Poroelasticity is the dominant energy dissipation mechanism in cartilage at the nano-scale

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INTRODUCTION: Recent studies of micro- and nano-scale mechanics of cartilage and chondrocyte pericellular matrix have begun to relate macromolecular structure to its mechanical response [1,2]. AFM-based indentation has revealed rate-dependent stiffness at the micro-scale [1]. While multi-scale elastic behavior has been studied, and poro-viscoelastic properties have been extensively documented at the tissue-level [3], time-dependent behavior and energy dissipation mechanisms of cartilage matrix at the nano-scale are not well understood. Here, we used AFM-based dynamic compression in conjunction with poroelastic finite element modeling to study the frequency-dependent behavior of cartilage using nano-scale oscillatory displacement amplitudes. We introduce the characteristic frequency \( f_{\text{peak}} \) at which the maximum energy dissipation occurs as an important parameter to characterize matrix time-dependent behavior. Use of micron-sized AFM probe tips with nano-scale oscillatory displacements over a 3-decade frequency range enabled clear identification of this characteristic frequency \( f_{\text{peak}} \).

The length-scale dependence of poroelastic behavior combined with judicious choice of probe tip geometry revealed flow-dependent and flow-independent behavior during matrix displacement amplitudes on the order of macromolecular dimensions and intermolecular pore-sizes.

METHODS Sample Preparation: Middle zone cartilage disks (9 mm diameter \( \times 0.5 \) mm thick) were harvested from the femoropatellar grooves of 1–2-week-old bovine calves and maintained in 0.154 M sterile phosphate buffered saline with protease inhibitors for less than 24 hours before testing. AFM-Based Nano-indentation and dynamic compression: Nano-indentation was performed using the MultiMode AFM with a PicoForce piezo and Nanoscope IV controller via the force mode (Veeco, Santa Barbra, CA). Gold-coated polystyrene colloidal probe tips (end radius, \( R \approx 12.5 \) \( \mu m \), nominal spring constant \( k \approx 4.0 \) N/m, Novascan Technologies, Ames, IA) were employed that were functionalized with a neutral hydroxylterminated self-assembled monolayer (OH-SAM, 11-methacrylodecane, Sigma-Aldrich, St. Louis, MO). The cantilever deflection sensitivity (nm/V) was calibrated on a N/m, Novascan Technologies, Ames, IA) were employed that were functionalized with a neutral hydroxylterminated self-assembled monolayer (OH-SAM, 11-methacrylodecane, Sigma-Aldrich, St. Louis, MO). The cantilever deflection sensitivity (nm/V) was calibrated on a

RESULTS: Sample Preparation:

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The amplitude of the sinusoidal displacement, \( \delta \), was chosen to be much less than the initial indentation (\( \delta < \delta_i \)). Cyclic loading was applied following two different pre-indentation depths of \( \delta_i = 2.4 \) \( \mu m \) and \( 3.3 \) \( \mu m \), which correspond to the contact distances of \( d = 7.9 \) \( \mu m \) and 9.3 \( \mu m \), respectively (Fig. 1). Modeling: was performed using the general purpose commercial finite element software ABAQUS (Version 6.9, SIMULIA, Providence, RI). Because of the symmetry of the problem, the specimen was modeled using axisymmetric, poroelastic transversely isotropic models. Using the general purpose commercial finite element software ABAQUS (Version 6.9, SIMULIA, Providence, RI). Because of the symmetry of the problem, the specimen was modeled using axisymmetric, poroelastic transversely isotropic models. Using the general purpose commercial finite element software ABAQUS (Version 6.9, SIMULIA, Providence, RI). Because of the symmetry of the problem, the specimen was modeled using axisymmetric, poroelastic transversely isotropic models. Using the general purpose commercial finite element software ABAQUS (Version 6.9, SIMULIA, Providence, RI).

DISCUSSION: The measured phase lag (related to energy dissipation) during sinusoidal loading of cartilage over a 3-decade wide frequency range suggests that poroelastic dissipation is the dominant loss mechanism even at displacement amplitudes ~15 nm (e.g., compared to intrinsic matrix viscoelastic effects). This conclusion is supported by the observed shift in the phase lag peak frequency, \( f_{\text{peak}} \), caused by the change in indentation depth, \( \delta_i \) and the associated change in contact distance, \( d \) (Fig. 2), consistent with the trends predicted by the linear poroelastic prediction: \( f_{\text{peak}} \propto 1/d^2 \) (Fig. 3). Comparing the isotropic and transversely isotropic poroelasticity models (Fig. 3), it was found that the transversely isotropic model better predicted the range of changes in both the magnitude and phase lag of the dynamic modulus, as has been observed at the tissue scale [6,8]. Ongoing studies focus on a range of probe tip diameters to further explore the robustness of the approach, and parametric analysis using the theoretical model.

REFERENCES:


ACKNOWLEDGMENTS: Supported by NSF Grant CMMI-0758651 and NIH-NIAMS Grant AR33236.