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Microindentation of Medial & Lateral Regions of Cartilage

Exploring the Mechanical Properties of Bovine Cartilage Utilizing the XZ-500 Extended Displacement Stage

Introduction

Quantitative mechanical testing techniques, particularly at small scales, have become of increasing interest as proven interdisciplinary techniques in the fields of biological sciences, engineering, and materials science. Exploring mechanical behavior and properties of relatively soft samples at the nano- and micro-scale is challenging; outcomes of such studies often attain limelight status among other characterization studies.

This study investigates mechanical properties of two different soft cartilage samples. Testing of these samples is of high relevance as researchers proceed to gain further knowledge in the areas of degenerative joint disease or wear-and-tear arthritis, and studies such as this are geared towards understanding the cartilage growth and healing mechanisms. However, challenges involved in nano- and micro-mechanical testing of such soft materials include the presence of strong adhesion and other interfacial forces, the ability to test within biologically relevant environmental conditions, and determination of the point of initial probe-sample contact. In this study, these challenges are confronted and a newer methodology of nano- to micro-mechanical testing is demonstrated with the help of the Hysitron **XZ-500 Extended Displacement Stage**.

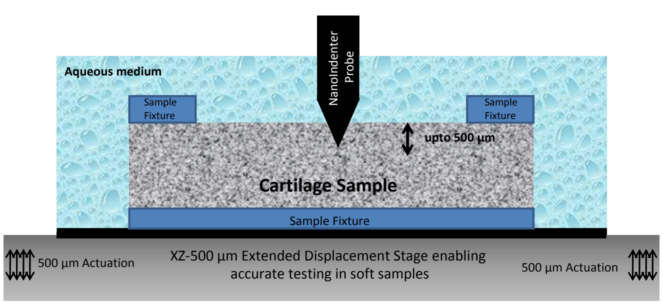

Figure 1: Schematic of the microindentation experimental set-up.

Materials & Methods

A Hysitron **TI 950 TriboIndenter®** equipped with the **XZ-500 Extended Displacement Stage** and a diamond 20 µm 90° conospherical probe was used to measure the reduced modulus of bovine medial and lateral cartilage samples. The **XZ-500 Extended Displacement Stage** provided a maximum actuation captability of 500 µm in the normal (indentation) direction at a resolution of ~1 nm.

Samples were obtained from 4-week old bovine knee (stifle) joints. Samples were prepared in the form of disks and were labeled

Lateral Femoral Condyle and Medial Femoral Condyle. Each disk was approximately 8 mm in diameter and 1 mm thick. All testing was performed in a phosphate-buffered saline solution. Samples were firmly held in place throughout the indentation process using a custom fixture that involved clamping the disk between two parallel metal plates with an opening to facilitate probe-sample contact. The mounted sample was magnetically affixed within a Petri dish, which was mounted onto the testing stage. The test setup is shown in the schematic in Figure 1. The displacement-controlled load function used for all indents included an initial 10 sec. lift-off segment to 10 µm, a 90 sec. loading segment to the peak displacement, a 60 sec. hold segment, and a 30 sec. unloading segment.

Results & Discussion

As can be observed from the X-axis in Figure 2, the displacement range of the **XZ-500 Extended Displacement Stage** was critical for achieving relatively large displacement depths in excess of 150 µm and in determing the initial point of probe-sample contact, which in turn impacts the accuracy of the results. The reduced modulus values of the Lateral Femoral Condyle cartilage disk sample varied from 495.7 kPa +/- 175.7 to 5511.2 kPa +/- 486 at 5 µm to 120 µm, respectively. The reduced modulus values of the Medial Femoral Condyle cartilage disk sample was much lower in comparison and varied from 200.6 kPa +/- 38.0 to 3469.1 kPa +/- 43.4 at 5 µm to 150 µm, respectively. Figure 3 shows a plot of the nature of the modulus variation with respect to the approximate displacement. The relatively large standard deviation is indicative of the inhomogeneity of the samples at this size scale. Larger probes, such as flat punch probes, can tend to average out these effects by contacting large areas.

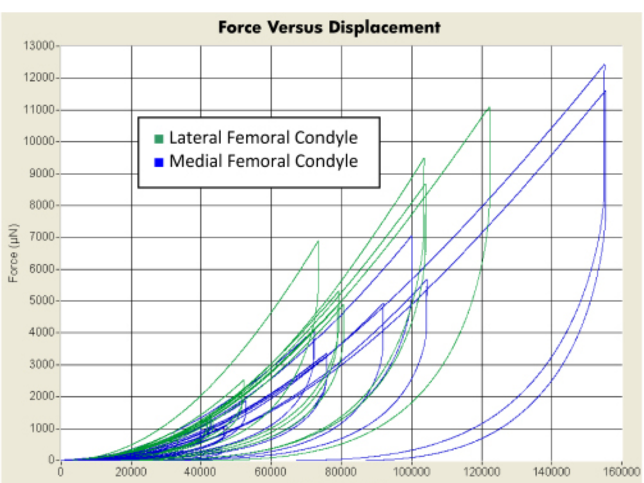

Figure 2: Force vs. displacement curves from indents on the Lateral and Medial Femoral Condyle samples.

Full range of motion from the nano- to micro-scale, in addition to the large range of displacement over which the probe can be actuated, makes the combination of this tool and technique not only unique, but also an important requirement in nano- and micro-mechanical testing of soft samples.

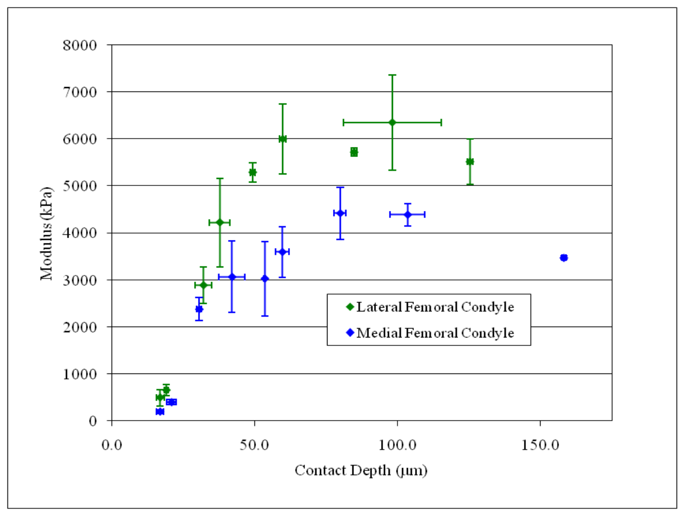

Figure 3: Plot showing the indentation modulus obtained at different contact depths on the Lateral and Medial Femoral Condyle sample.

Conclusion

A methodology more refined than previously used methods for nano- to micro-scale testing on soft cartilage samples has been successfully demonstrated. This methodology has also provided accurate and repeatable results on other similarly soft samples such as hydrogels, and can help advance research in behavioral mechanics of soft materials that are exceptionally relevant for the biomedical industry among others.

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