate in the TT, bifurcated 27 mm distal to and 24 mm proximal to the MMCA, respectively. The very distal bifurcation has not been described in the previous studies, as shown in Table 1. It should be noted that the number of specimens in the current study is relatively small, as compared to the other studies

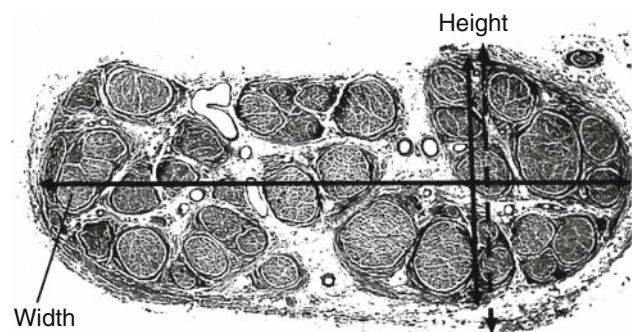


Fig. 3 The measurement of TN cross-sectional diameters. The dotted line shows an example of how the measurements are made with connective tissue. The increase in diameter caused by the connective tissue is included in Table 4 and is given as a number in braces following the measured nerve diameters without connective tissue

shown in Table 1. This is due to the time-consuming quantitative approach of the current work.

In all ten cases the MPN was located anterior to the LPN.

Calcaneal branch patterns

In five of the ten specimens analysed, two CBs were identified, in good agreement with the results of a study performed by Davis and Schon (1995) (Table 2). In three of these five cases, one of the two CBs descended separately from the TN, separating from the TN at a point more than 7–10 cm proximal to the MMCA. In these three cases, the second CB originated either from the posterior side of the TN in the TT (one case) or from the anterior side of the TN in the TT (two cases).

In the two cases without separately descending CBs, one CB originated from the posterior side of the TN in the TT and one from the anterior side. In one of these cases the anterior CB originated directly from the MPN. This origin is in accordance with the findings of Havel et al. (1988) and Davis and Schon (1995), but was not seen by Horwich (1938) and Dellon and Mackinnon (1984).

In the present study, the CB from the anterior side of the TN usually innervated the anterior part of the skin under the heel, while the posterior branch innervated the more posterior part of the skin.

In total five of the ten specimens had CBs descending separately from locations more than 7–10 cm proximal to the MMCA; in two of these cases the separately descending CB was the only CB.

In one case with a single CB, this branch originated from the medial part of the TN, between the locations of the MPN and LPN. This was also the only case where the location of the CB point was proximal to the TT. All other CB points were located in the TT (except for the cases with separately descending CBs).

External measures of the tibial nerve

These external measurements of the TN were made after fixating the nerve in formaldehyde, but before dehydrating and embedding the nerve in paraffin.

The total cross-sectional dimensions of the TN and of the separate CBs were measured at different locations along the nerve (Table 3). These five locations (0, 30, 45, 75 and 90 mm) are defined in the proximal direction, relative to the MMCA. The external measures include both the fascicles and the connective tissue.

The cross-sectional area of the TN was elliptic, and the height and the width of the ellipse were measured as shown in Fig. 3. While the TN was merely oval in the more proximal end of the specimens, it flattened close to the MMCA. There was a large variability in the cross-sectional size of the TN. At the MMCA, the width varied within 3.3–3.9 mm and the height within 5.7–8.7 mm ($n = 4$). At a location 70–75 mm proximal to the MMCA, the cross-sectional width varied within 2.4–3.5 mm and the height varied within 4.0–8.6 mm ($n = 10$).

Internal measurements and fascicular separation

Location of the bifurcation and branch points

The external measurements of the location of the bifurcation and branch points of the LPN, MPN and the CB are shown in Tables 4 and 5, together with the internal measurements of the cross-sectional size of the fascicles of the LPN, MPN and the CBs. In addition, the potential fascicular separation lengths of the MPN, the LPN and the CBs within the TN are shown in Tables 4 and 5.

