**Biomedical Physics & Engineering Express**

**PAPER**

*In vitro* multichannel single-unit recordings of action potentials from the mouse sciatic nerve

I Chen®, S J Ilham®, T Guo®, S Emadi and B Feng®

Department of Biomedical Engineering, University of Connecticut, Storrs, CT 06269, United States of America

E-mail: fengb@uconn.edu

Keywords: single-unit, extracellular recording, peripheral nerve, neuromodulation, sciatic nerve, ultrasonic stimulation, mouse

---

**Abstract**

Electrode arrays interfacing with peripheral nerves are essential for neuromodulation devices targeting peripheral organs to relieve symptoms. To modulate (i.e., single-unit recording and stimulating) individual peripheral nerve axons remains a technical challenge. Here, we report an *in vitro* setup to allow simultaneous single-unit recordings from multiple mouse sciatic nerve axons. The sciatic nerve (~30 mm) was harvested and transferred to a tissue chamber, the ~5 mm distal end pulled into an adjacent recording chamber filled with paraffin oil. A custom-built multi-wire electrode array was used to interface with split fine nerve filaments. Single-unit action potentials were evoked by electrical stimulation and recorded from 186 axons, of which 49.5% were classed A-type with conduction velocities (CV) greater than 1 m s⁻¹ and 50.5% were C-type (CV < 1 m s⁻¹). The single-unit recordings had no apparent bias towards A- or C-type axons, were robust and repeatable for over 60 min, and thus an ideal opportunity to assess different neuromodulation strategies targeting peripheral nerves. For instance, ultrasonic modulation of action potential transmission was assessed using the setup, indicating increased nerve conduction velocity following ultrasound stimulus. This setup can also be used to objectively assess the design of next-generation electrode arrays interfacing with peripheral nerves.

---

**Introduction**

Neuromodulation targeting the peripheral nervous system (PNS) has attracted growing research interest as evidenced by its effectiveness in managing a variety of disease conditions, including epilepsy, obesity, incontinence, urinary tract disorders, and chronic pain [1–4]. PNS neuromodulation generally requires the stimulation of and recording from peripheral nerves via single electrodes or electrode arrays, the applications of which in clinical settings are limited to either microneurographic needles [5, 6], a slanted Utah array [7], or transversal multichannel intrafascicular electrodes [8]. In contrast, electrode arrays that interface with the brain have been routinely used in pre-clinical studies to allow recordings of action potentials from individual neurons (i.e., single-unit recordings) and focal stimulation of a small volume of the brain tissue [9, 10]. Further, successful translation of the electrode–brain interface is evidenced by the wide application of deep-brain stimulators and recent success of electrode arrays that interface somatosensory and motor cortices in patients [11, 12]. The challenge presented for electrodes to successfully interface with, and especially record from peripheral nerves, relates to the extra layers of connective tissues associated with the nerves (which are absent in the central nervous system): the epineurium that encircles the nerve trunk and provides mechanical protection, the perineurium that wraps each nerve fascicle and forms tight-junctions (i.e., blood–nerve barrier), and the endoneurium that protects myelinated nerve fibers consisting of either myelinated axon or a Remak bundle of unmyelinated axons [13]. Electrodes outside the fascicles usually record a population of neural activities in the form of compound action potentials, consisting of a large peak from volleys of myelinated A-fibers and a much smaller peak from unmyelinated C-fibers, despite the greater number of C-fibers in peripheral nerves [14, 15]. Utah slanted arrays are one of the few clinically available intrafascicular electrode arrays that penetrate the...
epineurium [7, 16]. While capable of single-unit recordings, most action potentials recorded by the Utah slanted array are from myelinated A-type axons; unmyelinated C-type axons are underrepresented in recordings using intra-fascicular arrays (unpublished communications). Although efficient at recruiting individual muscles via focal stimulation [17], longitudinal and transverse intrafascicular multichannel electrode arrays generally do not record single-units from axons [8, 18, 19]. The needle electrodes used in microneurography allow reliable recordings from unmyelinated C-fibers in humans. Through intricate manipulation, the needle tip can penetrate the endoneurium to be in close proximity with C-type axons so as to achieve reliable extracellular recordings [6, 20, 21]. Micro lesions of recorded axons have been documented in some microneurographic recordings, indicative of its potential risk [22].

