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Optical measurement of blood flow changes in spinal cord injury

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Abstract. Little is known about cell death in spinal cord tissue following compression injury, despite compression being a key component of spinal injuries. Currently models are used to mimic compression injury in animals and the effects of the compression evaluated by observing the extent and duration of recovery of normal motor function in the days and weeks following the injury. A fibreoptic photoplethysmography system was used to investigate whether pulsation of the small arteries in the spinal cord occurred before, during and after compressive loads were applied to the tissue. It was found that the signal amplitudes were reduced and this reduction persisted for at least five minutes after the compression ceased. It is hoped that results from this preliminary study may improve knowledge of the mechanism of spinal cord injury.

1. Introduction

The mechanisms through which spinal cord injury (SCI) can occur are complex. Compression injury is a key component of natural spinal injuries and is caused by displacement of the vertebral bones or discs. The compression can give rise to haemorrhage, oedema and ischaemia which can cause further injury after the initial impact (Castro-Moure et al 1997, Amer & Levy 1999). Such secondary injury can cause serious permanent loss of function, however appropriate and rapid hospital treatment can potentially minimize this damage component. Several models have been developed to investigate various SCI mechanisms in adult rats. Such models mimic the effects of primary injury, mostly due to laceration and contusion (Crowe et al 1997). Other models, such as that used in the current study, provide quantitative information on the effect of compression on neuronal damage and subsequent recovery. A section of spinal cord is usually exposed in anaesthetized animals. The recovery of the animals is monitored in the days following several minutes’ controlled compression of the exposed section of the cord. The exact mechanism through which neuronal damage occurs during and after the compression is uncertain, and the extent to which ischaemia contributes to the injury is not known.

There are several methods available for assessing blood perfusion in tissue. Laser Doppler flowmetry (LDF) reports two indices that provide an indication of the concentration of moving erythrocytes and the erythrocyte velocity. LDF has been demonstrated as a potentially useful modality for assessing blood flow during compression injury in spinal cord (Westergren et al 2001). Photoplethysmography (PPG) reveals the degree of pulsation of the small arteries within the tissue, i.e. volume changes of the elastic blood vessels occurring during the cardiac cycle (Allen 2007), and has been used to quantify limb perfusion (Zaramella et al 2005). Although successful acquisition of reliable signals from central nervous tissue (human brain) has been
demonstrated (Phillips et al 2010), to date no investigations of PPG for assessing perfusion during or after spinal cord injury have been reported.

A study was undertaken to measure photoplethysmographic signals from the spinal cords of anaesthetized rats before, during and after compression. These animals were used for ongoing compression model studies and the apparatus was modified to incorporate a fibreoptic probe for measuring the PPG waveforms. This pilot study was performed to investigate whether PPG signals are useful for assessing whether blood supply is disrupted during compression of the spinal cord and in the period immediately after compression.

2. Materials and methods

2.1. Measurement system

The measurement system for this study (Phillips et al 2006) was developed and evaluated in our laboratory, based around a fibreoptic reflectance PPG probe comprising two optical fibres, one of which was used to transmit light from an LED with a peak emission wavelength 850 nm to the tissue and the other to return a fraction of the backscattered light to a photodiode. A signal processing system was interfaced to a 16-bit data acquisition card (National Instruments Inc. Austin, TX, USA) installed into a notebook computer. The optical fibre probe consisted of two silica optical fibres (a transmitting fibre and a receiving fibre) each with a core diameter of 400 μm, an outer cladding diameter of 730 μm and a numerical aperture (NA) of 0.39 (Ocean Optics Inc., Dunedin, FL, USA). The distal end of each fibre was cut and polished flat and the proximal end terminated with a male SMA connector. The fibres were coated in a protective PVC jacket, which is stripped away over a length of several centimetres from the distal end.

A signal processing system was interfaced to a 16-bit data acquisition card (National Instruments Inc., Austin, TX, USA) installed into a notebook computer. A LabVIEW (National Instruments Inc., Austin, TX, USA) was implemented to display and record the acquired PPG signals.

2.2. Static compression system

![Figure 1. Apparatus for rat spinal cord compression. (A) compression bar and weight. (B) section of spinal cord showing compression area (light gray). (C) Compression bar with optical fibres inserted. Adapted from Huang et al (2006).](image)

Apparatus developed for a well-established static compression system (Nystrom et al 1988, Li et
al 1996, Huang et al 2006) was adapted for this study. The apparatus consists of a stereotactic frame which holds the animal so that the spinal cord is maintained in a horizontal position. A metal bar measuring \((h \times l \times w)\) 1.5 mm x 5 mm x 3 mm attached to a platform on which a 50 g weight is placed is used for the compression as shown in Figure 1. The metal tube connecting the bar to the platform is supported within the stereotactic frame in such a way that it is free to move in the vertical direction only.

