Tutorial: a guide to techniques for analysing recordings from the peripheral nervous system

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TUTORIAL

Tutorial: a guide to techniques for analysing recordings from the peripheral nervous system

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Abstract

The nervous system, through a combination of conscious and automatic processes, enables the regulation of the body and its interactions with the environment. The peripheral nervous system is an excellent target for technologies that seek to modulate, restore or enhance these abilities as it carries sensory and motor information that most directly relates to a target organ or function. However, many applications require a combination of both an effective peripheral nerve interface (PNI) and effective signal processing techniques to provide selective and stable recordings. While there are many reviews on the design of PNIs, reviews of data analysis techniques and translational considerations are limited. Thus, this tutorial aims to support new and existing researchers in the understanding of the general guiding principles, and introduces a taxonomy for electrode configurations, techniques and translational models to consider.

1. Introduction

Neural control is at the heart of our agency in the world. Through a combination of conscious and automatic processes, the nervous system enables us to regulate ourselves and our interactions with our environment. As we seek to modulate, restore, or enhance these abilities, we have turned in recent decades to technologies that can interface effectively with the nervous system. Certain applications benefit from interfaces at the level of the central nervous system (CNS), for example, when dealing with brain disorders such as Parkinson's, psychiatric conditions [1, 2] or attempting to coordinate patterns of movement through spinal circuitry [3, 4]. In many cases, however, the peripheral nervous system (PNS) offers the advantage of carrying afferent or efferent information that most directly relates to the function of the target of interest.

For most of its history, this field has focused on restoring function lost as a result of amputation [5–7] or paralysis [8, 9]. More recently, the scope has broadened considerably (as reviewed in [10]), including significant interest in chronic disease applications involving the autonomic nervous system [11–13]. These developments have gone hand-in-hand with new progress in neural interfacing technologies and have spawned the related field of *bioelectronic medicine*.

Peripheral nerve interfaces (PNIs) can modulate neural activity through stimulation and/or monitor neural control and feedback through recording. Key potential applications of peripheral nerve recordings include the control of prosthetic limbs through extracted motor commands [14], the closed-loop control of functional electrical stimulation (FES) systems through extracted afferent information [15], and the neuromodulation of body functions through

the identification of autonomic control signal [16] and electrical disease biomarkers [17].

Achieving stable and functional recording in the PNS has been a persistent challenge in neural engineering for several reasons—many of which are distinct from those faced by CNS interfaces. First, in peripheral nerves, the nerve fibres (axons) are tightly packed into fascicles, which are in turn held together by connective tissue to form the nerve trunk [18]. The whole structure, in many cases, has a diameter on the order of hundreds of microns to a few millimetres. In this configuration, the axons are densely organised into a relatively small-diameter structure, making it difficult to isolate activity related to a particular function. Second, sparse firing patterns and the lack of large, synchronised populations result in small-amplitude signals. Third, many peripheral nerves experience significant movement during physical activities (e.g. during respiration in larger mammals), and are often located close to muscles whose bioelectrical activity creates substantial interference. Lastly, in the case of chronic implantation, encapsulation tissue can form in and around an electrode and alter the nerve–electrode interface and thus the amplitude and nature of the recorded signals [19].

Several device designs have been proposed to create reliable PNIs and these are divided into *intraneural* and *extraneural*. The former involves penetration of the device into the nerve trunk, while the latter relies on devices positioned on or near the surface of the trunk [20, 21]. These approaches have been considered in a trade-off between selectivity and invasiveness.

Regardless of the approach, multi-channel PNI designs are increasingly providing new possibilities to extract detailed information about the neural function. The availability of multiple channels provides signal processing opportunities for resolving ambiguities that cannot be dealt with in a single channel, and enables powerful machine learning or regression approaches [14, 22–29]. Several recent reviews have covered peripheral nerve electrode designs [20, 21, 30, 31], but reviews of the data analysis techniques necessary for the creation of effective PNIs are limited [32].

The objective of this tutorial article is to provide a resource that captures the key methods for the analysis of peripheral nerve recordings, and specifically examines the critical interplay between the type of electrode used, the signal analysis techniques applied, and the nature of the information extracted. Further guidance is also given on the importance of appropriate experimental models and the challenges associated with chronic implantation.

The structure of this paper is as follows. Section 2 will introduce general principles and typical approaches to making recordings. Section 3 will introduce the most common electrode geometries and discuss their impact on the nature of information extracted, section 4 will describe the most common signal processing approaches, section 5 will discuss experimental and translational considerations, and section 6 will identify future trends and technologies.

2. General principles

It is important to understand the general principles of a PNI recording system before considering the relative merits afforded by different configurations. The following sections will briefly introduce the properties of peripheral nerve recordings as well as general principles in acquiring and pre-processing these signals.

Figure 1 shows a simplified overview of a typical PNI. Electrodes are placed near, on, or in, the nerve trunk and are connected to low-noise amplifiers (LNAs) and a digital signal processing (DSP) system. The neural signals occurring within the nerve are thus amplified, digitised, and processed to provide an output that can take several forms including: spike trains, continuous time series, and images showing the source of the neural activity within the nerve trunk. This information can then be used to inform a prosthesis such as an artificial limb or a neuromodulation device. It is important first to have an understanding of the nature of neural signals, and so a brief overview will now be given.

2.1. Biophysics of neural signals

Individual axons produce action currents with magnitudes in the pico-ampere range, due to APs, which may be detected as neural signals. These small action currents give rise to small potentials (on the order of 1 μV), that are difficult to detect in the presence of noise and interference. There are two ways to obtain a detectable potential: an electrode must be tiny and in close proximity to the axon, so that the potential is produced across the spreading resistance from a node of Ranvier, or there must be a restricted extra-cellular space that creates a high resistance through which the small action currents flow. These two cases give rise to a taxonomy wherein an interface may be defined as operating with either unrestricted (e.g. intraneural) or restricted (e.g. extraneural) extracellular space.

Neural signals that occur spontaneously (i.e. without external stimulation or modulation) are composed of individual APs resulting from normal biological functions. Typically, neurons within different fascicles innervate unrelated tissue, and do not fire synchronously. Consequently, the observed neural signal is characterised by low amplitude and high frequency activity.

Spontaneous signals recorded extraneurally (i.e. within a constrained extracellular space) from the surface of the nerve trunk are rarely larger than 30 μV peak-to-peak [33], whereas intraneural signals recorded from inside the trunk can be over 100 μV

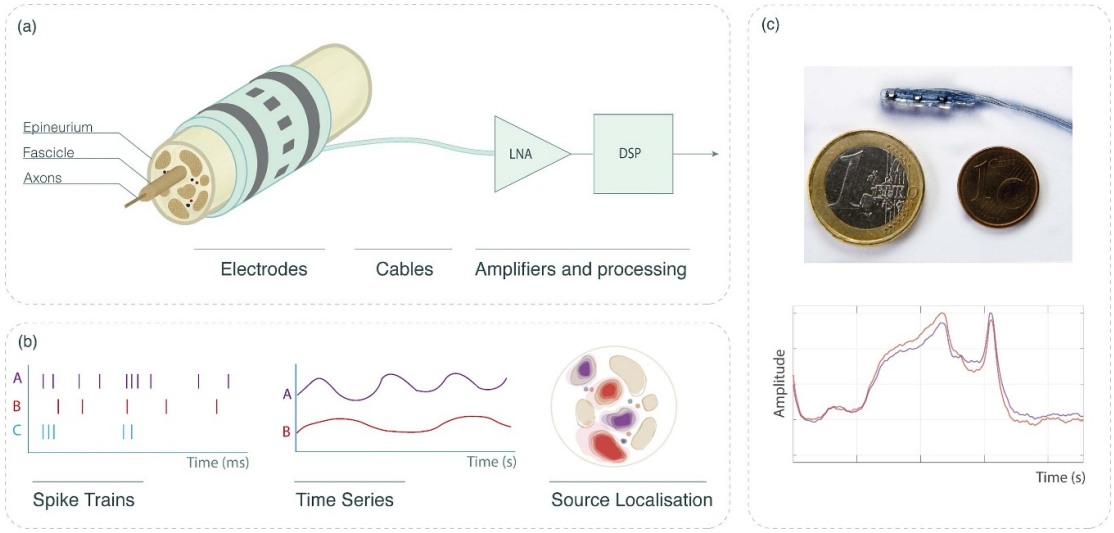


Figure 1. A simplified overview of a peripheral nerve recording system. Panel (a)—electrodes placed near the nerve are connected to low-noise amplifiers (LNAs) and then to a digital signal processing (DSP) system. The LNA and DSP stages may be implanted or secured externally depending on the experiment. Panel (b)—exemplar outputs from the recording system: spike trains, wherein individual action potentials (APs) can be identified and labelled; time series extracted by integrating some statistical feature over time; localised sources that can be aligned to a known fascicular structure. Panel (c)—photograph of a three-electrode nerve cuff and an example time domain signal recorded from such an electrode after processing.

peak-to-peak [34]. Most of the signal power is concentrated in the range of 300 Hz–5 kHz, with the peak below 3 kHz [35–37].