Single-unit recordings from peripheral nerves have been mainly conducted in animals in vitro or in vivo (e.g., recordings from vagus nerves [23, 24], aortic depressor nerves [25], spinal nerves [26, 27], splanchnic nerves [28], and pelvic nerves [29–31]). To achieve a high signal-to-noise ratio, the technically challenging approach taken was to carefully isolate nerve trunks, gently splitting the trunk into fine nerve bundles, and teasing a bundle into microns-thick fine filaments. Commercially available microelectrode arrays, most of which are designed to interface with the brain, are not suitable for recordings from teased peripheral nerve filaments. Thus, the aforementioned studies were limited to one-channel electrode, single-unit recordings from one to two easily identifiable axons in a single nerve filament. Successful nerve splitting is technically demanding and of low efficiency, and these appear to be factors that limit the broad application of single-unit recording/stimulation of peripheral nerves [26, 32]. In the present study, we aimed to increase the efficiency of single-unit recordings by developing an in vitro setup that permits interfacing of multiple peripheral nerve filaments with a custom-built multi-wire electrode array, resulting in simultaneous single-unit recordings from multiple peripheral nerve axons. Our recordings consist of a significant proportion of single-unit action potentials from unmyelinated C-fibers, which are usually underrepresented in extracellular recordings from peripheral nerves. Portions of the data have been previously reported in abstract form [33].

**Methods**

All experimental procedures were approved by the University of Connecticut Institutional Animal Care and Use Committee.

*In vitro perfusion chamber*

A custom-built tissue perfusion chamber, which also served as the platform for multi-channel single-unit recordings, consisted of a tissue compartment perfused at 2–6 ml min⁻¹ with oxygenated (95% O₂, 5% CO₂) Krebs solution (in mM: 117.9 NaCl, 4.7 KCl, 25 NaHCO₃, 1.3 NaH₂PO₄, 1.2 MgSO₄, 2.5 CaCl₂, and 11.1 D-glucose at room temperature) and an adjacent nerve recording compartment filled with paraffin oil (Thermo Fisher Scientific, Waltham, MA). The volume of the tissue compartment was limited to be no more than 6 ml to facilitate rapid replacement of chamber solutions during pharmacological tests (within 1 min for 6 ml min⁻¹ flow). The nerve was pulled into the recording compartment through a mouse hole under a plastic gate that separated the tissue and recording compartments. The bottom of the tissue compartment was covered by a layer of silicone for pinning down the nerve (Sylgard 182 silicone elastomer; Dow Corning, Midland, MI); a hydrophilic glass surface was placed at the bottom of the recording compartment filled with hydrophobic paraffin oil media to attract the hydrophilic nerve fibers via surface tension.

*Design of a multi-wire electrode array*

A custom fabricated 5-channel electrode array was utilized to interface with teased nerve filaments as shown in figure 1(C). The electrode array was built with a self-designed PCB board and micro-wires (Nichrome, 65 μm thick, A-M Systems Inc., Sequim, WA). One end of the PCB board was configured with connectors compatible with the recording device; the other end was configured with micro-wires through metal pads. Micro-wires were deployed parallel (100–150 μm apart) to each other and the insulation (Formvar) for the distal ~0.2 mm wires was peeled off via a fine-tipped soldering iron under a stereomicroscope to expose the conducting tips (figure 1). The electrode impedance of each channel when submerged in Krebs solution was below 100 kΩ at 1120 Hz measured by PZ-35-32 (Tucker Davis Technologies, FL). The shaft of the electrode array was insulated by a thin layer of silicone.

*Mouse sciatic nerve harvesting*

Sciatic nerves were harvested from male C57BL/6 mice aged 6–8 weeks, 20–30 g (Taconic, Germantown, NJ). Mice were anesthetized by isoflurane inhalation, euthanized by exsanguination via perforating the right atrium, and transcardially perfused with oxygenated Krebs solution. The whole length of the sciatic nerve (~30 mm) was harvested bilaterally from their proximal projection to the L4 spinal cord to their distal branches innervating gastrocnemius muscles. Sciatic nerves were then transferred to the custom-built perfusion compartment containing oxygenated Krebs solution at room temperature. One end of the sciatic...
nerve was pinned down in the tissue compartment. The ~5 mm distal end of the sciatic nerve was gently pulled over and laid onto a mirror in the recording compartment filled with paraffin oil.

**Single-unit recording**

Action potentials were evoked by electrically stimulating the proximal end of the sciatic nerve in the tissue compartment. To minimize stimulus artifacts, a suction electrode fabricated with a quartz glass capillary was used to deliver monopolar cathodal current pulses (0.2–0.8 mA, 0.2 ms duration, 0.5 Hz). To enhance the signal-to-noise ratio (SNR) of recordings, the connective tissue layers (epineurium and perineurium) were carefully dissected away before splitting the individual nerve fascicle into fine filaments (~25 μm thick). The ~5 mm distal nerve trunk in the recording chamber was treated with collagenase type 4 and split into fine filaments to interface with the multi-wire electrode array. The collagenase solution was subsequently removed and the nerve was rinsed thoroughly with oxygenated Krebs solution. Single-unit action potentials from all five electrodes were recorded simultaneously, digitized at 25 kHz, band-pass filtered (300–3000 Hz) and stored on a PC using an integrated neural recording system.
Transmission electron microscopy
Peripheral nerve tissues were processed for electron microscopy as described previously [34]. Briefly, tissues were fixed at 4 °C for 60 min in a 0.12 M phosphate buffer solution (PB, pH 7.2) containing 2.5% glutaraldehyde, 2% paraformaldehyde, and 3 mM MgCl₂. Following two rinses in PB, the tissues were further fixed with 1% Osmium tetroxide in 0.12 M PB at room temperature for 2 h in the dark. After an additional rinse in PB, the tissues were dehydrated through a series of 30, 50, 70, 95 and 100% ethyl alcohol for 10 min each followed by two dehydrated through a series of 30, 50, 70, 95 and 100% propylene oxide for 10 min each. After an additional rinse in PB, the tissues were embedded in an epoxy resin mixture [35] and polymerized at 60 °C for 48 h. Blocks were sectioned on an ultramicrotome (Leica, Bannockburn, IL) and collected on grids. The grids were then stained in 2% uranyl acetate and 2.5% Sato's lead citrate, washed with water, and dried at room temperature. The images were taken with an FEI Tecnai T12 transmission electron microscope equipped with an AMT 2 K XR40 CCD (4 megapixel) camera at an accelerating voltage of 80 KV.

