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# Probing intervertebral discs with PhotoAcoustics

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## ABSTRACT

Photoacoustic measurements are tested as a possible tool for non invasive quantification of Water/Collagen relative content in InterVertebral Discs (IVDs).

**Keywords:** PhotoAcoustic, InterVertebral Disc

## 1. INTRODUCTION

Intervertebral discs (IVD) are fibrocartilaginous organs that provide the linkage and mobility of the vertebrae of the spine. Healthy, they have a high water content, nearly 80% by volume, which diminishes with age thus decreasing intervertebral mobility and nutritional contributions to disc cells. Disc degeneration (DD) is thus a process in principle natural that continues throughout life but can, under the influence of various factors, accelerate suddenly and become pathological. Early detection of this pathology, at the first signs of back pain, is therefore of great interest to quickly establish effective treatments. We have recently shown the central role played in this pathology by the regulation of the water content of the IVD which proceeds from a delicate balance between mechanical stresses exerted by the adjacent vertebrae, electrochemical phenomena within the extracellular matrix (ECM) and the performance of the nutritional transport processes throughout the ECM. The multiphysical model developed makes it possible to estimate the viability of the organ but requires the knowledge of geometric, mechanical and physical properties of the IVDs to initialize the calculation. Because of its multiphysical content, PhotoAcoustic Tomography brings access to some of these parameters. Through spectroscopic photoacoustic measurements performed on porcine IVDs freshly excised, we demonstrate the accessibility to the collagen/water reative internal composition of the discs.

## 2. MATERIAL AND METHOD

### 2.1 Samples Preparation

The IVDs used for this study were harvested from lumbar spinal porcine segments (L2-L5) (Landrace, mean age 4 months, mean weight 40 kg) collected from animals euthanized at the CERIMED facility (Marseille, France). The experiments were conducted in accordance with the European regulations 2 related to the protection of animals in scientific experiments. Once collected, the IVDs were placed in an isotonic saline solution and maintained at 4°C. Immediately before the experiment, the lumbar spine was separated by transverse sawing through the middle of vertebral bodies. The sample was cleaned by removing the rear part of the vertebra (pedicles and processes). Each specimen was composed of one or two IVDs entrapped between two half-vertebra bodies and the anterior/posterior ligament (Fig. 1). During the experiment the discs are settled in a home-made wetted room that ensures alignment and prevent swelling by immersion in a 0.9M%NaCl solution. Through spectroscopic photoacoustic measurements performed on porcine IVDs freshly excised, we demonstrate the accessibility to the collagen/water reative internal composition of the discs.

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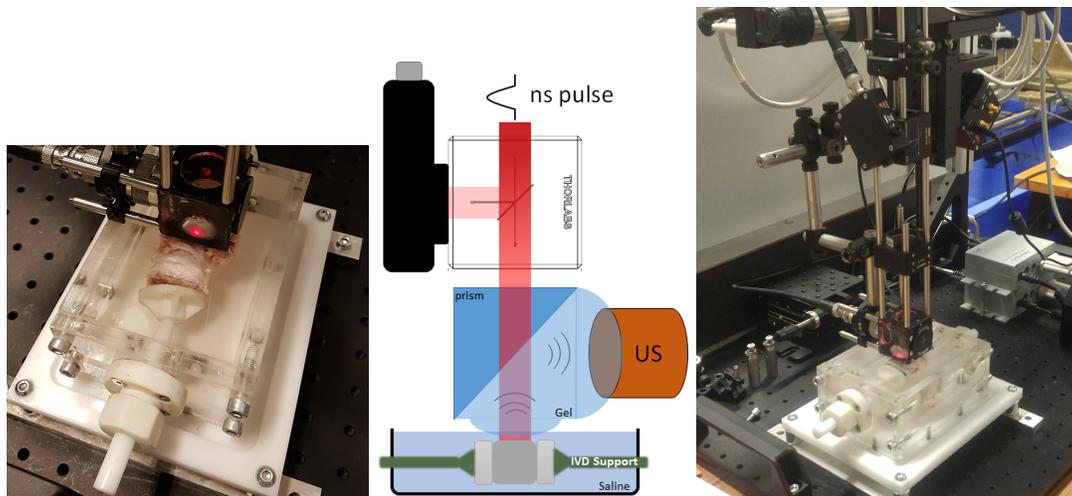


Figure 1. Experimental setup of PA invasive characterization of IVD

## 2.2 Experimental Setup

To evaluate the PA potential for invasive characterization of IVD, an experimental setup was built, in which a high energy tunable pulsed laser source (NT352A, Ekspla, Lithuania, up to 120 mJ, scanning wavelength range 670 - 2600 nm, 20 Hz pulse repetition rate) and a 3-axis cartesian micromanipulator were used. An optical fiber solution was chosen for flexibility. An optoacoustic combiner was developed to combine both optical and acoustic paths based on the concept of optical transparency and acoustic reflectivity. The collimated laser output enters the optoacoustic combiner after the extraction of real-time sample of 1% of the pulse energy (using beamsampler BSF10B, Thorlabs) and the sample was measured using a large band photodetector (DET10A, Thorlabs, Si Detector, 200 - 1100 nm). The generated acoustic signal propagates through the ballistic gel (Clear ballistics, USA) and reflected on the glass surface towards the ultrasound transducer (diameter 10 mm, focal length 1, central frequency 3.5 MHz, Olympus, France). The collected signal is then preamplified (preamplifier Olympus, France, gain 40 dB, cutting frequency 10 MHz) and driven into an oscilloscope (Tektronix, USA, 4 channels, bandpass width 200 MHz) for signal analyses. The whole acquisition process is controlled by a Labview interface program which is connected to the cartesian micromanipulator, OPO and oscilloscope.

