ADInstruments

Application Note

Life Science

Recording Natural Activity in Small Nerves

This application note describes the use of the PowerLab system, with a Bio Amp and Chart software, to record physiological activity in small nerves of anaesthetised animals.

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Introduction

Natural physiological activity in peripheral nerves is of interest in both research and teaching. Recordings from whole nerves show summed overlapping activity of thousands of fibres; individual spikes cannot be identified. To see individual spikes, the nerve must be divided into filaments or thinned by micro-dissection[1].

Equipment

The ML 132 Bio Amp is well suited for recording from small nerves. Differential amplifiers designed for nerve recording are available from other manufacturers, and may be used in place of the Bio Amp. Choice of data acquisition unit depends on whether individual spikes in myelinated fibres are to be resolved:

- E-series PowerLab units allow continuous sampling at up to 1000 samples/s.
- S-series PowerLab units have a high continuous sampling rate (up to 100,000 samples/s) that permits fast spikes to be recorded. Computed Input functions available on S-series units include ‘Abs smoothed’ and ‘RMS smoothed’, which are useful extra options for quantifying nerve activity.

Electrodes

A variety of electrodes may be used to record from living nerves. Hooks made of platinum or silver wire (approximately 0.4 mm in diameter) may be placed under the nerve, one near the cut end and the other 5–10 mm away. To maximise signal amplitude and prevent drying, the nerve and electrodes are usually placed in a paraffin pool or, more conveniently, surrounded by silicone gel.

Alternatively, the cut end of a whole nerve can be taken up into a closely-fitting suction electrode[2].

The animal should be connected to a common or ground electrode – often a strip of silver or a hook electrode, buried in the musculature.

Connections to Bio Amp

Connections should normally be made to the Bio Amp input socket, shown in Fig. 1. The socket has two isolated ground pins; it is convenient to use one for shielding, and to connect the common electrode from the animal to the other.

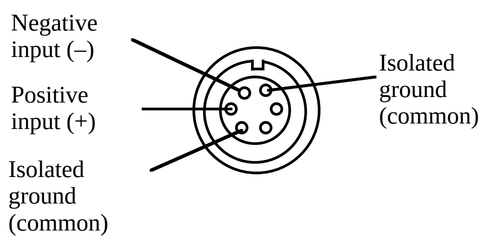


Figure 1. Pin connections to Bio Amp input socket (front view). The two Isolated ground pins are joined together internally.

Instead of connecting to the Bio Amp directly, it is possible to connect electrodes to the Bio Amp lead (Fig. 2). However, the capacitance between wires in this long cable is several hundred pF, which may significantly slow and attenuate fast spikes recorded with small electrodes.

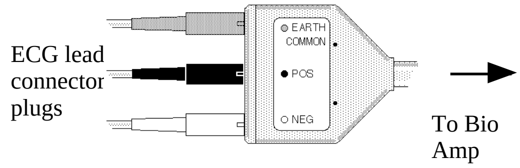


Figure 2. Pin connections of Bio Amp lead supplied with Bio Amp. Wires to electrodes can be soldered to cut-off ECG connector leads.