Ultrasound stimulation
A preliminary study of neuro-modulation by high intensity ultrasound was performed. A focused 1.1 MHz ultrasound transducer (H-101G, Sonic Concepts, WA) was used to generate ultrasound stimulation to the sciatic nerve in vitro. A power amplifier (E&B A-300, Electronics & Innovation, Rochester, NY) was used to drive the ultrasound transducer, which receives a repetitive pulse wave from a function generator (square pulse, 100 ns duration, BK Precision 4054, Yorba Linda, CA). A pulse repetition frequency (PRF) of 100/150/200/250 kHz was used. An adjustable attenuator (50DR-046, JFW Industries, Indianapolis, IN) was placed between the function generator and power amplifier for fine-tuning ultrasound intensity. Ultrasound was delivered to the nerve segment using a water cone filled with degassed water placed above the perfusion chamber. A needle hydrophone (HNR-1000, Onda Corp., Sunnyvale, CA) was used to establish the ultrasound intensity at the nerve trunk, which is quantified using the spatial peak temporal average intensity method (i.e., the ISPTA [36]). The ultrasound intensity delivered to the nerve ranged from 0 up to 100 W cm⁻².

Statistical analysis
Data were processed off-line using customized MATLAB programs (MathWorks R2016b). A root-mean-square (RMS) value of 5 ms pre-stimulation noise was calculated and five times that value was set as the detection threshold for action potential spikes. The time of first exceeding the threshold was deemed as the onset of the action potential. To avoid confusion, only easily discriminable single-unit spikes temporally separated from adjacent spikes in the record were analyzed. Conduction delays were measured as time between the onset of stimulus artifacts and the onset of recorded action potentials. Conduction velocity was computed from the conduction delay and distance between stimulating and recording electrodes. Data are presented as means ± SE. One-way ANOVA was performed as appropriate using SigmaPlot v9.0 (Systat Software, San Jose, CA). Differences were considered significant when p < 0.05.

Results
Design of the perfusion chamber for conducting the in vitro single-unit recordings is reported in section 2, and the schematic diagram and a picture of the in vitro recording setup are displayed in figures 1(A) and (B), respectively. The custom-built multi-wire electrode array (figure 1(C)) allows interfacing with split sciatic nerve filaments in the recording compartment for simultaneous single-unit recordings (figure 1(D)). Collagenase treatment did not cause apparent morphological changes of the sciatic nerve (figure 2(A)). After collagenase treatment, the epineurium and perineurium covering the distal 5 mm of the sciatic nerve were carefully removed, and the nerve trunk was split into fine filaments as displayed in figures 2(B) and (C). Electron microscopy images show the cross sections of nerve axons in the non-split sciatic nerve trunk (figure 2(D)) in contrast to axons in one split nerve filament (figure 2(E)). As indicated by arrows in figure 2(D), several unmyelinated axons form a Remak bundle, which is wrapped by the endoneurium. The myelinated axons, indicated by arrow heads in figure 2(D), are relatively larger in size and each is wrapped by the endoneurium. Fine nerve fiber splitting resulted in a limited number of axons in each nerve filament and disrupted endoneuriums as indicated by double arrows in figure 2(E). The dry-up of split nerve filaments in figure 2(E) was caused by the processing of tissue with volatile propylene oxide for electron microscopy.

Action potentials were evoked by a monopolar cathodal stimulus delivered via a suction electrode on one end of the sciatic nerve (0.2–0.8 mA, 0.2 ms duration, 0.5 Hz). Single-unit action potentials were successfully recorded from 186 axons. Typical single-unit recordings from the custom-built multi-wire electrode array are displayed in figure 3; five electrode wires were in contact with five split nerve filaments. The tight seal of the suction stimulating electrode with the nerve resulted in a stimulus artifact width <4 ms. As displayed in figure 4(A), the robustness of the recording is demonstrated by the almost overlapping records of single units (labeled 1 to 10 in figure 3)
evoked from 10 consecutive nerve stimulations (0.5 Hz). The standard deviations of the 10 consecutive conduction delays from each of the 186 axons were quantified and plotted in figure 4(B), which reveals that greater than 95% of the recorded conduction delays have normalized standard deviations less than 1.5%. The stability of the in vitro single-unit recordings was assessed by repeated recordings of 10 consecutive stimulus-evoked spikes at 5–30 min intervals for up to 60 min. Forty-three fibers were tested at 5 min after the baseline recordings and 17 fibers tested at 15, 30, and 60 min after baseline. As displayed in figure 5, the pooled data indicated that the normalized conduction delay did not change over time.

