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REVIEW | *50 Years of Microneurography: Insights into Neural Mechanisms in Humans*

Microneurography: how it started and how it works

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Vallbo ÅB. Microneurography: how it started and how it works. *J Neurophysiol* 120: 1415–1427, 2018. First published June 20, 2018; doi:10.1152/jn.00933.2017.—In the first section, this historical review describes endeavors to develop the method for recording normal nerve impulse traffic in humans, designated microneurography. The method was developed at the Department of Clinical Neurophysiology of the Academic Hospital in Uppsala, Sweden. Microneurography involves the impalement of a peripheral nerve with a tungsten needle electrode. Electrode position is adjusted by hand until the activity of interest is discriminated. Nothing similar had previously been tried in animal preparations, and thus the large number of preceding studies that recorded afferent activity in other mammals did not offer pertinent methodological guidance. For 2 years, the two scientists involved in the research impaled their own nerves with electrodes to test various kinds of needles and explore different neural systems, all the while carefully watching for signs of nerve damage. Temporary paresthesiae were common, whereas enduring sequelae never followed. Single-unit impulse trains could be discriminated, even those originating from unmyelinated fibers. An explanation for the discrimination of unitary impulses using a coarse electrode is inferred based on the electrical characteristics of the electrode placed in the flesh and the impulse shapes, as discussed in the second section of this paper. Microneurography and the microstimulation of single afferents, combined with psychophysical methods and behavioral tests, have generated new knowledge particularly regarding four neural systems, namely the proprioceptive system, the cutaneous mechanoreceptive system, the cutaneous nociceptive system, and the sympathetic efferent system to skin structures and muscular blood vessels. Examples of achievements based on microneurography are presented in the final section.

afferent impulses; human; microneurography; peripheral nerves; sensory mechanisms

INTRODUCTION

A prominent branch of neuroscience in the decades following the end of the second world war was the functional analyses of cutaneous and proprioceptive sense organs in nonhuman mammals, done by recording from single afferents in the dorsal roots or peripheral nerves. Accurate descriptions of the functional properties of sense organs were provided, allowing for convincing distinctions to be made between unit types. For instance, Iggo and Muir (1969) and Chambers et al. (1972) identified two kinds of slowly adapting mechanoreceptors in the skin, type I (SAI) and type II (SAII) afferents, and demonstrated their connection with Merkel and Ruffini endings. Successively, a fairly comprehensive description emerged of the various mechanoreceptive sense organs in the mammalian skin, due to contributions by several groups (Iggo 1974). Analyses of psychoneural relationships by Mountcastle et al. (1967) and Talbot et al. (1968) attracted great interest. Collateral analyses of psychophysical and neural responses to identical tactile stimuli, in humans and monkeys, strongly suggested that fundamental elements of human percepts may already be set at the level of the primary afferents (Mountcastle 1967; Talbot et al. 1968). The functional role of muscle spindles and the fusimotor system was another popular research area at the time. The complexity of the fusimotor system was demonstrated in elegant experiments by Matthews (1972) at Oxford. In addition, the role of the γ-system was widely discussed in relation to the control of self-generated movements. Our interest in trying to record from human nerves was strongly stimulated by the exciting achievements of these studies of the tactile and the fusimotor systems.

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The first section of this paper describes the process undertaken to establish the method for recording from human nerves. The second section discusses which factors are likely essential for single-unit discrimination using an electrode with a naked metal tip that is several times larger than the nerve fiber. In the third section, the potential of the method is illustrated by microneurography and microstimulation studies of various neural systems.

THE MICRONEUROGRAPHY PROCEDURE

Microneurography is, in principle, a very simple technique but in practice is quite demanding. In this method, a needle electrode is inserted percutaneously towards a nerve in a limb or the face, with the electrode freely floating in the flesh. Skillful manipulation is required, first, to impale the nerve fascicle and, second, to discriminate impulse trains in a single nerve fiber. All adjustments of electrode position are done by hand. This is possible because movements of the electrode tip at the insertion depth are much smaller than movements at the surface. An essential factor here is the high friction between the flesh and electrode shaft. Searching for neural activity can be aided by electrical stimulation down the electrode to assess its position in relation to the nerve and target fascicle. Anesthetics are not required during microneurography, as pain, if felt at all, is only minor-to-moderate and short-lasting. Hence, subjects are attentive during the experiment and are able to cooperate in psychophysical and psychomotor tests while nerve signals are being recorded. In addition to the recording of impulses, electrical stimulation through the recording electrode using repetitive pulses, i.e., intraneural microstimulation, may be used to explore the perceptual effects of impulse trains in an identified single afferent nerve fiber.

WHERE AND WHEN

The development of microneurography began in 1965 at the University Hospital in Uppsala, Sweden, in the Department of Clinical Neurophysiology. The technique was developed by two scientists, Karl-Erik Hagbarth and Åke Vallbo, who were both medical doctors and had received their PhDs and basic scientific training at the Nobel Institute of Neurophysiology at the Karolinska Institute in Stockholm, headed by Ragnar Granit. Hagbarth was an established scientist who had published important analyses of cutaneous reflex mechanisms in humans (Hagbarth and Kugelberg 1958). As head of the department, he was responsible for its administration and clinical routines, mainly consisting of electroencephalography and electromyography testing of neurological patients. Åke Vallbo joined the laboratory in 1965, soon after finishing his MD and PhD education, to focus on scientific matters.

The term microneurography was coined by Zotterman (1939), a physiology professor in Stockholm. He is known as the first to record impulses from unmyelinated nerve fibers, building on his cooperation with Edgar Adrian, who was awarded the Nobel Prize for demonstrating that messages carried by nerve fibers consist of a series of pulses of uniform size. We are grateful to Zotterman for his impactive innovation, which he dropped in passing during informal chatting. Much to our surprise, the term microneurography has come to be reserved for the technique of recording impulses in human nerves using percutaneously inserted needle electrodes.

It may seem trivial to point out that the past technology, as well as the issues and concepts discussed 50 years ago, were fundamentally different from those in our current times. On the other hand, it is not trivial, when looking at the historical aspects of science, to appreciate how scientific thinking and action were limited by the technical realities and state of knowledge at the time. Fifty years ago, functional brain imaging was more or less science fiction. Computers were rare in neurophysiology laboratories. They were slow and cumbersome to handle and thus were used by only a few groups for data collection and analysis of impulse trains. In contrast to modern times, most laboratories relied on analog technology. Impulse trains were usually displayed online on an oscilloscope screen for primary evaluation during the experiment and then stored on analog tape. Recordings were photographed for offline analysis, using a running film in a Grass camera facing the oscilloscope screen, while the speed of the film accounted for the time base. Such technology was present in the laboratory where our initial attempts to visualize afferent activity in human nerves were made. The equipment was designed primarily for clinical testing of neurological patients. It was used daily for electromyography and conduction velocity measurements and was only accessible for research purposes out of hours.

INITIAL STEPS

When I first joined the laboratory, Hagbarth told me that he had tested needle electrodes in his own ulnar and median nerves. Using maximal amplifier gain, he had occasionally noticed, through the loudspeaker, barely perceivable changes in the background noise when stroking the skin. He posited that the noise modulations might be of neural origin rather than electrical artifacts. However, the putative nerve activity was so small and capricious that he was not encouraged to spend further effort in this direction.

