Study of the Spinal Cord Activity in the Pig

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Abstract – Current imaging techniques on organs take advantage of the knowledge we have acquired. However, so far, few information on the functioning of the spinal cord (SC) can be found. In this endeavor, this study aimed to analyze optical properties of the spine and use these results to determine to what extent Near Infra-Red spectroscopy could be used to assess neural activity in the spine. In the first step, the transmittance spectrum of the pig's SC was obtained using a spectrophotometer for the dimensioning of the main blocks of an embedded system. Secondly, adapted light sources with custom probes were used to observe autonomic functions in the spine at rest and under stimulation. Results on the measured haemodynamics show the impact of global stimuli on a local section of the SC. The photoplethysmogram of the SC at rest showed an AC-to-DC ratio, below 1 %, which is a low contrast in comparison with the finger.

Keywords: Biomedical imaging, spinal cord, haemodynamics.

I. INTRODUCTION

The Spinal Cord (SC) is the input of sensory information and the output of the motor commands of the limbs and trunk. Its damage can have major consequences, affecting the life quality and the life expectancy of patients.

Existing monitoring techniques (Magnetic Resonance Imaging - MRI or scanner) only provide structural information of the spinal cord integrity. No functional data can be obtained. Other usual monitoring techniques, such as EEG, SSEP and MEP, are used to identify compromise during surgery. However, these methods cannot identify vascular problem before neurologic functions has been affected.

Measurement of haemodynamic variations in the SC, primarily measurement of oxygenation levels, is the main way of measuring neuronal activity, because such activity is linked to blood flow; underlying principle of functional MRI (fMRI). This approach is useful when evaluating the functional effects of a SC lesion, and in analyzing how these effects progress following surgical intervention, medication or rehabilitation therapy [1-3].

Although interesting, fMRI is still at the research level (not in clinical routines) and has some limitations. Indeed, monitoring is punctual (during total immobilization of the subject) and not chronic, under normal condition of life. Moreover, the spinal column, being magnetically inhomogeneous (causing a heterogeneous signal), affects the repeatability of measurements [4, 5]. In addition, the mechanical movement of the SC within the vertebrae, linked to the patient's breathing, remains a source of parasitic signals and imprecision. Lastly, temporal resolution in fMRI limits its use to observe events lasting down to few seconds. This specification doesn't interfere for major brain processes at a high level. However, it does not comply with the dynamics of the SC, where physiological responses last less than a second.

In this paper, in order to monitor the SC activity with an embedded system, we firstly analyze the in vivo absorption spectrum of the pig SC and we show, in a second time, the use of Near Infra-Red Spectroscopy (NIRS), as a complement to the fMRI approach, to track in real-time autonomic functions of the SC.

II. DIFFUSE OPTICAL IMAGING PRINCIPLE

A. Modified Beer-Lambert-Bouguer Law

Diffuse Optical Imaging (DOI) is a particularly attractive method to measure blood supply variations: a luminous flux is cast onto biological tissues and photons migrate inside along a trajectory, which is determined by the optical properties (*i.e.* absorption and scattering) of the material (see Fig. 1). The amount of light collected highly depends on the media absorption spectrum. When multiple chromophores are present, the absorption coefficient μ_a at wavelength λ is given by:

$$\mu_a(\lambda) = \sum_j \varepsilon_j(\lambda) C_j \qquad (1)$$

where ε_j is the molar extinction coefficient of the chromophore j [L.mol^{-1}.cm^{-1}] and C_j the molar concentration [mol.L^{-1}].

