# Size effects in the elasticity and viscoelasticity of bone

P. M. Buechner, R. S. Lakes

Abstract Size effects of large magnitude are observed in the torsional shear modulus and damping of bovine plexiform bone. Damping increases and stiffness decreases with specimen size over all sizes studied. Measurements were conducted in torsion using a laser-based micromechanics apparatus capable of viscoelastic studies over a range of frequencies up to 100 kHz, upon samples of various size, with no parasitic friction or other errors that could mimic any size effect. Torsional tan  $\delta$  at 1 Hz varies by about a factor of five over the size range 2.8–6.2 mm thick, and is more dependent on specimen thickness at 1 Hz than it is at higher frequency. The size effects are attributed to compliance and viscoelasticity of the interfaces between laminae. These laminae must be substantially stiffer than whole bone. Observed size effects are likely to play a role in understanding scaling laws of bones in living organisms.

## Introduction

Compact bone is often analyzed as a classical continuum even though it is a natural composite that exhibits a rich hierarchical structure. Compact bone in humans and large carnivores has a fibrous Haversian structure; compact bone in cows, and other large herbivores (Currey 1984), as well as dinosaurs (Currey 1962), has a lamellar plexiform structure. Osteons in Haversian bone are large hollow fibers (~200 µm dia.) composed of concentric lamellae (fine layers) and of pores (e.g., Haversian canals, roughly cylindrical, 10-70 μm in diameter). Laminae in plexiform bone are layers (~200 μm thick) also composed of smaller scale structures including pores, fibrils and mineral crystallites. Osteons and laminae are built of collagen fibers, and these fibers contain fibrils. At the ultrastructural level (nanoscale) fibrils are a composite of hydroxyapatite mineral microcrystals and the protein collagen (Hancox 1972). Spacing between hydroxyapatite crystals characteristically is of the order 1 nm. Bone cells (osteocytes) reside in ellipsoidal lacunar cavities having minor and major diameters typically from 5 to 10 µm and 10 to 30 µm, respectively. These constituents on different structural levels have been associated with various physical properties (Lakes 1993). The mineral crystals, for example, provide stiffness. Cement lines, the boundary between osteons, are made of a "ground substance" that behaves in a viscous-like manner (Lakes and Saha 1979; Martin and Burr 1989), contributing to viscoelasticity of the tissue. These boundaries, in addition to pores and channels, also contribute to the toughness of bone by blunting and re-directing cracks.

The present study was motivated by the observation that relatively large specimens of compact bone, examined in a study of stress-induced fluid flow (Buechner et al. 2001), exhibited mechanical damping substantially in excess of values previously reported in the literature for the same kind of

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P. M. Buechner, R. S. Lakes (☒)
Engineering Mechanics Program,
University of Wisconsin-Madison,
147 Engineering Research Building,
1500 Engineering Drive, Madison,
WI 53706-1687, USA
e-mail: lakes@engr.wisc.edu
Tel.: +1-608-2658697
Fax: +1-608-2637451

R.S. Lakes is also at the Department of Biomedical Engineering, Department of Engineering Physics, Materials Science Program and Rheology Research Center all at the University of Wisconsin.

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bone. After various artifacts and errors were excluded, the question of specimen size was considered. Substantial size effects (Lakes 1995) are known to occur in the elastic behavior of human compact bone. They have been attributed to relative motion between osteons and localized bending or twisting moments transmitted by individual osteons. In the present work, size effects are explored in the stiffness and damping properties of bovine plexiform bone.

## Materials and methods

Bovine femurs were obtained fresh, were refrigerated, and kept moist during all subsequent processing. Each specimen was rough cut using a band saw and fine cut using a low speed diamond saw; final specimens had rectangular sections. For orthotropic materials, the effective shear modulus, which governs the rigidity for both circular and rectangular sections, depends on two tensorial moduli (Lekhnitskii 1981), so there is no penalty associated with the rectangular section shape in terms of interpretation. Each specimen was taken from the cranial midshaft section of a bovine femur. During cutting, a syringe filled with Ringer's solution was used to keep all surfaces of the bone specimen saturated. Figure 1 shows the cross sectional microstructure to be classic plexiform structure. Initial dimensions were  $41.8 \times 6.4 \times 6.0$  mm. The sample long direction was aligned with the bone long direction and the transverse directions were aligned with the circumferential and radial directions. Following testing the specimen was cut down incrementally three times, while maintaining the proportions of all three dimensions. Further experimentation was done on another sample taken from a different bovine femur and prepared in a similar way.

