Materials Research Express

PAPER

Mechanical properties of cortical bones related to temperature and orientation of Haversian canals

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Keywords: mechanical properties, cortical bones, bone mechanics, compressive test, fracture

Abstract

The understanding of mechanical performances and microscopic failure mechanisms of cortical bones under service condition is necessary prerequisite of fracture prevention, which would support the development of bone tissue engineering and design of bionic bones. By using miniaturized horizontal in situ compression tester, the effects of both temperature and sampling orientations on the compressive strengths and fracture morphologies were investigated. The significant difference between fracture strengths and compressive strains at various temperatures indicated that the cortical bone was sensitive to temperature. Direct experimental evidences revealed the gradually fibrotic trend of fracture surfaces as a function of sampling orientation. Through the Haversian canals distribution analysis, the relationship between the distribution of Haversian canals and fracture path was established. Essentially, the competition between high density Haversian canals and stress concentration factor determines the initiation and propagation of cracks.

1. Introduction

As highly calcified hard tissue, cortical bones exhibit excellent comprehensive mechanical properties due to complex and ordered microstructures [1, 2]. The loading types and the micro-environment of cortical bones under service conditions are complicated and changeable, which induce diversified failure behaviors [3]. The relationship between the microstructure evolution and external conditions, including stress and temperature, need to be further investigated. Improving the testing ability of the microscopic deformation behavior and performance evolution of bone materials under service conditions is the key to reveal the failure mechanism. Regarding the testing of bone mechanical properties [4], static testing methods, including compressive [5], tensile [6], bending [7] and indentation [8], are frequently used, such as E Lefevre et al used microindentation to assess the mechanical properties of the extracellular matrix. Combining with the influencing factors, including porosity [3], sugar content [9] and mineral content [10], these methods could obtain basic strength, anisotropy, toughness, microstructure evolution, healing behavior and gradient of mechanical properties between cortical and cancellous bones. For instance, the plastic deformation behaviors of both single lamellae and macromorphic samples was revealed by the micropillar compression inside a scanning electron microscope [11]. The propagation behavior and orientation-dependent crack-growth resistance was also examined by in situ mechanical testing [1].

A large number of studies involved the effects of sampling depth and porosity on mechanical properties of cortical bones, and the finite element analysis of failure mechanism subjected to different loading conditions were also performed, such as F Khor et al analyzed the anisotropic three-point bending and axial torsion behaviors of cortical bones by using an effective human body models [12]. The in vitro environment could be established to achieve the mechanical tests under approximate service condition, such as K Merlo et al carried
out the indentation and fracture toughness tests under thermostatic (37 °C) ribose solution to assess the elastic modulus and fracture toughness [9]. Meanwhile, due to the ordered structures consisting of mineralized collagen fibrils with specific orientation and embedded hydroxyapatite crystals, the mechanical anisotropy of cortical bones related to axial or radial orientation was also investigated [13]. However, abundant evidences regarding the effects of sampling orientations on the compressive strength and fracture mechanism of cortical bones under service condition need to be provided. In addition, as typical internal inherent structure and continuous tubular system containing the nutrient vessels of bone, Haversian canals, which were proved to induce the anisotropy of cortical bones along different directions [14], might significantly influence the differences in mechanical properties. The orientation and distribution of Haversian canals on the compressive strength and fracture mechanism were seldom mentioned.

In this paper, based on a self-developed miniaturized compressive device, an approximate in vivo environment was established. Compressive properties and fracture morphologies related to the temperatures, sampling orientations and distribution of Haversian canals were investigated.

2. Experimental methods

Taking fresh tibia cortical bones from adult porcine as experimental materials, the sampling method is shown in figure 1(a). Within eight hours after slaughtering, the specimens were sliced off at various sampling orientations of 0°, 45°, and 90° along with the long axis of porcine tibia, respectively. Based on an identical sampling thickness along with the radial direction of cortical bone, uniform specimens ($l \times w \times t$: 10 × 5 × 2 mm) were prepared through mechanical polishing. In order to achieve the compressive tests under approximate in vivo environment, as shown in figure 1(b), the specimen was immersed in thermostatic (38.5 °C) Ringer’s solution [15]. Rectangular specimens (figure 1(c)) were used to obtain the stress-strain relationship. Meanwhile, to facilitate the observation of evolution behavior of fracture surfaces, cortical bone specimens with prefabricated double V-notched defects were prepared to obtain large stress concentration factor. According to previous study [16], through reasonable alteration of a horizontal in situ tensile/compressive tester, the functions, including solution environment, temperature control unit and compatibility with optical microscope, were comprehensively integrated. Seen from figure 1(d), the specimens were placed on a pair of support columns with symmetrical square grooves, which could guarantee the uniaxial alignment during the compressive process. The bottom of the container filled with Ringer’s solution was disposed at a water bath, which integrated with a group of ceramic heating rods to adjust the temperature. Meanwhile, by using a stereomicroscope (Nikon, SMZ-745) with high depth of field, the deformation behaviors of prefabricated defects could be real-timely monitored.