At, or distal to, the bifurcation point, the TN gave off a motor branch to the abductor digiti minimum. Usually this branch merged with the LPN and was included in the LPN fascicular group in the measurements, and thus it was included in the MPN group in a few cases only. In the cases

Table 2 Branching patterns of the calcaneal branch modified from Davis and Schon (1995)

Author (year)	Specimens (<i>N</i>)	Branches		Origin: location			Origin: nerve		
		One (%)	Multiple (%)	TT only (%)	<i>P</i> only (%)	Both (%)	TN (%)	LPN (%)	MPN (%)
Horwich (1938)	100	–	–	96	4	0	4	96	0
Dellon and Mackinnon (1984)	20	75	25	35	40	25	87	13	0
Rondhuis and Hudson (1986)	34	100	0	–	–	–	100	0	0
Havel et al. (1988)	68	79	21	56	37	7	78	29	6
Davis and Schon (1995)	20	40	60	30	35	35	95	10	15
Current study Andreassen Struijk et al. (2009)	10	50	50	40	30	30	100	0	10

TT tarsal tunnel, *P* proximal, *TN* tibial nerve, *LPN* lateral plantar nerve, *MPN* medial plantar nerve

Table 3 The external dimensions of the tibial nerve

Nerve	Size at MMCA (0 mm) (mm)	Size at 30 mm (mm)	Size at 45 mm (mm)	Size at (1): 70 mm or (2): 75 mm (mm)	Position/size size at 90 mm or more (mm)
N2					
Tibial	–	w:4.5, h:2.6	w:4.5, h:2.6	2—w:5.3, h:2.6	
Calcaneal	–	w:1.5, h:1.0	w:1.5, h:1.0	2—w:1.2, h:1.2	
N3					
Tibial	–	w:8.5, h:3.1	w:8.5, h:2.8	2—w:8.6, h:2.4	
N4					
Tibial	–	w:6.2, h:3.1	w:5.4, h:2.4	1—w:4.3, h:3.4	
N7					
Tibial	–	w:5.2, h:3.5	w:5.8, h:3.0	1—w:5.8, h:2.6	
N8					
Tibial	–	w:5.1, h:3.1	w:4.4, h:3.0	2—w:4.0, h:2.8	Position: 90 mm w:3.8, h:3.0
Calcaneal	w:2.2, h:1.3	Runs on surface of TN	Runs on surface of TN	Runs on surface of TN	Runs on surface of TN
Calcaneal	w:2.2, h:1.2	Runs in TN			
N9					
Tibial	w:7.2, h:3.7	w:5.0, h:3.7	w:4.6, h:3.7	2—w:4.7, h:3.3	Position: 92 mm w:4.2, h:2.7
Calcaneal	w:1.2, h:0.8	w:0.9, h:0.7	w:1.0, h:0.6	2—w:1.0, h:0.6	w:0.6, h:0.6
Calcaneal	w:1.0, h:1.0	Runs in TN			
LPN	w:5.2, h:2.8	Runs in TN			
MPN	w:5.9, h:2.8	Runs in TN			
N10					
Tibial	w:7.2, h:3.7	w:5.5, h:3.5	w:5.3, h:3.2	2—w:5.3, h:2.6	Position: 108 mm w:5.3, h:2.1
Calcaneal	w:1.4, h:1.0	w:1.2, h:0.7	w:1.0, h:0.8	2—w:0.9, h:0.7	Runs in TN
N11					
Tibial	w:5.7, h:3.9	w:4.4, h:3.6	w:4.9, h:3.5	2—w:4.0, h:3.5	Position: 97 mm w:3.6, h:3.2
N12					
Tibial	w:8.7, h:3.3	w:6.2, h:3.4	w:5.0, h:3.3	2—w:4.4, h:3.1	Position: 100 mm w:5.3, h:3.0
N13					
Tibial	–	w:3.1, h:4.9	w:5.2, h:3.2	2—w:4.9, h:2.9	Position: 100 mm w:5.8, h:2.3
Calcaneal	w:1.8, h:1.3	–	–	–	–
Calcaneal	w:1.0, h:0.7	Runs in TN			
LPN	w:4.8, h:2.2	Runs in TN			
MPN	w:5.3, h:2.1	Runs in TN			

Position is taken in the proximal direction, relative to the MMCA

where the TN bifurcated proximal to the MMCA, the measured size of the TN is rather large due to the distance between the LPN and MPN. In one nerve (N12), a small branch w:0.4 and h:0.3 mm branching off from the MPN, merged 7 mm proximal to the MMCA and was not included in the measurements.