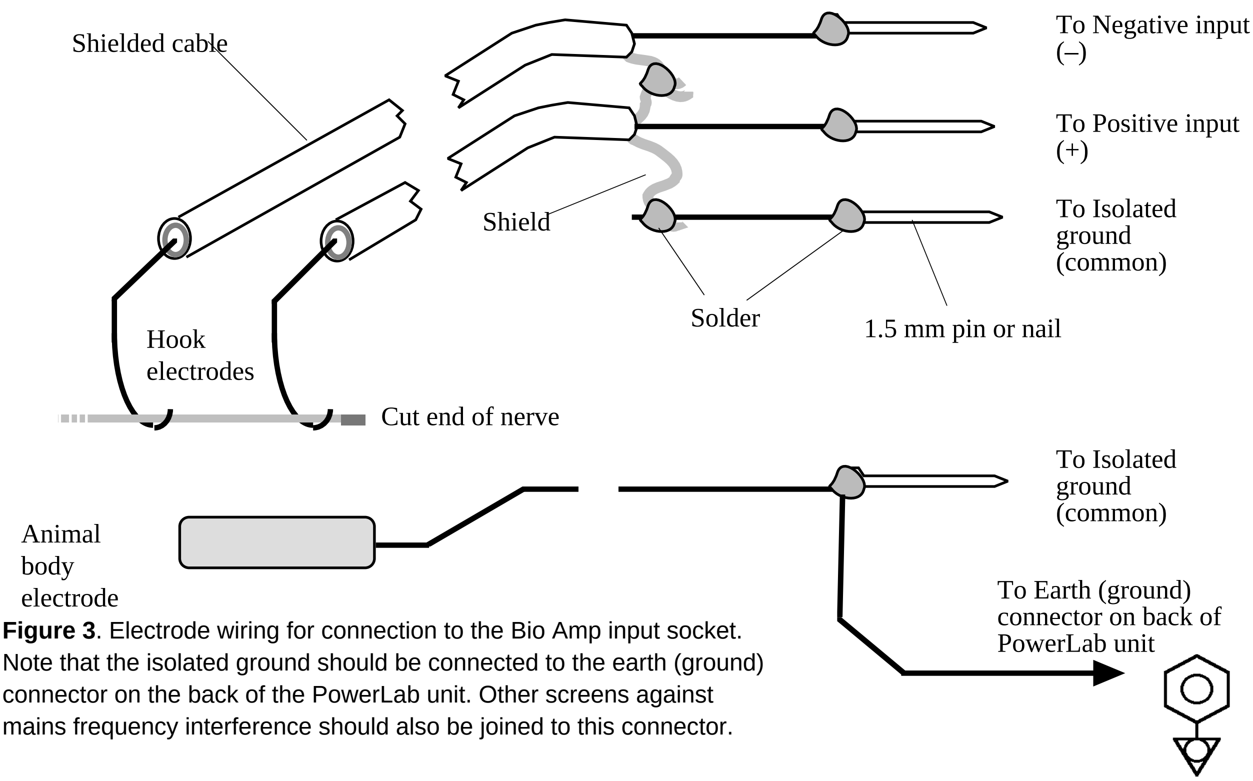
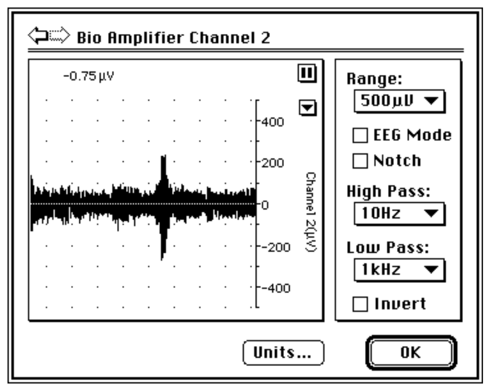


Figure 3. Electrode wiring for connection to the Bio Amp input socket. Note that the isolated ground should be connected to the earth (ground) connector on the back of the PowerLab unit. Other screens against mains frequency interference should also be joined to this connector.

The wire connections from electrodes to the Bio Amp should be made with shielded cable, as shown in Fig. 3. To minimise the capacitance of the cables, they should be kept as short as practicable (say 30–60 cm). The wires are soldered to pins or nails (1.5 mm diameter and 10–15 mm in length) that can be inserted in the Bio Amp input socket.

Figure 4. Suggested Bio Amp settings. Interference at the mains frequency may be reduced by turning the Notch filter on. For a sampling rate of 1000/s, the Low Pass filter frequency should be set to 500 Hz or 1 kHz as shown.



Setting up

Suggested initial Bio Amp settings are shown in Fig. 4. The High Pass filter frequency should be 10 Hz. The Range (that is, amplifier gain) should be adjusted so that neural activity is clearly visible in the display area, but does not reach the top or bottom limits.

A sampling rate of 1000/s in Chart is sufficient to record the summed activity of slowly-conducting fibres in a whole nerve, for example a renal sympathetic nerve (Figs 5 and 6).

For recording individual spikes in dissected nerve filaments, a sampling rate of 2000, 4000 or 10k/s is likely to be needed. E-series PowerLab units can record at these rates in 'burst' mode on one channel only; S-series units can record continuously on several channels. The Bio Amp's Low Pass filter frequency should be set to 2 kHz or 5 kHz for these faster recordings.

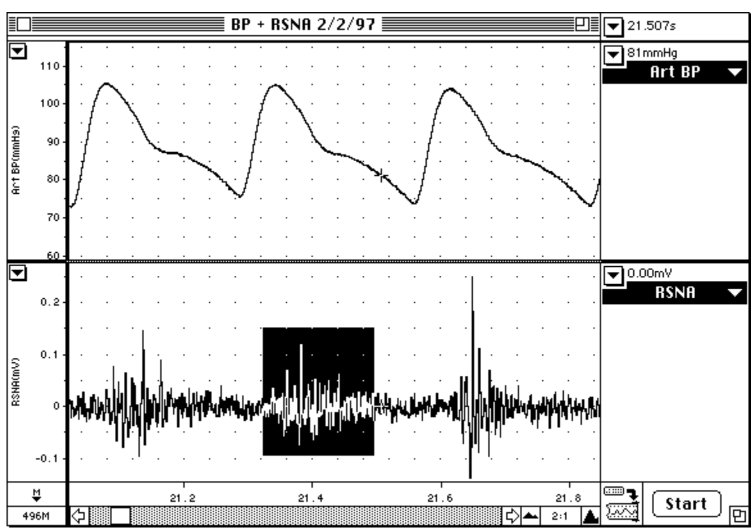


Figure 5. Rabbit blood pressure(Art BP)and renal sympathetic nerve activity (RSNA), recorded at 1000 samples/s. An enlarged view of the selected burst of activity is shown in Fig. 6.