Displayed in figure 6(A) is an example of conduction delays from fast-conducting A-fibers and slow-conducting C-fibers, which are used to calculate nerve conduction velocities. The histogram of conduction velocities (CV) from 186 axons is plotted in figure 6(B), which were recorded from 91 split nerve filaments in 15 sciatic nerves. Usually, there are single-unit recordings in three to five channels in the electrode array and each channel includes 1–4 easily discriminable single-unit spikes. The CV appears to
follow a bimodal distribution ($R^2 = 0.94$) separated by the threshold CV (1 m s$^{-1}$) that divides the A- and C-fibers. The long length of the nerve (∼30 mm) in the

Figure 3. Representative single-unit recordings from mouse sciatic nerve filaments. The custom-built multi-wire electrode array was in contact with five different nerve filaments, each ∼25 μm thick. The 100–150 μm distance between adjacent electrode wires ensure that each of the five channels of recordings are from different nerve axons. Easily discriminable single-units with peak-to-peak magnitude over 10 times the root-mean-square (rms) of the background noise are labeled from 1 to 10.

Figure 4. Robust and repeatable single-unit recordings from 186 mouse sciatic nerve axons. (A) Action potentials were evoked by 10 consecutive stimuli (0.5 Hz) applied to the axons presented in figure 3 (#1–10) overlaid upon one another, illustrating the minimum variation in action potential shapes and latencies. Ten consecutive conduction delays were recorded from each of the 186 axons and the normalized standard deviation of the conduction delays were plotted as a histogram (B); the standard deviation is less than 1.5% in 95% of the axons.

Figure 5. Stable single-unit recordings over 60 min. (A) Two representative recordings conducted 60 min apart from the same nerve filament. (B) There was no significant difference in normalized conduction delays recorded over time (one-way ANOVA, $F_{4,132} = 0.09, p = 0.99$). The number of axons tested at each time point is reported in the bar plot.

Figure 6. Histogram of conduction velocities (CV) of action potentials from 186 mouse sciatic nerve axons. (A) Representative recordings from fast-conducting A-fibers and slow-conducting C-fibers. (B) Of the 186 recorded axons, 49.5% have CV greater than 1 m s$^{-1}$ and are classed as myelinated A-fibers, whereas 50.5% have CV less than 1 m s$^{-1}$ and are classed as unmyelinated C-fibers. The CV histogram follows a bi-modal distribution (continuous curve, $R^2 = 0.94$).
in vivo setup along with the reduced stimulus artifact (<4 ms) permitted recordings from nerve fibers with CV up to 7.5 m s⁻¹. Of the 186 axons, 49.5% had CV over 1 m s⁻¹, which are classed as myelinated A-type axons in mice [31]. The other 50.5% axons had CV less than 1 m s⁻¹ and are classed as unmyelinated C-type axons.

The in vivo setup achieved robust and repeatable single-unit recordings from multiple sciatic nerve axons and we were able to record from a significant proportion of unmyelinated C-fibers, ideal features for an objective test bench for assessing the effect of neuromodulation of peripheral nerves. As a proof of concept, the effect of focused ultrasonic stimulation (US) was assessed using our in vivo setup. As displayed in figure 7(A), single-units evoked by 10 consecutive electrical stimuli (0.5 Hz) were recorded before, immediately after and 15 min after the delivery of focused US stimulation (up to 100 W cm⁻², ~30 s) to the nerve trunk between the stimulating and recording electrodes. A representative recording is displayed in figure 7(B) and summarized data from 15 axons are presented in figure 7(C), showing a significant reduction of the conduction delay immediately after US and complete recovery 15 min afterwards.

**Discussion**

In contrast to the widely available techniques to interface with individual neurons in the brain, studies on electrode arrays to interface with mammalian peripheral nerve axons are comparably scarce. Nerve cuff electrodes outside the peripheral nerve epineurium can only record compound action potentials contributed to by a large number of activated nerve axons [14, 37, 38]. Although penetrating the perineurium, intrafascicular electrode array recordings are biased towards myelinated A-type axons [7, 16]. A recent study employed a floating microelectrode array (Microprobes for Life Sciences) to record from rodent sciatic nerves, but the conduction velocities were not measured and the type of recorded fibers cannot be determined [39]. Needle electrodes used in micro-neurography are capable of penetrating the endoneurium to record from unmyelinated C-type axons, but are limited to single-electrode recordings [6, 40]. A microchannel electrode array fabricated on PDMS was used to interface with rodent dorsal rootlets which, unlike the relatively easy access to peripheral nerves, are protected by the vertebral column and enclosed by the dura mater [41, 42]. The recent advances in intracellular calcium indicators allow optical methods of recording rapid calcium transients as surrogate of neural action potentials. However, image recordings have not been conducted at peripheral nerve axons but only at neural somata in the DRG, and not all DRG neurons have detectable calcium transients [43].