Fascicular separability

There were major differences in the fascicular separability of the TN into the LPN, MPN and CB (Tables 4, 5). The separable length was 0–41 mm, measured from the MMCA in the proximal direction. In average the TN was separable into the LPN and MPN for a length of 18.7 mm ($n = 10$).

This number includes the nerves, in which the TN bifurcated proximal to the MMCA, which means that the LPN and MPN run as separate nerves in the distal part of the length used for the calculation of the average value shown above. In two cases (N7 and N12 in Table 4), there was no separability of the TN. In one case (N7 in Table 4) the merge point was found from the photographs to be distal to the bifurcation point measured externally. The reason for this is the shrinkage and loss of nerve tissue due to the processing of the nerve performed before the photographs were taken, as further described in the discussion.

Excluding the five CBs that were descending separately from locations more proximal than 70 mm from the MMCA, the CBs were running separately or were separable

Table 4 Size and location of the branch points of the TN, MPN and the LPN

Nerve	MPN, LPN external bifurcation point relative to MMCA [mm]	MPN, LPN internal merge point relative to MMCA [mm]	MPN size at MMCA [mm]	MPN size at merge point [mm]	LPN size at MMCA [mm]	LPN size at merge point [mm]	TN size at MMCA [mm]	TN size at merge point [mm]
N2	-7.5	30.5	w:3.8 (0.5), h:2.1 (0.4)	w:3.8 (0.7), h:2.6 (0.4)	w:2.4 (0.3), h:1.9 (0.2)	w:2.3 (1.0), h:1.6 (0.5)	w:6.6 (0.5), h:2.1 (0.4)	w:4.2 (0.3), h:3.8 (0.7)
N3	10.0	41.0	w:4.3 (1.2), h:2.1 (0.3)	w:3.4 (0.4), h:2.2 (0.7)	w:3.0 (0.7), h:2.4 (2.1)	w:2.4 (0.3), h:1.7 (0.8)	w:9.7 (0.3), h:2.4 (2.1)	w:5.7 (0.6), h:2.2 (0.7)
N4	23.6	38.0	w:3.9 (0.4), h:2.1 (0.3)	w:3.7 (0.3), h:2.6 (0.3)	w:2.5 (0.1), h:2.1 (0.1)	w:2.0 (0.6), h:1.8 (0.7)	w:6.5 (0.2), h:2.0 (0.3)	w:5.1 (0.6), h:2.7 (0.3)
N7	7.2	5.0	w:3.4 (0.1), h:2.4 (0.8)	w:2.7 (0.4), h:2.3 (0.6)	w:2.4 (0.7), h:2.2 (0.2)	w:2.4 (0.6), h:2.1 (0.1)	w:5.6 (0.2), h:2.3 (1.0)	w:4.6 (1.3), h:2.5 (0.3)
N8	-15.5	24.5	w:3.0 (1.0), h:2.4 (0.5)	w:2.9 (0.3), h:1.5 (0.7)	w:5.7 (0.9), h:2.5 (0.8)	w:3.9 (0.79), h:2.5 (0.5)	w:7.0 (1.0), h:3.7 (1.3)	w:4.6 (0.3), h:2.9 (1.2)
N9	3.8	22.0	w:2.6 (1.8), h:2.7 (1.1)	w:3.7 (1.4), h:2.5 (0.3)	w:2.5 (1.4), h:2.3 (0.7)	w:2.8 (0.3), h:2.2 (0.3)	w:6.1 (2.1), h:2.5 (1.4)	w:4.7 (1.1), h:3.5 (0.7)
N10	-6.7	5.0	w:3.4 (0.3), h:2.5 (0.5)	w:3.2 (0.6), h:2.3 (1.2)	w:2.6 (0.5), h:1.8 (1.6)	w:2.4 (0.5), h:2.2 (0.9)	w:5.8 (0.6), h:2.3 (1.0)	w:5.3 (0.8), h:2.3 (1.1)
N11	-27.2	5.0	w:2.5 (1.0), h:1.2 (0.7)	w:2.6 (1.3), h:1.7 (0.5)	w:3.3 (1.0), h:2.3 (0.7)	w:3.5 (0.7), h:1.3 (2.5)	w:4.2 (0.6), h:3.7 (0.7)	w:4.2 (0.4), h:3.4 (0.6)
N12	-14.0	Distal to MMCA	-	-	-	-	At 7.0, w:6.6 (0.4), h:3.2 (0.8)	At 7.0, w:5.0 (0.3), h:3.0 (0.7)
N13	3.0	21.0	w:5.7 (0.4), h:1.7 (0.8)	w:4.5 (0.7), h:2.3 (1.7)	w:3.8 (0.6), h:2.4 (0.3)	w:3.0 (1.1), h:2.6 (0.3)	w:9.5 (0.8), h:3.4 (0.3)	w:6.6 (0.7), h:3.0 (1.1)

