Innervation Territories of Mechanically Activated C Nociceptor Units in Human Skin

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1Department of Clinical Neurophysiology, University Hospital, S-75185 Uppsala, Sweden; and 2Department of Physiology and Experimental Pathophysiology, University of Erlangen, Nürnberg, Germany

Schmidt, Roland, Martin Schmelz, Matthias Ringkamp, Hermann O. Handwerker, and H. Erik Torebjörk. Innervation territories of mechanically activated C nociceptor units in human skin. J. Neurophysiol. 78: 2641–2648, 1997. Innervation territories of single mechanically activated C nociceptors in the skin of the leg and foot were explored in normal human subjects. Microneurographic recordings were obtained in the peroneal nerve from 70 mechano-heat responsive (CMH) and 7 mechano-(but not heat) responsive (CM) units. Units were identified by their constant long-latency response to intracutaneous electrical stimulation of their terminals. Responsiveness to mechanical, heat, or transcutaneous electrical stimuli was verified by transient slowing of conduction velocity after activation by such stimuli. We determined their thresholds to mechanical stimuli (mean 33.7 mN, median 30 mN, range 3–750 mN) and heat (mean 42.5°C, median 42.5°C, range 37–49°C). Most mechano-receptive fields (mRFs) were found on the foot dorsum (60 units) and some on the lower leg (14 units) and toes (3 units). Most units had one continuous mRF, but 10 units had more complex fields. Areas of mRFs mapped with a von Frey filament (750 mN) ranged from 10 to 363 mm² (mean, 106 mm²). The mRFs were oval or irregularly shaped with greatest diameters ranging from 3 to 45 mm. Mean areas of mRFs were largest on the lower leg (198 mm²), smaller on the foot dorsum (88 mm²), and smallest on the toes (35 mm²). Forty-nine of the 77 units had identical mRFs and electro-receptive fields (eRFs). Twenty-six units had larger eRFs than mRFs, whereas the opposite was found for two units only. Areas of eRFs ranged from 16 to 511 mm² (mean 121 mm²). An estimate of the innervation density based on the present data and the presumed number of C fibers in cutaneous fascicles of the peroneal nerve suggests a considerable overlap of nociceptive endings in the skin. Such overlapping nociceptor innervation in the skin allows for substantial spatial summation in response to punctate noxious stimuli, which may be a prerequisite for high accuracy in localization of painful events from a C-fiber input. The reduction in size of innervation territories distally allows for finer discrimination of spatial dimensions of noxious stimuli distally as compared with proximal regions of the extremities. Mean maximal diameters of the mechano-receptive fields of CMH and CM units on the lower leg (22.3 mm) and foot (15.3 mm) are of similar size as the radius of axon reflex flares evoked by noxious mechanical stimuli in these regions.

INTRODUCTION

In the late 19th century Max von Frey discovered in human skin discrete pain sensitive spots, the ‘‘pain points’’ (Schmerzpunkte) when probing the skin at threshold intensity for sensory detection with extremely fine sharpened needles or with thin spicules from plants. This discovery was the basis for distinguishing a sense of pain from other sensory modalities of the skin (von Frey 1894, 1896, 1922). Stru falsone of von Frey’s pupils, found approximately 150–200 pain points/cm² on the lower arm and leg and also on the back of the hand and of the foot (Stru Hold 1924). He also reported that the number of pain points increased when increasing the force of the spicules from 1 to 2 g, implying that the definition of a pain point is highly dependent on the stimulus used, and that there is a very dense representation for pain in the skin.