We decided to start with an approach that seemed safer and technically less demanding than inserting needle electrodes into nerves. Like many other scientists interested in motor control, we were particularly intrigued by muscle spindles and the role of the complex fusimotor system in natural movements. Our experimental ideas were inspired by an influential hypothesis at the time, the follow-up-length servo hypothesis, which proposed that α-motor neurons are not always excited directly from supraspinal centers but indirectly via the stretch reflex loop (Merton 1953). This idea invited imaginative speculation that the γ-system may modulate spindle sensitivity in relation to motor intention, learning, and mental imaging of voluntary movements.

Our experimental plan was to use a juxtaneural electrode to record a compound action potential made up of the synchronous firing of spindle primary afferents in response to tendon taps. Our hypothesis was that changes in spindle sensitivity due to γ-activity would appear as changes in the size of the compound afferent volley. We were led to use this approach based on a colleague's recording, using a juxtaneural needle, of a compound action potential from small cutaneous nerves in response to tapping a finger nail (Sears 1959). Moreover, if the afferent volleys in response to a tendon tap were too small to come out in single shots, the technique of averaging could be applied to increase the signal-to-noise ratio. However, our

endeavors failed altogether and subsequent microneurography recordings demonstrated why. The afferent response to tendon taps does not summate to form a compound action potential, simply because the activity is too dispersed in time, in contrast to the response to tapping a nail.

OBSTACLES: TECHNICAL, CONCEPTUAL, AND MEDICAL-ETHICAL

While contemplating the failure to record afferent volleys in response to tendon taps, we decided to return to the intraneural needle approach, hoping that polishing that technique might improve the signal-to-noise ratio. We foresaw a number of problems and obstacles. A prominent concern was whether it would be possible to record nerve signals of reasonable quality. Discussions with more senior scientists were largely discouraging, as they argued that recording with needle electrodes was not only impossible but dangerous as well. Another kind of obstacle was technical; what kind of electrode would be optimal? We started with the assumption that the electrode should be like those used in studies of brain structures in animals. Another obvious obstacle was of a medical-ethical nature. How large was the risk of causing serious damage to the nerve? These obstacles will be discussed in greater detail in the following sections.

TECHNICAL OBSTACLES

To appreciate the technical situation we were facing in the need to produce a suitable needle electrode, it is pertinent to know that the numerous studies of afferents in non-human mammals mainly used the nerve splitting technique. In such an experiment, the skin is typically incised and a filament of the nerve or dorsal root is placed on silver hook electrodes in a pool of paraffin oil. The filament is then successively split and tested until impulses from a single afferent are discriminated. In contrast, recordings in peripheral nerves or dorsal roots by means of metal needle electrodes had not been reported thus far. Hence, the studies of single afferent recording in nonhuman mammals did not offer useful guidance for the design of needle electrodes suited to recording from peripheral nerves.

A unique study from 1960 may illustrate that microneurography seemed far-fetched to many researchers (Hensel and Boman 1960). In this study, the invasive nerve splitting approach was applied to human subjects. Under short-lasting general anesthesia, the skin was opened over the superficial branch of the radial nerve at the wrist, a filament of the nerve was cut, and a paraffin oil pool was arranged, much as was done in animal studies. Excellent single-unit impulse trains were recorded, but the method was not used in subsequent studies.

Although metal needle electrodes had not been used to record from the peripheral nerves of experimental animals, they were commonly used in studies of central structures. Electrodes made of various metals were described in the literature, and their properties were widely discussed. After testing a number of materials, we concluded that tungsten was the optimal choice. One important advantage was tungsten's mechanical properties. Tungsten is hard but semiflexible and nonbrittle. Because of this, even a very delicate tip does not break, but bends, when subjected to lateral forces, minimizing the risk of metal fragments being left in subjects. In contrast, steel is brittle, and platina-iridium and silver are too soft. One modification we tested was the application of a layer of silver or gold on the free tungsten surface, but this did not seem to improve the electrical properties.

In many studies of brain structures in experimental animals, the electrode impedance and size of the naked metal area were widely discussed. Our interpretation was that high impedance and minimal tip size would be essential to optimize selectivity and signal-to-noise ratio. We were quite occupied with this idea for a time and worked accordingly. However, it soon became apparent that tungsten needles with slender taper and delicate tips, as used in studies of brain activity in mammals, could not penetrate human skin without being seriously deformed. We explored the possibility of protecting the tip within a hypodermic needle for insertion through the skin. However, this proved to be a dead end for a number of reasons, besides being a very cumbersome procedure.

Various kinds of needle electrode coatings had been described in the literature, e.g., Insl-X (Hubel 1957), glass, epoxy varnish, and Hostaflon TF (Konietzny and Hensel 1974). We found that Insl-x did not stick well enough to the metal. Hostaflon was highly recommended due to minimal friction with body structures. However, this property was not found to be particularly advantageous in microneurography experiments, in addition to the method of application being cumbersome. Glass-coated platina-iridium electrodes were recommended for their excellent electrical properties, but we never tried them, as we worried that glass fragments might be left in subjects. However, such electrodes were tried by colleagues at the Karolinska Institute in Stockholm, and two studies were published before the researchers stopped recording from human nerves altogether (Freyschuss and Knutsson 1971; Knutsson and Widén 1967). We never discussed electrode technology with our colleagues in Stockholmn nor did we learn why they stopped nerve fiber recording.

After many trials with various kinds of coatings, we finally decided on epoxy. However, this was not ideal because it did not adhere well enough to the tungsten surface to withstand the mechanical strain when the electrode is forced through the skin. Invariably, the epoxy coating was pushed upwards from the tip like a sleeve, leaving a larger free metal area. We tried various measures to overcome this flaw, e.g., applying the coating in numerous very thin layers, varying baking time, or chemically roughing the tungsten surface before coating. However, our attempts failed to solve the problem. Incidentally, the decoating problem remains unsolved today, and epoxy-coated tungsten electrodes fail to maintain high impedance, whether they are custom made or commercially bought; impedance regularly falls to <500 kΩ when inserted through the skin.

At this stage, it seemed that we had to give up the idea of impaling human nerves with electrodes sufficiently delicate to discriminate the impulses of single units, to judge from descriptions in the literature of the technique required for single-unit discrimination in brain and spinal cord recordings. Apparently, the only way forward was to produce much sturdier electrodes that would be able to withstand the mechanical strain without deformation when pushed through the skin. We hoped that the heavy electrodes would at least be useful for multiunit recordings from large afferents. Our hope was supported by sporadic observations of noise modulations when we applied superficial touch or deep pressure within the innerva-

tion zone of the nerve. However, the signal-to-noise ratio was still rather low. Moreover, it was puzzling and frustrating to find that the faint sound modulations indicating nerve activity appeared only occasionally during an experimental session, even when the electrode seemed to be well placed in the nerve. To understand this and other basic aspects of the recording situation, we turned to animal experiments. Tests with microneurography needle electrodes inserted into exposed cat nerves under full visual control clarified one fundamental point: the fact that the penetration of the perineurium of a fascicle is a necessary step to detect neural activity and this is not achieved as easily as expected. It turned out that the needle electrode can actually pass right through the nerve several times without impaling a single fascicle. The structural and physical properties of nerves explain this phenomenon: the perineurium of a fascicle is a very tough, fibrous sheath, whereas the interconnecting tissue between fascicles is very loose. Hence, impalement requires reasonably accurate hit toward the center of the fascicle. Otherwise, the fascicle will roll and slip away from the needle tip, which is not extremely sharp but slightly rounded, with a diameter of 3–5 μm. In other respects, the use of exposed cat nerves did not shed further light on basic aspects of microneurography recordings nor did we find the testing of electrode properties in animal preparations to be very helpful. As such, we continued to use our own nerves to test the various modified electrode designs in an attempt to improve the signal-to-noise ratio. Electrode testing consumed a significant amount of time, particularly as each test involved the tedious procedure of achieving intrafascicular positioning.

