



Dietary Supplements Do Not Improve Bone Quality in C57BL/6J Female Mice



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Introduction

Dietary supplements are marketed for ability to improve health outcomes. Many dietary supplements are components of the bone extracellular matrix[1], and supplementation may increase bone quality through reducing collagen degradation or the incorporation of elements that may improve bone hydration such as glycosaminoglycans. Results have been varied as to whether specific dietary supplements improve bone quality in the relatively few studies that have examined their effect [2,3]

We hypothesize that material properties of bone will be improved with treatment of dietary supplements in mice

Study Design

Animals

C57BL/6J female mice at 11 weeks of age were given dietary supplements 5X a week for 8 weeks (n=10/group)

Mice were given the following supplements

- Control (Milli-Q water)
- Collagen (1 g/kg)
- Chondroitin sulfate (250 mg/kg)
- Fish Oil (1 g/kg)
- Glucosamine (300 mg/kg)

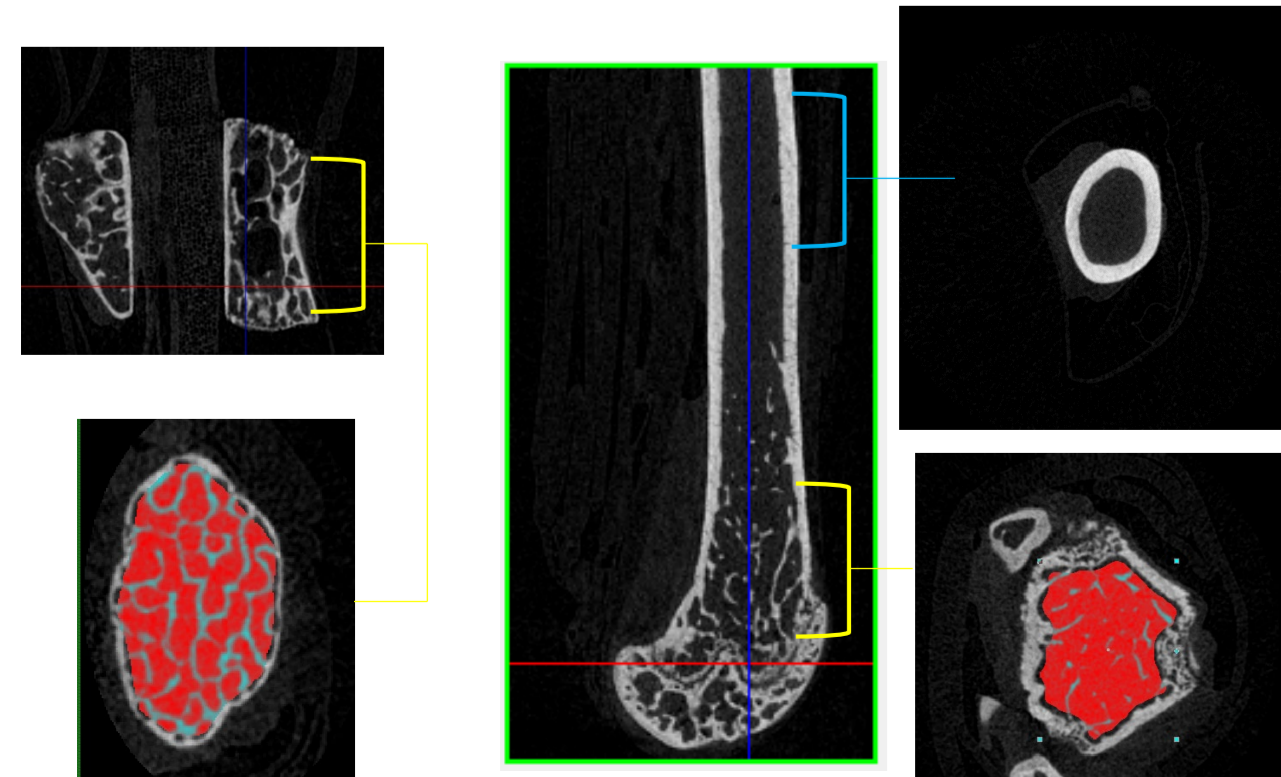
Dietary supplements were suspended in Milli-Q water and/or deposited directly onto a Bacon Softie® food supplement