The resulting attenuation of incident light can be approximated using a variation of the Beer-Lambert-Bouguer law, describing

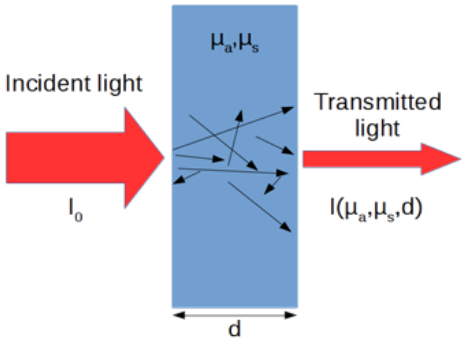

Figure 1. Optical transmission in heterogeneous media

the cumulative effects of absorption and scattering properties in:

$$I(\lambda) = I_0 e^{-(\mu_a(\lambda)+\mu_s(\lambda))d} \qquad (2)$$

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where I_0 is the intensity of incident light, I the intensity of the output light and d the optical path length of each chromophore. As a random process of a photon bouncing of a particle, only statistic values are used for the scattering coefficient μ_s.
In blood perfused tissues, compounds of hemoglobin are the main chromophores targeted for monitoring [6].

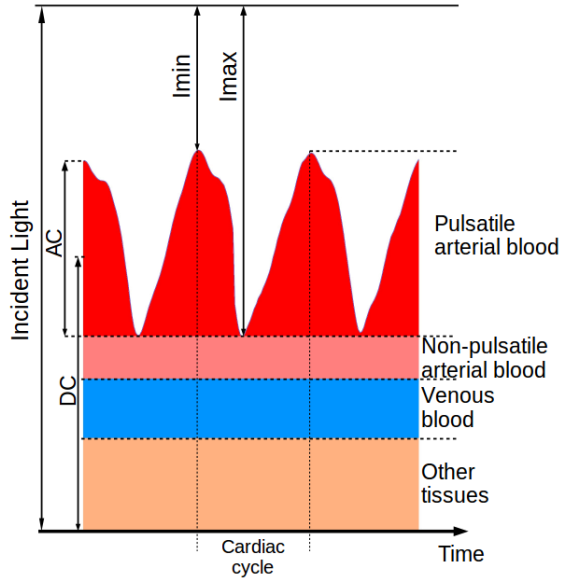

Figure 2. Absorption of light in tissues (scale not respected, AC component dilated)

B. Photoplethysmographic monitoring

Recent wave of health monitoring devices embed heart rate activity sensors and oxygen-level recording. Most of these sensors base the monitoring on the analysis of the photoplethysmogram (PPG) signal, with more or less accuracy [7]. Acquisition is made possible by few low cost and low power optoelectronic components (LED and photodetector, typically) and its analysis can give information from overall condition to local disorders [8-12].
When light is cast onto living tissues, photons are absorbed by numerous media, including skin, flesh, bone and blood. The pulsating nature of arterial blood makes it distinguishable from the rest, because it exhibits a small amount of modulation due to heart pulsation. Fig 2. shows an example of a PPG signal where the pulsatile component is referred to as the AC part and the dominant steady detected signal as DC part. Previous studies [13] have shown that the DC component corresponds to changes in blood volume, and the AC component the vascular compliance and resistance against the pulse wave. The latter also gives the heart rate with its period and its amplitude reflects the stroke volume. Those values help investigating overall condition through cardiac output changes. On the finger, the AC is usually 1 - 10 % of the DC component, which makes it delicate to compute an accurate oxygen-level value [6]. The non-pulsatile elements in the path of a photon give few information regarding haemodynamics. Thus, they considered as parasitic elements that must be taken carefully into account by the front-end and the PPG signal post-processing.