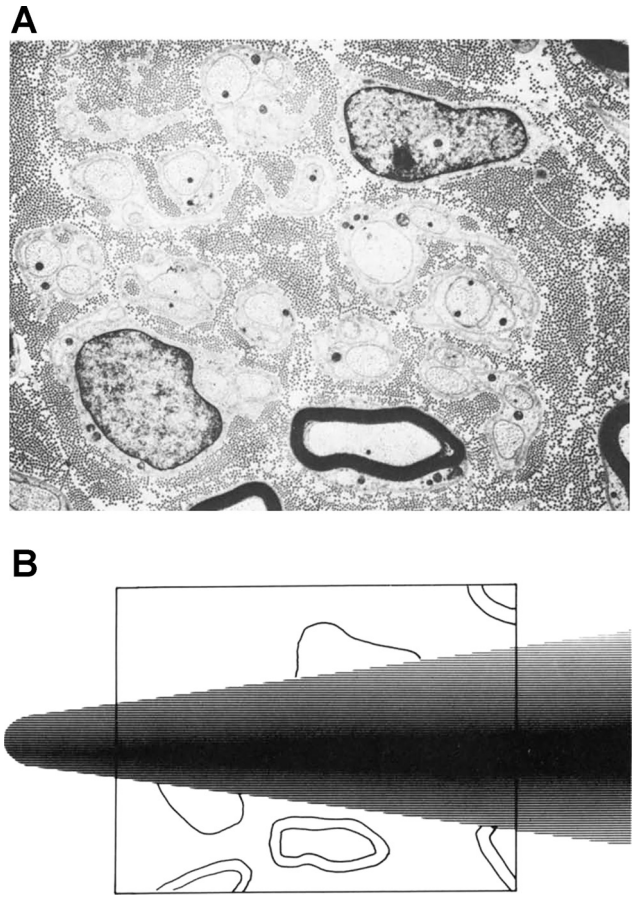

Fig. 1. Size relationships between the microneurography electrode and nerve fibers *A*: cross section of a human sural nerve. *Bottom*: small myelinated nerve fiber and a Schwann cell nucleus. *Top*: another Schwann cell nucleus. Between these large structures, a diagonal band is present that contains a number of unmyelinated nerve fibers scattered among connective tissue fibers. *B*: a scaled-down drawing of the picture in *A,* with a typical microneurography needle electrode superimposed. The black cone represents the naked tungsten area that extends 30 μm from the tip. The diameter of the electrode shaft is 200 μm. [Adapted from Wall and McMahon (1985) with permission from Wolters Kluwer].

CONCEPTUAL OBSTACLES AND UNEXPECTED FINDINGS

One fundamental concern was the feasibility of recording neural activity with high enough quality to answer essential physiological questions. Competent colleagues argued that the extracellular currents produced by nerve fiber impulses would be too small to give rise to measurable potential changes, particularly because resistance in the extracellular space is very low. However, further efforts were encouraged by our recurring experience that faint signals of neural origin in response to tactile stimuli appeared more and more frequently. Moreover, with experience we were able to refine the experimental procedure and determine essential factors for successful electrode manipulation. Various modifications to the electrode design were tried without much success in improving the signal-to-noise ratio, although occasional sessions invited wishful interpretations.

At this stage, we had accepted that the technique would allow for multiunit recording but probably not single-unit discrimination. This seemed reasonable considering the dimensional relationships between microneurography needle electrodes and nerve fibers. Figure 1*A* shows a section of a human sural nerve with one myelinated fiber and a number of unmyelinated fibers. Two Schwann cell nuclei are seen as well. Figure 1*B* shows a scaled-down diagram of the photograph in Fig. 1*A*, with the naked metal section of a typical microneurography needle superimposed. The black cone represents the uncoated region of the electrode, which in this case is 30-μm long. The shaft of the electrode, on the other hand, is 200 μm in diameter. The illustration highlights the disproportional size relationship between the bare tungsten metal area and the nerve fibers. As described previously, the large uncoated area is not a purposeful decision but is the result of decoating due to high friction against the skin and subcutaneous structures. The impedance of a typical electrode is on the order of a few hundred megaohms, as measured with a 1-kHz 20 mV sinusoid using an AC bridge.

While we were collecting multiunit data and testing various electrode modifications, two inconspicuous features attracted our attention during offline inspection of the film strips. These features marked fundamental and unexpected breakthroughs. One was a series of minimal deflections occurring periodically with the regularity and time characteristics of single-unit impulses. Here, we suddenly saw the momentous opening of a door allowing the exact analyses of signals from sense organs and psychoneural correlation analyses within a human individual. It soon became evident that our first instance of single-unit discrimination was not a unique and lucky event. Unitary impulse trains appeared occasionally but in increasing frequency. This turned out to be more a matter of perseverance and experimental skill than electrode design.

Another feature that we initially interpreted to be trivial noise modulations turned out to be similarly exciting. Generally, the combination of noise and background neural activity amounted to ~20 μV peak-to-peak. We became increasingly aware of and puzzled by the fact that amplitude was not always time invariant but was characterized by faint and rhythmic modulations, which initially were barely perceivable through the speakers. Moreover, they were present in some experimental sessions but lacking altogether in others. When present, they occurred intermittently but with some regularity.

Once we had excluded electronic artifacts like variations in the mains and in DC current leakage of the amplifier input stage, we realized that the rhythmic modulations must be of neural origin, hence representing activity in an efferent system. It seemed that the only reasonable candidate could be γ-motor fibers innervating the intrafusal fibers of muscle spindles. As something similar had never been described in reduced animal preparations, it seemed we had made a promising discovery that could provide exciting insights into the mysterious γ-system. This idea was consistent with our finding that the modulations were present in fascicles innervating muscles but lacking in skin fascicles.

The true origin of the rhythmic modulations, however, only became clear once we had opened our minds to an incredible alternative: that it was possible to record the activity of the small unmyelinated fibers of the autonomic nervous system with our heavy microneurography electrode. Electrocardiogram and respiratory muscle activity recordings, along with the neural signal, showed a close time relationship between heart and respiratory activity on the one hand and the modulations of the microneurography signal on the other. The exciting conclusion was obvious: the rhythmic modulations were produced by efferent sympathetic activity in unmyelinated nerve fibers. Looking back at our initial stubbornness regarding the origin of these modulations, it is comforting to read a comment by Andrew Huxley regarding his and Alan Hodgkin's analyses of ionic current in the excitable nerve membrane: "It is easy to fail to think of an idea that with hindsight seems very obvious" (Huxley 2002).