Dynamic material damping properties were studied upon two specimens of bovine femoral bone in torsion. Linear viscoelasticity in the frequency domain is quantified by  $\delta$ , the phase angle between stress and strain for a load that is sinusoidal in time. Tan  $\delta$  is the ratio of the imaginary to real part of the complex modulus; it is proportional to the ratio of energy dissipated to energy stored in a cycle of deformation. The instrumental method (Brodt et al. 1995; Lakes and Quackenbush 1996), broadband viscoelastic spectroscopy (BVS), is capable of measuring the viscoelastic behavior of materials over eleven decades of time and frequency in torsion. Oscillatory torsional loads were applied by driving a sinusoidally varying voltage across one of two sets of Helmholtz coils using either a function generator (Stanford Research model DS345), or a lock-in amplifier (Stanford Research Systems SR850). This Helmholtz coil imposed a magnetic field on the permanent magnet and transmitted an axial torque on the specimen. The angular displacement of the specimen was measured using laser light reflected from a mirror mounted on the magnet to a split-diode light detector. The detector signal was amplified with a wide-band differential amplifier. Torque was inferred from the Helmholtz coil current as follows. Calibration experiments were done using the well-characterized type 6061 Al alloy, which has wellknown moduli. Viscoelastic properties (modulus and  $tan \delta$ ) of the specimen below resonance were inferred from amplitude and phase measurements from the lock-in amplifier and were confirmed via phase measurements upon elliptic Lissajous figures of torque signal vs. angular displacement signal. Peak surface shear strain, inferred from the end angular displacement, was  $2 \times 10^{-7}$  or less, well within the linear range of behavior for bone. Experiments were conducted at constant coil current, hence constant torque amplitude. Damping at resonant frequencies was inferred via the usual resonant half-width method or by fitting of a Lorentzian curve.

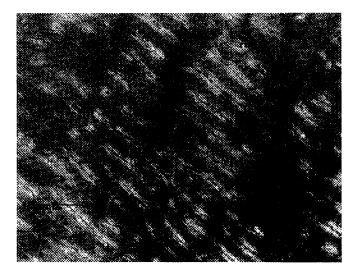


Fig. 1. Structure of bovine plexiform bone under transmitted polarized light. Scale bar: 425  $\,\mu m$ . The structure contains laminae oriented circumferentially. In the machined specimens, the laminae were oriented along the principal direction of the cut prism

The resonance half-width method involves measuring the width of the frequency-response curve near resonance to infer tan  $\delta$ :

$$\tan \delta = \frac{1}{\sqrt{3}} \frac{\Delta \omega}{\omega_0} \quad , \tag{1}$$

where  $\Delta\omega$  is the full width of the resonance curve at half maximum. This is valid if  $\tan\delta$  is relatively small, less than about 0.2. Owing to the range of frequency and progressive cutting down of specimens, these measurements are considerably more painstaking than the more common measurements of modulus for a single specimen geometry.

## Results and discussion

Experimental torsional tan  $\delta$  results in Fig. 2 disclose the effect of sample size and frequency. Torsional tan  $\delta$  at 1 Hz varies by about a factor of five over the size range 2.8–6.2 mm in square cross section, and is more dependent on specimen thickness than it is at higher frequency. The data gap near 1 kHz is due to coupling with bending modes caused by bone anisotropy. Noise in the torsion data appeared at frequencies corresponding to bending natural frequencies independently measured in bending experiments. For comparison, prior results for 3-mm diameter specimens of compact bone, at physiological frequencies from 0.1 to 10 Hz, disclosed tan  $\delta$  on the order of 0.01–0.02 (Lakes and Katz 1979) for both human Haversian and bovine plexiform bone. The present results for comparable size specimens of bovine plexiform bone are consistent with the prior results. The present results also show substantially larger tan  $\delta$  for larger size specimens as shown in Fig. 2.