Position is taken in the proximal direction, relative to the MMCA. In the cases where the CB descended separately from the TN, measurements were made for both the CB and the TN (see also Table 5). The cross-sectional measurements at the MMCA were made within 0–4 mm postal to the MMCA. The sizes are given as width: fascicular size (connective tissue) and height: fascicular size (connective tissue), see Fig. 3

Table 5 Size and location of the branch points of the CB

Nerve	Origin and external location of branch point, relative to MMCA [mm]	Internal location of merge point relative to MMCA [mm]	Cross-sectional size at the MMCA [mm]	Cross-sectional size at the merge point [mm]	Number of CBs
N2	Separately descending	Proximal to 77.0	w:1.1 (0.2), h:0.8 (0.5)	w:0.9 (0.6), h:0.5 (0.7)	1
N3	8.8 TN posterior	Proximal to 68.0	w:1.6 (2.6), h:1.1 (1.9)	w:1.2 (1.1), h:0.7 (1.9)	1
N4	23.6 TN central	21.0	w:1.8 (0.2), h:0.9 (0.3)	w:1.5 (0.4), h:0.8 (0.2)	1
N7	-3.5 MPN	14.5	w:1.8 (0.5), h:1.5 (0.3)	w:1.3 (0.2), h:1.0 (0.1)	2
N8	12.8 TN posterior	14.5	w:2.0 (0.8), h:1.5 (0.5)	w:1.9 (0.3), h:1.5 (0.2)	2
	TN surface	84.0	w:0.6 (0.1), h:0.3 (1.4)	w:0.8 (0.3), h:0.4 (0.4)	
N9	2.0 TN anterior	44.0	w:1.1 (1.7), h:0.8 (1.0)	w:0.7 (0.1), h:0.5 (0.2)	2
	Separately descending	Proximal to 89.0	w:0.7 (0.6), h:0.5 (0.4)	w:0.5 (0.9), h:0.3 (0.7)	
N10	10.0 TN posterior	22.0	w:1.8 (0.4), h:1.2 (0.2)	w:1.1 (0.3), h:1.0 (0.2)	1
	75.5 TN posterior	83.0	w:1.2 (0.2), h:0.8 (0.3)	w:1.0 (0.2), h:0.6 (0.2)	
N11	-4.9 TN posterior	14.0	w:0.4 (0.3), h:0.3 (0.6)	w:0.3 (0.3), h:0.2 (0.2)	1
N12	-3 TN anterior	7.0	w:1.5 (1.4), h:1.1 (0.6)	w:1.9 (0.7), h:0.7 (0.6)	2
	-12 TN posterior	distal to MMC		-	
N13	Separately descending	Proximal to 96.0	w:1.2 (0.2), h:0.6 (0.2)	w:0.8 (0.8), h:0.8 (0.4)	2
	10.0 TN anterior	18.0	w:0.7 (0.4), h:0.5 (0.3)	w:0.7 (0.7), h:0.5 (0.5)	

Position is taken in the proximal direction, relative to the MMCA

from the TN in up to 0–44 mm from the MMCA. On average the separable length was 17.5 mm. In one case (N4 in Table 5) the merge point was found from the photographs to be distal to the branching point measured externally. The reason for this is the shrinkage and loss of nerve tissue due to the processing of the nerve performed before the photographs were taken, as further described in the discussion. Only one branch showed no separability.