III. EXPERIMENTAL RESULTS

A. Materials and Methods

Due to its anatomic proximity with humans, in structure and vascularization, a pig subject had been chosen for the tests. Up to this date, no studies have shown large animal model results with custom opto-electrical embedded probes, demonstrating the feasibility of this imaging technique on subjects of size comparable to humans, for a potential transfer.
The measurements were performed with veterinaries of the XP-MED society, after obtaining permission from the local ethical committee of Cr2i (INRA, Jouy-en-Josas, France) under authorization number 03-1405.
This study is composed of two highly related in vivo experiments on a pig's spine. Taking into account the disparity of biological parameters between two subjects of the same species, optical characterization must precede biomedical imaging.
The spine of a female FBM pig, 11 months of age and weighting 38 kg was tested in our experiments. The animal rested on its belly, stabilized under anesthesia and ventilation during the tests with the Primus anesthesia workstation from Dräger Medical, Inc. The vertebral column was exposed from T11 to L3, where the anterior part of the blood supply of the spine is accessible [14]. Fig. 3 shows the optical path scheme in the vertebra, which the experiments are based on. The emitter (E) and the receiver (R) are in contact with the sides of the spinous process, called laminae.
The luminous flux propagates through multiple biological layers of the SC, including the vertebral body, the dura mater, the cerebrospinal fluid, veins, arteries and the white and gray matters. Each layer modulates the spectrum of the sent light.

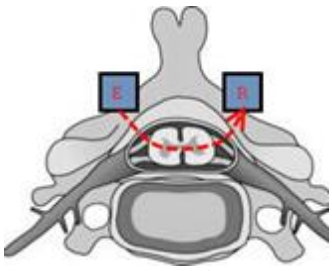

Figure 3. Scheme of the optical imaging in verterbra

B. Spectral Characterization

In the first part of this study, spectral characterization of the SC has been made prior to medical imaging.
The SC is a complex multilayered structure, where optical reflections are present at each interface and interact with absorption and scattering in each layer.
Contrary to the brain, information about optical properties of the SC are scarce. Many papers talk about specific tissues, rather associated to the brain, such as grey matter, dura matter, bones, etc. [15, 16]. Very few papers [17-21] talk about optical reflectance in specific conditions. To our knowledge, only one curve of the optical transmittance can be found in literature [22] for ex vivo rats. The complexity of the measurement is the principal reasons to the quasi-absence of data. Moreover, until now, the study of this organ did not show as much interest in the medical world as the study of the brain.

In order to develop an optimized embedded system, with low power especially, to monitor the SC activity of a pig, thanks to DOI, it is of first importance to quantify the transmittance of the SC in both the visible and NIR ranges. With these measurements, wavelengths of interest can be put in evidence, firstly. Secondly, the order of magnitude of the attenuation factor can be quantified, which permits to size the instrumentation chain, with the best tradeoff between power consumption and SNR [23].

Spectral characterization of the pig SC has been realized in various conditions with portable spectrophotometer (AvaSpecULS2048XLUSB2, Avantes Inc.), associated to white light source (AvaLight-HAL, Avantes Inc.) with two optical fibers. As shown in Fig. 4, fibers are applied to each side of the SC.

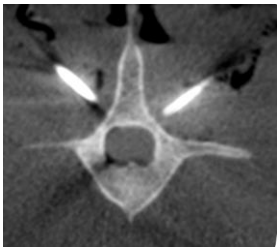

Figure 4. In vivo scan of the SC aligned with the fibers

In Fig. 5, the optical transmittance versus wavelength is presented in two different cases in the range of 500 nm to 950 nm, with a resolution of 3 nm. Highly noisy data at lower wavelength than 500 nm aren't shown, mainly in raison of low illumination of the employed light source. Wavelengths higher than 950 nm are not interesting for us, because water (which corresponds to 77 % of the SC) highly absorbs the illumination flow. The y-axis is in relative unit, because in vivo measurements are delicate on a so peculiar geometry that the vertebra. In case 1, the both fibers are directly on each lamina. Distance between fibers is then close to 23 mm. The maximum transmittance is then 2.8×10^{-5}. In case 2, bones are suppressed and fibers are put on each side of dura matter. Transmittance is then improved by a factor 10, with a maximum value of 2.4×10^{-4}. Note that, between 500 and 600 nm, lobes due to

