Characterizing Mechanical Properties of Cartilage in Situ

The Challenge:

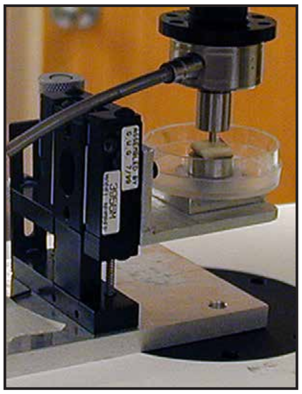
Using Indentation Testing to Characterize Cartilage

Background

Articular cartilage is often characterized as an isotropic elastic material with no interstitial fluid flow during instantaneous and equilibrium conditions, and indentation testing is commonly used to deduce Young's modulus under these assumptions. With indentation testing, specimen shape is greatly simplified and avoids the need for large tissue volume. The test can also be performed on small animals, making indentation a particularly appealing test method. A limitation of this type of testing is that only one of the two independent elastic constants can be deduced from the single indentation test. Poisson's ratio is usually assumed or measured by an independent test. The goal of this work was to develop a method that would allow determination of both elastic constants from indentation testing alone.

Meeting the Challenge

Researchers Dr. Hui Jin and Professor Jack Lewis (University of Minnesota Department of Orthopaedic Surgery) used the dynamic loading capabilities of the ElectroForce® 3200 instrument to characterize the mechanical properties of articular cartilage by investigating novel test methods. This article is provided courtesy of Dr. Jin and Professor Lewis.



Modeling of Cartilage

The method for reducing data from an indentation test depends on the material model assumed, the two most common being a single phase, isotropic, homogeneous linear elastic model, and a biphasic isotropic, homogeneous linear elastic solid and incompressible, inviscid fluid model.

The single phase linear elastic model is assumed when modeling either the short time response or the long time equilibrium response. Although an isotropic, homogeneous elastic model is a significant approximation of the real cartilage behavior, it is used to avoid the greater complexity of the more realistic models when desired, or in situations when experimental conditions make more detailed experimental testing of the cartilage impractical. For the linear elastic model, the material parameters are Young's modulus, E, or shear modulus, G, and Poisson's ratio, v, where both contribute to the indentation stiffness (the ratio of indenting load to indenting depth).

In previous cartilage testing studies, other configurations have been used along with indentation. Hayes (1971) and Hori (1976) used confined compression and torsion tests to obtain the Poisson's ratio. Kempson (1971) measured Poisson's ratio in a tensile test of full thickness cartilage cut parallel to the surface. Jurvelin (1997), Wong (2000) and Korhonen (2002) measured Poisson's ratio by directly measuring the lateral expansion of an unconfined compression test.

A method for determining all the material constants by a single indentation test was introduced in the context of the biphasic theory (Mak, 1987; Mow, 1989). Subsequent studies have reported the values of Poisson's ratios (Setton, 1992; Schenck, 1994; Hale, 1993; Athanasiou, 1994). The Poisson's ratio in the biphasic theory is that of the equilibrium state (Mow, 1989).

The linear elastic biphasic theory has been modified by making the solid phase linear viscoelastic (Mak, 1986). The long time response has been assigned to the biphasic fluid flow; the short time response has been assigned to the viscoelastic solid (DiSilvestro, 2001). Garcia (2000) extended the linear biphasic theory to a transverselyisotropic solid and inviscid fluid, including large deformations.

Materials and Methods

The method is based on the use of a flat-ended cylindrical rigid punch with radius *a* indenting on the surface of a linear elastic layer with thickness h, shear modulus G, and Poisson's ratio, ν (Hayes, 1972). The layer is bonded onto a flat rigid substrate.

The indentation stiffness, defined as the ratio of the indenting load p to the indenting depth, ω, in the linear elastic case may be expressed as:

$$\text{(1)} \qquad \frac{p}{\omega} = \frac{4Ga}{1-\nu}\,\kappa(a/h,\nu)$$

where, $\kappa(a/h,\nu)$ is a correction factor that accounts for the finite layer effect. Under the same condition, and following the same procedure, indenting the sample twice using two different sized indenters, results in:

$$\text{(2)} \qquad \frac{\frac{p}{\omega}_1}{\frac{p}{\omega}_2} = \frac{a_1}{a_2}\,\frac{\kappa(a_1/h,\nu)}{\kappa(a_2/h,\nu)}$$