Quantification of neural activity

Activity recorded from nerves is generally biphasic (Fig. 6). Amplitude quantification takes place in two steps: inversion of the negative-going deflections (either by taking the absolute value of the waveform, or by squaring it), and then some form of averaging over time. Several methods are available in Chart. Some are off-line calculations, and some are on-line computed functions.

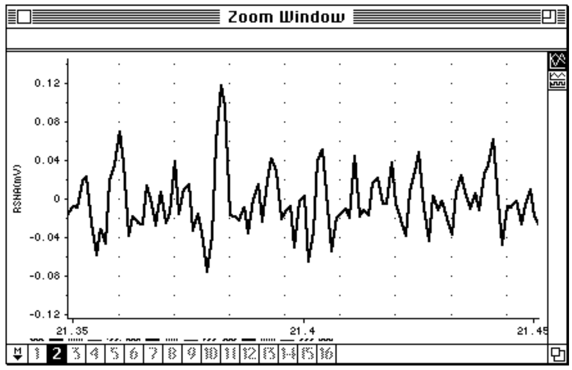


Figure 6. Zoomed view of sympathetic nerve activity from Fig. 5. Spikes from many fibres are superimposed and overlapped.

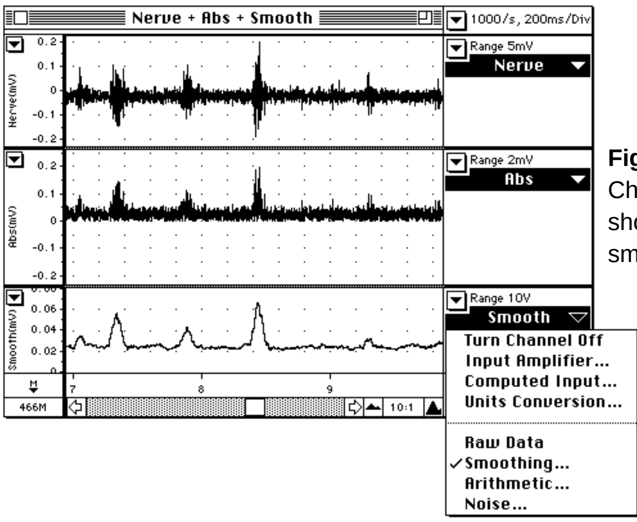


Figure 7. Quantification of nerve activity. Channel 1 shows raw trace. Channel 2 shows absolute value. Channel 3 has smoothed absolute value.

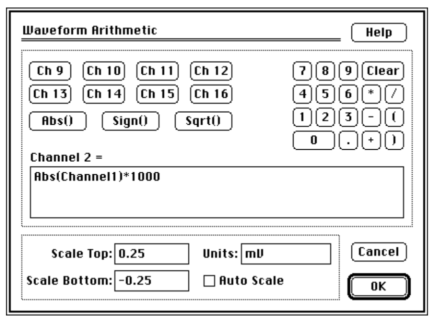


Figure 8. Waveform Arithmetic dialog box, showing settings to calculate the absolute value of Channel 1 and display it in Channel 2.

Off-line calculations

Off-line calculations have the advantage that they can be applied after the experimental results have been recorded. The settings can be adjusted by trial and error for best results.

- Absolute value by Waveform Arithmetic then Smoothing. See Figs 7, 8, and 9. This method uses standard Chart calculations, and has the advantage (for teaching) of displaying the intermediate absolute value of the waveform.
- The 'Noise' Chart extension (Figs 10 and 11) is more versatile and precise in the time averaging settings. It can display either the standard deviation of the signal or its variance.

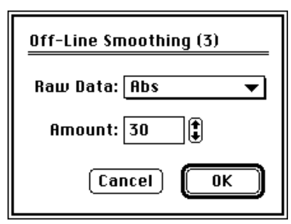


Figure 9. Smoothing dialog box, showing settings to calculate Channel 3 in Fig. 7.