3. Experiments and discussions

The compressive stress-strain ($\sigma$-$\varepsilon$) curves of rectangle specimens with various sampling orientations $\theta$ were obtained. Considering the service temperature would induce significant change in kinetic parameters of demineralization [17], due to the ordered structures of mineralized collagen fibrils, the demineralization rate was depended on the temperature. Although some studies have revealed the effect of temperature on strength cortical bone [17–19], the quantitative relation between the compressive strength and temperature under service
condition was lacking, especially considering most studies of bone strength were performed at RT. Therefore, the compressive $\sigma$-$\varepsilon$ relation related to RT and 38.5 °C were focused. At RT, on basis of a strain rate of $10^{-4}$ s$^{-1}$, seen from figure 2(a), the compressive strength $\sigma_b$ and strain after fracture $\varepsilon$ exhibited obvious correlation with $\theta$. Specifically, when $\theta$ equaled to the longitudinal direction of cortical bone, $\sigma_b$ reached its maximum value. Also, $\sigma_b$ manifested as non-monotonic decreasing trend as a function of $\theta$. Meanwhile, $\theta$ corresponding to maximum and minimum $\varepsilon$ were 90° and 45°, respectively. The irregular downtrends of $\sigma_b$ and $\varepsilon$ might be determined by the heterogeneous microstructures. Especially, the distribution of Haversian canals probably acted as critical in influencing factors. Similar tendencies of $\sigma_b$ and $\varepsilon$ were obtained regarding cortical bone specimens at body temperature of 38.5 °C. As shown in figure 2(b), specimen with $\theta$ of 45° exhibited the weakest resistance to plastic deformation and compressive fracture. Regarding the effects of sampling orientation on the fracture behavior [20], J C Behiri et al has revealed a variation in orientation from 0° to 90° produced average increases in critical stress intensity factor from 3.2 to 6.5 MN·m$^{-3/2}$. In order to reduce the error, 10 specimens for each $\theta$ were tested. At RT, $\sigma_b$ firstly decreased from 125.6 ± 21.4 MPa at $\theta$ of 0° to 83.9 ± 10.2 MPa at 45° then increased to 95.6 ± 12.5 MPa at 90°. Similarly, at 38.5 °C, $\sigma_b$ exhibited relatively lower strengths of 82.7 ± 11.3 MPa, 52.1 ± 13.1 MPa and 67.6 ± 10.9 MPa, respectively, corresponding to $\theta$ of 0°, 45° and 90°. The significant difference between $\sigma_b$ at both temperatures indicated that the strength of organic matters inside the cortical bone was sensitive to temperature. The effect of temperature on bone strength has been explained by the strength weakening mechanism on basis of a single critical flaw and Griffith energy release hypothesis [21]. Considering that the osteon and bone lamella were approximately parallel to the long axis of porcine tibia, corresponding to $\theta$ of 0° and 90°, the external stresses were parallel and perpendicular to a group of osteon and bone lamella, respectively. Therefore, the bone specimens exhibited strong compressive resistances. Regarding the specimen with $\theta$ of 45°, the interlaminar delamination and interfacial shearing of osteon and bone lamella would weaken the strength and deformation, which accorded to our research regarding the fracture strength and consumption of plastic work [22] under combined stress. Furthermore, seen from the fracture morphologies (inserts in figure 2), at RT, with $\theta$ of 0°, the fracture morphology mainly consisted of flat shearing surface...
interspersed with locally lamellar flocculation features. When $\theta$ increased to 45°, holistic fibrosis and local micro cracks were observed, and the orientation of the fibrosis exhibited holistic anisotropy [23]. With $\theta$ of 90°, uplifted lamellar fiber structure was the main morphological feature. The fiber orientations were not entirely parallel to the fracture surface, a number of fibers with consistent orientation began to warp. According to previous study [24], lamellae composed of calcified collagen fiber bundles and both delamination and fiber debonding were also observed. The gradually increased sampling orientations promoted the morphological change from planarization to fibrotic. The gradually uplifted fibers exhibited a trend from stickiness to gradual peeling, which was similar with the observation obtained by SEM fractography of human cortical bone [25].
Furthermore, the Haversian canals distribution based on various $\theta$ were obtained through micro-CT analysis (North Star Imaging, CT-X5000). The scanning voltage and precision were 80 kV and 0.5 $\mu$m, respectively. Seen from figure 3, for arbitrary $\theta$, the intersection lines of fracture surfaces were closely related to the distribution of Haversian canals. Combined with the machine vision analysis and statistical analysis of Haversian canals quantities at different sections along the radial and axial directions, the density distribution of Haversian canals at different regions were obtained. Specifically, corresponding to $\theta$ of $0^\circ$, $45^\circ$ and $90^\circ$, respectively, the regions with high density Haversian canals were consistent with the intersection lines of fracture surfaces. The expansion direction of the fracture surface was from the region with higher to lower density Haversian canals. Especially, the orientations with largest density gradient of Haversian canals were the main regions where cracks nucleated. The competition between stress concentration factor and density gradient of Haversian canals would determine the position of crack initiation.

4. Conclusions

The temperature-dependent compressive properties of cortical bones were directly revealed. As the sampling orientations gradually increased, the fracture morphologies gradually changed from planarization to fibrotic and from stickiness to peeling. The inhibition effect of high density and uniformly oriented Haversian canals on crack propagation was obvious. Therefore, the widespread existence and evidently uneven distribution of Haversian canals inside cortical bone were preconditions of the consistency between the fracture surface and the region with high density Haversian canals. Previous study also revealed that the micro cracks gradually propagated from the calcified interstitial tissue through the osteon to the Haversian canals [26]. Therefore, the competition between the region with high density Haversian canals and notches with significant stress concentration factor determined the position of crack initiation.

Acknowledgments

This work is funded by the National Natural Science Foundation of China (51875241, U1601203), National Key R&D Program of China (2018YFF010124, YS2018YFA070002) and Jilin Province Science and Technology Development Plan (20190302078GX, 20180201126GX).

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