The disc was immersed in a saline solution tank for acoustic coupling. Measurements are then performed at user-defined positions, if needed, an automatic scan in a defined zone with a given step size can be performed. From the set of 2D scanning measurements, measurements of the maximum intensity PA (MIP) signal were obtained. Importantly, an on-line monitoring of pulse laser energy is implemented to normalize the measured PA amplitudes spectra and overcome the fluctuations in the measured PA signal.

## 2.3 Signal analysis

The photoacoustic (PA) response of a IVD is directly correlated to its local absorption, surrounding media and spatial orientation from the incident laser beam illuminating the zone of interest. The amplitude of the time-resolved measured PA signal is controlled by the initial generated pressure  $P_0$ , source of the acoustic response, directly proportional to the optical fluence,  $\phi(\lambda)$ , locally deposited:  $P_0 = \Gamma \mu_a(\lambda) \phi(\lambda)$ .  $\Gamma$ , the Grueneisen coefficient depends on the mechanical properties of the medium, the absorption coefficient  $\mu_a(\lambda)$  of the solution is related to the vis-NIR electronic molar extinction coefficient  $\epsilon_i(\lambda)$  of the molecular species constituting the medium and their concentrations  $c_i$ , as follows:  $\mu_a(\lambda) = \sum_i \epsilon_i(\lambda) c_i$ . Hence, knowing the physics of propagation of optical and acoustic waves in the considered media, one can reconstruct the absorption coefficient. IVDs are essentially composed of collagen fiber and water, density of collagen decreasing as one goes to the center of the disc. Spectroscopic measurements were then performed by tuning the laser wavelength from 700 to 1000 nm for different positions to highlight the impact of water content of IVD.

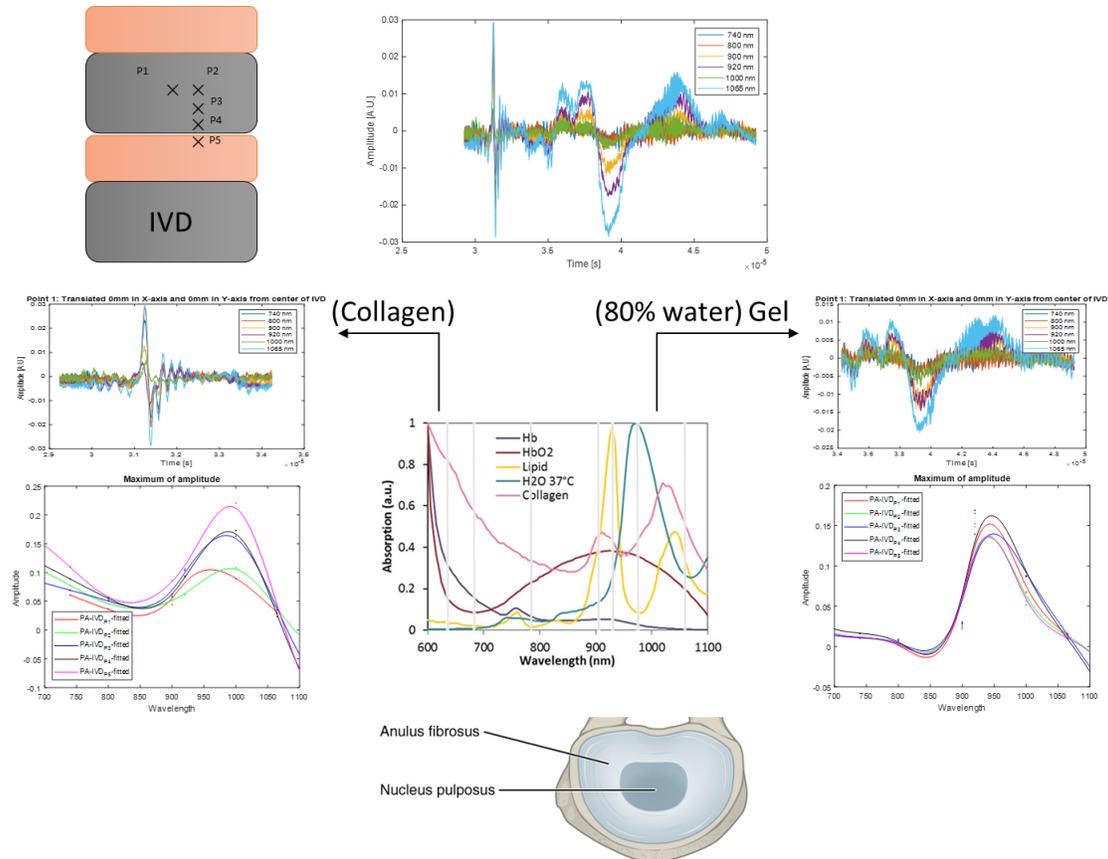


Figure 2. Example of measurements

### 3. SPECTROSCOPIC PHOTOACOUSTIC MEASUREMENTS

A series of test measurements have been performed on different sites of an IVD (see schema Fig. 2, top left). PA time of flight measurements (Fig. 2, top right) clearly exhibit two time windows corresponding to two domains in depth:

- at short times, close to the surface, corresponding to the dense collagen region: the MIP spectral profile is closer to the collagen spectrum;
- at longer times, deeper, the medium clearly shows a spectral response of high water content medium.

### 4. CONCLUSION

The relative collagen/water composition of the IVDs can be accessed non destructively through PA spectroscopic measurements. Next steps include the study of the modification of the PA signal under mechanical constraints.

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