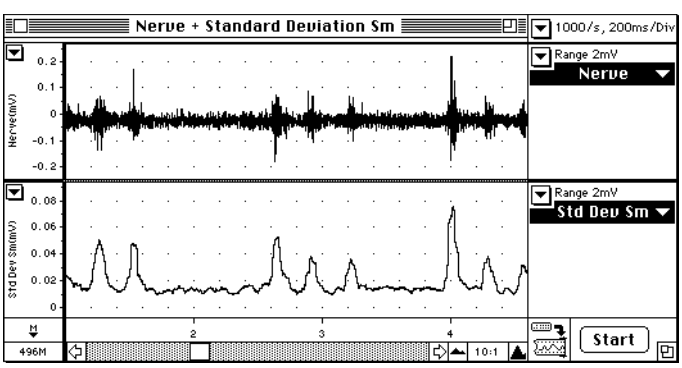


Figure 10. Quantification of nerve activity with 'Noise' Chart extension. Channel 1 shows raw trace. Channel 2 shows the smoothed standard deviation.

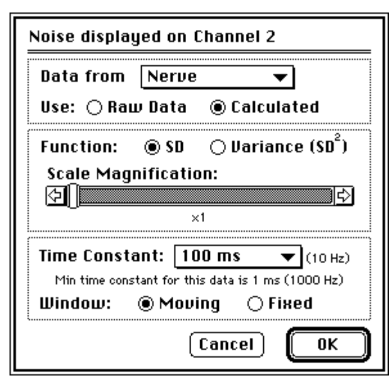


Figure 11. The 'Noise' dialog box.

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On-line computed functions

In these methods, the measure of nerve activity is computed at the time of data acquisition. The raw signal is sampled internally at 2000/s, independently of Chart's sampling rate, and the smoothed or integrated measure is computed from it. It is not necessary to record the raw nerve activity itself (which requires a high sampling rate). The computed input function can be sampled quite slowly (20–100/s), so that files are small and manageable, even when they contain recordings made over long times.

- Abs Smoothed computed input function (S–series PowerLabs only). See Fig. 12.
- RMS Smoothed (S–series PowerLabs only). RMS means root-mean-squared; the computations are similar to those of the 'Noise' Chart extension.
- Integral(Absolute Value) computed input function. See Fig. 13.

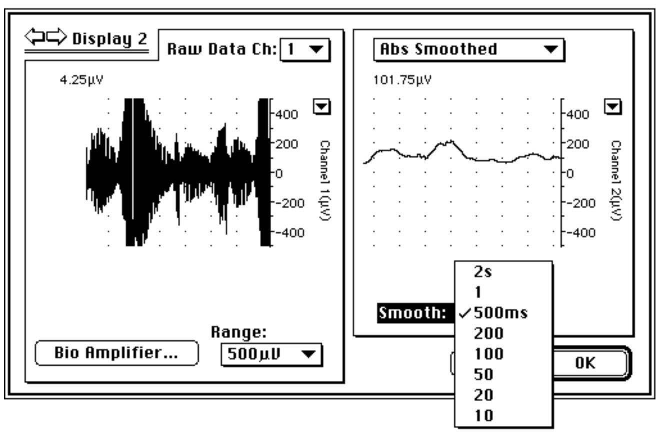

Figure 12. Computed Input dialog box for the 'Abs smoothed' function. The pop-up menu shows a 500 ms smoothing time.

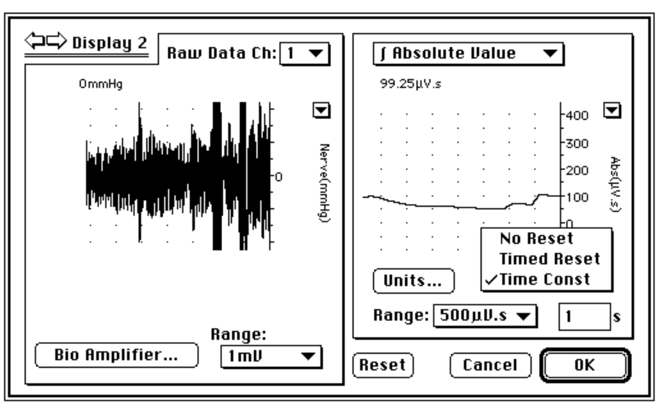

Figure 13. Computed Input dialog box for the 'Integral (Absolute Value)' function. The pop-up menu has been used to select Timed Reset, and a time constant of 1 s has been entered. (Note: in Chart versions 3.5.4 and earlier, 1 s is the minimum time constant value).

References

1. Bronk, D.W. and Stella, G., 'Afferent impulses in the carotid sinus nerve', *Journal of Cellular and Comparative Physiology* 1: 113–130 (1932)
2. Stys, P.K., 'Suction electrode recording from nerve and fiber tracts', In *Practical Electrophysiological Methods*, Kettenmann, H. and Grantyn, R. eds, pp189-194. (Wiley-Liss, New York, 1992)