 MCB, vol.2, no.2, pp.63-68, 2005

Interfacial Strength of Cement Lines in Human Cortical Bone

X. Neil Dong[1,2], Xiaohui Zhang[1] and X. Edward Guo[1]

Abstract: In human cortical bone, cement lines (or reversal lines) separate osteons from the interstitial bone tissue, which consists of remnants of primary lamellar bone or fragments of remodeled osteons. There have been experimental evidences of the cement line involvement in the failure process of bone such as fatigue and damage. However, there are almost no experimental data on interfacial properties of cement lines in human cortical bone. The objective of this study is to design and assemble a precision and computer controlled osteon pushout microtesting system, and to experimentally determine the interfacial strength of cement lines in human cortical bone by performing osteon pushout tests. Thirty specimens were prepared from humeral diaphyses of four human subjects. Twenty specimens were tested under the condition of a small hole in the supporting plate, in which the cement line debonding occurred. The cement line interfacial strength ranged from 5.38 MPa to 10.85 MPa with an average of 7.31±1.73 MPa. On the other hand, ten specimens were tested under the condition of a large hole in the supporting plate, in which the shear failure inside osteons was observed. The specimens tested under the condition of the large hole resulted in an average shear strength of 73.71±15.06 MPa, ranging from 45.97 MPa to 93.74 MPa. Therefore, our results suggest that the cement line interface between osteon and interstitial bone tissue is weaker than that between bone tissue lamellae.

keyword: Cortical Bone, Bone Mechanics, Bone Microstructure, Cement Line, Interface, Strength.

1 Introduction

Cement lines are boundaries between secondary osteons and the surrounding interstitial lamellae in cortical bone as well as between trabecular packets in trabecular bone. Several investigators have examined the morphology and composition of cement lines in bone [Burr, Schaffler, and Frederickson (1988), Frasca (1981), Schaffler, Burr, and Frederickson (1987), Zhou, Chernecky, and Davies (1994)]. These studies indicate that cement lines are about 1-5 μm thick, less mineralized with a greater calcium/phosphorus ratio than the surround bone tissue and contain no major collagen fibers. Given the morphology and biochemical composition, it has been long hypothesized that cement lines serve as weak interfaces in bone tissue [Burr, Schaffler, and Frederickson (1988), Carter, Hayes, and Schurman (1976), Katz (1980), Piekarski (1970), Schaffler, Burr, and Frederickson (1987)]. Furthermore, experimental evidences of cement lines involvement in the damage and failure process of both cortical bone and trabecular bone have been observed [Carter and Hayes (1977), Choi and Goldstein (1992), Jepsen, Davy, and Krzypow (1999), Lakes and Saha (1979), Hiller, Stover, Gibson, Gibeling, Prater, Hazelwood, Yeh, and Martin (2003)]. However, there are almost no data on mechanical properties of cement line of human cortical bone, especially the interface strength of cement lines. Therefore, the objective of this study is to design and assemble a new osteon pushout microtesting system, and to experimentally determine the interfacic strength of cement lines in human cortical bone by performing osteon pushout microtesting experiments.

Although osteon pushout tests have been employed to measure the shear strength of osteonic lamellae in the past, the debonding at cement lines was never observed in these experiments. Ascenzi and Bonucci [Ascenzi and Bonucci (1972)] measured the shear strength of osteons using a 160-μm-diameter cylindrical punch to push out the lamellae from cortical bone specimen with a thickness of 300 μm, which was supported by a plate containing an 800-μm-diameter circular hole. The osteons did not fail at the cement line, but along the margin of the punch. It had been thought that possible reason could be the significant bending involved in the test region due to the relative large hole in the supporting plate, which is

[1] Bone Bioengineering Laboratory, Department of Biomedical Engineering, Columbia University, New York, NY 10027, U.S.A.
[2] Orthopaedic Research Laboratories, University of California at Davis, Sacramento, CA 95817, U.S.A.

almost three times of the average sized osteon [Martin and Burr (1989)]. Recent simulation results from finite element analyses of osteon pushout experiments showed that the reduced shear stress at the cement line in the testing configuration with a large supporting hole is the primary reason of shearing failure between bone tissue lamellae, instead of cement line debonding [Dong and Guo (2004)].