To the best of our knowledge, this study is the first to achieve simultaneous single-unit recordings from mammalian peripheral nerves with no apparent bias towards myelinated A-fibers. Of the 186 axons studied, more than half were from unmyelinated fibers with slow conduction velocity, a much larger proportion than previous reports of intrafascicular recordings using penetrating electrode arrays [7, 16]. Anatomically, the small diameter of C-fibers relative to A-fibers contributes to the weaker total transmembrane currents associated with action potentials, generating comparably smaller extracellular source signals [44, 45]. Thus, it is common to encounter myelinated A-type fibers more frequently than C-type when conducting extracellular recordings from peripheral nerves [46, 47]. The proportion of our recorded unmyelinated fibers is almost comparable to the lower range of the C-fiber proportions reported by
anatomic studies (60–70%) [13], supporting the efficiency of our approach to record from unmyelinated peripheral axons. The maximal conduction velocity (CV) recorded in our in vitro preparation was limited to <7.5 m s⁻¹ by the total length of the sciatic nerve (30 mm) and width of stimulus artifact (4 ms). The histogram of the recorded CV follows a continuous bimodal distribution contributed by A-type and C-type fibers; the distribution tapers as CV is greater than 6.5 m s⁻¹. Previous reports showed that most myelinated axons in mouse spinal saphenous and sural nerves have CVs below 10 m s⁻¹ when recorded at 37 °C [48], comparable to the maximum CV recorded in our setup at room temperature.

Regarding the strength of recorded extracellular action potential spikes from peripheral nerve axons, mathematical models and experimental data have indicated that the amplitude of the single-unit action potential increases inversely as a function of the distance between the recording electrodes and nerve axon [45, 49–52], and directly as a function of axon CV, which correlates with axon diameter and myelination [53, 54]. Thus, the efficiency of our methods to record from unmyelinated axons is likely due to the short distance between the axons and recording electrodes, which is presumably contributed to by the fine nerve splitting to disrupt the endoneurium layers that separate unmyelinated axons from electrodes. Compared to the concentration and duration used to dissociate DRG neurons for patch-clamp recordings [55–58], the strength of collagenase treatment used in the current study is mild and does not cause apparent morphological changes to the nerve trunk. However, collagenase treatment contributes to the increased rate of successful single-unit recordings by reducing the circumferential mechanical strength of the epineurium, and thus facilitates the nerve splitting process by mitigating constrictive damage to the split filaments. In addition to effectively reduced recording noise [59], the large contact area between the micro-wire recording electrode surface and individual split nerve filament increases the likelihood of forming a focal small distance between the axon and electrode. Besides nerve-electrode distance and axon CV, the impedance between the recording and reference electrodes also positively correlates with the amplitude of recorded single-unit action potentials [60]. Thus in this study, the recording amplitude was further enhanced as paraffin oil in the recording compartment effectively increased the impedance of the recording electrode relative to the reference electrode. Following a similar design concept, novel microelectrode arrays can be developed to interface with peripheral nerve axons and achieve reliable recordings which can be conveniently tested using this in vitro setup.

In this study, action potentials generated by suprathreshold electrical stimulation can be reliably recorded as evidenced by negligible variations in conduction delays following consecutive stimuli. Further, the conduction delays recorded from the same axon are robust and repeatable for at least 60 min (the longest time tested in this study), providing an ideal testing bench to assess a variety of neuromodulation and interventional schemes targeting peripheral nerves. As a proof of concept, we tested the effect of focused ultrasound applied briefly between the stimulus and recording electrodes, which caused a significant increase in nerve conduction velocity immediately after ultrasound application that completely recovered within 15 min. The effects of ultrasonic modulation on peripheral neural transmission have been studied previously and the results are contradictory. Tsui et al reported increased nerve conduction velocity following ultrasonic stimulation [61], consistent with our current findings, whereas others reported that ultrasound reduced nerve conduction velocity and attenuated action potential transmission [51, 52]. It is worth mentioning that all prior studies of ultrasonic modulation were based upon measuring changes in compound action potential shapes and amplitude, which are not solely determined by action potential transmission but also are affected by changes in recording conditions likely occurring following mechanical disturbances from ultrasound (e.g., change of nerve–electrode distance). In comparison, the recordings in the present experiments avoided those recording-related confounding factors. With carefully designed experiments, the current setup has the potential to provide convincing data to resolve the aforementioned controversies on ultrasonic neuromodulation of peripheral nerves. In addition, the ability to record multiple single-unit action potentials simultaneously will allow the application of this setup to efficiently study other neuromodulatory strategies including, but not limited to electrical, infrared light, and chemical/pharmacological.