Torsional stiffness size effects for the present bovine plexiform bone samples as well as previously studied human Haversian bone, human osteons (Ascenzi et al. 1994), several sets of microsamples (Yang and Lakes 1981) and a whole human femur (Huiskes et al. 1981) are shown in Fig. 3. Substantial size effects in effective shear modulus of the bovine plexiform bone begin at considerably larger specimen sizes than in human Haversian bone. We remark that all the effective shear moduli in Fig. 3 were calculated using classical elasticity, hence the term effective. Size effects in  $\delta$  determined in the present study exceed a factor of five over the range of sizes studied. For comparison, Haversian bone (Lakes 1995) exhibits an asymptotic region to the right in Fig. 3 over which size effects are minimal and classical theory is approximately applicable. Plexiform bone, by contrast, exhibits no such asymptotic region even for the largest specimens. We hypothesize that a classical asymptotic region would be approached in plexiform bone if specimens of substantially greater thickness and of the same structure were available. The rationale is that the classical continuum representation of a material depends on the structure size to be much smaller than the specimen size. Indeed, it is appreciated that to fully delineate these size effects, experiments upon many specimens are called for.

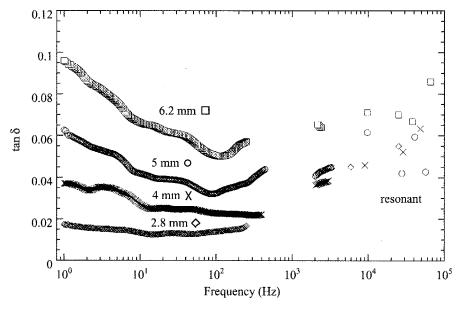


Fig. 2. Torsional damping  $\tan \delta$  of bovine plexiform bone as it depends on specimen thickness and frequency. Size effects are more substantial at the lower frequencies, in the physiological range

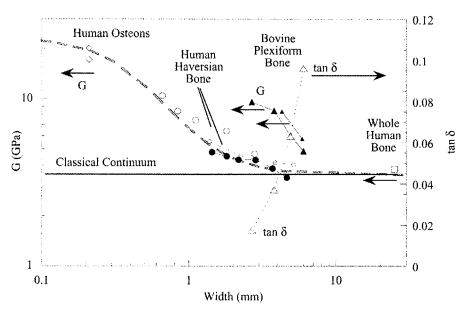


Fig. 3. Shear modulus G and damping  $\tan \delta$  at 1 Hz vs. specimen thickness. For a classical continuum, both G and  $\tan \delta$  are independent of thickness (horizontal line). Size effects in the stiffness (shear modulus) of bovine plexiform bone (solid triangles) in the present results occur over a range of specimen sizes considerably larger than size effects in the stiffness of human bone. Open diamonds, osteons (Ascenzi et al. 1994); open circles and solid circles for several data sets, microsamples (Yang and Lakes 1981); open squares, a whole bone (Huiskes et al. 1981). Damping in bovine bone (open triangles) varies by a factor of more than five for the size range studied, and exhibits no sign of approach to an asymptotic value

Since each specimen must be repeatedly cut down and examined over a range of frequency, such a program would be very painstaking. Even so, prior size-effect studies with many specimens of human bone (both quasistatic and dynamic resonant) indicate the size effects are much larger than the scatter between specimens.

These results show that a classically elastic or viscoelastic continuum view of bone does not correspond to reality since classical continua allow no size effects. Consequently, significant deviations of observed stress from classical theory will occur for very slender samples of bone as well as slender bones, or for small holes or cracks (Park and Lakes 1986) in bone in which strain gradients are large. Size effects may also occur in functionally gradient materials, however in the present bone specimens, there was no systematic variation of density observed with specimen size. Size effects are also known in the plasticity of metals (Fleck et al. 1994) on a scale of tens of  $\mu$ m as well as in the elasticity of synthetic cellular solids (Anderson and Lakes 1994). The present size effects are of larger magnitude than size effects in metals or foams.

