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In - Vivo Diffuse Damage in Human Cortical Bone Does Not Compromise Bone Toughness

Timothy L. Norman
Cedarville University, tnorman@cedarville.edu

G. P. Parsamian

C. Coleman

IN-VIVO DIFFUSE DAMAGE IN HUMAN CORTICAL BONE DOES NOT COMPROMISE BONE TOUGHNESS

+*Norman, T (A-NIH); *Parsamian, G; *Coleman, C
+*West Virginia University, Morgantown, WV. Musculoskeletal Research Center, Health Sciences Center, Department of Orthopedics, 304-293-1072, Fax: 304-293-7070, tnorman@wvu.edu

Introduction: Microdamage accumulation processes have been proposed to act as a stimulus for bone remodeling[1] and serve as one of the major factors contributing to increased skeletal fragility.[2] Recent studies have demonstrated that bone exhibits stiffness losses prior to the appearance of microscopic cracks.[3] Diffuse areas of staining (or diffuse damage) at the submicroscopic level has supported the idea that damage initiates at the ultrastructural level.[3,4,5,6] Even though diffuse damage has been implicated in the initiation of microcracks and bone remodeling, the incidence of in-vivo diffuse damage and the mechanics of its origin and propagation are not known. The objective of this study was to examine the incidence of in-vivo diffuse damage in human cortical bone from the midshaft of the tibia and proximal femur. The relationship between in-vivo diffuse damage and bone fragility as measured by fracture toughness was also examined.

Methods: Twenty-seven fresh human tibias and femurs were harvested from 15 male (average age = 70.5 ± 10.2 yrs.) and 14 female cadavers (average age = 79.3 ± 9.4 yrs.). Bulk sections were obtained from the mid-diaphyseal region of the tibia and from the proximal femur and stained according to published procedures.[7] A 250 μm thick transverse slice was then removed from the center of each section, ground to 150 μm thick and polished to remove surface scratches. A Zeiss LSM-510 Laser Scanning Confocal Microscope (LSCM) system, equipped with an argon ion laser and attached to a Zeiss Axiovert 100M Fluorescence microscope, was used for damage assessment. The samples were scanned by the monochromatic 488 nm laser beam exciting BF fluorescence and the emission was recorded by a detector after passing the LP 505 filter. A Planar-Neofluar 20×/0.5 objective was used to produce surface scans of 460.6×460.6 μm^2 size. Five fields were randomly chosen from each cortex and measurements were averaged per specimen. Diffuse damage areas were identified as the areas containing a network of small bright cracks surrounded with pooled blurry stained regions. Those areas were subsequently circumscribed and the diffuse damage area density (Df.Dm.Ar.) parameter defined as the ratio of the total damaged area (Dm.Ar.) and bone area (B.Ar.)(Df.Dm.Ar.=Dm.Ar./B.Ar., mm^2/mm^2) was obtained. Damage measurements were correlated with age, gender and with tension (mode I) and shear (mode II) fracture toughness.[8] The primary statistical analysis was the analysis of variance (ANOVA). The statistical package JMP™ (SAS Institute, Cary, NC) was used to perform all analyses.

Results: The statistical analysis revealed that there was no significant difference between the Df.Dm.Ar in males and females. Df.Dm.Ar. of the pooled data tended to increase with age, but the increase was not significant (Figure 1). The average Df.Dm.Ar. from the pooled data was 3.00 % (SD = ± 1.56 %). Fracture toughness also did not vary with the in-vivo diffuse damage (Figure 2).

Discussion: Although there was a trend for Df.Dm.Ar to increase with age, the increase was not significant. This finding differs from previous studies that investigated microdamage and reported that microdamage increased with age in both the tibias and femurs of males and females[8,9] and in the femoral neck.[10] This study shows that in-vivo diffuse damage occurrence is small, averaging approximately 3 percent of total bone area. In contrast, in-vivo microdamage density of human cortical bone ranged from about 0.1 #/mm^2 to about 0.3 #/mm^2 (10 to 30 cracks per square centimeter) for an age range of 50 to 90 years, respectively.[8] Previous work investigating the influence of microdamage on bone fragility as measured by fracture toughness has indicated that in-vivo microdamage has a weak but significant inverse relationship with fracture toughness.[11] However, given the relatively small amount of in-vivo diffuse damage described in this study, it is not surprising that there would not be a significant relationship between diffuse damage and fracture toughness in the current study. It is also not likely that diffuse damage compromises bone's strength or stiffness in-vivo. Clearly, however, diffuse damage does influence bone properties in acceleration.[2-6] It is concluded that in-vivo diffuse damage in human cortical bone is small and does not influence bone's ability to resist fracture in-vivo.

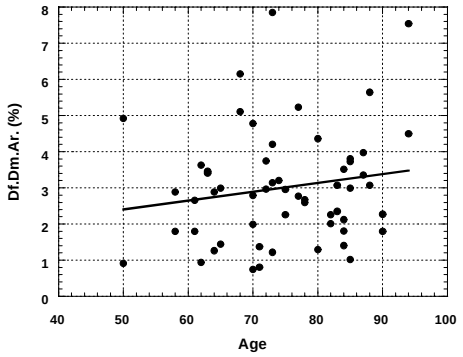

Figure 1. Average Df.Dm.Ar. increased with age, but the increase was not significant.

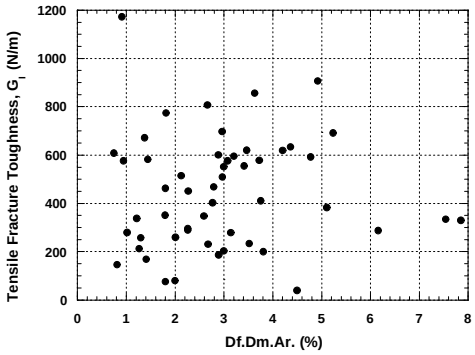

Figure 2. Toughness does not change with diffuse damage

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