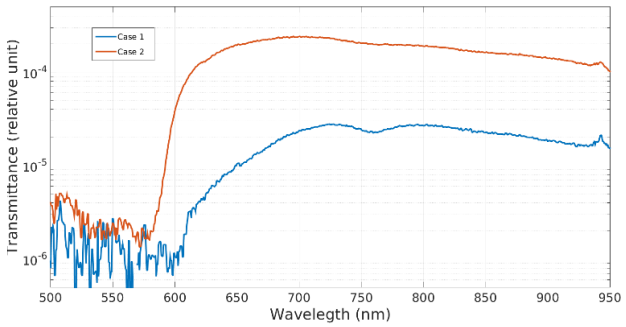

Figure 5. In vivo transmittance versus wavelength of pig SC in two different cases

oxyhemoglobin is weakly visible. Around 750 nm, we can see the effect of fat, and, finally, around 950 nm, peak is due to artefact. In the both cases, wavelengths between 600 nm and 940 nm are good candidate to monitor the SC.

C. PPG Monitoring

In the second part of this study, DOI technique was applied on the pig SC. For this purpose, low noise probes designed according to typical PPG measurement were used. Those probes exhibit response times below 5 µs with a transimpedance amplification of 4.7×10^{6}, allowing the observation of fast varying physiological phenomena. Taking into account results from previous the optical experiments, illumination used LEDs at 624 nm and 880 nm. The latter is used to show the resulting PPG signal detected by transmission through the spine in the vertebral canal. Since frequencies out of the [0.4 - 5] Hz are of no use in this experiments [24], they are removed in the analog signal pre-conditioning achieved before digital conversion and the signal post-processing. The overall system functions on battery to reduce parasitic signals and a LabVIEW interface is used to record real-time data.

Monitoring was performed on the T14 vertebra to track changes impacting the lower spinal cord blood supply [25]. In comparison with the finger, the PPG at rest (the baseline) showed an AC-to-DC ratio, also called contrast, below 1 %. The low density of vascularization in the vertebral column, in comparison to finger, is to be taken responsible as well as the presence of the vertebra, counting for the DC value.

Effects of various stimuli were observed in the PPG signal. Forced haemodynamic changes were induced by means of small electrical stimulation on the femoral nerve and i.v. epinephrine injection. We present in Fig. 6 the resulting signal detected by transmission through the spine under chemical stimulation. The neurotransmitter modifies the pulse by increasing the cardiac output. As expected, tachycardia appears few seconds after the injection with a heart rate starting at 84 bpm, ramping up to 132 bpm. The AC amplitude in the PPG signal also increases, showing an augmentation of the stroke volume. Those results, validated by the measurements, taken on the tongue of the animal, displayed in parallel by the Infinity Gamma XXL (Dräger Medical, Inc.), demonstrate the impact on the blood supply on the lower spine of general stimulation.

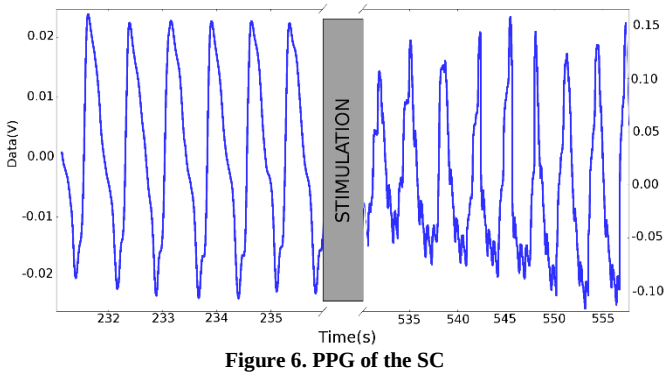

Figure 6. PPG of the SC

IV. CONCLUSION

We have shown in this study that NIRS can be used for imaging functional activity of the pig SC.

Preliminary optical measures were conducted to qualify the transmittance spectrum of the spinal cord in the vertebral column.

Haemodyamic variations have also been identified with DOI, thanks to optimized home-made opto-electrical probes.

The in vivo results show that it is possible to acquire, in real time, the PPG signal of these tissues and assess metabolic conditions of the monitored organ through haemodynamics. However, the obtaining of these signals, in such media, represents a challenge in the case of an implanted system, due to the poor contrast.

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