Femurs and vertebrae (VB) were collected and stored at -20°C in PBS-soaked gauze

Micro-computed Tomography

Bones scanned on Skyscan 1172
9.8 µm voxel size
Hydroxyapatite phantoms scanned for calibration
Femur cortical ROI: 1 mm after bottom of trochanter
Trabecular ROI: 1 mm above end of growth plate

VB ROI: full VB from VB fully formed to growth plate



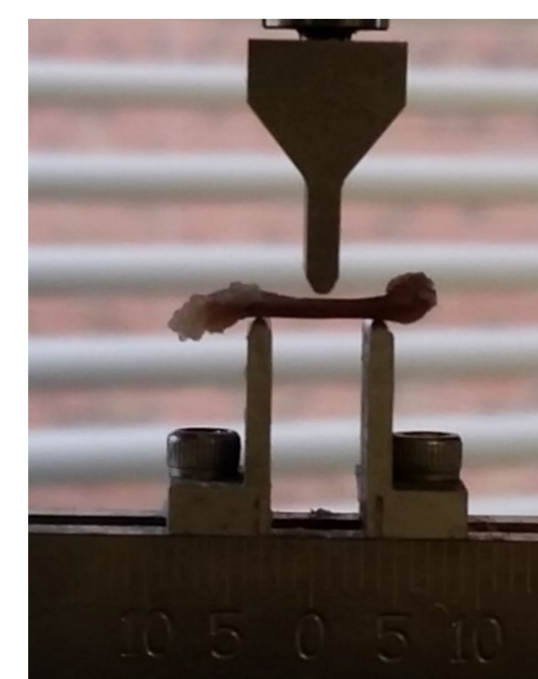
ROIs of VB
ROIs of femur

Mechanical Testing

3-pt bending tests for femurs
Displacement = 0.025 mm/s, bones tested until failure
Geometry from CT and beam equations used to calculate tissue-level material properties