In terms of specimens (choosing the most separable CBs in cases of multiple CBs) the maximal separability of a CB was in the range of 7–23.6 mm, excluding the five specimens with separately descending CBs and the one with a merge point more proximal than 68 mm from the MMCA (N3, in Table 5). In average this gave a separable length of 14.8 mm. This means that in six (60%) of the specimens the separability length was longer than 68 mm, and for the remaining four (40%), the maximum separability length was on average 14.8 mm, and no specimens showed no separability of any CB.

Cross-sectional size of the TN

At the MMCA, the width of the cross-sectional area of the TN varied within 4.2 (0.6)–9.7 (0.3) mm (the number in the parentheses gives the additional size of the connective tissue) and the height varied within 2.0 (0.3)–3.7 (1.3) mm. The averages and standard deviations given as: averages/standard deviation were for the width: 6.8/1.7 (0.7/0.6) mm and for the height: 2.8/0.7 (0.9/0.7) mm.

At the merge point of the MPN and LPN, the width varied within 4.2 (0.3)–6.6 (0.7) mm and the height within 2.2 (0.7)–3.8 (0.7) mm. The averages and standard deviations given as: averages/standard deviation were for the width: 5/0.7 (0.6/0.3) mm, and for the height: 2.9/0.5 (0.7/0.3) mm (Table 4).

The thickness of the connective tissue varied between 0.2 and 2.5 mm, but was usually <1 mm, and was often unevenly distributed around the nerve.

Cross-sectional size of the MPN fascicle group

At the MMCA, the width of the MPN fascicle group varied within 2.5 (1.0)–5.7 (0.4) mm, and the height varied within 1.2 (0.7)–2.7 (1.1) mm. The average/standard deviations of the width were 3.6/1.0 (0.7/0.5) mm and of the height: 2.1/0.5 (0.6/0.3) mm. At the merge point, the width varied within 2.6 (1.3)–4.5 (0.7) mm, and the height varied within 1.5(0.7)–2.6 (0.4). The average/standard deviations of the width were 3.4/0.6 (0.7/0.4) mm and of the height: 2.2/0.4 (0.7/0.5) mm (Table 4).

Cross-sectional size of the LPN fascicle group

At the MMCA, the width of the LPN fascicle group varied within 2.4 (0.3)–5.7 (0.9) mm, and the height within 1.8 (1.6)–2.5(0.8) mm. The average/standard deviations of the width were 3.1/1.1 (0.7/0.4) and of the height: 2.2/0.2 (0.7/0.7) mm. At the merge point, the width varied within 2.0

(0.6)–3.9 (0.7) mm and the height within 1.3 (2.5)–2.6 (0.3) mm. The average/standard deviations of the width were 2.7/0.6 (0.6/0.3) mm and of the height: 2.0/0.4 (0.7/0.7) mm (Table 4).

Cross-sectional size of calcaneal fascicle group

At the MMCA, the width of the CBs varied within 0.4 (0.3)–2.0 (0.8) mm and the height within 0.3 (0.6)–1.5 (0.5) mm. The average/standard deviations of the width were 1.3/0.5 (0.7/0.7) and of the height: 0.9/0.4 (0.6/0.5).

At the merge point (proximal end of the specimen for the separately descending CBs), the width varied within 0.3 (0.3)–1.9 (0.7) mm and the height within 0.2 (0.2)–1.5 (0.2) mm. The average/standard deviations of the width were 1.0/0.5 (0.5/0.3) and of the height: 0.7/0.3 (0.5/0.5) mm (Table 5).

Discussion

For the majority of the specimens, the bifurcation of the TN into the LPN and MPN was located in the tarsal tunnel (eight specimens, 80%), which agrees well with Davis and Schon (1995). In one case, the TN bifurcated distally to the tarsal tunnel, which has not been reported in the previous studies (Table 1).

The TN gave off more than one CB in five (50%) of the cases. This is in contrast to older studies of the CB branching patterns, but agrees well with the more recent study of Davis and Schon (1995).

