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Anisotropic mechanical properties of ovine femoral periosteum and the effects of cryopreservation

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ABSTRACT

The mechanical properties of periosteum are not well characterized. An understanding of these properties is critical to predict the environment of pluripotent and osteochondroprogenitor cells that reside within the periosteum and that have been shown recently to exhibit a remarkably rapid capacity to generate bone de novo. Furthermore, the effects of cryopreservation on periosteal mechanical properties are currently unknown. We hypothesized that the periosteum is pre-stressed in situ and that the periosteum exhibits anisotropic material properties, e.g. the elastic modulus of the periosteum depends significantly on the direction of loading. We measured the change in area, axial length, and circumferential length of anterior, posterior, medial, and lateral fresh periosteal samples removed from underlying bone (t=0-16 h) as well as the average strain in axially and circumferentially oriented anterior periosteal samples subjected to tensile strain (0.004 mm/s) until failure. The elastic modulus was calculated from the resulting stress-strain curves. Tensile testing was repeated with axially aligned samples that had been slowly cryopreserved for comparison to fresh samples. Periosteal samples from all aspects shrank 44-54%, 33-47%, and 9-19% in area, axial length, and circumferential length, respectively. At any given time, the periosteum shrank significantly more in the axial direction than the circumferential direction. Tensile testing showed that the periosteum is highly anisotropic. When loaded axially, a compliant toe region of the stress-strain curve $(1.93 \pm 0.14 \text{ MPa})$ is followed by a stiffer region until failure (25.67 + 6.87 MPa). When loaded circumferentially, no toe region is observable and the periosteum remained compliant until failure (4.41 ± 1.21 MPa). Cryopreservation had no significant effect on the elastic modulus of the periosteum. As the periosteum serves as the bounding envelope of the femur, anisotropy in periosteal properties may play a key role in modulating bone growth, healing and adaptation, in health, disease, and trauma.

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1. Introduction

The material properties of ovine femoral periosteum are not well characterized. An understanding of these properties is critical to predict the environment of pluripotent and osteochondroprogenitor cells that reside within the periosteum and that have been shown recently to exhibit a remarkably rapid capacity to generate bone *de novo* (Knothe Tate et al., 2007; Knothe and Springfield, 2005). Furthermore, as the periosteum serves as the bounding envelope of the femur, anisotropy in periosteal properties may play a key role in modulating bone growth, healing and adaptation, in health, disease and trauma.

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Histological studies of periosteal architecture reveal an axially aligned collagen structure in the mid-diaphysis (Foolen et al., 2008) and a high density of elastin (Allen and Burr, 2005; Allen et al., 2004), suggesting that long bone periosteum is elastic and anisotropic. Previously published studies describe the mechanical testing and materials characterization of periosteum from long bones of the frozen pig metacarpus (Popowics et al., 2002), fresh chick tibiotarsus (Bertram et al., 1998), and frozen bovine tibia (Uchiyama et al., 1998). As a whole these studies show that long bone periosteum is pre-stressed (Bertram et al., 1998; Popowics et al., 2002) and exhibits non-linear behavior in response to tension (Bertram et al., 1998; Popowics et al., 2002; Uchiyama et al., 1998). Upon release from the underlying bone, a significant recoiling force or pre-stress is measured in the periosteum (Bertram et al., 1998). Studies on flat bones, e.g. the pig mandible and pig zygomatic arch, suggest that the pre-stress is anisotropic (i.e. more free shrinkage is observed along the length of the bone compared to the orthogonal direction) (Popowics et al., 2002).

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