Therefore, one of the critical steps in performing a successful osteon pushout test is to match the size of a selected osteon with that of the hole in the supporting plate. In this study, a microtesting machine is designed to implement osteon pushout tests under two different testing configurations. In the first testing configuration, a condition of "small hole", the size of the hole in the supporting plate was matched as closely as possible to the size of the selected osteon. In the second testing configuration, a condition of "large hole", the size of the hole in the supporting plate was chosen as 900 μm.

2 Materials and Methods

2.1 Specimen Preparation

A total of thirty bone specimens were prepared from humeri of four human subjects (3 males, 1 female, 47-57 years old), which were free of bone diseases. First, bone slabs with a thickness of 10 mm were sectioned from the middle diaphysis of the humerus, perpendicular to its longitudinal axis. These slabs were then further cut into posterior, anterior, lateral and medial quadrants using a low speed diamond blade saw (Isomet, Buehler Instruments, Lake Bluff, IL). Next, a 4x4x10 mm parallelepiped was obtained from each quadrant. The lateral sides of each parallelepiped were polished and examined under a light microscope to identify the longitudinal direction of osteons. Perpendicular to the longitudinal axis of these osteons, 4x4x1 mm bone specimen slices were then cut using the low speed saw. All the above procedures were performed under wet condition. Under constant water irrigation, both sides of the bone specimen slice were polished manually using a series of silicon carbide papers with progressive grades (1200, 2400, 4000 grits, Struers, Inc., Westlake, OH). Final polishing of specimens was also performed manually using a soft cloth with 3 μm diamond slurry (Metadi© Suspension, Buehler, Lake Bluff, IL) until the lamellar structure of the bone specimen was clearly visible with minimal surface scratches under the reflected light microscope. After polishing, specimens were cleansed in a distilled water ultrasonic bath to remove polishing debris. The thickness of each specimen, ranging from 300 to 800 μm, was then measured by a digital caliber three times.

2.2 Microtesting Device for Osteon Pushout

The experimental method for determining cement line properties employed a newly designed osteon pushout microtesting machine. The new osteon microtesting system (Fig. 1, upper panel) consisted of a motorized XY stage (Prior 138, Prior Scientific, NY/NJ Scientific, Inc.), an Olympus BX51 upright light microscope, a custom-made microtesting indenter device (Fig. 1, lower left) and an XY table for sample mounting and positioning (Fig. 1, lower right). The motorized XY stage had two step-motors in two horizontal directions with a spatial resolution of ± 1 μm.

The microtesting indenter device (Fig. 1, lower left) consisted of a linear actuator (Polytec PI, Inc., Auburn, MA, PI M-227.10), a load transducer (Sensotec, Columbus, OH, Model 31), a capacitive displacement transducer (Micro Epsilon, America, Raleigh, NC, CapaNCDT620), and a microindenter (National Jet Company, LaVale, Maryland). Depending on the indenter used, the diameter of the microindenter ranged from 150 μm to 250 μm. The microtesting indenter was mounted on a rod clamp with a damped rod. The rod clamp and the microscope were mounted on the same bread board.

The XY table was mounted on the motorized stage. In the center of the XY table, there was a support plate with a small hole (Fig. 1, lower right). The size of the hole in the supporting plate ranged from 200 to 900 μm. The prepared bone specimen was placed on the top of the support plate, and the bone sample was held tightly on the support plate by four heads and moved by manually adjusting the micrometers attached to the head.

2.3 Osteon Pushout Experiment

The specimen was placed on the specimen supporting plate and viewed under a light microscope, which was connected to a video camera. Under the microscope, an osteon of appropriate size was chosen first, and then appropriately sized hole and microindenter were chosen. Then, the osteon was aligned exactly in the center of the hole on the support plate by adjusting the micrometers.