In Eq. (2), Poisson's ratio is the only unknown and can be obtained by solving this nonlinear equation. Once the Poisson's ratio is obtained, the shear modulus G can be determined from:

$$\text{(3)} \qquad E = 2G(1+\nu)$$

The method was validated first by simulating the indentation on a finite element model in which material properties were assumed, a load-deflection test simulated, and then this data entered into Eq. (2). The predicted E and ν agreed with the input values, supporting the numerical solution algorithm for Eq. (2). The method was next tested on a polyurethane rubber 2 mm thick (Measurements Group, Inc. Raleigh, NC USA) and an elastic foam 5.5 mm thick that was removed from a computer mouse pad. The Young's modulus of each material was first measured from an unconfined compression test. Material specimens (radius 5 mm, thickness 2 mm for the rubber and 5.5 mm for the foam) were compressed between two metal plates on the ElectroForce 3200 instrument at a rate of 0.2 mm/min to 10% compressive strain. The plate-to-sample interfaces were lubricated with silicon grease to reduce friction.

The materials was then tested by indentation on the ElectroForce 3200 instrument. Three flat-ended cylindrical indenters 2 mm, 4 mm and 6 mm in diameter were used in the indentation tests. Data from the tests was used in Eq. (2) to predict E and ν. Results are shown in Table 1, along with the manufacturer's nominal data and the E measured by the compression tests. There was good agreement between the three sets of data.

Material	Indenter Size (mm)	Poisson's ratio from indentation	Young's Modulus (Mpa)	
			Indentation	Unconfined Compression
Elastic Foam	13.0/6.0	0.347	0.432	0.418
Urethane Rubber	6.0/4.0	0.489	4.420	4.712
	6.0/2.0	0.489	4.416	
	4.0/2.0	0.490	4.402	
			4.413	
	Average	0.489	4.413	

Table 1: Elastic properties of urethane rubber and an elastic foam obtained from indentation (two different sized indenters) and unconfined compression tests. Manufacturer's nominal E for urethane rubber is 4.5 MPa and for ν=0.5.

Comparison to Experimental Results

After validation testing, the method was applied to bovine articular cartilage. Seven square-shaped samples (20 x 20 mm) were cut from the relatively flat part (medial and lateral facets) of seven fresh adult cow patellae with normal appearing cartilage. Each sample included the full thickness of the cartilage, subchondral bone and a layer of cancellous bone that served as the rigid substrate in the indentation.

The same impermeable indenters used to test the polyurethane rubber were used in the cartilage tests. Samples were thawed and then bonded onto the bottom of the holding cup that was attached to the adjustable holder using cyanoacrylate cement. The bath was then filled with phosphate buffered saline (PBS).

Displacement controlled cyclical loading (displacement peak/valley = -0.25/-0.125 mm, at 0.5 Hz, for 20 cycles) was performed to precondition the sample. The sample was given 40 minutes to recover. The indenter was then brought into contact with the sample surface, and a 0.15 mm indenting depth was applied at a nominal loading rate of 1.5 mm/sec, followed by a 1200 second displacement hold. Between tests, each sample was given 40 minutes to recover from the previous loading. Indentation testing from the 3 indenters was used in Eq. (2) to predict E and ν at both the very fast rise region (instantaneous) and the end of the hold period (equilibrium).

Predicted E and ν for the bovine cartilage samples are shown in Table 2. On-going work suggests that, for cartilage modeled as an isotropic, homogeneous, linear elastic material in instantaneous and equilibrium states, the introduced indentation method may be used to determine the associated effective Poisson's ratio and elastic moduli. Continued development of the method may be especially suited to the determination of cartilage material properties in small animals where other test methods are impractical.

	Poisson's Ratio	Poisson's Ratio	E(MPa)	E(MPa)
	t=0	t=∞	t=0	t=∞
	0.485	0.483	2.811	0.917
	0.539	0.508	1.09	0.15
	0.511	0.495	1.566	0.277
	0.535	0.526	1.283	0.311
	0.465	0.326	2.077	0.518
	0.478	0.401	1.573	0.328
	0.508	0.504	2.145	0.646
AVG	0.503	0.463286	1.792143	0.449571
SD	0.02829	0.0727	0.59092	0.263078

Table 2: Effective Poisson's Ratio and Young's Modulus for bovine cartilage as determined by indentation tests with seven different cartilage specimens. Each specimen was tested with three different sized indenters, and data was reduced for three combinations of indenter diameters.

Summary

The ElectroForce 3200 test instrument is well suited for characterizing soft, viscoelastic biological materials such as articular cartilage. Researchers Dr. Hui Jin and Professor Jack Lewis (University of Minnesota Department of Orthopedic Surgery) continue their quest to characterize the mechanical properties of articular cartilage by investigating novel test methods using the dynamic loading capabilities of ElectroForce systems technology.

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