Decades later, with the advent of powerful recording techniques, recordings from single nociceptive afferents were achieved, in particular from polymodal C-fiber units, first in animal experiments, and then, after the development of microneurography, also from human volunteers (Torebjörk and Hallin 1970). It soon became clear that these nociceptor units do not form single sensory spots in the skin, but have extended innervation territories (receptive fields), often comprising several particularly sensitive spots (Torebjörk 1974; van Hees and Gybels 1981). In parallel, histological studies revealed that thin afferent nerve fibers show extended terminal branching and hence cover complex innervation territories (Kruger 1996; Messlinger 1996). Furthermore, even the ‘‘naked nerve endings’’ of individual branches are by no means uniform structures. They are characterized by multiple transduction sites, the ‘‘sensory beads’’ that are distributed over the whole terminal axon separated by nonreceptive membrane segments covered by Schwann cells (Messlinger 1996). The complex geometry of these sensory beads may perhaps be regarded as a structural substrate for the sensitive spots within the receptive fields of nociceptor units. However, not the individual transduction site but the whole innervation territory of individual nociceptors is the functional unit which is important not only for the spatial resolution of the nociceptive system, but also for the efferent functions of the nociceptor units, in particular the axon reflex mechanisms.

The size and shape of the receptive fields of nociceptor units in human skin have not been studied extensively. Van Hees and Gybels (1981) reported that 33 polymodal C nociceptors on the dorsum of the hand (recorded from the radial nerve) had circular or elliptical receptive fields with mean axes of 6 and 7 mm. The largest receptive field was 10 × 15 mm and the smallest 2 × 3 mm. There are only anecdotal reports on the dimensions of receptive fields of polymodal C-nociceptor units (recorded in the peroneal nerve) on the skin of the lower leg and foot. In one study it was stated...
that the extensions of the receptive fields varied from 1 × 1 to 6 × 17 mm (Torebjörk 1974). In another study (Jörnum et al. 1989) a mean area of the receptive fields was reported to be 57 ± 26 (SD) mm². No descriptions of the receptive fields in different regions were given. Here we describe for the first time the detailed organization of the innervation territories of a representative sample of mechano-responsive (CM) and mechano-heat responsive (CMH) C-nociceptor units in the skin of the lower leg, foot, and toes in humans.

METHODS

Subjects

The experiments were performed in 37 healthy human subjects, 31 men and 6 women (19–36 yr). All subjects gave their informed consent and the study was approved by the ethics committees of the Universities of Uppsala and Erlangen, Nürnberg.

Search for and identification of C fibers

Microneurography techniques were employed for recording from cutaneous C fibers in the peroneal nerve, innervating the skin on the lower leg and the dorsum of the foot and toes. The search procedure for single C fibers and the mapping of their receptive fields has been described before in detail (Schmelz et al. 1994, 1995; Schmidt et al. 1995; Torebjörk 1974). Briefly, a tungsten microelectrode was inserted into a skin fascicle of the peroneal nerve at knee or ankle level. The innervation territory of the impaled fascicle was identified by multifiber responses of large mechano-responsive fibers to the touching or scratching of the skin. Transcutaneous electrical stimuli from insulated stimulators (0.3 Hz, 0.3 ms, 40–100 V constant voltage pulses, Grass S88 stimulator, or 5–10 mA constant current pulses, World Precision Instruments, A 360) were applied to the skin within the innervation territory of the impaled fascicle (lower leg or dorsum of foot and toes) via a pointed steel electrode (tip area 1 mm²), until a C-fiber response was obtained. Two tungsten microelectrodes (0.2 mm diam) were inserted intracutaneously ~5–8 mm apart at the skin site from where the C-fiber response had been elicited. Pulses of moderate intensity (0.3 ms and 4–20 V) suprathreshold for C-fiber activation were then applied through these needles at regular intervals of 4 s throughout the experiment. The C-unit responses to the intracutaneous electrical stimulation were recorded on-line by a PC computer via a interface card (DAP, Microstar) by using the SPIKE/SPIDI software package (Forster and Handwerker 1990). Traces triggered by the intracutaneous pulses were displayed successively from top to bottom on the computer screen. C units were identified by their constant long latency in response to regular electrical stimulation as in previous studies (Schmelz et al. 1994, 1995; Schmidt et al. 1995). Responsiveness to additional natural or transcutaneous electrical stimuli was detected by a transient increase in latency resulting from the slowing of conduction velocity after extra impulses in the activated C unit. It has been proven in a previous study (Schmelz et al. 1995) that this ‘‘marking’’ method is sensitive enough to allow detection of just one extra impulse in a stimulated C fiber. This means that this powerful technique is useful for classification of C units, for threshold determinations, and for mapping the extensions of the innervation territories. Because of the long conduction distances (20–50 cm) slight differences in conduction velocities caused latency separation of multiple C fibers recorded in one intraneural site. Furthermore, with the marking method there was never any ambiguity as to which unit responded to a particular stimulus, even if two or more units had adjacent or even overlapping innervation territories.

