ESTABLISHING AND VALIDATING AN OSTEOPOROTIC MODEL USING BOVINE TIBIA

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Introduction

Osteoporosis (OP) is the most common bone disease; it is associated with more than 9 million fractures annually worldwide [1]. Research into OP continues to be a key area, but current models of OP can be expensive and/or hard to obtain. Whilst ex vivo human specimens require ethical approval for their use and are associated with higher variability than animal specimens, alternatives such as artificial bone models fail to adequately represent animal bone properties. In vivo OP animal models can be made, however the costs associated with these are often prohibitive, with variable results. Previous authors [2] have used bovine vertebrae as a model and successfully modified them using acid degradation protocols to mimic osteoporosis. However no long bone model of OP has been created using these techniques.

Aim: establish and validate (biomechanically) a bovine model of OP; determine which bovine long bone is most appropriate for the model and optimise preparation and degradation techniques.

Methods

Bovine long bones from 4-5 month old calves were sourced from a local abattoir. Three calf bones from 4 different diaphyseal regions were sectioned into 15 mm specimens, producing 18 specimens for each region. Bone mineral density (BMD) was assessed with quantitative CT scanning (X Tec, XT H 225 ST, Nikon Metrology UK Ltd, Derby UK) before and after degradation techniques were implemented.

Different preparation methods were used (n=24 in each condition): reverse osmosis water, phosphate buffered solution, fresh (<4hr of harvest), 0.6M, 1.2M and 2.4M hydrochloric acid. Half of the samples were dried for 4 hrs at 63^{o}C to assess dehydration affects.

Biomechanical testing of samples was performed using an Instron machine (Instron, High Wycombe, UK). A 3.5 mm cortical screw (Stryker, Newbury, UK), was inserted to 0.5 Nm, with axial tensile loading at 5 mm/min recording using Bluehill 3 (Bluehill, Instron, High Wycombe, UK) at 10 Hz until maximum force was demonstrated. Following tests for normality, paired t-test and Pearson correlation coefficients were analysed, with the study powered to 90%, to detect a 5% significance.

Results

The bone density of calf long bones was (mean±SD) 1.82±0.08 g/cm^3. Long calf bones did not show changes in density across the four diaphyseal regions (range: 1.79+/-0.07 to 1.84 +/-0.08 g/cm^3 (p=0.869) Acid degradation produces significant (p<0.01) reductions in BMD with decreases from normal of 23%, 31% and 33% for 0.6 M, 1.2 M and 2.4 M respectively (Figure 1), and 25%, 25% and 29% for the dried samples (p<0.01)

Pullout testing demonstrates a significant reduction in pullout force with decreasing bone density (R^2=0.839 p=0.01). For dried samples the trend continued, but not at a significant level (R^2=0.331, P=0.233).

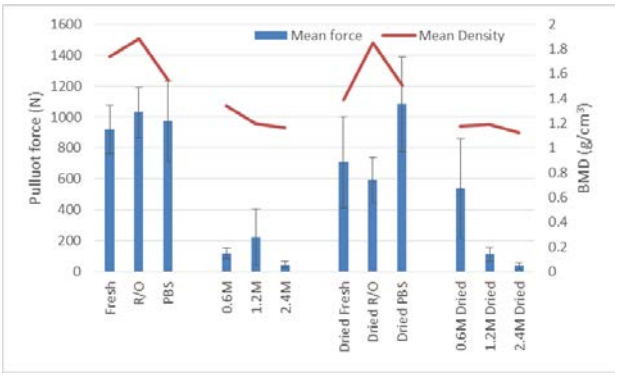

Figure 1: Pullout force (N) for each bone preparation method with respective bone mineral density (g/cm^3)

Discussion

Calf bones provide a suitable model for biomechanical and OP research. Significant reductions in BMD are produced using acid degradation protocols, with associated reductions in mechanical strength of the material. This validated model establishes a readily available substrate that can be used to mimic the conditions seen in OP following simple degradation methods.

References

1. Johnell et al. Osteoporos Int, 17:1726-33, 2006
2. Akbay et al. Eur Spine J. 17(3):468-73, 2008

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