Proximal to the MMCA, the LPN and MPN fascicle groups were separable for an average length of 18.7 mm. The maximum separability length was 41 mm, and in two cases the TN was not separable at all.

In six (60%) of the cases, the specimen had a CB descending separately from the TN from a location more than 68–90 mm proximal to the MMCA. Excluding the separately descending CBs, the separability length for the remaining four (40%) cases was 7–23.6 mm.

The dimensions of the nervous tissue are expected to change during the processing of the nerves. At least two factors may be involved. Firstly, the nerves shrink during fixation, dehydration and embedding. Secondly, cutting the nerves into 3-mm-wide segments followed by sectioning in the microtome may cause additional loss of tissue, which should be taken into account when interpreting the numbers in Tables 2, 3, 4 and 5. Tissue shrinkage related to the formalin fixation occurs within 14 h and is estimated to be in the order of 4%, as suggested by a study on prostate cancer (Schned et al. 1996). Further, the axial shrinkage of the nerves due to dehydration was estimated by measuring the length of eight nerve specimens before and after

dehydration. The average shrinkage was $5.3 \pm 1.4\%$ (SD). The loss of nerve tissue due to the mechanical processing was evaluated in six specimens and estimated to be $5.9 \pm 3\%$ (SD). In one nerve the contribution to this average from (1) cutting the nerve into 3-mm pieces and (2) slicing the nerves with the microtome was found to be 50%. The total loss/shrinkage of the nerves was calculated to be $11.8 \pm 4.8\%$ (SD $n = 6$) in the axial direction with an estimated additional 4% due to the fixation in formalin. Thus, the total axial shrinkage is estimated to 15.8%. Assuming that the cross-sectional loss is similar to the measured axial loss due to dehydration and fixation, a total cross-sectional shrinkage of 9.3% is estimated. Additional shrinkage due to the staining procedure might have taken place. Still, the cross-sectional shrinkage may be smaller than this since the expansion of the 4- μm -thick slices in the water bath, after slicing the nerve with the microtome, causes swelling and expansion of the tissue. Therefore, in the case of prostate cancer, a microscope-ready slide showed only a 4.3% shrinkage in total after fixation in formalin and embedding in paraffin (Schned et al. 1996).

The study of the fascicular patterns of the LPN, MPN and CB fascicular groups showed no interfascicular communication among the groups distal to the merge points of the nerves, but within the groups there was clearly a merging and splitting of the fascicles. This is in accordance with the results of Terzis and Smith (1990), opposing the descriptions by Sunderland (1978), who suggest exceeding inter fascicular communication. In the majority of the cases, the fascicles belonging to a certain group tended to stay at the same side of the nerve throughout the length of the specimen.

This length in which the TN was separable into the LPN and MPN is in the lower range of what is desirable for implantation of nerve cuff electrodes, since the cuff should preferably not be implanted in a place where there is too much motion of the surrounding tissue, as is the case near the MMCA during ankle extension and flexion. Further, the S/N ratio of the recorded nerve signal increases with cuff length. Therefore, the cuff should have a length in the range of at least 15–30 mm, depending of the diameter of the nerve fibres (Andreasen and Struijk 2002, 2003b; Stein et al. 1975). The fact that half of the specimens had CBs that were separable for more than 70 mm gives a good possibility of implanting a cuff with two channels, one for the CB and one for the TN. The fascicular pattern with the fascicles tending to descend with a consistent cross-sectional position within the TN suggests that a cuff with multiple electrodes (Struijk et al. 1996) may be used for selective recording from the TN, even proximal to the merge points.

In general there was a high variability in the separability of the TN, also at the fascicular level, which agrees with

the finding of significant fascicular branching patterns of peripheral nerves by Kudoh and Sakai (2007).

Therefore, a MRI scanner (Farooki et al. 2001) may be useful to obtain knowledge about the TN branch patterns for a particular case prior to a cuff implantation.

Acknowledgments We thank Mette Bille for her great assistance with embedding and slicing the nerves, and Jens Søndergaard Petersen, MD, for skilful assistance with the dissections. Further, we thank the European Commission, for funding this work through the BIO-MED-2 program BMH-CT96-0897.

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