A schematic illustration of the marking technique, used for mapping the mechano-receptive fields (mRFs) and the electro-receptive fields (eRFs) of C-afferent units is shown in Fig. 1.

Mapping of mechano-receptive fields

The extension of the (mRF) was mapped as described before (Schmelz et al. 1994, 1995). For this purpose the tip of a pointed steel electrode was moistened with sodium chloride or electrode gel and gently contacted to the skin for unipolar electrical stimulation, useful for classifying C units, for threshold determinations, and for detection of just one extra impulse in a stimulated C fiber. This means that this powerful technique is sensitive enough to allow detection of just one extra impulse in a stimulated C fiber. This means that this powerful technique is useful for classification of C units, for threshold determinations, and for mapping the extensions of the innervation territories. Because of the long conduction distances (20–50 cm) slight differences in conduction velocities caused latency separation of multiple C fibers recorded in one intraneural site. Furthermore, with the marking method there was never any ambiguity as to which unit responded to a particular stimulus, even if two or more units had adjacent or even overlapping innervation territories.

FIG. 1. Schematic illustration of the technique used for mapping the mechano-receptive field (mRF) and electro-receptive field (eRF). A: (○), responsiveness to mechanical stimuli on skin with a stiff von Frey filament and responsiveness to transcutaneous electrical stimulation (black region). (●), responsiveness to electrical stimulation only (gray region). (◇), unresponsiveness to mechanical as well as electrical stimulation. Successive single C-unit responses displayed from top to bottom (◇) to transcutaneous electrical stimulation in innervation territory at regular intervals of 4 s (shock artifacts under arrow). Mechanical or electrical stimulation outside innervation territory evoked no response in the unit, as observed by its constant latency (◇, no response). Stimuli within mRF (○) and eRF (●) elicited extra discharges (white potentials on black background) causing latency increases in the unit under study. This proved that the unit was activated by stimuli applied to those particular spots.

Mapping of electro-receptive fields

The extension of the eRF was mapped as described before (Schmelz et al. 1994, 1995). For this purpose the tip of a pointed steel electrode was moistened with sodium chloride or electrode gel and gently contacted to the skin for unipolar electrical stimulation, without mechanically activating the unit under study. A metal plate on the skin >5 cm away was used as reference electrode. The stimulation points were spaced by ~2 mm. A skin area with a radius of at least 2.5 cm around the intracutaneous needle electrodes was searched to assure that the whole mRF was also mapped for units with discontinuous mRFs. Each point from which mechanical stimuli activated the respective C unit was marked on the skin in blue.

After the experiments, the marks were transferred to a transpar-
ent sheet and the differently marked mRF and eRF of each unit were analyzed after scanning the drawing into computer and determining the areas by a suitable computer program. The maximal diameter, the diameter perpendicular to the maximal diameter, extent in longitudinal direction, and extent in transverse direction were also measured.

**Mechanical thresholds**

Mechanical thresholds were obtained by testing sensitivity to different von Frey filaments within the mRF. The whole receptive field was checked and the lowest threshold was noted. A detailed analysis of different thresholds which might occur inside the borders of the receptive fields (Torebjörk 1974) was not performed. In the present paper, only afferent C fibers with a mechanical threshold of <0.75 N are included. The receptive field organization of other types of C fibers will be described elsewhere (for classification of C-nociceptor units in human skin, cf. Schmidt et al. 1995).

**Heat thresholds**

For characterizing units as CMH, i.e., mechano-heat responsive or as CM, i.e., mechano-responsive but not heat responsive (Schmidt et al. 1995), heat stimuli were delivered from a halogen lamp feedback controlled from a thermocouple attached to the skin (Beck et al. 1974). Skin temperature was increased by 0.25°C/s, from an adapting temperature of 31°C to tolerance level of about 50°C. The temperature leading to activation of the unit was noted. Because the electrical stimuli were delivered every 4th s, a response to heating could possibly be overestimated by 0–1°C.