The small amplitude of the neural signal creates significant challenges, especially against the background of instrumentation noise (on the order of 2–4 μV root-mean-square (RMS)). Adding considerably to this challenge is the interference from nearby muscles (electromyographic (EMG) activity), which can be an order of magnitude larger than the neural signal when both are recorded using a monopolar reference. The EMG bandwidth (approximately 5–500 Hz) has partial overlap with the neural signal, precluding effective removal using linear filtering without loss of information.

Neural signals may also be directly evoked, or modulated, by mechanical, chemical, or electrical stimulation. When this occurs, many axons produce APs simultaneously, and the resulting neural signal (the evoked compound AP (eCAP)) is the result of the superposition of these APs. In this case the amplitude of the eCAP will be much larger than that of a single AP ($\sim$100 μV for cuffs), see figure 6(a) for an exemplar recording of both spontaneous APs and eCAPs.

2.2. Amplification and acquisition

At the amplification and acquisition stage, a well-chosen reference montage can be used to minimise the contributions of interfering sources. The most widespread example of this approach is the tripolar arrangement, in which the signals at the end electrodes are averaged and used as a [33]. This configuration helps suppress EMG interference by taking advantage of the linearisation of these electric fields along the length of the recording array [38], and can be implemented through several alternative differential recording arrangements [39]. For intraneural recordings both monopolar and bipolar approaches have been reported [40, 41]. Examples of the monopolar, bipolar, and tripolar amplifier configurations are given in figure 2.

Acquisition is generally less critical, and can be performed by most good quality analogue-to-digital converters with sufficient sampling rates (typically >30 kHz), although techniques that measure the conduction velocity of the neural signals may require supra-nyquist sample rates to adequately sample fast APs if the inter-electrode distance is small.

2.3. Signal pre-processing and denoising

Noise and interference in neural recordings can arise from a several sources such as interfering muscle activity [38], movement artefacts causing audiophonic noise or triboelectric noise, noise from a high impedance ground, or electromagnetic noise [26]. Using the appropriate signal processing techniques can help minimise noise and interference.

At the data pre-processing stage, bandpass filtering is commonly applied to isolate the neural signal, with a high-pass frequency in the 250 Hz–1 kHz range and a low-pass frequency in the 3–7.5 kHz range. If the objective is simply to detect the presence of neural activity, a rectified-bin-integration (RBI) approach can be applied, in which windows that contain neural activity, as well as noise, produce higher values than those that only contain noise [42]. This approach, however, is accompanied by a loss of temporal resolution that may preclude the use of many of the techniques described in the following sections.

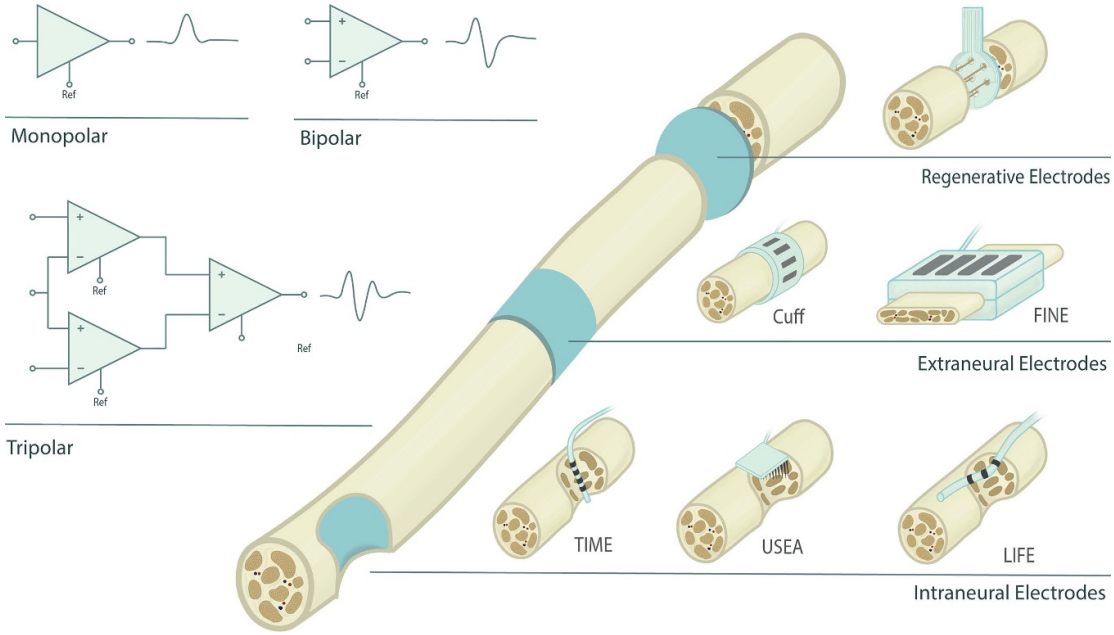


Figure 2. Amplifiers are typically configured as either monopolar, bipolar, or tripolar. The choice of configuration has an impact on the morphology and amplitude of the recorded signal. In each case a reference electrode is required that is usually placed distal to the recording site. Arrays of electrodes can be connected as individual monopoles or as isolated or shared dipoles or tripoles. Electrodes may be intraneural (TIME, USEA, LIFE), extraneural (Cuff, FINE) or regenerative [20, 21].

An additional and more sophisticated pre-processing option is wavelet denoising, which relies on transforming the noisy data into an orthogonal time-frequency domain, thresholding the wavelet coefficients to remove the noise, then transforming back to the original time domain. Selection of the mother wavelet and thresholds are key considerations, but reports have varied about which choices are optimal for neural signals [35, 41, 43].

3. Electrodes

The geometry and configuration of the electrodes used to obtain a recording are critical in determining the type of signals that can be measured. Selecting the most appropriate electrode material, structure, configuration, and geometry for a specific application requires an in-depth understanding of the advantages and disadvantages of each. Ciancio *et al* [44] have summarised these requirements, and from their work and the wider literature it is possible to identify two key characteristics of a PNI.

Selectivity—Selectivity refers to the ability of the PNI to stimulate, or record from, specific axons, fascicles, or nerves, whilst being insensitive to off-target axons, fascicles, or nerves. Depending on the application, the desired stimulation selectivity may differ from the recording selectivity; for example, two different electrodes may be used to stimulate and record in the same prosthesis and the overall selectivity may vary from one application [7] to another [45].

Stability—Equally important is the stability of the PNI, both for acute and chronic applications. It should be stable over time and should inflict as little physiological or histological damage to the tissue as possible [46]. These properties are governed by factors such as the mismatch of mechanical properties between the tissue and the electrode, as well as the immunological reaction of the tissue to different materials and surface treatments [47].

3.1. Electrode location

3.1.1. Extraneural electrodes

Extraneural electrodes (those that constrain the extracellular space) are placed either in the vicinity of the nerve trunk or in direct contact with the epineurium (the outer sheath of the nerve trunk). These types were originally microelectrodes, or simple hooks, with the interface placed in an oil bath, and have evolved into cuffs [48] and the flat interface nerve electrode (FINE) [47]. The former type inherently records APs from one or a few axons, the latter is more-or-less sensitive to all axons within the lumen. The extracellular space is constrained by ensuring that the interface is snug around the nerve trunk, thus the resistance of the extracellular space is increased along with the detected potentials arising from the small action currents. Thus extraneural interfaces provide an interface with generally low selectivity [21]. Their primary advantage is stability as they do not penetrate the epineurium and thus are less likely to cause immediate damage to the nerve.

One exception to this is the microchannel interface, in which the nerve trunk may be micro-dissected into fascicles that may then be placed within channels that constrain the extracellular space [49]. This results

in fewer axons per channel and typically higher signal amplitudes ($\sim$100 μV).