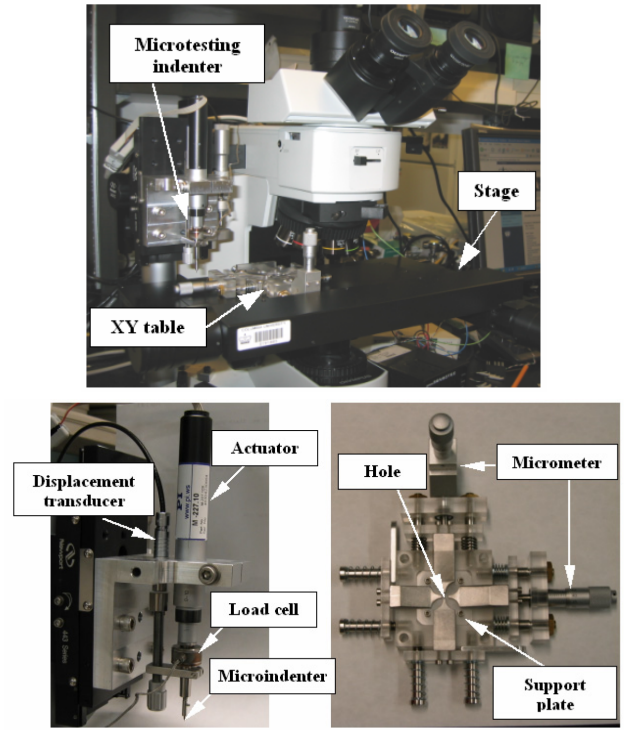

Figure 1 : Osteon pushout microtesting system (upper panel). Microtesting indenter device (lower left); XY table (lower right). A bone specimen is mounted on the top of the support plate, and the chosen osteon is aligned with the hole in the center of the support plate under the microscope by adjusting micrometers. The stage is used to translate the XY table from the microscope to the microtesting indenter such that the chosen osteon is just below the micro indenter and then the pushout test is performed.

Afterwards, the XY table with the specimen was translated to the testing position so that the chosen osteon was aligned exactly underneath the microindenter and then the pushout test was performed. The movement of the stage and the actuator, and the data acquisition (using DAQ board PCI-MIO-16XE-10, National Instruments, Austin, TX) were all controlled by the LabView program (LabView 6.1, National Instruments, Austin, TX) on a personal computer.

Osteon pushout tests were performed in two different testing configurations. Among those thirty specimens prepared, twenty specimens were tested under the condition of "small hole" and the rest ten specimens were tested under the condition of "large hole". In both testing configurations, the specimen was subjected to pushout testing with a constant displacement rate of 0.4 μm/sec. From the load data, the peak load P_{max} was determined. For specimens tested under the "small hole" condition, the interfacial strength of the cement line was determined by

$$\tau = \frac{P_{\max}}{\pi p D h} \tag{1}$$

Where D was the diameter of the osteon, h was the specimen thickness and p is the percentage of debonding at the cement line. For specimens tested under the "large hole" condition, the shear strength of the bone lamellae was determined using the same Equation (1) by replacing D with the diameter of the microindenter.

After microtesting, and the specimen was polished again using a soft cloth with diamond slur to remove surface scratches created during the pushout test. The specimens were then examined under a reflected/transmitted light microscope for details of cement line debonding or interlamellar shear failure. The percentage of deboning at the cement line was measured.

3 Results

Under microscopic examinations, sixteen specimens among twenty specimens tested under the condition of "small hole" indicated cement line debonding: ten specimens were debonded completely at the cement line while six specimens were partially debonded (Fig. 2). Tests on the remaining four specimens were not successful due to the misalignment of the indenter. The cement line interfacial strength ranged from 5.38 MPa to 13.65 MPa with an average of 7.96±1.99 MPa (n=16). Among these specimens, the average interfacic strength of complete debonding was 7.10 MPa, which was slightly lower than that of partial debonding (Table 1). The load-time curve showed a typical response of an osteon pushout test (Fig. 3).

Among ten specimens tested under the condition of "large hole", eight specimens were tested successfully while the remaining two specimens were excluded as no significant load drop was observed during the pushout test (the test had to be terminated because of the range of the linear actuator). Under microscopic examinations, no