SINGLE-UNIT DISCRIMINATION OF A AND C FIBER IMPULSES

The dimensional relationship between the microneurography needle electrode and nerve fiber continued to puzzle us. Figure 1 is taken from "a critical review" of the microneurography technique, particularly single-unit discrimination and microstimulation (Wall and McMahon 1985). In it, the authors stress that recording "one or a few units remains to be explained given the relative size of the electrodes and of the axons." Their skepticism regarding the technique was probably representative of the thoughts of many senior scientists. The authors prompt the reader to contemplate Fig. 1 so that he "may judge for himself the likelihood of contacting a single fiber let alone the suggestion of intramyelin impalement." Moreover, they claim it to be obvious that "the metal on the outside of the axon would short out the signal." Their explanation for single-unit discrimination is that "the vast majority of the axons in the region of the electrode are pressure blocked" due to long-lasting electrode manipulation "so that only one or a few fibers are capable of conducting impulses into the area." This explanation, however, was refuted soon afterwards, largely on the basis of experimental data (Torebjörk et al. 1987).

For a discussion of single-unit discrimination, some additional factors and observations are relevant. One significant fact is that an electrode placed in the flesh is not equivalent to a resistance but rather emulates a capacitance of ~1 nF (Westling 1972). This is consistent with the obvious fact that carriers of electric charges are of different nature in bodily fluid and in metal, i.e., ions and electrons and the carriers cannot cross the interface between the bodily fluid and the metal of the electrode. Hence, there is no resistive leak from the large metal area to short out the signal. Electrical charges on one side of the interface attract or repulse charges on the other side, much as in a capacitance. Signal amplitude seen by the amplifier is a matter of the number of charges filling up the capacitance, whereas the physical size of the naked metal area has little bearing on single-unit discrimination except by defining the size of the capacitance.

The shapes of the nerve impulses also offer important hints. Impulses in unmyelinated C fibers are invariably triphasic, as would be expected with an extracellular electrode close to the excitable membrane. The triphasic complex shown in Fig. 2*A* depicts the transmembrane current as seen from the outside. The most prominent phase is in the negative direction, representing the inward current associated with the rising phase of the intracellular action potential. A similar triphasic complex is occasionally seen with myelinated nerve fibers, indicating that the electrode tip is close to the regenerative membrane at a node of Ranvier. Obviously, the rarely seen triphasic impulse shape is expected from the length relationship between the node and internode of the myelinated fiber. The most common impulse shape in myelinated nerve fibers is biphasic, as de-

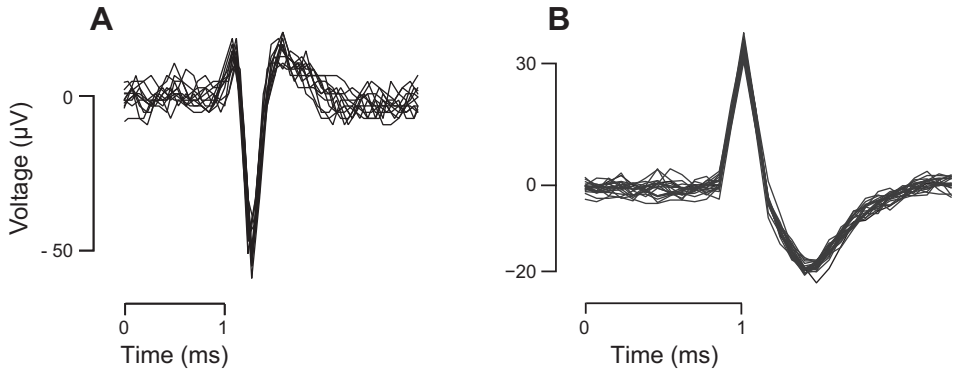

Fig. 2. Shapes of impulses recorded with a microneurography electrode Typical shapes of unmyelinated (*A*) and myelinated (*B*) fibers recorded from human nerves with the microneurography technique. [Adapted from Vallbo et al. (2016)].

picted in Fig. 2*B*. The most prominent deflection here is in the positive direction. The fact that the electrode records the rising phase of the intracellular action potential as a positive deflection indicates that either the electrode is picking up the intracellular action potential or, more likely, the electrode is seeing the outward transmembrane current passing through the myelin during the rising phase of the intracellular action potential. The large amplitude of the extracellular recording indicates a peculiar relationship between fiber and electrode. Most likely, the myelin is locally reduced in thickness due to pressure from the electrode, which would promote excessive transmembrane current locally at the point of contact. This interpretation implies that the discrimination of impulses in a single myelinated afferent requires delicate manipulation to attain not only a tight apposition between fiber and bare tungsten surface but also a suitable deformation of the fiber structure at the spot.

An impulse shape with two peaks, as shown in Fig. 3, *2 top middle insets*, is also fairly common in myelinated fibers. This shape usually appears after some minutes of recording a single peaked impulse. The two peaks originate from activities in two adjacent nodes. Action potentials in the two nodes are unduly separated in time because of slow conduction due to fiber injury, which causes excessive current leak at the site of damage and fiber run down. These descriptions of impulse morphology in myelinated fibers are supported by time-variant changes in shape that often occur during long-lasting recordings (Fig. 3, *top*). Initially, the major deflection is single peak, suggesting reasonably normal conduction. After some time, two peaks appear, indicating a local delay. The two become increasingly separated in time (Fig. 3, *2 top middle insets*) until the second peak may disappear altogether (Fig. 3, *right inset*). The dot pattern in the upper panel illustrates the successive increase in inter-peak interval, and finally the blockage of impulse conduction. The block occurs simultaneously with the transition from a dual to single peak impulse shape, implying that the downstream node of the two has failed to produce an action potential. The illustration in Fig. 3 originates from a recording of impulses in a muscle afferent, firing at a frequency of ~10 impulses/s, as shown at *bottom*.

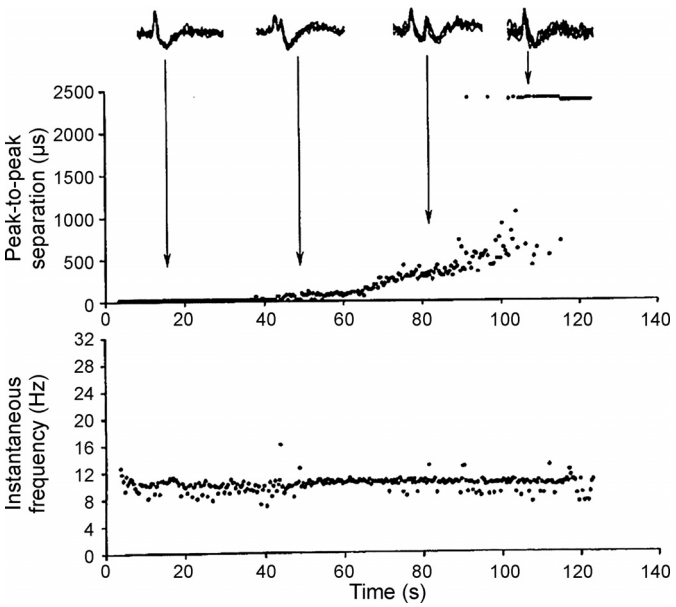

Fig. 3. Time-dependent changes to impulse shape and blockage in a myelinated nerve fiber The recordings at *top* show sample spikes on an expanded time scale to demonstrate how impulse morphology changes from a single peak to a dual peak and then back to a single peak. The 2 peaks represent activity in 2 adjacent nodes. The peaks are separated due to a conduction delay caused by electrode induced injury. *Bottom*: interpeak interval is successively increased until the 2nd peak disappears as a result of a conduction blockage at the recording site, i.e., the downstream node failed to produce an action potential. Recordings were collected from a human muscle spindle afferent firing spontaneously at a rate of ~10 impulses/s, as shown in the lower panel. [Adapted with permission (Inglis et al. 1998; ©The Physiological Society 513.1 1–314 (1998). ISSN 0022 3751)].