3.1.2. Intraneural electrodes

Intraneural electrodes are normally implanted directly inside the fascicles, penetrating the perineurium, and show better selectivity than extraneural electrodes as they have closer contact with the fascicles. Examples of these include thin-film longitudinal intrafascicular electrodes (LIFEs) [50], and transverse intrafascicular multi-channel electrodes (TIMEs) [51]. In terms of stability, their invasiveness is higher than extraneural electrodes and the implantation itself may cause damage [21, 52, 53].

3.1.3. Regenerative interfaces

Regenerative interfaces are often designed to interface with small groups of axons, allowing for selective stimulation and recording with the best possible level of selectivity. Instead of penetrating the nerve with the electrode, the nerve is transected and then supported to regrow through a structure containing electrode channels [54]. In one embodiment, the structure resembles a sieve consisting of a piece of material with multiple micropores covered with a conductive material. After transection, each end of the nerve is placed on either side of the sieve and the axons grow through the micropores. A schematic representation of this is shown in figure 2.

It has been shown that neurons will regenerate through the sieve structure, and the sieves are functional as both recording and stimulation devices with high selectivity. However, regenerative electrodes may result in incomplete or constrained regeneration of axons, leading to difficulties with chronic implantation [55, 56].

3.1.4. Electrode configuration

Several recent papers review the biocompatibility and stability of different electrode materials and structures [31, 57]. However, from the point of view of neural recordings, it is helpful to consider a taxonomy of geometries—as it is the spatial relationship between the signal source and the electrodes, alongside the constraining of the extracellular space, that often defines the recording selectivity and capability.

Electrodes may be grouped to form *point measurements* (single channel), *one-dimensional linear arrays* (organised circumferentially, transversely, or longitudinally), or *two-dimensional linear arrays*. Figure 3 illustrates these geometries using an extraneural cuff interface. The substrate (i.e. insulating tube) of the cuff and FINE interfaces constrains the extracellular space and serves to maintain the spatial relationship between the electrodes. Intraneural electrodes may, or may not, have stable fixation and so while the geometries are applicable to all interfaces, the spatial stability should also be considered.

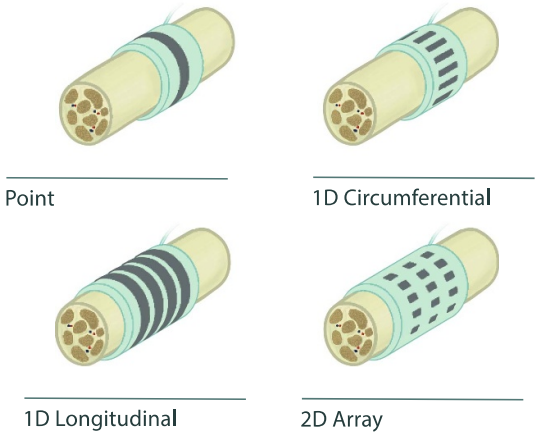


Figure 3. Examples of the four electrode geometries that give rise to either single channel or multichannel recordings represented in a cuff electrode. The reference (not shown) is normally a separate electrode located in the tissue.

3.2. Point measurements

One-dimensional point measurements are by far the most common and lend themselves to a wide array of signal processing techniques. Point measurements, in this paper, refer to measurements produced from a single recording channel (i.e. an observation at a single spatial location) that may have been referenced by a monopolar, bipolar, or tripolar configuration.

3.3. One-dimensional arrays

One-dimensional arrays can be formed by placing multiple electrodes either longitudinally, transversely, or circumferentially. Thus, both temporal and spatial classification becomes possible.

Temporal classification—relies on the fact that the propagation of the AP may be observed by placing an array of electrodes located longitudinally along the length of the nerve. Different properties of the APs, such as conduction velocity (which is proportional to axon diameter), may then be used to discriminate APs from individual axons, or types of axons [58, 59]. These approaches do not require any prior knowledge about the AP morphology, and many can improve the signal-to-noise ratio (SNR) of the recording by averaging over multiple recording channels.

Spatial classification—relies on the fact that an array of electrodes arranged circumferentially can selectively identify activity from within different fascicles or axons, based on the spatial location of each electrode with respect to the neural source. Passive recording approaches include source localisation and types of beamforming, many of which do not require prior knowledge about the expected morphology of the AP but do require high SNRs to localise activity to individual fascicles [60].

3.4. Two-dimensional arrays

Circumferentially and longitudinally spaced electrodes can be combined to form a two-dimensional

structure that enables the observation of APs in both space and time [61].

Spatiotemporal classification—combines the benefits of longitudinal and circumferential arrays for a more comprehensive and robust characterisation of the APs. Classification can be performed using templates, or by training a convolutional neural network (CNN) to recognise the spatiotemporal patterns associated with specific neural activity [22].

4. Analysis techniques

Section 3 introduced a taxonomy of electrode geometries and explained how the choice of geometry impacts the available signal processing methods—including temporal, spatial and spatiotemporal. Several key analysis techniques will now be introduced and discussed in the context of *content extraction*. The information of interest in peripheral nerve recordings can be broken down more broadly into two categories: *anatomical* and *functional*.

4.1. Anatomical content

Anatomical content pertains to the size, shape, type, and positions of structures within the nerve. In terms of PNI, anatomical information can be provided by making observations of the propagation direction (afferent versus efferent), axon type (e.g. Aδ, C fibres), and the spatial location of neural sources.

Throughout this paper, *direction sensitivity* refers to the ability to discriminate afferent versus efferent, *velocity* refers to the conduction velocity of each AP, and *location of neural sources* refers to the determination of the spatial location of the source of the neural signal within the nerve. Anatomical content is most readily obtained using extraneural electrodes, as they provide a macro view of the activity within the entire nerve, as opposed to intraneural electrodes that offer a more microscopic (and thus spatially localised) view.

4.2. Functional content

Functional content within a nerve pertains to a specific organ or system. For example, signals within the ulnar nerve may encode sensation (via cutaneous afferents) from the hand and forelimb. The overall presence (e.g. a non-selective power measurement) of neural activity can also be considered functional content. Functional interpretation of a signal can be made without knowledge of the anatomical underpinnings (e.g. where exactly in the nerve an axon is located) but does require knowledge of the nerve's innervation.

A common approach to extracting functional content is to identify single unit activity (i.e. APs that result from specific axons) in a process called spike sorting, wherein individual APs are grouped into clusters based on morphology. The resulting single unit labels (spike trains) can then be used directly in a neuroprosthesis without reference to any anatomical organisation. Spike sorting is most used in intraneural electrode configurations wherein the SNR is high and individual APs can be observed. Figure 4 illustrates this process for exemplar data with three active axons.

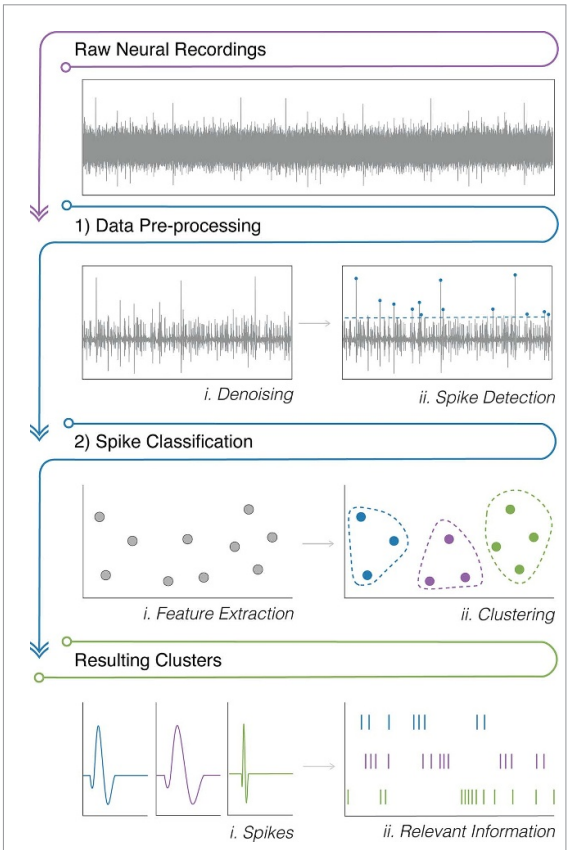
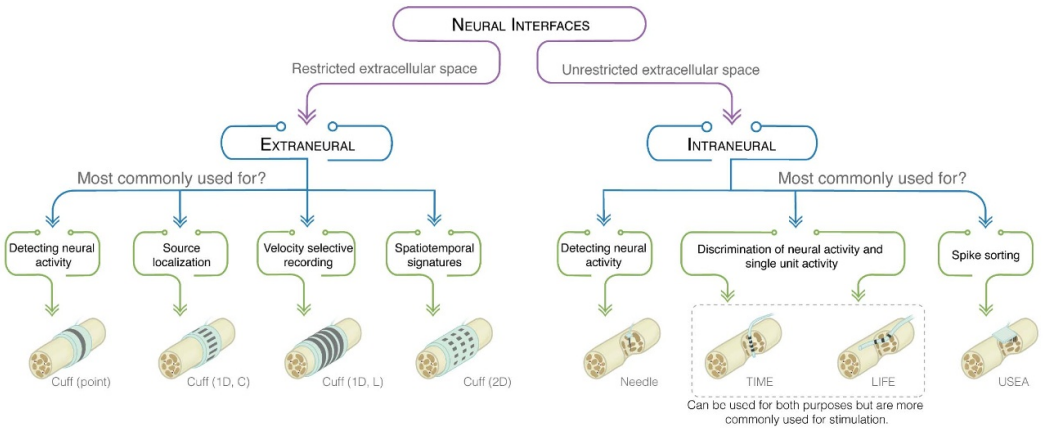