**Location of units**

To determine the location of units the cutaneous innervation territory of the peroneal nerve was divided into three parts: the dorsum of the toes except the ungual phalanges and the lateral part of the little toe; the dorsum of the foot up to the malleoli; and the distal lateral half of the lower leg. The upper half of the lower leg was covered by the preamplifier equipment and could not be used for recording. Each mechano-afferent unit was classified as belonging to one of these skin areas. The sizes of these three parts of the leg were estimated in one female and two male subjects (162, 181, and 192 cm) by laying a transparent plastic foil over the skin and drawing the approximate outlines of the peroneal nerve innervated territory on the transparency and measuring the areas by an appropriate computer program.

**Pain locognosia test**

The ability of six healthy male subjects to locate painful punctate mechanical stimuli on the dorsum of the foot and on the lateral part was determined under conditions of intact nerve fiber conduction and during ischemic A fiber block when only C fibers were conducting. The experiment was modified from a method introduced by Nordenbos (1972) and employed by others (Hamburger 1980; Jörum et al. 1989; Koltzenburg et al. 1993; Ochoa and Torebjörk 1989; Schady et al. 1983). The subject had his eyes closed and a moderately sharp Babinsky needle was pressed against the skin on the dorsum of the foot or on the lateral part of lower leg until the subject experienced pain. The contact spot was marked with red ink. The subject wore red goggles so he was unable to see the red mark on the skin. He opened his eyes after each stimulus and marked with black ink the spot where he thought he had been stimulated. The distance between the red and black spots was measured to the nearest millimeter. On the other leg a sphygmomanometer cuff was inflated well above systolic blood pressure around the calf 5-cm distal to the fibular head. After approximately 30 min of ischemia, when the subject had lost the capacity to perceive touch (A β-fiber block) and cold (A δ-fiber block), but could still perceive warmth and delayed pain (intact conduction in C fibers), a similar test procedure was performed as on the control leg. This locognosia test was repeated 10 times at the lower leg and 10 times at the dorsum of the foot both in the control leg and in the ischemic leg when C fibers only were conducting.

**RESULTS**

**Sample of C units**

Seventy-seven mechanically activated C nociceptor units were recorded from the peroneal nerve, 74 units at knee level, and 3 units at ankle level. Sixty-seven units were recorded from male and 10 from female subjects.

**Location of C units**

The location of the innervation territories of the 77 C units is schematically shown in Fig. 2. Of the 74 units recorded at knee level, 14 were located laterally on the lower leg, 57 on the dorsum of the foot, and 3 on the dorsum of toes. All three units recorded from the superficial peroneal nerve at ankle level were located on the dorsum of the foot. As seen in Fig. 2, several C units were found on the medial part of the foot dorsum, outside of the “conventional” cutaneous innervation territory of the peroneal nerve as often shown in textbooks of neuroanatomy. Of the units recorded in women, eight were found on the foot and two on the lower leg. Thus the distributions were similar in men and women. Thirty-seven of the units were recorded from hairy skin areas and 40 from hairless skin. All units in hairless skin areas were situated on the dorsum of the foot or on the toes.
Mechanical and heat thresholds

The mean mechanical threshold was 33.7 mN (median, 30 mN; range, 3.4–750 mN). The mechanical thresholds were not significantly related to the size of the mRFs (Spearman rank correlation, $n = 73, R = -0.06, P = 0.63$), to the location on leg or foot (Kruskall-Wallis analysis of variance (ANOVA), $P = 0.82$), or to the presence or absence of hair on skin (Mann Whitney $U$ test, $P = 1$). Seventy units were classified as CMH units, responding to heating up to 50°C. The mean heat threshold for 65 heat-sensitive units for which an exact threshold could be obtained was 42.5°C (median, 42.5°C; range, 37–49°C), again without significant relation to receptive field size (Spearman rank correlation, $R = 0.05, P = 0.7$), location on leg or foot (Kruskall Wallis ANOVA, $P = 0.9$), or location in hairy vs. hairless skin (Mann Whitney $U$-test, $P = 0.27$). The remaining seven units were heat insensitive and classified as CM. As in a previous study (Schmidt et al. 1995), no difference was observed in mechanical thresholds between CM and CMH units (Mann Whitney $U$ test, $P = 0.27$). A weak relation between mechanical and heat thresholds for the CMH units was suggested by nonparametric tests (Spearman rank correlation, $n = 62, R = 0.26, P = 0.044$), implying that a small part of the variability in thresholds might be the result of a common factor such as receptor depth. However, simple product-moment correlation between mechanical and heat thresholds (Pearson correlation, $r = 0.18$) was not significant. The distribution of mechanical versus heat thresholds for the CMH units is shown in Fig. 3.