Conclusions

We developed a novel in vitro setup to allow simultaneous single-unit recordings from multiple mouse sciatic nerve axons that are not biased towards recordings of fiber type. An enhanced action potential amplitude from extracellular recordings was achieved by reducing the axon-recording electrode distance via collagenase treatment, fine fiber splitting, and increased contact area between electrodes and nerve axons. This in vitro setup can be used as a test bench to objectively assess the design of next-generation electrode arrays for interfacing with peripheral nerves. This setup can also serve as an objective and convenient platform to study a variety of neuromodulation strategies that target peripheral nerves, including electrical, infrared light, ultrasonic and pharmacological.
Acknowledgments

This study is supported by NIH grant DK100460 awarded to BF. We acknowledge Drs Maritza Abril and Marie E Cantino for their technical support with tissue preparation and electron microscopy imaging at the Biosciences Electron Microscopy Laboratory (BEML) of the University of Connecticut.

ORCID

L Chen https://orcid.org/0000-0001-9018-7734
S J Ilham https://orcid.org/0000-0003-2150-9538
T Guo https://orcid.org/0000-0003-0977-0573
B Feng https://orcid.org/0000-0001-9277-211X

References

[1] Chakravarthy K, Nava A, Christo P J and Williams K 2016 Review of recent advances in peripheral nerve stimulation (PNS) Curr. Pain Headache Rep. 20 60
[2] Joussain C and Denys P 2015 Electrical management of neurogenic lower urinary tract disorders Ann. Phys. Rehabil. Med. 58 245–50
[3] Poo M R, Ecker J, Grossi U, Dang N, Scott S M, O’Neill P R, Jones J P and Knoblauch C H 2014 A systematic review of sacral nerve stimulation mechanisms in the treatment of fecal incontinence and constipation Neurogastroenterol. Motil.: Offl. J. Eur. Gastrointest. Motil. Soc. 26 1222–37
[4] Gorozcenik T and Pang D 2014 Peripheral neuromodulation: a review Curr. Pain Headache Rep. 18 412
[5] Namer B and Handwerker H O 2009 Translational nociceptor research as guide to human pain perceptions and pathophysiology Exp. Brain Res. 196 163–72
[6] Schmelz M and Schmidt R 2010 Microneurographic single-unit recordings to assess receptive properties of afferent human C-fibers Neurosci. Lett. 470 158–61
[7] Davis T S, Wark H A, Hutchinson D T, Warren D J, O’Neill K, Scheinblum T, Clark G A, Normann R A and Gregor B 2016 Restoring motor control and sensory feedback in people with upper extremity amputations using arrays of 96 microelectrodes implanted in the median and ulnar nerves J. Neural Eng. 13 036001
[8] Raspovic S et al 2014 Restoring natural sensory feedback in real-time bidirectional hand prostheses Sci. Trans. Med. 6 222ra19
[9] Jochem T, Denison T and Wolf P 2009 Integrated circuit amplifiers for multi-electrode intracortical recording J. Neural Eng. 6 012001
[10] Li and Andrews R J 2007 Tridimensional nanoelectrode array for precise deep brain stimulation: prospects of a new technology based on carbon nanofiber arrays Acta Neurochir. Suppl. 97 537–45
[11] Wodlinger B, Downey E J, Tyler- Kabara E C, Schwartz A B, Boninger M L and Collinger J L 2013 Ten-dimensional anthropomorphic arm control in a human brain-machine interface: difficulties, solutions, and limitations J. Neural Eng. 12 016011
[12] Fleshers N S, Collinger J L, Foldes S T, Weiss J M, Downey E J, Tyler- Kabara E C, Bensaia S J, Schwartz A B, Boninger M L and Gaunt R A 2016 Intracortical microstimulation of human somatosensory cortex Sci. Trans. Med. 8 361a141
[13] Schmalbruch H 1986 Fiber composition of the rat sciatic nerve Anatomičesk. Rev. 215 71–81
[14] Schuetter M, Donaldson N, Seethooh V and Taylor J 2013 Fibre-selective recording from the peripheral nerves of frogs using a multi-electrode cuff J. Neural Eng. 10 036016
[15] Yilmaz-Rastoder E, Gold M S, Hough K A, Gebhart G F and Williams B A 2012 Effect of adjuvant drugs on the action of local anesthetics in isolated rat sciatic nerves Regional Anesthesia Pain Med. 37 403–9
[16] Clark G A, Wendelen S, Page D M, Davis T, Wark H A, Normann R A, Warren D J and Hutchinson D T 2014 Using multiple high-count electrode arrays in human median and ulnar nerves to restore sensorimotor function after previous transradial amputation of the hand 2014 36th Annu. Int. Conf. IEEE Engineering in Medicine and Biology Society (EMBC) pp 1977–80