Figure 4. Typical spike sorting pipeline. Raw recordings are filtered, spikes are detected, extracted, and clustered.

More generally, multi-unit single axon AP trains, eCAPs, or signal windows from either intraneural or extraneural recordings can be associated with a particular function of interest through classification or clustering approaches. In this paper the task of associating neural signals with different functional events is termed *discrimination of neural pathways*.

In some cases, certain information can overlap anatomical and functional content. For example, the location of neural sources refers to the spatial location of the source within the nerve, but with some *a priori* information, a functional aspect can be determined. In the popular rat sciatic nerve model, identifying neural sources in the tibial nerve would correspond to dorsiflexion of the ankle while identifying neural activity in the peroneal nerve would correspond to plantarflexion of the ankle.

Deciding what information (anatomical vs functional) to obtain depends largely on the application of interest. If the intended use is focused on understanding the underlying physiology, anatomical content will likely be more applicable. On the other hand, functional content may be of more interest if the intended use is to obtain a control signal (i.e. to use in a neuroprosthetic device).



	Type of Information / Electrode Geometry	Low SNR				Medium SNR		High SNR	
		Cuff - Point	Cuff – 1D C	Cuff – 1D L	Cuff – 2D	TIME	LIFE	MEA	Needle
Anatomical / Functional	Directionality			★	✓	✓	✓	✓	
	Velocity			★	✓		?	?	
	Location of Neural Sources		★		✓	?		?	
	Nerve Imaging		★		?				
	Single Unit Identification					✓	✓	★	✓
	Discrimination of Neural Pathways	✓	✓	✓	★	✓	✓	✓	✓
	Detection of Neural Activity	★	✓	✓	✓	✓	✓	✓	✓

★ - indicates the most common usage, ✓ - indicates demonstrated usage, ? – indicates potential usage but not seen in the literature

Techniques / Methods	Applicable Electrode Geometry				Anatomical + Functional Content				Functional Content		
	Point	1D Longitudinal	1D Transverse / Circumferentially	2D	Directionality	Velocity	Location of Neural Sources	Nerve Imaging	Single Unit Identification	Discrimination of Neural Pathways	Detection of Neural Activity
Time series Analysis	X	X	X	X						X	X
Choice of Reference Configuration	X	X	X	X	X						X
Template Matching	X	X	X	X	X				X		X
Velocity Selective Recordings		X		X	X	X					X
Source Localization			X	X			X				X
Spatiotemporal Signatures		X	X	X	X	X				X	X
Electrical Impedance Tomography			X	X				X		X	X

Figure 5. Taxonomy and selection guide for electrode geometry, applicable information that can be obtained from the selected geometry and the type of signal processing technique that could be applied.

4.3. Content extraction techniques

Once the information of interest is known, the appropriate electrode configuration and data analysis techniques can be employed. Figure 5 introduces a taxonomy of the different methods and the applicable electrode configurations that can be used to determine anatomical or functional content. For example, if one is interested in imaging (producing a cross sectional image with the location of neural sources identified), then a 1D–circumferential cuff could be employed alongside the electrical impedance tomography (EIT).

4.3.1. Time series analysis

The most common way of analysing neural signals still lies in using time-domain, frequency-domain, or statistical features without necessarily relying on any aspect of the anatomical information (i.e. fibre velocity, spatial location) directly.

Time domain techniques to smooth the neural signal over a window can be used to detect neural activity [62]. The most common approach is the RBI operation, where the signal is rectified, binned into time windows, and integrated. This technique extracts the signal's envelope and enables a simpler and smoother signal for analysis compared to the noisier raw recordings. This signal can then be thresholded to determine if a neural source of interest is active.

Other techniques involve calculating features within the window for identifying neural activity. The mean absolute value, RMS, and variance are some examples of time-domain features, whereas features derived from the power spectral density are examples of frequency-domain features that can be used for identifying neural activity [45, 60, 63].

Another approach for identifying neural activity is to observe the statistical properties of the signal and noise. For example, the autocorrelation matrix of white noise will be diagonal because the samples are completely uncorrelated. Therefore, the eigen-decomposition of this matrix will yield a single

non-zero eigenvalue. In contrast, a recording containing both neural data and noise will have off-diagonal elements in the autocorrelation matrix, and thus more than one eigenvalue. The difference between the greatest and smallest eigenvalue can be used to detect neural signals [42].

These techniques are simple and effective for detecting neural activity but alone are often inadequate for obtaining more sophisticated functional or anatomical content. However, if the dominant application is to detect neural activity, these techniques are typically sufficient and can be quickly and easily implemented.

4.3.2. Choice of reference

Reference choice has a significant impact on the recording quality of a PNI as it can attenuate interfering signals and can also be used to determine the direction of propagation (i.e. afferent vs efferent signals) without the need for other techniques or methods.

In extraneural recordings, a bipolar recording [64, 65] will exhibit a reverse signal shape when comparing afferent and efferent activity, but this configuration is more susceptible to noise sources than the more common tripolar configuration. Recent work by Sabetian and Yoo suggests the use of a tetrapolar configuration [24] (a bipolar recording of the outputs of two consecutive tripoles), which demonstrated improved SNR over a bipolar or tripolar configuration alone and can be used to identify an afferent signal versus an efferent signal.

4.3.3. Template matching

Template matching is a technique that involves comparing a signal to a known template. This technique is typically used in spike sorting approaches to separate detected APs [66, 67]. Spike templates that represent each neuron are created and can be used to classify new APs based on their similarity (i.e. shape). This approach can also be used to discriminate neural activity when incorporated into a matched filter approach [61].

The main advantage of template matching is that it is amenable to implementation on an online system, but its effectiveness is reduced if the templates are similar or the number of distinct sources increases. Another drawback occurs when AP shapes overlap in time [68] and thus choosing templates that represent the underlying neural activity may not be straightforward. The use of multi-contact electrodes has helped mitigate the effects of this issue.

4.3.4. Velocity selective recording

The concept of velocity discrimination is founded on the fact that the conduction velocity of an AP is a function of the axon properties, all of which are assumed to be either time invariant (e.g. myelin thickness, diameter, membrane properties) or tightly regulated (e.g. temperature, ionic concentrations) [69]. The axon diameter and myelin's presence (or lack thereof) are the main factors that determine the difference in conduction velocity from one axon to another.

The velocity of an AP can be computed by delaying the signals recorded from each element of a longitudinal array relative to one another by an interval that corresponds to the conduction velocity before summing together [70, 71]. One advantage of this process is the ability to distinguish afferent and efferent neural activity by simply applying a negative delay.

The first demonstrations of velocity selective recording were made using electrical stimulation to recruit large amplitude eCAPs, in worm [71] frog, pig, and rat [27, 28, 72–74]. The *delay-and-add* process is used to provide a spectral representation of eCAP recordings, where the spectrum is presented with respect to velocity rather than frequency. Figure 6(a) shows a time domain eCAP recording and the corresponding velocity spectrum.