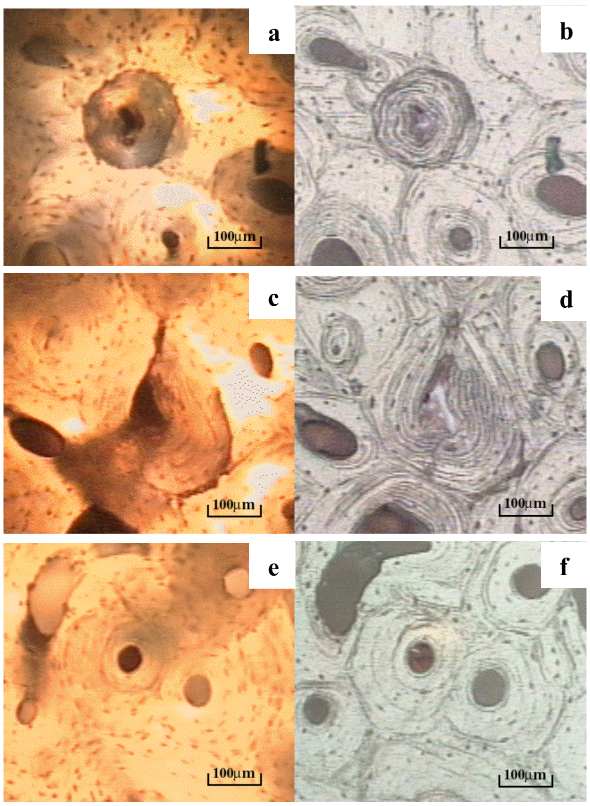

Figure 2 : Micrographs of tested specimens. Figure a and b show the complete debonding at the cement line (a: transmitted light microscopy; b: reflected light microscopy). Figures c and d demonstrate the partial debonding at the cement line (c: transmitted light microscopy; d: reflected light microscopy). Figure e and f indicated micrographs of a specimen tested on the condition of large hole indicate interlamellar shear failure of bone lamellae inside the osteon (e: transmitted light microscopy; f: reflected light microscopy).

cement line debonding was observed in these specimens tested under the condition of "large hole". A portion of bone lamellae inside the osteon was pushed out (Fig. 2). From the load-time data, the shear strength of osteonic lamellae ranged from 45.97 MPa to 93.74 MPa with an average of 73.71±15.06 MPa (n=8).

4 Discussion

For the first time, the interfacial strength of cement line in human cortical bone has been determined in this study.

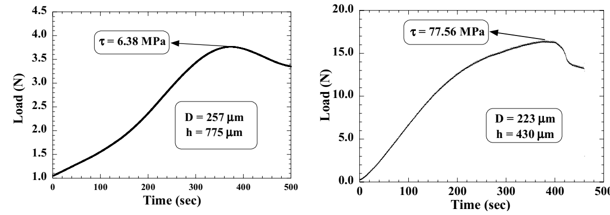

Figure 3 : Typical load-time curves of osteon pushout tests (a: the condition of "small hole"; b: the condition of "large hole"). D is the diameter of the selected osteon. H is the thickness of the cortical bone specimen. τ is the calculated interfacial strength for curve a and shear strength for curve b using Eq. 1.

The measured cement line interfacial strength was 7.96 MPa and was significantly lower than the shear strength of osteonic lamellae (73.71 MPa). Therefore, our experimental results suggest that the cement is indeed a weak interface in human cortical bone. The interlamellar shear strength (73.71 MPa) determined using the condition of "large hole" is higher than the reported value (56 MPa) by Ascenzi and Bouncci (1972) in human femurs. Anatomic difference may reconcile this difference. Interestingly, our data almost coincide exactly with reported 74 MPa of a similar, early study [Frost, Roth, and Villanueva (1961)] although their test configuration was quite different from ours. In their study, simple shear tests without regard to individual osteons or cement lines were performed using a 900μm punch and 70 μm thick cortical specimens from a human femur. Furthermore, our interlamellar shear strength is only slightly higher than the reported, continuum shear strength of human cortical bone (68 MPa) [Reilly and Burstein (1975)].

In summary, our experimental data show that the debonding strength of cement lines is lower than the shear strength of bone tissue lamellae. These data suggest and support the weak interface hypothesis of the cement line.

Acknowledgement: This work was supported by NIH R21 AR49613. Authors also would like to thank Mr. Johnny Tang for his help in preparing specimens.

References:

Ascenzi, A; Bonucci, E. (1972): The shearing properties of single osteons. *Anatomical Record* 172: 499-510.