With unmyelinated fibers, no similar observations have been made to explain the unexpected finding that unitary impulses can be discriminated with an extracellular electrode that is many times larger than the fiber. It seems reasonable to hypothesize that a tight contact at a minute, single spot is not sufficient for single-unit discrimination because only little current to the capacitance would be produced at the point of contact between fibber and electrode. Such spatial relations are probably the case with multiunit recordings of sympathetic efferent activity which usually does not exceed 15–20 μV. A reasonable hypothesis would be that a longer stretch of fiber in tight contact with the electrode is essential for single-unit discrimination. This would imply a larger regenerative membrane area, producing a larger current to the capacitance, which would be sufficient to produce a potential change of 30–50 μV that would stand out from the background. A larger contact area would be obtained by a relatively long stretch of fiber in close apposition with the electrode circumference. An additional requirement would be for the fiber to be in good enough condition to produce an impulse that propagates over the total contact length. Incidentally, it should be appreciated that the time shift would be negligible over the length of fiber in contact with the electrode.

The assumption that the particular relationships between the electrode and nerve fiber outlined above are essential for single-unit discrimination fits with our general experience that the discrimination of unitary impulses is seldom achieved immediately after a fascicle has been impaled but usually requires a fairly long period of electrode manipulation. To attain single-unit discrimination is, of course, a matter of trial and error. Through experience, a talented experimenter successively acquires greater skill to minimally adjust the electrode position on the basis of faint sound indicators from the loudspeaker.

MEDICAL-ETHICAL OBSTACLES

Another particular concern during our endeavor to develop the method of microneurography was the risk of medical complications. A number of pertinent questions in this regard needed answers, as various kinds of medical complications seemed possible. We were fully aware that mechanical damage to the nerve fibers was bound to occur when the needle electrode was forced through a fascicle. The longer the search period involving continual movements of the needle, the larger the number of damaged and severed fibers. Would this give rise to serious consequences? We reasoned that the conditions were optimal for the successful regeneration of severed fibers because spatial relationships within the fascicle remained relatively intact, with many of the severed fibers being in end-to-end positions. However, was there a risk of intraneural bleed-

ing or edema that might give rise to the formation of intraneural scar tissue with long-lasting neurological symptoms? How large was the risk of bacterial or viral infection with serious sequelae? Was there even a risk of adverse immunological reactions due to disintegration of the myelin and axonal structures?

There were two ways to handle the medical-ethical issues. One approach would be to run the microneurography procedure in animal experiments and assess the degree of fiber degeneration and other possible intraneural changes using histological analyses. We did not consider this approach, partly because our histological competence was low and the laboratory facilities were not available. In retrospect, an ethical committee would probably have demanded animal tests before the method was applied to human subjects. However, the ethical committee system had not yet been established at the time. Incidentally, histological and histochemical analyses of animal nerves submitted to needle electrode impalements were later pursued by other groups. They have demonstrated that a fair proportion of nerve fibers are affected and that reparative processes are still going on after 4 wk (Fried et al. 1989; Rice et al. 1994).

Rather than exploring possible adverse effects using animal experiments, we decided to pursue the microneurography experiments using our own nerves. For about 2 years, the two of us utilized the large limb nerves that are reasonably accessible, the bilateral median, ulnar, and peroneal nerves. A common symptom after a microneurography experiment was hypersensitivity to local pressure at the site of impalement. A firm pressure elicited clear paresthesiae, which were also provoked when we moved the joint into an extreme position that stretched of the nerve. These phenomena started to appear a few days after the experiment and lasted for up to a couple of weeks. However, after careful watching for other subjective symptoms as well as electromyographic tests, we recorded no enduring adverse effects. Altogether, the experience was encouraging from a medical point of view. On the other hand, observations of hypersensitivity to local pressure led to our rule that recording should not be pursued more than once a month at the same recording site. It should be emphasized that microneurography experiments have since been performed in a number of laboratories, with no reports of enduring adverse effects (Gandevia and Hales 1997; Ochoa 2004). Thus it can be stated that the method is safe, as long as the experiments are performed with the recommended care and consideration. Experience has made it clear that it is essential to give detailed guidance to new scientists to minimize mechanical damage of the nerve. For instance, the electrode tip may be hooked by hitting a bone. Intraneural manipulations with a hooked electrode will produce excessive pain and probably considerable nerve fiber injury. Another aspect worth emphasizing is that inconsiderate psychological attitudes and rude jokes can be more harmful to the subject than might be foreseen because subjects are generally more tense and worried about serious damage than may be evident.

INITIAL PUBLICATIONS

The data collected from 2 years of microneurography exploration using our own nerves resulted in five short notes, a congress report, and four full papers that were published in 1968 and 1969. The first official presentation of our microneurography recordings was given at the Scandinavian EEG Society in Copenhagen in 1966. An abstract report was published in *Electroencephalography Clinical Neurophysiology* (Vallbo and Hagbarth 1967). The four full papers represent three areas of inquiry that have become major fields in microneurography research: the muscular proprioceptive system, the cutaneous mechanoreceptive system, and the sympathetic efferent system to skin and muscles (Hagbarth and Vallbo 1968a, 1968b, 1969; Vallbo and Hagbarth 1968). Soon after our first single-unit recordings, two PhD students in the laboratory opened up another major line of research, the cutaneous nociceptive system, largely based on their finding that unitary impulses in unmyelinated afferents can be discriminated (Torebjörk and Hallin 1970) .

After my 2 years of joint experimental work with Hagbarth, I moved to the newly established University of Umeå. An attractive aspect of breaking up our partnership was building a laboratory of my own, designed exclusively for microneurography and optimized for the kind of research I was keen on pursuing. Hagbarth and I continued to cooperate part-time for another couple of years and kept in touch for discussions over many years, until his death in 2005 at the age of 78.