However, in the analysis of spontaneous neural activity, APs are not necessarily coincident. Thus, the overall recorded amplitudes are significantly reduced compared to the case of the eCAP and the velocity spectrum, whilst a useful tool for the analysis of eCAPs, is not suitable for the problem of naturally evoked or occurring (spontaneous) CAPs. Figure 6(b) illustrates an exemplar recording of spontaneous neural activity and demonstrates the stark contrast to the eCAP of figure 6(a). Accordingly, methods have been developed to process spontaneous neural activity that make use of a blend of array processing and image processing techniques to convert the recordings into conventional spike trains [58].

4.3.5. Source localisation and beamforming

Most, if not all, techniques for locating neural sources derive from source localisation and beamforming techniques that are related to inverse problems in electroencephalography [75]. Briefly, the inverse problem of bioelectric source localisation is based on the equation:

$$\boldsymbol{d} = \boldsymbol{L}\boldsymbol{j} + \boldsymbol{\varepsilon} \tag{1}$$

where $\boldsymbol{d}$ is an $M \times 1$ vector containing the recorded data from M electrode contacts, $\boldsymbol{j}$ is an $N \times 1$ vector whose entries represent the magnitude of the current dipoles distributed in the region under consideration, and $\boldsymbol{L}$, known as the *lead field* matrix, is an $M \times N$ matrix whose entries represents the influence of a unit current dipole on the potential recorded at a particular electrode. $\boldsymbol{\varepsilon}$ is an $M \times 1$ vector of additive noise.

The objective is to recover $\boldsymbol{j}$ based on the measurements of $\boldsymbol{d}$ and estimate of $\boldsymbol{L}$.

Beamforming is a signal processing technique that uses spatial filters that combine recordings made at different spatial locations to enhance (via constructive interference) the selectivity of the recordings.

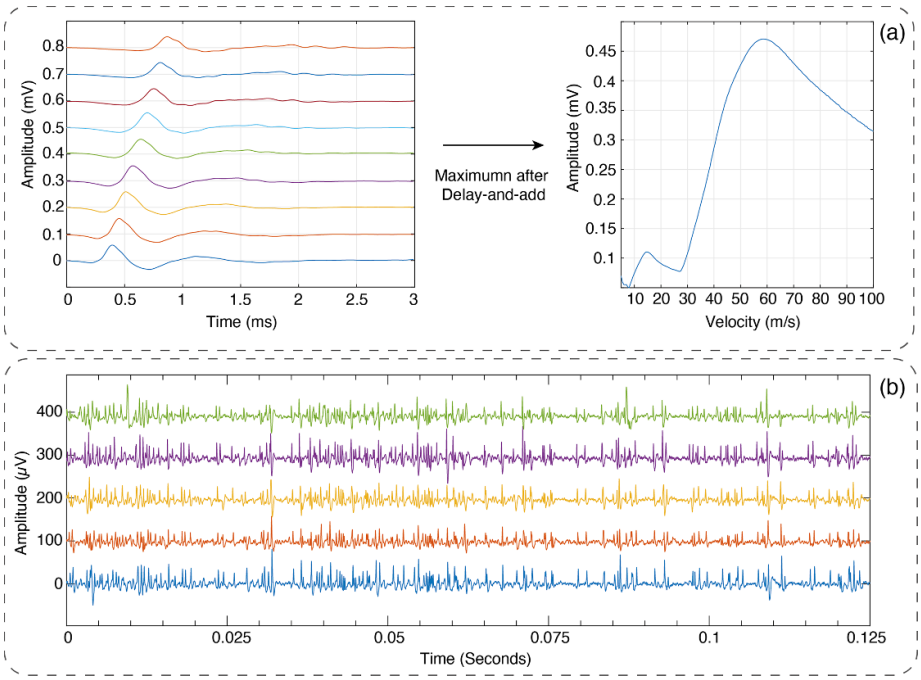


Figure 6. An example of an eCAP, the left panel shows the time domain propagation (and dispersion) of the eCAP along a recording array. The eCAP is formed by the summation of many lower amplitude APs that constructively interfere. Also present, but less clear, is a second slower eCAP that occurs approximately 1 ms after the first. The right panel shows the intrinsic velocity spectrum computed from the same recording using the delay-and-add process, showing the two velocity peaks that correspond to each eCAP. (b) An example of spontaneous neural activity, which by contrast with the eCAP contains individual APs (with much lower amplitudes) propagating and overlapping (but not dispersing) along the recording array. Reprinted from [58], Copyright (2020), with permission from Elsevier.

When tuned correctly, this can be used to localise neural sources within the nerve. The advantage of this technique is that it can provide both anatomical (the location of the neural activity within the nerve) and, with some *a priori* information, functional content. However, solving the inverse problem is non-trivial and in small nerves the neural sources of interest are much closer in distance, reducing the ability to localise them.

Most source localisation approaches have been attempted with extraneural electrodes [25, 26, 29, 76–82]. An approach using FINE electrodes has shown the most promising results, demonstrating the feasibility of real-time implementation in chronic recordings involving canines [26, 82].

4.3.6. Spatiotemporal signatures

The concept of a neural spatiotemporal signature was first introduced in [22, 61, 83]. This technique involves extracting neural recordings from a group of fibres associated with a particular function (e.g. a motor command to a single muscle) using a 2D array of electrode contacts (longitudinally and circumferentially). This observed spatiotemporal signature can be associated with the neural pathway that produced it. Thus by identifying the correct spatiotemporal signature, discrimination of neural activity can be achieved (i.e. afferent versus efferent, flexion versus extension, etc).

The main advantage of this technique comes from the direct integration of the temporal and spatial information allowing for a more comprehensive characterisation of neural activity. These spatiotemporal signatures have been used as an input to a CNN which demonstrated the ability to discriminate naturally evoked CAPs (nCAPs) and be used to reconstruct firing patterns of different neural pathways [22]. This network is known as the extraneural spatiotemporal compound AP extraction network or ESCAPE-NET. The overall ESCAPE framework can be seen in figure 7 alongside an example of a spatiotemporal signature.

This technique has shown promising results in an acute rat model. A recent simulation study [84] aimed at mimicking chronic conditions suggested that the selectivity of the spatiotemporal signature can be maintained by establishing a recalibration schedule for the classifier.

4.3.7. Electrical impedance tomography (EIT)

EIT is a method that can provide fascicular-level selectivity with an extraneural approach using electrodes distributed around the nerve. EIT is an emerging medical imaging technique in which changes in the impedance of a conductive volume, such as a nerve, may be imaged using an array of external electrodes [85, 86]. In this method, a flexible, cylindrical, multi-electrode cuff is placed around a nerve, and the

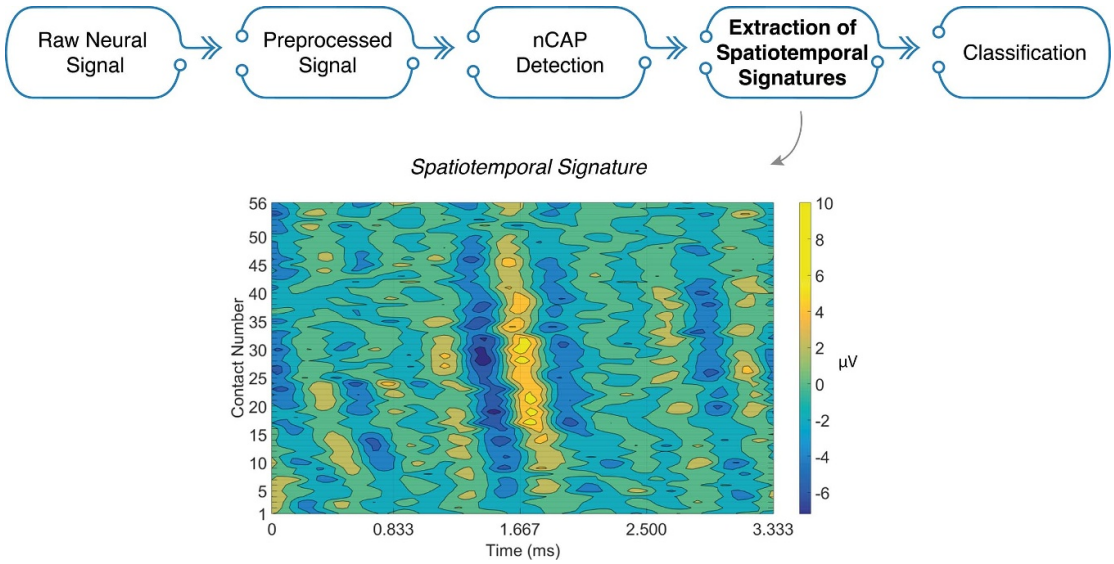


Figure 7. The ESCAPE framework, which consists of the following steps. Raw neural signal: a neural signal is recorded using an implanted extraneural electrode. Pre-processing: the raw neural signal is then pre-processed with denoising and referencing techniques to improve the SNR. Naturally evoked CAP (nCAP) detection: improved SNR signal is then used to locate nCAP locations. Extraction of spatiotemporal signatures: nCAPs are extracted from the pre-processed signal and represented as spatiotemporal signatures. Classification: a classifier uses these spatiotemporal signatures to discriminate neural pathways of interest. Adapted from [22].