Table 1 : Parameters of bone specimens tested under the condition of a small hole

#	D_O	D_H	D_P	h	F	p	τ
A1	238	244.25	228.75	549.3	2.496	100	6.207
A2	262.75	285.5	236.25	494.7	3.196	100	7.826
A3	210.5	217.8	172	475.7	2.485	100	7.900
A4	257.5	408	222.5	775	4	100	6.379
A7	187.25	417.5	170.5	574	2.896	100	8.575
F2	210	220	150	342.3	1.817	100	8.044
A10	252	278.25	200	669	3.903	100	7.368
A11	216.25	278	180	722.7	2.64	100	5.376
A12	250.75	410	200	650.7	2.842	100	5.543
E2	341.75	408	270	592.3	4.93	100	7.751
A5	296.3	408	254	650	4	91	7.257
A8	245.75	401	184.75	807.3	4.320	94	7.351
E4	260	408	200	440.3	3.9	79	13.648
E7	313.5	408	257	530	4.183	89	8.989
E11	326.8	401	274.25	497.3	3.837	73	10.315
F1	329.5	410	250	517.7	4.077	86	8.890

D_O: diameter of the osteon (μm)
D_H: diameter of the hole in the supporting plate (μm)
D_P: diameter of the punch (μm)
h :thickness of the cortical bone specimen (μm)
F: ultimate load of the push out test (N)
p: percentage of debonding at the cement line(%)
τ: interfacial strength of the cement line (MPa)
The average interfacial strength for complete debonding is 7.10 MPa. The average interfacial strength for partial debonding is 9.41 MPa. The overall average interface strength is 7.96±1.99 MPa.

Burr, DB; Schaffler, MB; Frederickson, RG. (1988): Composition of the cement line and its possible mechanical role as a local interface in human compact bone. *Journal of Biomechanics* 21: 939-45.

Carter, DR; Hayes, WC. (1977): Compact bone fatigue damage: a microscopic examination. *Clinical Orthopaedics & Related Research* 127: 265-74.

Carter, DR; Hayes, WC; Schurman, DJ. (1976): Fatigue life of compact bone–II. Effects of microstructure and density. *Journal of Biomechanics* 9: 211-8.

Choi, K; Goldstein, SA. (1992): A comparison of the fatigue behavior of human trabecular and cortical bone tissue. *Journal of Biomechanics* 25: 1371-81.

Dong, XN; Guo XE, (2004): Geometric Determinants to Cement Line Debonding and Osteonal Lamellae Failure in Osteon Pushout Tests. *Journal of Biomechanical Engineering* 126: 378-390.

Frasca, P. (1981): Scanning-electron microscopy studies of 'ground substance' in the cement lines, resting lines, hypercalcified rings and reversal lines of human cortical bone. *Acta Anatomica* 109: 115-21.

Frost, HM; Roth, H; Villanueva, AR. (1961): Physical characteristics of bone. Part III: A semimicro measurement of unit shear stress. *Henry Ford Hospoital Medical Journal* 9: 157-62.

Hiller, LP; Stover, SM; Gibson, VA; Gibeling, JC; Prater, CS; Hazelwood, SJ; Yeh, OC; Martin, RB. (2003) Osteon pullout in the equine third metacarpal bone: effects of ex vivo fatigue. Journal of Orthopaedic Research, 21: 481–488.

Jepsen, KJ; Davy, DT; Krzypow, DJ. (1999): The role of the lamellar interface during torsional yielding of human cortical bone. *Journal of Biomechanics* 32: 303-10.

Katz, JL. (1980): Anisotropy of Young's modulus of bone. *Nature* 283: 106-7.

Lakes, R; Saha, S. (1979): Cement line motion in bone. *Science* 204: 501-3.

Martin, RB; Burr, DB. (1989): *Structure, function and adaptation of compact bone*. New York: Raven Press.

Piekarski, K. (1970): Fracture of Bone. *Journal of Applied Physics* 41: 215-23.

Reilly, DT; Burstein, AH. (1975): The elastic and ultimate properties of compact bone tissue. *Journal of Biomechanics* 8: 393-405.

Schaffler, MB; Burr, DB; Frederickson, RG. (1987): Morphology of the osteonal cement line in human bone. *Anatomical Record* 217: 223-8.

Zhou, H; Chernecky, R; Davies, JE. (1994): Deposition of cement at reversal lines in rat femoral bone. *Journal of Bone & Mineral Research* 9: 367-74.