CONTRIBUTIONS BASED ON MICRONEUROGRAPHY

The remainder of the article will briefly outline studies from various fields to give the reader an idea of the potential and pertinence of the microneurography technique. It is important to point out that this account will be limited in scope and personal in selection. It is by no means an attempt to give a comprehensive presentation of the achievements attained during 50 years of microneurography research. Consequently many review articles are given in the reference list but relatively few original articles. References to additional reviews may be found in an article by Gandevia and Hales (1997). Descriptions of electronic equipment designed for microneurography recording and microsimulation is not considered in the present paper. References are given to a number of articles that discuss recording amplifier design as well as other technological issues including post-processing practices (Edin et al. 1988; Gandevia and Hales 1997; McKeon and Burke 1980; Westling 1972). Particularly the article by Gandevia and Hales brings up and discusses many essential aspects of the technique.

Moreover, it should be pointed out that the following presentation is not an attempt to rate contributions on the basis of scientific quality and significance.

SYMPATHETIC EFFERENT ACTIVITY

During the first few years following the first successful microneurography experiments, it was already obvious that the field would grow along several lines. We realized that there were too many potential avenues of inquiry for the two of us to tackle alone, and thus we were happy to see Wallin's interest in the sympathetic system, which he developed masterfully in collaboration with scientists from several continents (Joyner et al. 2010; Wallin and Fagius 1988).

The sympathetic branch of microneurography research is unique because of its relevance to cardiovascular physiology and pathophysiology and because it attracted a great deal of

interest from scientists in the field of circulation. Studies of sympathetic efferent activity have largely used multiunit recordings, which are less demanding than the single-unit recordings used in studies on other systems. In fact, sympathetic activity does usually not require long-lasting searching but is often encountered soon after nerve impalement. These two features make the recording of sympathetic activity manageable in combination with more demanding methods of measuring cardiovascular variables such as central venous pressure or arterial blood pressure via intravascular catheters.

An early and fundamental finding was that sympathetic activity differs in several respects between skin and muscle nerves, indicating separate control systems (Wallin and Fagius 1988). In skin nerves, the activity is highly reactive to sensory stimuli, particularly those with emotional significance (Delius et al. 1972b). For instance, a sudden arousal stimulus such as an unexpected handclap behind the subject gives rise to a burst of activity in the cutaneous nerves. In muscle nerves, on the other hand, the activity is under heavy baroreceptor control, causing it to vary with the continuous variations in arterial blood pressure that normally occur in phase with the cardiac and respiratory cycles (Delius et al. 1972a).

The indications of different control systems that were evident in recordings of skin and muscle nerves clashed with the prevailing view that the sympathetic system is diffusely organized and therefore should produce broadly similar efferent activity towards various target organs.

The sympathetic input to muscular vessels attracted particular interest because of its potential role in the control of blood pressure. An initial hypothesis was that sympathetic efferent activity would be a key factor determining mean arterial blood pressure and would even account for pathological hypertension. This idea seemed reasonable considering two established facts: the fact that the sympathetic efferent input to muscular blood vessels is largely vasoconstrictive and that the vascular bed of the skeletal muscles accounts for a major part of the total peripheral resistance under resting conditions.

Experimental testing of this hypothesis revealed interesting findings but failed to support it. One exciting finding was that the amount of sympathetic efferent activity in the resting state varies considerably between individuals, whereas it remains largely constant within an individual over a long period (Fagius and Wallin 1993). It was further found that differences between individuals were not associated with expected differences in mean arterial blood pressure, a finding that refuted the original hypothesis. A significant factor behind this counterintuitive finding was identified when it was found that subjects with high sympathetic efferent activity had low cardiac output and vice versa. The mechanisms controlling the reverse relationship between the amount of sympathetic activity and cardiac output remain to be clarified.

It should be emphasized that these findings were collected from a select group of subjects made up of normotensive young males up to the age of ~40. Studies of other groups revealed a number of interesting differences. For instance, the reverse relationship between sympathetic efferent activity and cardiac output as seen in young men was not found in young women. This and other findings suggest that physiological strategies to regulate blood pressure are not identical between men and women.

The most exotic application of the microneurography method was a study on sympathetic activity conducted during a space flight in the Space Shuttle Columbia. The purpose of the study was to investigate the mechanisms involved in the control of arterial blood pressure. It was known that microgravity causes hypovolemia and that astronauts often suffer from orthostatic hypotension and presyncope after returning to Earth. Microneurographic recordings performed on the space shuttle by astronauts from the US and Japan indicated that sympathetic activity to the vascular bed of striated muscles was, in fact, moderately enhanced during the flight (Ertl et al. 2002). Moreover, it was demonstrated that the response to simulated orthostatic stress was enhanced, indicating that sympathetic reflexes worked in principle normally to compensate for the orthostatic effect of hypovolemia.

MUSCLE AFFERENTS

Studies in the proprioceptive branch of microneurography were less rewarding than expected, in the sense that they failed to support the ideas that had inspired researchers beforehand – e.g., the follow-up-length servo hypothesis (Merton 1953) and the idea that mental attention and motor intention might modify the activity of the γ-system while the main muscle remained silent (Gandevia et al. 1997; Hagbarth et al. 1975; Vallbo 1971). In contrast, the findings indicated that the fusimotor system to the small muscle fibers within the muscle spindle is activated when the parent muscle contracts but remains inactive as long as the parent muscle is relaxed. This conclusion was based on simultaneous recordings of afferent impulses from individual spindle afferents and the electrical activity of skeletal muscle fibers (EMG). A similar pattern was found in behaving animals as well (Prochazka et al. 1988).

At the same time, definite differences between humans and behaving cats were found with regard to the relative intensity of activity in the two motor systems. With cat hindlimb muscles, the balance between EMG intensity and muscle spindle impulse rate was found to vary with the motor task, indicating fusimotor independence (Prochazka et al. 1988). Convincing evidence for similar independence has not been found with human hand and finger muscles. It remains to be assessed whether the different findings can be accounted for by a genuine physiological difference or different experimental conditions. In microneurography experiments, restricted conditions are required to maintain recording stability, implying that the motor task cannot be as widely varied as with behaving animal where free movements are allowed. The effects on the fusimotor activity of the subject's mental set, e.g., with regard to precision demand and attention to the motor task, were tested in several laboratories. No support for fusimotor independence was found when the mental set of human subjects was modified (Gandevia et al. 1997; Vallbo and al-Falahe 1990).

Tests of spindle afferent activity during finger movements revealed a phenomenon not previously observed: spindle afferents monitor the minute and unperceivable discontinuities at 8–10 Hz, which are due to a pulsatile component of the motor command during voluntary movements (Gross et al. 2002; Vallbo and Wessberg 1993; Wessberg and Vallbo 1995). Modulations of afferent activity at 8–10 Hz suggest that muscle spindles may play a role in relation to a putative

~10-Hz clock, which has been postulated as a component of a cerebral system that organizes motor output (Gross et al. 2002; Lisman 2015; Llinás 1991).

CUTANEOUS NOCICEPTIVE C-AFFERENTS

The nociceptive field of microneurography was opened by Torebjörk and Hallin 1970, who originally demonstrated that single-unit impulses could be discriminated not only with large myelinated afferents but with the small unmyelinated C-fibers as well. The nociceptive field was much supported by the "marking" technique. This technique relies on the fact that unmyelinated nerve fibers exhibit a postactivation transmission delay in the wake of preceding impulses (Torebjork and Hallin 1976). The delay may be pronounced and may last for several seconds. In the standard "marking" experiment, intradermal needle electrodes are inserted into the skin at a location where physical stimuli indicate afferent terminals, as detected by the intraneural electrode. Repetitive electrical pulses are delivered at a low frequency through the intradermal electrodes to monitor the baseline transmission time from the terminals to the intraneural recoding site. When a short-lasting natural stimulus is added, an increase in the transmission time indicates that the afferent has responded to the natural stimulus.