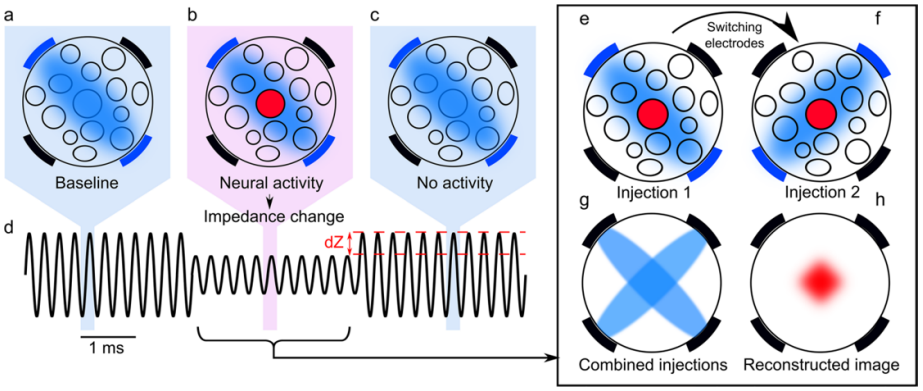


Figure 8. The principle of fast neural EIT in a 'model' peripheral nerve. (a)–(d) The impedance change occurs during neural activity (b) with respect to the background (a) and (c), which is measured by passing a constant current through a pair of external peri-neural electrodes and recording the voltage response on the remaining electrodes (d). Typically, cuffs have 16–32 electrodes; just 4 are shown here for explanatory purposes. (e) and (f) The pair is then switched from one to another and the paradigm is repeated covering all possible electrode combinations. (g) These measured transfer impedances can be reconstructed into tomographic images using principles similar to x-ray back projection. (h) Reconstructed images of the neural activity have a resolution of less than 100 μm and 0.3 ms with current methods using 6 kHz applied current and 16 electrodes in a nerve 1.5 mm in diameter. Reproduced from [86].

imaging technique of fast EIT is applied to image the activity within the fascicles. Changes in impedance caused by small decreases in bulk tissue resistance occur as ion channels open and close, and these can be detected (with some averaging) using the external electrodes.

Mathematically, EIT is similar in principle to inverse source localisation. However, EIT has significant advantages including: more independent data (for N electrodes, there are $O(N^2)$ independent measurements at a time compared to O(N) for inverse source localisation), a potentially unique solution, no field cancellation problem, and no theoretical limitations on the accuracy [87–89].

The principle of operation is that small currents (typically 30 μA at 6 kHz) are injected between different pairs of electrodes, while the resulting voltage is measured at every other pair. This process is repeated over all electrodes to produce a set of measurements that can be re-constructed into an *image* of the nerve. Figure 8 illustrates the principle of fast neural EIT in a model peripheral nerve. An impedance change associated with neural activity occurs in figure 8(b) relative to the baseline cases in figures 8(a) and (c).

This change in impedance can be observed as a change in the measured voltage of a pair of electrodes to a current applied in another pair of electrodes in figure 8(d). Multiple measurements of this change can be used to produce an image of the activity in figures 8(e) and (h). EIT is an effective recording approach for eCAPs, wherein the averaging process can readily be performed, but has yet to be demonstrated with spontaneous neural signals.

The signal processing and recording methods presented in this paper are given as examples, and new methods will undoubtedly be developed in due course. However, the taxonomy in figure 5 should provide the reader with a powerful tool for selecting the electrode geometry and thus the *type* of information that can be extracted by the different methods. Having now covered the fundamentals of neural recording and introduced a range of electrode geometries and signal processing methods, the following section will cover the translational considerations and experimental challenges associated with animal models.

5. Translational considerations

The previous sections have introduced the fundamentals of peripheral nerve recordings, from the biophysics of the APs, through the electrode geometry, and then the signal processing. This section will introduce the animal models typically used to develop and test PNIs and discuss the potential translational issues using the context of the cervical vagus nerve as an example target.

5.1. Experimental models

Most of the development of neural interfaces is performed using animal models, in either *acute* or *chronic* experiments. There are both ethical and scientific reasons for using animal models; however, these models also lead to several associated translational challenges. A recent detailed review of animal models used for PNI development can be found in Aman *et al* [91].

5.1.1. Acute experiments

In acute surgical experiments, the electrodes (recording, stimulation, or both) are implanted in anesthetised animals and the target nerve is explored over the course of a few hours whilst the animal remains unconscious [17, 25, 72, 83, 92, 93]. The animal is then terminated at the end of the experiment. Acute experiments are beneficial because they provide a stable platform for interrogating the nervous system that includes any and all organs or tissues of interest, whilst removing several of the challenges associated with chronic experiments such as movement artefacts and the need for either percutaneous connectors or a wireless telemetry system [38].

However, the anaesthetic regime employed for the surgery has an influence on the electrophysiology that remains unclear, even though the molecular mechanisms of anaesthetics are well characterised. For example, it is known that both isoflurane and ketamine, two widely used anaesthetic paradigms, differentially impact sensory processing in the mouse primary visual cortex [94]. The complex interactions between the anaesthetic agents, and their compound effect on the electrophysiology of the animal, remain problematic and may hinder the direct translation of results to awake animals.

5.1.2. Chronic experiments

In chronic experiments the electrodes are implanted under general anaesthesia, but the animals are then *recovered* for a period of time (from days to months) [26, 95–97]. Recording may take place during the implantation surgery, while the animal is awake and freely moving, or during a termination surgery. The former and the latter have the advantage of not requiring percutaneous connectors or a wireless telemetry system but suffer the same drawbacks as acute experiments. Recordings made whilst the animals are awake and freely moving are, for many applications, the most representative but also by far the most challenging. One advantage of using percutaneous connectors is that measurements of the electrode impedances may be made directly, and then related to the recorded signal amplitudes.

It is important that an appropriate species be chosen for the model, based not just on the ease of the experiment but also on any translational opportunities or issues. If the goal of the experiment is to elucidate some fundamental electrophysiologic property or phenomena, then a well-characterised species such as mice or Xenopus Laevis frogs may be suitable. Likewise, the development of electrode materials and the associated instrumentation may not depend greatly on the species and so mice or rats may be chosen based on reasons of cost or simplicity. However, if the goal—at any point in time—is to translate the research into humans, then the choice of model is critical. The difference between the peripheral nerves of humans and other mammals is not obvious; differences may exist in gross anatomy and geometry of the nerves, the type and level of fascicular structure and vascularisation, and the distribution of myelinated versus unmyelinated axons [98, 99].