As for the significance of macroscopic viscoelasticity, damping occurs in intact bone, but increases with microdamage (Jepsen and Davy 1997). Damping has been considered as an in vivo tool for assessment of bone integrity (Dimarogonas et al. 1993); the change in bone's damping factor was much more substantial than the change in bone density in relation to strength. Damping of bone will have the effect of attenuating impacts during locomotion. Size effects on the scale observed here imply a need to revise current extrapolation of results for laboratory specimens to whole bones; including extrapolation based on finite element analyses, which virtually always presume classical behavior.

# Role of the microstructure; continuum models

Structural mechanisms for the size effects are as follows. A thin layer of a protein-polysaccharide substance occurs at interfaces such as the cement lines between osteons in human Haversian bone. In bovine plexiform bone, interfaces also occur between laminae, but their composition is not well characterized. Cement lines are compliant as inferred from ultrasonic measurements (Katz 1980). Both cement lines and laminar interfaces are viscous like based on observations of relative motion across these interfaces following prolonged deformation (Lakes and Saha 1979; Park and Lakes 1986). Naïvely one may consider larger specimens to contain more such interfaces than smaller ones, giving rise to higher damping and lower stiffness. However, the number of interfaces per area does not depend on specimen size. Since interfaces between osteons or laminae can move with respect to each other, they can carry a bending or twisting moment as well as a force. Such individual moments are not accounted for in classical elasticity as a continuum description. As for porosity, it is not considered to be a

substantial cause for the effects since synthetic cellular solids (Lakes 1986; Anderson and Lakes 1994) disclosed size effects of considerably smaller magnitude than those in bone.

For many purposes it is convenient to view complex materials such as bone in a continuum formulation. The rationale is that continuum material properties such as elastic moduli can be extracted from experiments on bulk samples. The continuum view is useful for making engineering predictions and for visualizing global response of materials. The structural view is relevant to the underlying causes of the behavior. By contrast in the structural view one may not have access to the material properties of each structural element, particularly in biological materials. As for the continuum view, distributed moments and associated phenomena are accounted for in non-classical (generalized continuum) formulations such as Cosserat elasticity (Eringen 1968) and viscoelasticity (Eringen 1967) as disclosed by experiment (Yang and Lakes 1981; Park and Lakes 1986; Lakes 1995). The rationale for use of generalized continuum theories is that they account for size effects in torsion and bending. Classical elasticity and viscoelasticity give rise to no size effects: the Young's and shear moduli (and the damping in a viscoelastic material) are independent of specimen diameter. The constitutive equations for a linear isotropic Cosserat elastic solid are (Eringen 1968)

$$\sigma_{kl} = \lambda \varepsilon_{rr} \delta_{kl} + (2\mu + \kappa) \varepsilon_{kl} + \kappa e_{klm} (r_m - \phi_m)$$
(2)

$$m_{kl} = \alpha \phi_{r,r} \delta_{kl} + \beta \phi_{k,l} + \gamma \phi_{l,k} \tag{3}$$

in which  $\sigma_{kl}$  is the force stress (which is a symmetric tensor in classical elasticity but is asymmetric here),  $m_{kl}$  is the couple stress (or moment per unit area),  $\varepsilon_{kl} = (u_{k,l} + u_{l,k})/2$  is the small strain, u is the displacement, and  $e_{klm}$  is the permutation symbol. The microrotation  $\phi_k$  in Cosserat elasticity is kinematically distinct from the macrorotation  $r_k = (e_{k \text{lm}} u_{m,l})/2$ . The usual Einstein summation convention for repeated indices is used and the comma denotes differentiation with respect to spatial coordinates. There are six independent elastic constants required to describe an isotropic Cosserat elastic solid,  $\alpha$ ,  $\beta$ ,  $\gamma$ ,  $\kappa$ ,  $\lambda$ , and  $\mu$ . Certain combinations of these constants give rise to characteristic lengths in the continuum formulation. For example, the characteristic length for torsion is  $lt = [(\beta + \gamma)/(2\mu + \kappa)]1/2$ ; the characteristic length for bending is  $l_b = [\gamma/2(2\mu + \kappa)]^{1/2}$ . Young's modulus is  $E = (2\mu + \kappa)(3\lambda + 2\mu + \kappa)/(2\lambda + 2\mu + \kappa)$  and the shear modulus is  $G = (2\mu + \kappa)/2$ . In a viscoelastic material, the elastic constants and hence also the characteristic lengths, are replaced by time-dependent or frequency-dependent viscoelastic functions (Eringen 1967). In viscoelastic materials such as bone it is well known that the moduli depend on frequency, and there is energy dissipation quantified by tan  $\delta$ . The characteristic lengths in the Cosserat continuum govern size effects. Since the size effect in torsion is more prominent at low frequency, the corresponding characteristic length is larger at low frequency and smaller at high frequency.