This technique offers a number of advantages. First, the discrimination of individual afferents does not require isolation on the basis of impulse shape and amplitude, which is particularly demanding with unmyelinated nerve fibers. In fact, impulses with very low amplitudes can be discriminated on the basis of transmission time. Second, the approach is very effective, since the functional properties of several afferents can be assessed in the same recording session. This is because the electrical pulses usually excite several fibers that are easily discriminated based on their different transmission times.

Studies in several laboratories have identified and characterized four types of putative nociceptive unmyelinated afferents on the basis of their responsiveness to mechanical and temperature stimuli (Torebjörk et al. 1996): polymodal afferents (CMH), which make up the largest group; mechanosensitive afferents (CM); heat-sensitive afferents (CH); and silent afferents (CM_iH_i). The silent afferents are largely unresponsive to either heat or mechanical stimuli but can be sensitized by inflammatory processes in the skin. This kind of peripheral sensitization is very likely significant for common types of hyperalgesia albeit central sensitization is probably essential as well.

Not only have nociceptive C fibers been identified and characterized using the marking technique, but putative itch afferents and sympathetic efferents as well.

TACTILE SYSTEM OF THE HUMAN HAND

In studies of the cutaneous mechanoreceptive system, it seemed particularly attractive to focus on the human hand. First, the tactile mechanisms of the primate hand had been widely studied as a basis for assessment of psycho-neural relations. Second, the hand has a prominent role in active touch, involving an intriguing cooperation between sensory and motor mechanisms (Gordon 1978; Mountcastle 2005; Wing et al. 1996). A reasonable starting point for the study of the hand's tactile system seemed to be the development of a reasonably comprehensive model of the tactile apparatus of the glabrous skin. It was shown that 17,000 tactile afferents innervate the palmar aspect of the hand (Johansson and Vallbo 1979b). The nerve fibers are myelinated and fast conducting at ~60 m/s. The tactile units are of four distinct types, differing with regard to functional properties, spatial distribution, and absolute density within the different regions of the palmar skin. Putatively, the nerve fibers are connected to the Meissner, Merkel, Ruffini, and Pacini end organs. Interestingly, unmyelinated tactile afferents were never encountered in the glabrous skin, in contrast to hairy skin, where they are abundant. A similar difference has been found between the hairy skin and pad skin of other mammals.

The basic description of the tactile apparatus of the glabrous skin invited hypotheses regarding the roles of the separate unit types and provided a foundation for hypotheses on psychoneural relations. The description also inspired more complex experiments, including microstimulation and brain imaging studies, as well as studies of the motor functions of the hand.

A significant source of inspiration for the study of the human hand's tactile mechanisms was the work of Mountcastle (1967) and Talbot et al. (1968), who analyzed the relationships between the neural responses in monkeys and psychophysical responses in humans to identical stimuli. These brilliant studies should be remembered as pioneering steps in the search to bridge the gap between biophysical events in the nervous system and mental events within the human mind, not to be overshadowed by later studies based on more advanced technologies and analytical methods. When microneurographic recording turned out to be successful, the method was obviously attractive for use in psychoneural correlation analyses, considering that the technique offered the option of analyzing intraindividual data in contrast to group data from two different species, as in earlier studies.

Microneurography and microstimulation were combined to shed light on several issues in the field of sensory physiology, e.g., the detection of threshold stimuli, the scaling of stimulus intensity, the perception of spatial location, the temporal quality of tactile stimuli, and the specificity and quality of percepts (Johansson and Vallbo 1979a; Knibestöl and Vallbo 1980; Torebjörk et al. 1987; Vallbo 1989).

With regard to the quality of a percept elicited by a natural stimulus, two different ideas had been much debated over the years: the specificity theory and the pattern theory. Advocates of the latter did not accept any fixed relationship between sensory unit specialization and perceptual response, instead claiming that "every discriminably different somaesthetic perception is produced by a unique pattern of nerve impulses" from several kinds of sense organs (Melzack and Wall 1962). A prediction made by the pattern theory is that "more than one fiber carrying single nerve impulses is essential for central cells to detect the characteristics of a sensory stimulus" (Melzack and Wall 1962). This prediction was subsequently refuted by microneurography experiments in which microstimulation with electrical pulses of single units showed that impulse trains in individual mechanoreceptive afferents from the glabrous skin may be clearly perceived as minute, but distinctly defined, skin deformations. The characteristics of the percept were readily distinguished with regard to location, spatial extent, duration, and quality. Moreover, these attributes matched the spatial and functional properties of the afferent unit with regard to receptive field size and location, as well as the dynamic

response properties. Thus repetitive stimulation of the slowly adapting Merkel unit was felt as a sustained deformation, whereas similar stimulation of the fast adapting Meissner unit was felt as an intermittent tapping or a flutter (Ochoa and Torebjörk 1983; Vallbo et al. 1984).These findings reject the pattern theory as a universal principle. They instead support the idea of sensory specificity, implying that the basic attributes of percepts may be accounted for by the selective sensitivity of stimulated sense organs. This is not to deny that the brain uses complex patterns of sensory input in several kinds of afferents to construct percepts of natural stimuli.

Incidentally microstimulation of individual Meissner as well as Merkel afferents from the glabrous skin may give rise to robust fMRI responses in somatosensory cortex (Trulsson et al. 2001).

Ruffini units (SAII) are unique among the four mechanoreceptive unit types in the glabrous skin as the microstimulation of a single SAII afferent does not evoke a sensation. It seems that this is consistent with other findings suggesting that the main role of SAII units is in proprioception rather than in discriminative touch as discussed later in this article.

With regard to sensory detection, the combination of microneurography and microstimulation efficiently demonstrated that psychophysical detection can be based on the minimum biophysical quantum, i.e., a single impulse in a single afferent. However, this is not universally the case. It is true for Meissner afferents in certain areas of the glabrous skin of the hand, whereas larger sensory input is required for other areas and with Merkel and Pacini afferents (Ochoa and Torebjörk 1983; Torebjörk et al. 1987; Vallbo et al. 1984).

TACTILE AFFERENTS IN THE CONTROL OF DEXTEROUS MANIPULATIONS

The cutaneous mechanoreceptive system of the hand is of particular interest considering the paramount importance of the hand in tactile exploration and the manipulation of physical objects. These functions are already essential in early childhood, as illustrated by the young child eagerly striving to grasp and examine any foreign object within reach, thereby fulfilling the brain's desire to build up an inner model of the physical world.

The complex mechanisms underlying the control of dexterous manipulations have been extensively explored by Johansson and Flanagan (2009) and Flanagan et al. (2006). They demonstrated that several sophisticated control functions are dependent on the high sensitivity of the tactile sense organs within the glabrous skin, as will be illustrated by the following examples.