A good example of the difference between species can be found by considering the cervical vagus nerve. The vagus is the tenth cranial nerve and for many years has been the focus of significant interest as a neuromodulation target for the treatment of diseases as broad-ranging as intractable epilepsy [100, 101], diabetes [102, 103], and rheumatoid arthritis [104–106]. Figure 9 shows the comparative anatomy of the cervical vagus nerve between mouse, rat,

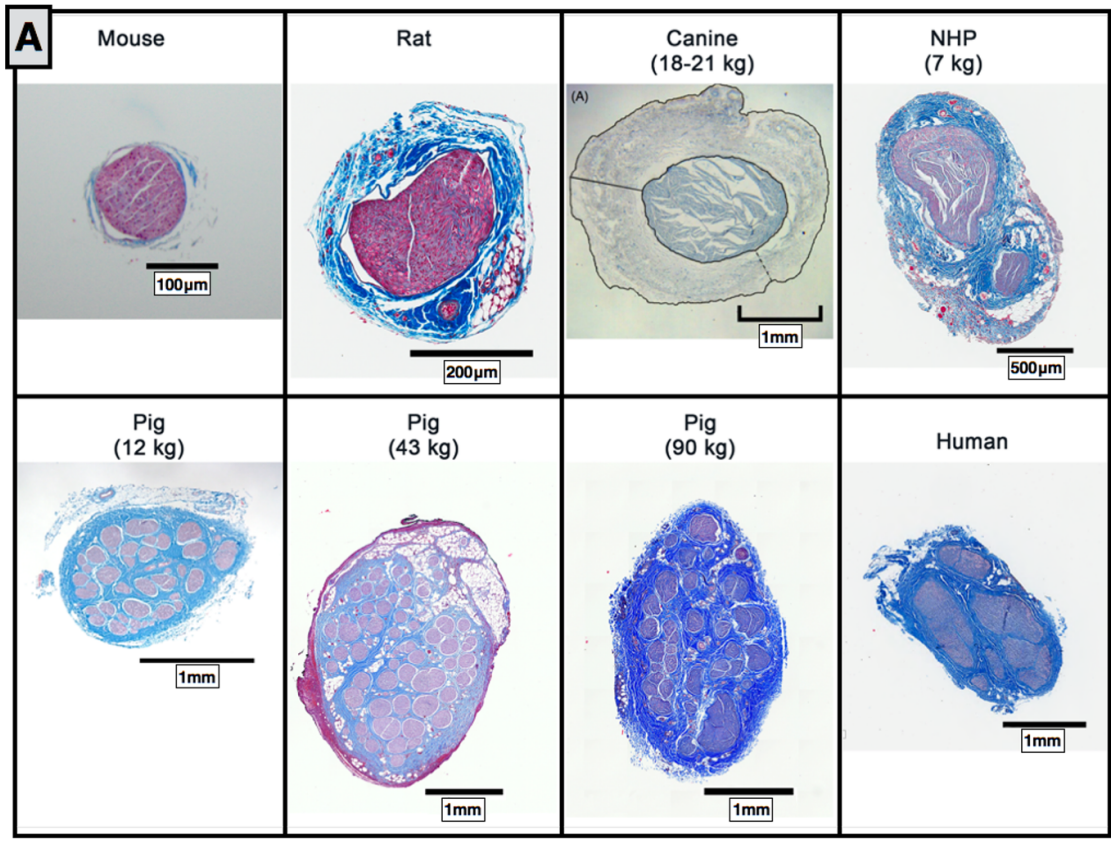


Figure 9. Comparative anatomy of the cervical vagus nerve between mouse, rat, canine, non-human primate (NHP), pig and human illustrating significant variation in both size and fascicular structure between species. Reproduced from [90]. © The Author(s). CC BY 4.0.

canine, non-human primate (NHP), pig and human [90]. Each of these will be discussed in the context of the individual animal model.

5.1.3. Rodents and small animals

Rodents, typically mice or rats, have been the staple model for biomedical research for a long time. They are well characterised, easy to care for, and have the benefit of being available in a multitude of different inbred or outbred strains. From an experimental perspective, they can be anaesthetised and maintained without complex anaesthetic equipment. Indeed in rats and mice it is commonplace to use only a single intraperitoneal administration of an agent such as urethane [107]. The peripheral nerves are, of course, physically much smaller than in humans. On the one hand this can make the surgical approach to the nerves relatively straightforward, as there is minimal tissue or bone to remove. On the other hand, it makes the design and placement of electrodes far more difficult, thus limiting the number of electrodes that can be placed as well as the separation between stimulation and recording electrodes. The latter point is important as an increased separation between the stimulation and recordings sites is one method for reducing or eliminating stimulation artefacts from recordings.

Animals such as guinea pigs, cats, and rabbits are an attractive middle ground between rodents and larger animals but are much less explored in the literature on electrophysiology. Cats have seen the greatest attention [65, 108, 109]. Aside from potential inexperience in working with these species, there is likely to be a stronger perceived ethical concern associated with species that are companion animals. Interestingly, this has led to the implantation of neuromodulation devices in companion animals as a therapy, for example the Brindley device has been implanted in companion dogs to restore urinary bladder control [110].

Turning to the cervical vagus as an example target, it is smaller in mice ($\emptyset \approx 100\,\mu$m) than in rats ($\emptyset \approx 200\,\mu$m), and typically consists of only 1–2 fascicles (figure 9(a)). The canine vagus is larger again ($\emptyset \approx 2-3$ mm) and has the thickest epineurium across most models, resulting in a greater distance from either the stimulation or recording electrodes placed on the epineural surface to the axons. This in turn would likely alter the stimulation thresholds and the amplitudes of the recorded neural signals. In the case of mice, rats, and canines, the fascicular structure is far less complex than that in humans—further complicating any translation of design parameters from one to the other.

5.1.4. Large animals

Large animals—in this context sheep and pigs—have long been common-place in neuromodulation research, although their prevalence is in part limited by their comparatively complex requirements for both housing and surgical management. Unlike rodents, sheep and pigs are generally not as well characterised, and are typically obtained from commercial farmers or kept in small groups for breeding purposes. Their housing and husbandry requirements are not necessarily more difficult than those of rodents but do require greater space and nutritional resources. Pigs in particular are frequently reported in experimental research and have been for many decades [72, 73, 101, 111].

Compared to rodents, the surgical approaches to the peripheral nerves of pigs and sheep can be more complex and time consuming, although the gross anatomy and the approach, in general, have the potential to be more directly translated to humans. Anaesthetic induction and maintenance is complex, and almost always requires a dedicated anaesthetist and mechanical ventilation providing volatile anaesthetics such as sevoflurane or isoflurane [72]. Agents such as fentanyl, propofol, and ketamine may be administered before or during surgery, typically as a bolus, although continuous rate infusion of propofol is possible [112]. The impact of these agents on the electrophysiology of the animals is more challenging in part due to the number of agents administered.

Returning to the vagus nerve as an example target, it is of similar size ($\emptyset \approx 2$ mm) in pigs, sheep, and humans [113]. The number of fascicles in the vagus nerve of pigs and sheep is on the order of 30, whereas in humans it is closer to 10. There is a similar thickness of epineurium and level of vascularisation across the three species. Thus, pigs and sheep represent appropriate models for developing electrodes, instrumentation, and surgical approaches that might be translated into humans. The distance between the axons and electrodes placed on the epineurium is likely to be similar, so stimulation thresholds and recorded signal amplitudes ought to be comparable.

5.1.5. Humans and non-human primates (NHPs)

Experiments involving either humans or NHPs are the least common, due in large part to the ethical and regulatory issues surrounding them. In humans, most experiments are performed as part of a larger rehabilitation package wherein the patient is implanted with a therapeutic device that may also collect data, or as part of a clinical trial for an innovative technology. Good examples of this include PNIs that are used for either controlling upper-limb prosthesis or for proving sensory feedback from a prosthesis—such experimental interfaces have been implanted in humans for several weeks or months [6, 7, 114, 115].

NHPs have predominantly been used in experiments that target the CNS, wherein single unit recordings can be made from awake, behaving animals using high-density electrode arrays [116]. The surgical approaches used in NHPs are most like those used in humans and other large mammals, an attribute that both aids potential clinical translation but also raises the costs and complexities of experimentation. One significant benefit with most NHPs is that they can be trained to perform purposive motor tasks (such as reaching and grasping), and this has made them popular in various areas of sensorimotor research, for example in the development of PNIs for the control of prosthetic limbs [117] or the use of optogenetic modulation to improve motor functionality [118]. However, there is often significant training time (and thus cost) associated with the development of purposive experimental paradigms [91].

Returning to the vagus nerve as an example target, the vagus nerve in NHPs is smaller than that of the human ($\emptyset \approx 500$ μm) with notably less complex fascicular organisation [113]. The number of fascicles in the vagus nerve of NHPs is typically only 1–2, but there is a similar (relative) thickness of epineurium and level of vascularisation. Both the marked differences in vagus nerve diameter and fascicular organisation make it challenging to appropriately design a translatable interface for this nerve, although the NHP model can be valuable for the testing of chronic implant stability. From both an economic and ethical standpoint, NHPs should only be used when development of an interface is sufficiently advanced so that the benefits of this model, like complex movement behaviour and similarity to human biomechanics, are scientifically mandatory.