What size specimen is sufficiently large that the bone tissue may be viewed as a classical continuum? The answer depends on the type of stress field. Referring to Fig. 3 for torsion rigidity, the shear modulus of human bone is within 10% of its asymptotic value (corresponding to a whole bone) for a specimen diameter of about 4 mm. For bovine bone, we did not have specimens sufficiently large within the cortical thickness to approach an asymptotic modulus. Therefore modulus studies of whole bones or at least machined cylindrical shells of bone may be needed for the classical continuum view to hold. As for strain distribution, Park and Lakes (1986) showed that for 6-mm wide prismatic bars of bone in torsion, substantial strain appeared at the corner of the cross section where classically the strain should be exactly zero. As for mechanical damping, Fig. 2 shows no convergence of curves for different size specimens at low physiologic frequencies. Therefore results of damping studies of machined laboratory specimens less than the cortex thickness are not likely to be representative of whole bone behavior.

The characteristic lengths are associated with structural length scales in the structural view. Since human bone is transversely isotropic, the isotropic formulation was successful in modeling the experimental results for restricted classes of deformation (torsion of specimens of circular and square section). In the case of bovine bone, which has orthotropic symmetry, an anisotropic generalized continuum model would be required.

The biological significance of interfaces in bone is that cement lines and other weak interfaces, in addition to pores and channels, impart a degree of toughness to bone, since they act as crack stoppers. Weak interfaces dissipate fracture energy via a pull-out mechanism (Piekarski 1970). Compliant interfaces such as cement lines and interlaminar interfaces are likely candidates as mechanisms for the observed size effects in stiffness and damping. In bone, concentration of strain is reduced below values predicted by classical elasticity. The beneficial redistribution of strain (Mindlin and Tiersten 1962) has been observed (Park and Lakes 1986) and found to be consistent with predictions of generalized continuum mechanics based on prior size-effect studies.

As for the structure-property role of plexiform bone in comparison with Haversian bone, consider that given time, plexiform bone in large organisms such as cows tends to remodel into Haversian bone. Haversian remodeling occurs particularly early in the life of humans, primates, and carnivorous mammals (Currey 1984). The reason for this remodeling is obscure since laboratory size specimens of plexiform bone are mechanically superior to Haversian bone. The trend of bovine bone shear modulus vs. size in Fig. 3, could lead to a value below that of Haversian bone. Therefore, at the scale of the whole bone, it is possible that Haversian bone is superior, at least in torsion, to plexiform bone. Experiments at the whole bone scale are called for to explore this possibility. Finally, the observed size effects are likely to play a role in understanding scaling laws (West et al. 1997; Bennett and Taylor 1995; McMahon 1973) of bones in organisms, particularly large herbivorous mammals and dinosaurs (Currey 1962; Rensberger 2000). On a yet larger scale, laminations are also observed in the context of geological structures, in which the role of non-classical elastic behavior has been considered (Adhikary and Dyskin 1997).

# Conclusion

Large size effects are observed in the stiffness and damping of bovine plexiform bone. Torsional  $\tan \delta$  at 1 Hz varies by about a factor of five over the size range; size effects are less pronounced at higher frequency. Therefore results of modulus and damping studies of machined laboratory specimens of bovine bone are not likely to be representative of whole bone behavior at low physiologic frequencies. The size effects are attributed to compliance and viscoelasticity of the interfaces between laminae.

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