Basic elements of object manipulation include the grasping, lifting, and then holding of an object while the intended task is being performed. It was shown that the grip force applied to lift a familiar object is largely determined by sensorimotor memories (Johansson and Westling 1988), whereas lifting a foreign object engages control mechanisms that are dependent on tactile afferents. When a foreign object is initially grasped, the person's sensorimotor systems measure the friction between the skin and the object before the object is lifted (Johansson and Westling 1984). The rate of grip force increase is then set on the basis of the friction. The early assessment of friction is apparently based on the degree of slipping and creeping within the contact area associated with finger pulp deformation. Slipping and creeping are encoded by tactile afferents; for instance, the Meissner units within the contact area respond more intensely to low than high friction (Johansson and Flanagan 2009).

When an object is being held and manipulated, tangential forces can vary considerably, which may threaten the hold. Any minute slippage between skin and object is monitored by the sensitive afferents within the contact area. Although not perceived consciously, the response of the tactile afferents triggers a short latency reflex that increases the grip force to maintain a secure hold (Johansson and Westling 1987).

It has been shown that the tactile afferents of the glabrous skin are involved in a number of other sensorimotor mechanisms that are active during the manipulation of objects, as described in original studies and review articles (Johansson and Flanagan 2009).

UNMYELINATED TACTILE AFFERENTS AND THE SOCIAL TOUCH HYPOTHESIS

The mechanoreceptive apparatus of the skin had long been studied as a system mediating discriminative touch and contributing to the control of motor actions. However, microneurography studies have added another two, and fundamentally different, roles: the social touch and proprioceptive functions. An expansion beyond discriminative touch started with the observation that the unmyelinated tactile afferents [C-tactil (CT)] in the hairy skin are not essential for the tickle sensation, as originally suggested by Zotterman (1939) and Vallbo et al. (1999, 2016). On the basis of microneurography recordings, and later, functional brain imaging as well as psychophysical tests, it was proposed that the essential role of the CT afferents is to support positive social touch (Olausson et al. 2010, 2016). It has been demonstrated that CT afferents are tuned to slowly moving touch stimuli, as in caressing gestures. Moreover, significant correlations have been demonstrated between activity in CT afferents and subjective estimates of pleasantness when two different stimulus parameters, the speed of an object moving over the skin surface and the temperature of a slowly moving object, were varied (Ackerley et al. 2014; Löken et al. 2009). It seems that these two sets of correlations provide support for the social touch hypothesis, which argues that the role of CT afferents, within the realm of perception, is to boost the feeling of pleasantness when you are touched by a friendly human being and hence may have the role to improve bonding between individuals.

The cerebral projection also seems to be consistent with the social touch hypothesis, as the CT afferents project preferentially to the insular cortex, which is known a gateway to the emotional systems of the brain, whereas the somatosensory cortex is not a prominent target of the CT afferents (Olausson et al. 2002, 2008). A system of afferents similar to the CT system in humans has been described in rodents. It has been shown that rodent C low threshold mechanoreceptors have a rewarding effect, as demonstrated in behavioral studies (Vrontou et al. 2013).

PROPRIOCEPTIVE FUNCTIONS OF CUTANEOUS AFFERENTS

Another role beyond discriminative touch was demonstrated by Edin, who showed that the slowly adapting type II units

(SAII, putative Ruffini) have an impressive kinesthetic potential (Edin 2001; Edin and Johansson 1995). Voluntary movements are accurately monitored by the SAII type afferents in the hairy skin. Moreover, psychophysical tests indicated that signals from cutaneous afferents are essential for normal perception of joint position. Hence, it seems justified to regard the mechanoreceptive apparatus of the skin not as a solely exteroceptive system but as a proprioceptive system as well.

MINOR BRANCHES OF MICRONEUROGRAPHY

In addition to the fields outlined above, a few additional areas of interest have generated new information. Intricate experiments have demonstrated that intraoral mechanoreceptive afferents in the periodontal ligament, akin to cutaneous SAII units (Ruffini), beautifully monitor the size and the directions of forces acting on individual teeth (Trulsson et al. 1992). Another study used microstimulation to characterize the contraction properties of individual motor units in small hand muscles (Thomas et al. 1990). Thermoreceptive afferents have been the subject of a limited number of investigations, which may be due to the particular difficulty of recording from the small myelinated afferents (Adriaensen et al. 1983; Hallin et al. 1982).

LOOKING BACK AFTER 50 YEARS OF MICRONEUROGRAPHY

Fifty years ago, integrative neurophysiology was dominated by experiments using reduced preparations, e.g., anesthetized and decerebrated mammals. However, at the time, interest in dissecting afferent systems in conscious human subjects and behaving animals was slowly growing. With this background, it seemed very tempting in the 1960s to seek an experimental method for recording neural signals in the peripheral nerves of attentive human subjects and particularly to achieve ultimate precision by identifying nerve impulses in single afferents.

An obvious potential advantage of the microneurography technique was that it could be used to describe the peripheral organization of the cutaneous sensory system in humans and to explore the functional properties of the system's components. As shown by the examples presented above, these investigations detailed the components and relationships within the tactile as well as nociceptive systems. The description of the sensory apparatus of the glabrous skin of the hand was comprehensive compared with corresponding accounts from studies in other mammals.

Moreover, the examples presented above illustrate that new findings have inspired scientists to explore previously dormant fields brought into the light due to the technique, e.g., the roles of tactile afferents in the control of dexterous hand functions and the functions of the sympathetic efferent activity to skin and striated muscles.

The unique potential of the experimental setup involving nerve impulse recording in a cooperative human subject has been illustrated by several studies, e.g., psychoneural correlation studies and analyses of proprioceptive mechanisms in voluntary movements. It seems that many of the conclusions reached by studies using this setup would have been difficult to obtain with other experimental designs.

A new technique can not only generate new facts and reveal hidden relationships but can also modify the landscape of concepts and ideas. In the realm of tactile sensibility, microneurography has refuted the pattern hypothesis as a general principle in sensation and has supported the idea of sensory specificity. Moreover, in the motor control domain, the method has refuted the follow-up-length servo hypothesis as well as the preconception that mental factors like motor intentions or attention are regularly important for modifying muscle spindle sensitivity through the fusimotor system.

A new technique may not only modify, refute, or support existing ideas but may foster new ones as well. The social touch idea was proposed on the basis of psychoneural observations of the slow tactile system in the hairy skin. This hypothesis is novel in its proposal that the main role of the slow tactile system is to detect and promote motivational and emotional responses to a specific kind of gesture by a friendly individual. The social touch hypothesis has provided inspiration for studies not only in humans but in other mammals as well (Vrontou et al. 2013).

Several branches of microneurography are based on the recording of neural events in a cooperative and attentive human subject. Fifty years ago, a combination of this nature was rare. Obviously, developments in brain imaging and other techniques that can be used with human subjects have radically changed the scene. However, the studies listed above may serve to show that microneurography fits in well with the expansion of integrative neuroscience and the interest in exploring the human nervous system by means of new and powerful methods and analytical tools that seemed impossible to imagine 50 years ago.

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AUTHOR CONTRIBUTIONS

Å.B.V. drafted manuscript; Å.B.V. edited and revised manuscript; Å.B.V. approved final version of manuscript.

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