LOAD AND STREAMING POTENTIAL RESPONSES OF ARTICULAR CARTILAGE AS A FUNCTION OF COMPRESSION SPEED DURING INDENTATION

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INTRODUCTION: Indentation measurements are not widely used for the evaluation of the electromechanical properties of articular cartilage. The complexity of the boundary conditions renders difficult the extraction of intrinsic parameters, which are easily found in confined or unconfined conditions. However, there are two important reasons for indentation measurements. They simulate more closely the way articular cartilage is solicitated during joint function and they are well suited for *in-vivo* characterization of cartilage functional properties. In this study, the dependence of the load and the streaming potentials over a large range of indentation speed is evaluated.

METHOD: Cartilage samples (n=4, 2x2cm) with a thick layer of subchondral bone (1cm) were harvested from the load bearing region of bovine shoulders and were tested either fresh or after storage at -80°C. For each sample, a series of ramp-release indentations (300µm amplitude) was performed at speeds ranging from 10 to 5000µm/s at the same position. The compression speeds were tested randomly. Five witness ramp-release compressions (100µm/s, 300µm) were done at different times in order to assess any alteration in the sample during the experiment. The cartilage surface was always oriented perpendicularly to the compression axis via angular actuators and the position of the surface was evaluated to within 3µm using a soft contact procedure. A 15 min delay was imposed between indentations. An array of 37 platinum electrodes (d = 100µm, spaced by 500µm) distributed over the surface of an hemispherical, smooth and impermeable indenter ($r_{curv.}$=4.65mm), was used to monitor the streaming potential signals at 600Hz. This indenter was mounted on a 10 Kg load cell attached to the vertical linear motion actuator of a mechanical tester (MACH-1 Mechanical Tester, Bio Syntech Canada Inc.).

RESULTS: Fig. 1 shows the load and the maximum streaming potential measured at the maximum amplitude of indentation (300µm) as a function of the compression speed for three cartilage samples tested in 0.15M PBS. Sample thicknesses were 1.19, 1.29 and 1.41mm (±3%) with a corresponding GAG content of 27.4, 23.0 and 18.6µg/mg of wet cartilage (measured using the DMMB assay). By integrating the potential over the indenter surface and with the load at 150µm, the streaming potential coupling coefficient (Ke) was calculated to be -8.0±0.3, -8.8±0.3 and -9.4±0.6 mV/MPa respectively (average for the different speeds). Fig. 2 shows the load and the maximum streaming potential measured at 300µm as a function of the compression speed at a single position for four NaCl concentrations on the same sample. The thickness at the tested position was 1.72 mm at 0.5M and 1.76 mm for the other concentrations. The Ke (at 150µm) were -20.0±0.6, -18.5±1.2, -7.8±0.4 and -2.5±0.2 mV/MPa respectively for the 0.01, 0.05, 0.15 and 0.5M NaCl concentrations. Fig. 3 describes the streaming potential distribution over the surface of the indenter (perpendicularly to the compression axis) for selected compression speeds at 150µm compression. The slope of these curves is proportional to the local fluid velocity relative to the extracellular matrix. All the results are presented as function of the compression speed rather than in function of the strain rate, since strain is not linear with the compression amplitude using spherical indenters.

DISCUSSION: As shown in Fig. 1 and 2, the streaming potential and the load at the maximum compression amplitude increase with the compression speed for compression speed below 500µm/s. At higher speeds, both streaming potentials and load become constant. Thus, it seems that there is a limiting strain-rate over which the cartilage response to compression is dictated by its instantaneous response. This attainment of an elastic response at a certain threshold compression speed (500µm/s in our case) has not been previously reported before, to our knowledge, in indentation geometry. A most critical observation in our study was that streaming potentials followed load nearly identically as function of compression speed suggesting that strain-rate dependent stiffening and eventual leveling off was coincident with fluid pressurization, supporting poroelastic mechanisms of load-bearing in the entire range of tested compression speeds.[1] In Fig. 2B, it is shown that an increase in salt concentration leads to a small reduction of load possibly due to a permeability increase with ionic strength.[2] The effect of the NaCl concentration on the streaming potential is more striking (Fig. 2B) due to the higher electrical shielding of charged GAG as salt concentration increases. The Ke measured at the different concentrations correlates well with previously reported values in this concentration range.[2,3] The distribution of the streaming potentials (or equivalently the pore pressure) over the indenter surface in Fig. 3 indicates that, for high speed compression, the streaming potential and therefore fluid pressure are almost constant everywhere over the indenter surface and fluid flow is confined near the point where the indenter, the cartilage and the bath are in contact. The potential profile at 2000µm/s is superimposed over that at 1000µm/s, correlating with the load plateau observed over 500µm/s. For the lower speeds, the potential profile indicates the presence of fluid flow even near the center of the indenter.

CONCLUSION: Electromechanical measurements using a spherical indenter have revealed compression velocity dependent stiffening and leveling off that is concomitant with streaming potential behavior. Spatial profiles of the streaming potentials and their dependence on salt concentration were consistent with poroelastic behavior, and explain certain aspects of physiological high strain-rate loading of articular cartilage.

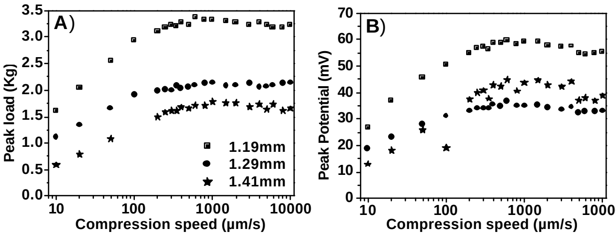

Fig.1. A) Peak load, and B) peak potential vs. speed and thickness.

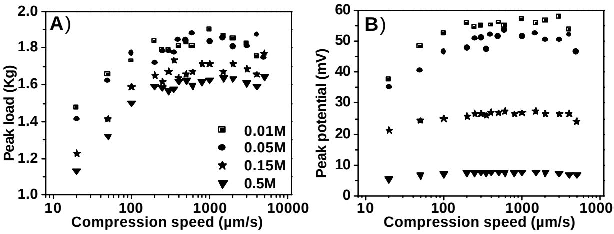

Fig.2. A) Peak load, and B) peak potential vs. speed and concentration.

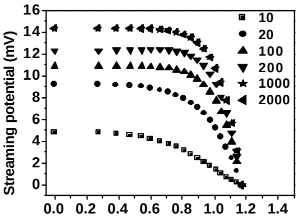

Fig.3. Fitted potential distribution along the indenter vs. speed.

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