[17] Badia J, Borettus T, Andreu D, Azevedo-Coste C, Stiegitz T and Navarro X 2011 Comparative analysis of transverse intrafascicular multichannel, longitudinal intrafascicular and multipolar cuff electrodes for the selective stimulation of nerve fascicles J. Neural Eng. 8 036023
[18] Borettus T, Badia J, Pascual-Font A, Schuettler M, Navarro X, Yoshida K and Stiegitz T 2010 A transverse intrafascicular multichannel electrode (TIME) to interface with the peripheral nerve Biosens. Bioelectron. 26 62–9
[19] Badia J, Raspovic S, Carpaneto J, Micera S and Navarro X 2016 Spatial and functional selectivity of peripheral nerve signal recording with the transversal intrafascicular multichannel electrode (TIME) IEEE Trans. Neural Syst. and Rehabil. Eng.: Publ. IEEE Eng. Med. Biol. Soc. 24 30–7
[20] Serra J, Bostock H and Navarro X 2010 Microneurography in rats: a minimally invasive method to record single C-fiber action potentials from peripheral nerves in vivo Neurosci. Lett. 470 168–74
[21] Donadio V and Liguori R 2015 Microneurographic recording from unmethylated nerve fibers in neurological disorders: an update Clin. Neurophysiol.: Offl. J. Int. Fed. Clin. Neurophysiol. 126 437–45
[22] Gandevia S C and Hales J P 1997 The methodology and scope of human microneurography J. Neurosci. Methods 74 123–36
[23] Yu X, Hu Y and Yu S 2014 Effects of acid on vagal nociceptive afferent subtypes in guinea pig esophagus Am. J. Physiol.: Gastrointest. Liver Physiol. 307 G471–8
[24] Liu Z, Hu Y, Yu X, Ji J, Fan X, Tae C M, Myers A C, Pastuhova P J, Li X and Yu S 2015 Allergen challenge sensitizes TRPA1 in vagal sensory neurons and afferent C-fiber subtypes in guinea pig esophagus Am. J. Physiol. Gastrointest. Liver Physiol. 308 G482–8
[25] Feng B, Li B Y, Nauman E A and Schild J H 2007 Theoretical and electrophysiological evidence for axial loading about aortic baroreceptor nerve terminals in rats Am. J. Physiol. Heart Circ. Physiol. 293 113659–72
[26] Zümermann K, Hein A, Hager U, Katzmann J S, Turnquist P B, Clapham D E and Reeh P W 2009 Phenotyping sensory nerve endings in vitro in the mouse Nat. Protoc. 4 174–96
[27] Bishop T, Marchand F, Young A R, Lewin G R and McMahon S B 2010 Ultrafine- B–induced mechanical hyperalgesia: a role for peripheral sensitisation Pain 150 141–52
[28] Gregersen H, Jiang W, Liao D and Grundy D 2013 Evidence for stress-dependent mechanoreceptors linking intestinal biomechanics and sensory signal transduction Exp. Physiol. 98 123–33
[29] Feng B, Kiyatkin M E, La J H and Gebhart G F 2012 Activation of guanylate cyclase C receptors attenuates peripheral drive from mouse colorectal afferents Neuroscience Meeting Planner, 473.17/HH7 (New Orleans, LA: Society for Neuroscience)
[30] Feng B, La J H, Schwartz E S, Tanaka T, McMurray T P and Gebhart G F 2012 Long-term sensitization of mechanosensitive and -insensitive afferents in mice with persistent colorectal hypersensitivity Am. J. Physiol. Gastrointest. Liver Physiol. 302 G676–83
[31] Feng B and Gebhart G F 2011 Characterization of silent afferents in the pelvic and splanchic innervations of the mouse colorectum Am. J. Physiol. Gastrointest. Liver Physiol. 300 G170–80
[32] Feng B and Gebhart G F 2015 In vitro functional characterization of mouse colorectal afferent endings J. Vis. Exp. (95) 52310
[33] Chen L and Feng B 2016 In vitro multichannel single-unit recordings of action potentials from mouse sciatric nerve Proc. of the Biomedical Engineering Society 2016 Annual Fall Meeting (Minneapolis, MN)
[34] Li Y, Qiao G E, Feng B, Zhao R B, Lu Y J and Schild J H 2008 Electrophysiological and neuroanatomical evidence of sexual dimorphism in aortic baroreceptor and vagal afferents in rat Am. J. Physiol. Regulat., Integrat. Comparat. Physiol. 295 R1301–10
[35] Glauer A 1991 Epoxy resins: an update on their selection and use Micros. AnaL 25 15–20
[36] Harris G R 1985 A discussion of procedures for ultrasonic intensity and power calculations from miniature hydrophone measurements Ultrasound Med. Biol. 11 803–17
[37] Rutten W L 2002 Selective electrical interfaces with the nervous system Annu. Rev. Biomed. Eng. 4 407–52
[38] Rieger R, Schuettert M and Chuang S C 2014 A device for emulating cuff recordings of action potentials propagating along peripheral nerves IEEE Trans. Neural Syst. Rehabilit. Eng.: Publ. IEEE Eng. Med. Biol. Soc. 22 937–45