Conduction velocities

The conduction velocities of the 74 units recorded at knee level were on average 0.92 m/s (range, 1.15–0.55 m/s). There was no significant correlation between size of the mRFs and conduction velocity in the C-afferent fibers (partial correlation coefficient, 0.06, $P = 0.61$). However, slower conduction velocities were encountered at shorter distances between receptive field and recording site (partial correlation coefficient 0.27, $P < 0.02$). Similar relation to recording distance has been previously shown (Schmelz et al. 1995), indicating tapering of C axon diameter toward the nerve endings.

Shape of receptive fields

The borders of the receptive fields were very distinct. Thus, moving the mapping instruments just 1–2 mm outside the border of a receptive zone failed to elicit any response. The size and shape of the receptive fields varied considerably, as seen in Fig. 4. In this figure black zones represent mRFs and gray zones represent further extensions detected by electrical transcutaneous stimulation (gray). Top: fields on proximal and distal parts of the lower leg. Middle: fields on proximal, middle, and distal parts of the dorsum of the foot. Bottom: fields on the toes. Fields are oriented in figure according to the longitudinal axis of leg and foot (distal = down) and fields recorded from the left side have been mirrored with respect to the vertical axis and displayed with lateral side to the left. Encircled fields belong to same parent axon.
TABLE 1. Areas of CMH and CM innervation territories

<table>
<thead>
<tr>
<th>Unit Location</th>
<th>Number of Units</th>
<th>m/eRF</th>
<th>Mean</th>
<th>Median</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leg</td>
<td>14</td>
<td>mRF</td>
<td>198</td>
<td>183</td>
<td>94–363</td>
</tr>
<tr>
<td></td>
<td></td>
<td>eRF</td>
<td>213</td>
<td>183</td>
<td>100–511</td>
</tr>
<tr>
<td>Foot</td>
<td>60</td>
<td>mRF</td>
<td>88</td>
<td>83</td>
<td>10–212</td>
</tr>
<tr>
<td></td>
<td></td>
<td>eRF</td>
<td>103</td>
<td>96</td>
<td>35–295</td>
</tr>
<tr>
<td>Toes</td>
<td>3</td>
<td>mRF</td>
<td>35</td>
<td>42</td>
<td>16–46</td>
</tr>
<tr>
<td></td>
<td></td>
<td>eRF</td>
<td>41</td>
<td>46</td>
<td>16–60</td>
</tr>
<tr>
<td>All regions</td>
<td>77</td>
<td>mRF</td>
<td>106</td>
<td>94</td>
<td>10–363</td>
</tr>
<tr>
<td></td>
<td></td>
<td>eRF</td>
<td>121</td>
<td>100</td>
<td>16–511</td>
</tr>
</tbody>
</table>

Values are in mm$^2$. CMH, mechano-heat responsive; CM, mechano-responsive; m/eRF, mechano- or electro-receptive field.

(8 on the dorsum of the foot, 2 on the lower leg, Fisher’s exact test: lower leg, foot, and toes, $P = 0.98$).

Mechano-receptive fields

As seen in Table 1, the mean mRF area for the whole unit population was 106 mm$^2$ (median, 94 mm$^2$). The size of the mRFs of the units varied with skin region, being largest on the lower leg (mean, 198 mm$^2$; range, 94–363 mm$^2$), smaller on the dorsum of the foot (88 mm$^2$, 10–212 mm$^2$), and smallest on the toes (35 mm$^2$, 16–46 mm$^2$). The difference between mRFs of units in different skin regions was significant (Kruskall-Wallis ANOVA, $P < 0.0001$). Post hoc tests (Mann-Whitney $U$ test) revealed that there was a significant difference between lower leg and foot ($P < 0.0001$), between lower leg and toes ($P = 0.008$), and between foot dorsum and toes ($P = 0.011$). These differences remain significant after correction for multiple comparisons.