A dedicated fiberoptic probe was constructed, consisting of two optical fibres, their centres laterally separated by a distance of 1.0 mm. The fibres were passed along the metal tube attached to the compression bar. The fibre ends were inserted into the bar so they were flush with the lower surface of the bar as shown in Figure 1(C). The lower surface of the bar is curved to fit the contour of the rat spinal cord thus avoiding any localized compression of the tissue. The proximal ends of the fibres were connected to the measurement system. The volume of sensitivity of the probe is almost certainly confined to the region of spinal cord tissue compressed by the bar.

2.3. Measurements

All experimental protocols of this study were approved by the animal care committee of Queen Mary University of London in accordance with the UK Animals (Scientific Procedures) Act 1986 and international guidelines on the ethical use of animals. The spinal cords of 6 female Sprague–Dawley rats weighing approximately 250 g were used for this study. Animals were deeply anaesthetized in a fume box with a mixture of 5% halothane (Meril, Essex, UK) in addition to a mixture of oxygen and nitrous oxide (1 : 1 ratio) at a flow rate of 750–1000 mL/min. Subsequent anaesthesia throughout the procedure was maintained using 1.5–2% halothane with oxygen and nitrous oxide at unchanged ratio delivered through a nosepiece. The skin and muscle overlying the spinal column were incised and a laminectomy was then performed at T12, leaving the dura undisturbed. The T11 and T13 spinal processes were clamped in a spinal compression frame, and the bar was placed in light contact with the spinal cord by suspending the base of the compression platform (area 2 x 5 mm, Fig. 1B) onto the exposed T12 cord dura under microscopic control. Measurements were recorded from the PPG system for 5 minutes. A weight of 50 g was then applied statically to the platform for exactly 5 minutes, during which time PPG signal recording continued. The weight was then removed and PPG signals recorded for a further 5 minutes. The platform was then removed, the muscle layers were sutured and the skin layers closed with wound clips.

The amplitudes of the acquired PPG signals were measured using a discrete Fourier transform algorithm. The PPG amplitudes from each subject were averaged over three five-minute epochs named: ‘Baseline’, ‘Compression’ and ‘Recovery’. The PPG amplitudes from the ‘Compression’ and ‘Recovery’ epochs were compared with the amplitude recorded during ‘Baseline’.

3. Results

Good quality PPG signals were achieved consistently from all six subjects. An example of the waveform obtained for the entire 15-minute measurement period is shown in Figure 2. It can be seen that in this example, the amplitude of the PPG signal decreased dramatically on compression. The PPG signal was attenuated for the duration of the compression. When the compression was relieved, the PPG amplitude increased to roughly two-thirds of its baseline value and then gradually increased. After five minutes the PPG amplitude was approximately equal to the baseline value.
Figure 2. 15-minute recording of PPG from the spinal cord before, during and after compression.

Figure 3 shows a graph of the PPG amplitude averaged over all three epochs for all six subjects. It can be seen that all six subjects showed a reduction in average PPG amplitude from baseline following compression of the cord. Four out of the six subjects showed a subsequent increase in PPG amplitude when the compression weight was removed.

The mean reduction in PPG amplitude on compression was –55.9% of the baseline value for all subjects. After removal of the compression weight, the mean PPG amplitude returned to –21.9% of the baseline value. Although recovery of the mean PPG amplitude was seen after removal of the compression weight for the population as a whole, a paired Student’s t-test...
demonstrated that the mean increase in amplitude was not significant ($P=0.085$). All six rats recovered to normal motor function within three days, suggesting that the compression of the spinal cord was not sufficient to cause permanent injury.

4. Discussion and conclusion

It was found that good quality infrared PPG signals could be obtained from the spinal cord dura using a fibreoptic probe during static compression model measurements in rats. Compression of the spinal cord resulted in reduced pulsation indicated by attenuation of the PPG signal amplitude. This suggests that ischaemia may occur during compressive injury. The reduction in PPG amplitude persisted even after removal of the compressive load, suggesting a further potential mechanism for secondary injury. Although the amplitude of the PPG signal gives an indication of the degree of arterial pulsation in the tissue vasculature, it should be noted that no information regarding the blood flow rate may be inferred from the PPG signal (Almond et al 1988).

The next stage in the development of this project will be a trial using a larger number of subjects exposed to different compression loads. The spinal cord, like the brain, is one of the most vascular tissues in the human body and also among the most vulnerable to small changes in blood oxygenation. A comparison of PPG amplitude attenuation and recovery of motor function may hopefully shed light on the likely extent of ischaemia in compressive spinal cord injury. Addition of a second wavelength to the measurement system should allow measurement of arterial oxygen saturation of blood within the tissue, which will indicate whether hypoxia is present during and after compression.

References

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