5.2. Chronic recordings

There are several points during a chronic experiment when neural recordings can be made: during implantation surgery, whilst awake and freely moving, and during termination surgery. There is intrinsic value in each of these, and many breakthroughs in the chronic stability of the neural interfaces have been made without recording from an awake animal—see for example the recording of bladder activity in rats [49]. However, it is often the goal to record neural signals whilst the subject is awake and freely moving. The challenges associated with doing this are multiple and span from husbandry and care to electrical interference and noise.

Fundamentally, the recording system needs to measure the potentials at the electrodes and communicate this to a remote device for processing or logging. The most straightforward way of achieving this is via percutaneous connectors; implanted cables are routed from the electrodes to the connector, and external instrumentation can then be connected on an ad-hoc basis [26]. Challenges include healing of the wound surrounding the connector, risk of damage due to normal or abnormal animal behaviour, and mounting the connector at a suitable anatomic site.

Electrical issues can arise from the length of the cable that may be required between the electrodes and the amplifier, or from triboelectric effects from the cables themselves [26].

Implanting the instrumentation is an attractive alternative to percutaneous connectors, for example using a telemeter [119]. In this scenario, the instrumentation, potentially including a power source and a wireless communication system, is packaged in an implantable device that is located near the recording electrodes. Data storage is likely to be limited, and in most cases a receiving device must be placed on the skin over the site of the telemeter to enable data collection.

Finally, there are fundamental challenges associated with the movement of the animal. Modern amplifiers are available with very high common-mode rejection ratios (CMRRs), and so in theory, should be more than capable of rejecting common-mode signals from nearby muscles. In practice, however, differential electrode impedances and the filter networks that might be placed before the amplifiers, will degrade the CMRR. Thus, in recordings from awake and freely moving animals, it remains very challenging to ensure that recordings are not contaminated by interference from nearby muscles.

6. Future directions

6.1. Closed-loop interfaces

Information extracted from peripheral nerve activity can be used to improve functional or health outcomes. This relationship typically takes the form of closed-loop stimulation, where the timing, pattern and/or location of stimulation are continuously adjusted based on the measured neural activity. The translational success of peripheral nerve recording techniques is therefore closely tied to closed-loop neuromodulation systems. Several studies in animal models have demonstrated the control of FES based on limb position or state estimates derived from peripheral nerve recordings, using both extraneural [120, 121] and intraneural [122] recordings. Early examples in humans included the regulation of grasping strength in a hand neuroprosthesis based on volar digital nerve feedback [8], and foot-drop correction using sural nerve recordings [123], both using nerve cuff recordings.

More recent work in humans demonstrated decoding of motor intention combined with sensory stimulation, using slanted microelectrode arrays [14]. While most of these efforts focused on neuroprosthetic applications, a study by Plachta *et al* is noteworthy for demonstrating an application related to bioelectronics medicine, namely the control of blood pressure using selective recording and stimulation of the vagus nerve [16].

Despite these examples, the number of studies that have demonstrated closed-loop stimulation based on peripheral trunk recordings remains low, although a substantial number of studies cite this concept as their motivation. This gap can be partially attributed to the challenges with obtaining informative and stable peripheral nerve recordings. Another notable challenge is the need for strategies for removing stimulation artefacts to effectively coordinate recording and stimulation. Artefact removal in this context may require a combination of hardware (e.g. blanking systems to prevent amplifier saturation during stimulation) and signal processing components [124], which increases the complexity of the instrumentation required and contributes to the low number of studies. Nonetheless, the variety of techniques described in this article and the recent acceleration of the field suggest that we are at a turning point in this regard and artefact rejection signal processing schemes that demonstrate a 25–40 dB rejection in the artefact are now available [125].

6.2. Autonomic nerve applications

Most of the techniques described in this manuscript have been developed for sensorimotor applications, such as the control of prosthetic limbs. However, there is a growing interest in the ability to record from smaller fibres within the autonomic nervous system (such fibres are frequently of interest in pain research). Recent advancements in hardware (e.g. more compact electrodes and multiple electrode contacts) have led to opportunities for recording from these small autonomic nerves. Thinly myelinated and un-myelinated axons (e.g. C-fibres) conduct APs more slowly (<2 m s^{-1}) than larger fibres, and the resulting AP and CAP amplitudes are much smaller. Thus, it is challenging to record C-fibre activity without employing a very selective interface (e.g. micro-needles [126]) or using supra-maximal stimulation to recruit all of the C-fibres simultaneously and thus maximise the amplitude of the resulting eCAP.

Microneurography, being the easiest way to identify single C-fibre APs, employs a needle that is able to record single or compound APs [127]. This approach allows for unambiguous recording and recognition of C-fibre APs. Microneurography is beneficial for developing a better mechanistic understanding of peripheral nerve encoding and modulation but is not suitable in most cases for chronic use.

Novel electrode designs have also been developed for the purposes of recording from small-fibre nerves, but novel signal processing techniques for obtaining small-fibre activity are limited [128–130]. This is mainly because validation of small-fibre activity is extremely challenging due to the low signal amplitude and the slow conduction velocity. Velocity selective recording (VSR) may be of significant benefit in this application as the enhancement of the SNR of the recorded signal will maximise the likelihood of observing APs from small-fibre activity, and both

the conduction velocity and the direction can be validated. After locating these small-fibre APs using VSR, template matching and spatiotemporal signatures may be able to separate activities of interest, but no studies using signal processing techniques have shown this possibility.

Interfacing with these slower-conducting nerve fibres will require new or adapted recording and analysis methods. These techniques will need to either enhance the SNRs of the recorded signals to address the low signal amplitudes or be able to isolate small-fibre activity. It is likely that a combination of novel electrode design and signal processing approaches will be needed to overcome these challenges to facilitate the growing interest in recording from the autonomic nervous system.

6.3. Alternative paradigms

Electrical recording remains the dominant modality for recording from the PNS. Whilst neuromodulation via direct electrical stimulation is common (including in clinical applications), few, if any, clinical devices record from peripheral nerves. This is despite a clear need driven by the desire to achieve closed-loop, time-invariant, neural interfaces within the PNS.

There has been some (limited) engagement with active modalities (such as EIT) and with optogenetics [131, 132], but conventional electrical recording using either intraneural or extraneural electrodes remains the dominant modality. Many nerve interfaces (such as cuffs) are claimed to be chronically implantable—however, these claims require further verification. Stimulation electrodes need only be placed close to the target nerves, and so a cuff can be placed quite loosely around the nerve. Conversely, recording electrodes need to be placed in direct contact with the epineurium (to maximise recorded signal amplitudes), and so have the potential to cause considerable damage when implanted long-term. There are few [26, 48] studies describing the results of chronic implantation of recording electrodes.

At the same time, closed-loop neuromodulation is complicated by the fact that large stimulation artefacts—caused by current flowing from the stimulation electrodes onto the recording electrodes—often saturate the recording amplifiers. Some solutions to this problem exist in the form of either signal processing or electrode balancing, but the problem remains.

A novel approach is to record the magnetic fields produced by the axons rather than the electric field. This would have the benefit of not requiring any direct contact with the nerve, as the magnitude of the observed magnetic field is not related to the contact impedance, instead the location is driven by the SNR required. This concept of magnetoneurography has been demonstrated both *ex-vivo* and *in-vitro*. Okawa *et al* used large arrays of superconducting quantum interference devices to record neural activity transcutaneously from both the brachial plexus and the carpal tunnel area during electrical stimulation [133, 134]. In the case of the brachial plexus, it does not appear that any stimulation artefact was present, but in the case of the carpal tunnel area, the authors were forced to use the most distal stimulation site and to employ an artefact rejection tool (common spatial pattern). Barbieri *et al* performed an *in-vitro* demonstration of recording from mouse muscle, and also developed a model for the magnetic field contribution from the two axial components in the case of a muscle bundle [113]. They recorded using giant-magnetoresistance (GMR) sensors and stimulated with a suction pipette. Despite the proximity of the stimulation electrode and the GMR sensors, they reported no stimulation artefacts.

7. Conclusions

Interest in recording from the PNS continues to be growing, and as more advanced techniques are developed, it becomes more challenging to know which method to adopt. Thus, this tutorial has introduced a taxonomy of electrode configurations and recording techniques that will support the selection process and can be augmented as new methods are developed. The important aspects of selecting an appropriate animal model, as well the challenges associated with acute and chronic recordings, have been introduced. This tutorial provides a good foundation in peripheral nerve recordings for both the novice and the experienced researcher as the field continues to develop.

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Ethical statement

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