Figure 5 shows the areas of the 70 mRFs for units recorded at knee level related to distance between innervation territory and recording site. The areas of the receptive fields are significantly smaller for the longer distances (i.e., distally) (Spearman rank correlation, $R = -0.38$; $P < 0.001$).

As can be seen from Table 2, the mRFs usually were not very elongated. When mRFs were elongated, the direction of the largest diameter was more or less random; i.e., there was no clear tendency for the units to be elongated in either the longitudinal or the transverse direction. The maximal diameter of mRFs had a range of 3.0–45 mm. The mean maximal diameter was 22.3 mm in the lower leg, 15.3 mm on the foot dorsum, and 7.7 mm on the toes.

Electro-receptive fields

Forty-nine of the 77 units had identical, completely overlapping mechano-receptive and electro-receptive innervation territories (Fig. 6). Twenty-six units had larger eRFs than mRFs. The contrary, i.e., a slightly larger mRF than eRF, was observed for two units. Such millimetric mecanopositive extensions were found at the border of the eRFs and may have been caused by stretching the skin by the fairly stiff von Frey filament. As detailed in Fig. 4, the extended innervation territories detected by electrical stimulation sometimes filled in gaps in the irregular contours of mRFs or made scattered mRFs more homogenous, but distinct eRFs separate from the mRFs were also found. The expansion of the innervation territory detected by electrical stimulation was considerable in a few units, but in most instances the eRF only indicated a marginal expansion of the mRF. As shown in Table 1, the mean area of the eRFs was 121 mm$^2$ (median 100 mm$^2$), again being largest in the lower leg (mean, 213 mm$^2$; range, 100–511 mm$^2$), smaller on the foot (103 mm$^2$, 35–295 mm$^2$) and smallest on the toes (41 mm$^2$, 16–60 mm$^2$). The difference between eRFs of units in different skin regions was significant (Kruskall-Wallis ANOVA, $P < 0.0001$). Post hoc tests (Mann-Whitney) revealed that the difference was significant between lower leg and foot ($P < 0.0001$), between lower leg and toes ($P = 0.008$), and also between foot dorsum and toes ($P = 0.012$). These differences remain significant after correction for multiple comparisons.

Pain locognosia test

The error in localizing a punctate painful mechanical stimulus from a Babinsky needle was 16 ± 2 mm in the lower leg and 13 ± 3 mm on the foot. During A-fiber block, when C fibers only were conducting, the localization error was 22 ± 2 mm in the lower leg and 20 ± 5 mm on the foot. The error was significantly greater in the leg as compared with the foot and during A-fiber block as compared with the unblocked state (2-way ANOVA repeated measures $P < 0.01, P < 0.01$).

Discussion

The most conspicuous finding of this study was the gradient in size of innervation territories for CM and CMH nociceptors, with significantly smaller fields on toes and foot as compared with the lower leg. This finding was consistent, regardless of the mapping procedure with mechanical or electrical stimuli. In this respect the afferent C fibers, subserving sensory functions, seem to have a different terminal organization from the sympathetic fibers innervating the same skin regions. Thus, although the mean eRFs of the afferent C fibers (121 mm$^2$) and the mean eRFs of sympathetic fibers (126 mm$^2$) were very similar, there was no pattern of the sympathetic fibers having smaller innervation territories on toes and foot as compared with the lower leg.

Another feature was the observation of much larger mRFs in this study on human CMH and CM units (mean, 106 mm$^2$) as compared with the mRFs of mechano-heat sensitive
C units in the arm and leg of the monkey (mean, 14 mm², as reported by Treede et al. 1990 or 17 mm² as reported by Davis et al. 1993). This is perhaps not surprising, considering the difference in size between humans and Macaca fascicularis and can hardly be explained by the use of a supra-threshold von Frey filament in our study. In other respects the receptive properties were quite similar. Thus, the mean filament of 750 mN may have given the impression that the

Mean diameters in mm; numbers in brackets are ranges.