[39] Vassadevan S, Patel K and Welle C 2017 Bodenst model for assessing the long term safety and performance of peripheral nerve recording electrodes J. Neural Eng. 14 016008
[40] Obreja O and Schmelz M 2010 Single-fiber recordings of unmethylated afferents in pig Neurosci. Lett. 470 175–9
[41] Chew D J et al 2013 A microchannel neuroprosthesis for bladder control after spinal cord injury in rat Sci. Trans. Med. 5 210ra155
[42] Delivopoulos E, Chew D J, Minev I R, Favetti J W and Lacour S P 2012 Concurrent recordings of bladder afferents from multiple nerves using a microfabricated PDMS microchannel electrode array Lab Chip 12 2540–51
[43] Smith-Edwards K M, DeBerry J J, Saloman J L, Davis B M and Woodbury C J 2016 Profound alteration in cutaneous primary afferent activity produced by inflammatory mediators eLife 5 e05207
[44] Dalkilic N and Pehlivan F 2002 Comparison of fiber diameter distributions deduced by modeling compound action potentials recorded by extracellular and suction techniques Int. J. Neurosci. 112 913–30
[45] Qiao S and Yoshida K 2013 Influence of unit distance and conduction velocity on the spectra of extracellular action potentials recorded with intrascleral electrodes Med. Eng. Phys. 35 116–24
[46] Thoren P, Munch P A and Brown A M 1999 Mechanisms for activation of aortic baroreceptor C-fibres in rabbits and rats Acta Physiol. Scand. 166 167–74
[47] McCully B H, Brooks V L and Andresen M C 2012 Diet-induced obesity severely impairs myelinated aortic baroreceptor reflex responses Am. J. Physiol. Heart. Circ. Physiol. 362 H2083–91
[48] Koltzenburg M, Stucky C L and Lewin G R 1997 Receptive properties of mouse sensory neurons innervating hairy skin J. Neurophysiol. 78 1841–50 (PMID: 9325333)
[49] Qiao S, Odoemene O and Yoshida K 2012 Determination of electrode to nerve fiber distance and nerve conduction velocity through spectral analysis of the extracellular action potentials recorded from earthworm giant fibers Med. Biol. Eng. Comput. 50 867–75
[50] Anastassiou C A, Perin R, Buzsaki G, Markram H and Koch C 2015 Cell type- and activity-dependent extracellular correlates of intracellular spiking J. Neurophysiol. 114 608–23
[51] Juan E J, Gonzalez R, Albors G, Ward M P and Izazoequ P 2014 Vagus nerve modulation using focused pulsed ultrasound: potential applications and preliminary observations in a rat Int. J. Imaging Sys. Technol. 24 67–71
[52] Wahab R A, Choi M, Liu Y, Krauthamer V, Zderic V and Myers M R 2012 Mechanical bioeffects of pulsed high intensity focused ultrasound on a simple neural model Med. Phys. 39 4274–83
[53] Struijk J J 1997 The extracellular potential of a myelinated nerve fiber in an unbounded medium and in nerve cuff models Biophys. J. 72 657–69
[54] Gold C, Henze D A, Koch C and Buzsaki G 2006 On the origin of the extracellular action potential waveform: a modeling study J. Neurophysiol. 95 3113–28
[55] La J H and Gebhart G F 2011 Colitis decreases mechanosensitive K2P channel expression and function in mouse colon sensory neurons Am. J. Physiol. Gastrointest. Liver Physiol. 301 G165–74
[56] Shinoda M, La J H, Bielefeldt K and Gebhart G F 2010 Altered purinergic signaling in colorectal dorsal root ganglion neurons contributes to colorectal hypersensitivity J. Neurophysiol. 104 3113–23
[57] Chen X, Molliver D C and Gebhart G F 2010 The P2Y2 receptor sensitizes mouse bladder sensory neurons and facilitates purinergic currents J. Neurosci.: Official J. Soc. Neurosci. 30 2365–72
[58] Beyak M J, Rani B, Krol K M, Kawaja M D and Vanmer S J 2004 Two TTX-resistant Na currents in mouse colonic dorsal root ganglia neurons and their role in colitis-induced hyperexcitability Am. J. Physiol. Gastrointest. Liver Physiol. 287 G485–55
[59] Lempka S F, Johnson M D, Mofitt M A, Otto K J, Kipke D R and McIntyre C C 2011 Theoretical analysis of intracortical microelectrode recordings J. Neural Eng. 8 045006
[60] Wang L, Riss M, Buitrago J O and Claverol-Tinture E 2012 Biophysics of microchannel-enabled neuron-electrode interfaces J. Neural Eng. 9 026010
[61] Tsai P H, Wang S H and Huang C C 2005 In vitro effects of ultrasound with different energies on the conduction properties of neural tissue Ultrasoundics 43 560–5