C in humans, similar

TABLE 2. Shape of mechano-receptive fields

<table>
<thead>
<tr>
<th>Unit Location</th>
<th>Number of Units</th>
<th>Maximal Diam</th>
<th>Perpendicular to Max Diam</th>
<th>Transverse</th>
<th>Longitudinal</th>
</tr>
</thead>
<tbody>
<tr>
<td>Leg</td>
<td>14</td>
<td>22.3 (14–39)</td>
<td>14.9 (10–23)</td>
<td>19.6 (12–38)</td>
<td>17.4 (11–26)</td>
</tr>
<tr>
<td>Foot</td>
<td>60</td>
<td>15.3 (3–45)</td>
<td>10.3 (2–18)</td>
<td>12.9 (3–45)</td>
<td>12.6 (2–24)</td>
</tr>
<tr>
<td>Toes</td>
<td>3</td>
<td>7.7 (5–9)</td>
<td>7.0 (4–9)</td>
<td>7.3 (5–9)</td>
<td>6.7 (4–8)</td>
</tr>
<tr>
<td>All regions</td>
<td>77</td>
<td>16.3 (3–45)</td>
<td>11.0 (2–23)</td>
<td>13.9 (3–45)</td>
<td>13.2 (2–26)</td>
</tr>
</tbody>
</table>

FIG. 6. Relation between mRFs and eRFs. Most of the fields were identical in area and shape (49 units). Twenty-six units had larger eRFs than mRFs. Two units had marginally larger mRFs than eRFs.

FIG. 7. Mean mechanical thresholds of 24 ± 30 mN in the monkey, they would appear if weaker filaments had been used for the receptive properties were quite similar. Thus, the mean filament of 750 mN may have given the impression that the

suggested that these extensions really are parts of the terminal innervation territory of the studied unit and do not represent an artifact induced by the transcutaneous electrical stimulation. This validates the transcutaneous electrical stimulation procedure as a useful tool for mapping the skin innervation territories of afferent silent C units, which are unresponsive to mechanical and heat stimuli (Schmidt et al. 1995) and for mapping the cutaneous innervation territories of sympathetic C units, which are not activated directly by natural stimuli applied to the skin. It is obvious, however, that any kind of transcutaneous stimulation procedure (mechanical, thermal, or electrical) will not detect deep nerve endings that are out of reach of the stimulus and hence are not activated.
Over the last 3 years we have sampled 324 afferent and sympathetic C units. Of these, ~60% were classified as CMH and CM units (Schmidt et al. 1995; Torebjörk et al. 1996). It is tempting to use these figures for a rough estimate of the innervation density of CM and CMH units in the leg and foot in humans. Let us assume that the cutaneous fascicles of the peroneal nerve have a similar C fiber density as the sural nerve, i.e., 20,000–40,000 C fibers/mm² fascicular area (mean 30,000) (Johnson et al. 1994; Ochoa and Mair 1969). According to Sunderland (1978) the total fascicular area of the peroneal nerve at knee level ranges from 2.6 to 5.7 mm², with a mean of 4.0 mm² (n = 11). L. Eriksson (unpublished observations) has recently measured the fascicular cross-sectional area of the peroneal nerve at knee level in three human corpses and found a range of 3.4–4.5 mm², mean 4.0 mm². From the estimates of Sunderland (1978), ~40% of the cross-sectional area of the peroneal nerve at the neck of the fibula consists of fascicles which innervate skin regions that are relevant for our recordings. Thus, assuming a mean cutaneous fascicular area of 1.6 mm² and a mean C-fiber density of 30,000/mm², the mean total number of unmyelinated fibers would be on the order of 50,000 in the relevant, cutaneous part of the peroneal nerve. CMH and CM units would constitute about 60% of these (Torebjörk et al. 1996), i.e., ~30,000 fibers. The cutaneous innervation territory of this nerve, from where we can obtain recordings, covers an area of ~200 cm², giving a mean innervation density on the order of 150 units/cm². However, our data indicate that there are considerable differences in regional densities. Of the 74 units recorded in the peroneal nerve at knee level, 14 (19%) were found on the lower leg, 57 (77%) on the dorsum of the foot, and 3 (4%) on the toes, thus implying that the distribution of units was significantly unequal (χ² test, P < 0.0001). Because these skin areas become smaller from proximal to distal by a factor of 4.4:4.3:1, the average innervation density can be calculated to 65 units/cm² on the lower leg, 200 units/cm² on the dorsum of the foot, and 60 units/cm² on the toes. If we make the conservative assumptions instead that the relevant fascicular area is 1 mm², that the C-fiber density is 20,000/mm², that the proportion of C-mechano-nociceptor fibers is 50% and that the innervation area is 300 cm², the mean innervation density will then be on the order of 33 units/cm² (14 units/cm² on the leg, 57 units/cm² on the foot, and 13 units/cm² on the toes). Although these rough estimates reflect a denser innervation on the foot than on the lower leg, it is also apparent that a sampling error probably exists such that units from the toes may be underrepresented, possibly because of difficulties in finding and mapping units on the small and curved surfaces of the toes. Similar estimates on the innervation density of C-polymodal nociceptors with small (1–2 mm diam) receptive fields in rat skin has yielded figures of 2 units/mm² on the hind leg and 8 units/mm² on the foot (Lynn and Baranowski 1987), suggesting an increase in nociceptor innervation density distally in the extremity, in concordance with our observations in humans. It may appear that the large innervation territories observed for human C nociceptors allows for lesser density of receptive fields in human as compared with rat skin, which according to the data of Lynn and Baranowski (1987) would be on the order of 200–800 units/cm². However, considering the gross approximations inherent in the calculations of innervation densities, it is not surprising if the results obtained under different assumptions and for different species would differ considerably.

The mean unit overlap, or number of layers of units at a given skin site can be calculated as the mean area of all CM and CMH unitary innervation territories, in a skin area divided by the size of that particular skin area. By this computation and by using the assumptions listed above, we come up with ~50–230 layers on the dorsum of the foot and 27–125 layers on the lower leg. Regardless of the low precision of our estimations, the data show that the skin has a dense innervation of multiple layers of nociceptive endings. This overlapping innervation, allowing for substantial spatial summation in response to noxious stimuli, together with a somatotopic organization for noxious events throughout the CNS up to the primary somatosensory cortex (Andersson et al. 1997) is a prerequisite for a fairly high accuracy in localizing painful events from a pure C-fiber input, both from the lower leg and the foot, as shown here, and from the foot and hand, as shown previously (Jürum et al. 1989, Kolzenburg et al. 1993; Ochoa and Torebjörk 1989). In addition, the reduction in size of the innervation territories distally will allow for finer discrimination of spatial dimensions of noxious stimuli distally in the extremities as compared with more central regions of the body.

It would be of interest to discuss the sizes of the innervation territories in relation to the flare sizes in response to punctate painful mechanical stimuli applied to the dorsum of the foot and the lower leg. It appears that the mechanically induced flare is weaker and smaller than the flare induced for instance by histamine or capsaicin, indicating different mechanisms of vasodilatation. Furthermore, preliminary unpublished data from our group suggest that the flare size, measured as Laser-Doppler increase in blood fluxes after punctate mechanical noxious stimulation of the skin, has a mean radius on the order of 25 mm on the lateral side of the lower leg and 20 mm on the dorsum of the foot. These observations are compatible with the mean maximal diameters of the mRFs documented in this study (23.3 mm on the lower leg and 15.5 mm on the foot). If the axon reflex underlying the flare also involves “silent branches” detected by transcutaneous electrical stimulation, the average innervation territories of CM and CMH nociceptor units would be larger and would fit quite well with the areas of the flare responses to mechanical stimuli.

It is an interesting issue for future research to explore as to whether or not axon reflex flares are induced differentially by different classes of cutaneous nociceptors, as has been suggested from observations on flare reactions induced specifically by activation of C-heat nociceptors in the skin of the pig (Lynn et al. 1996).

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