Compression tests for vertebrae

Displacement = 0.025 mm/s, bones tested until displacement limit or force limit

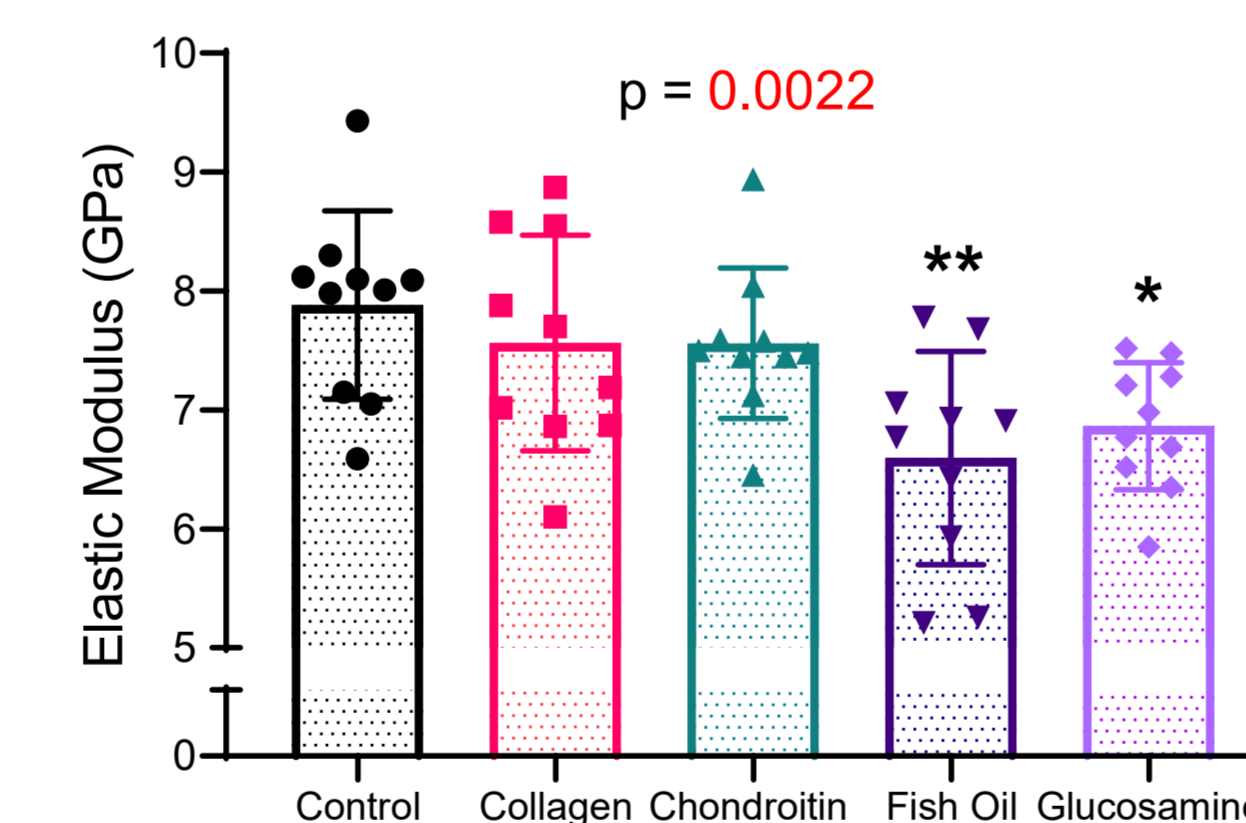
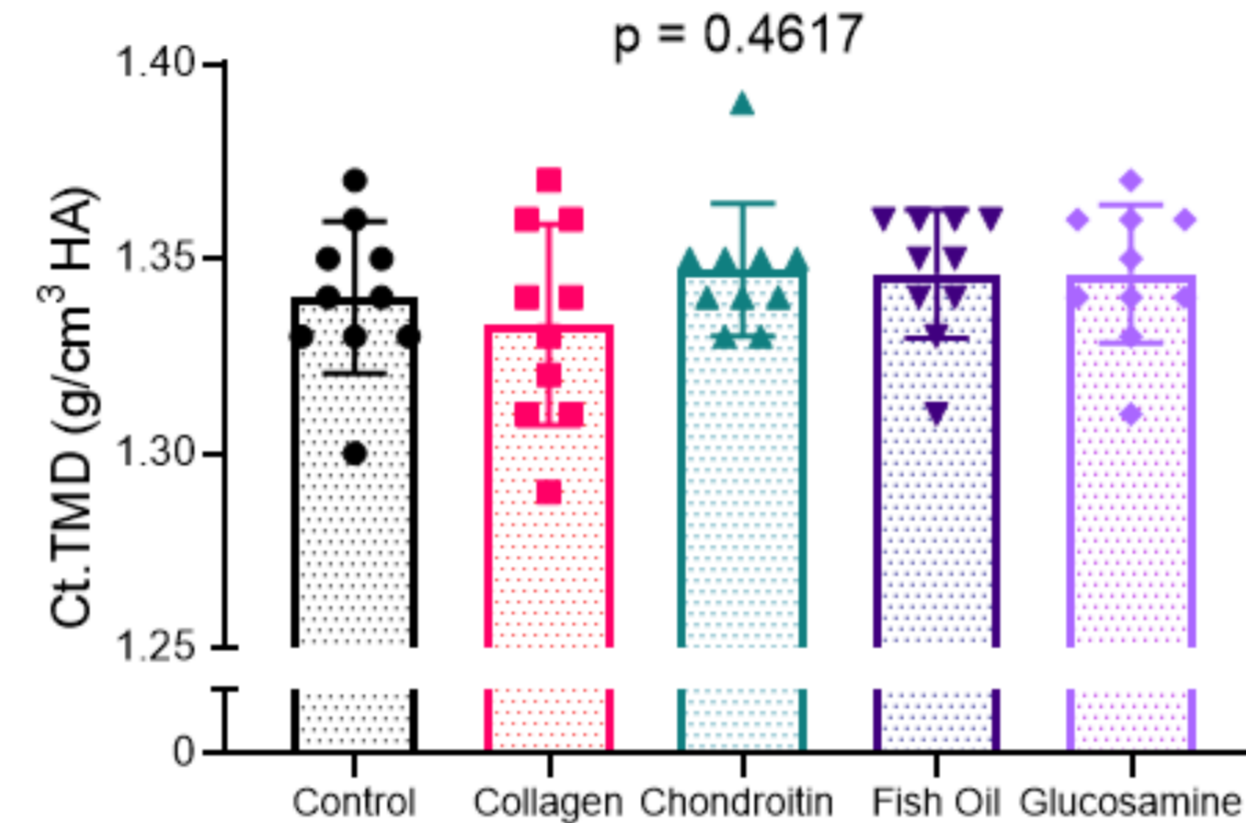
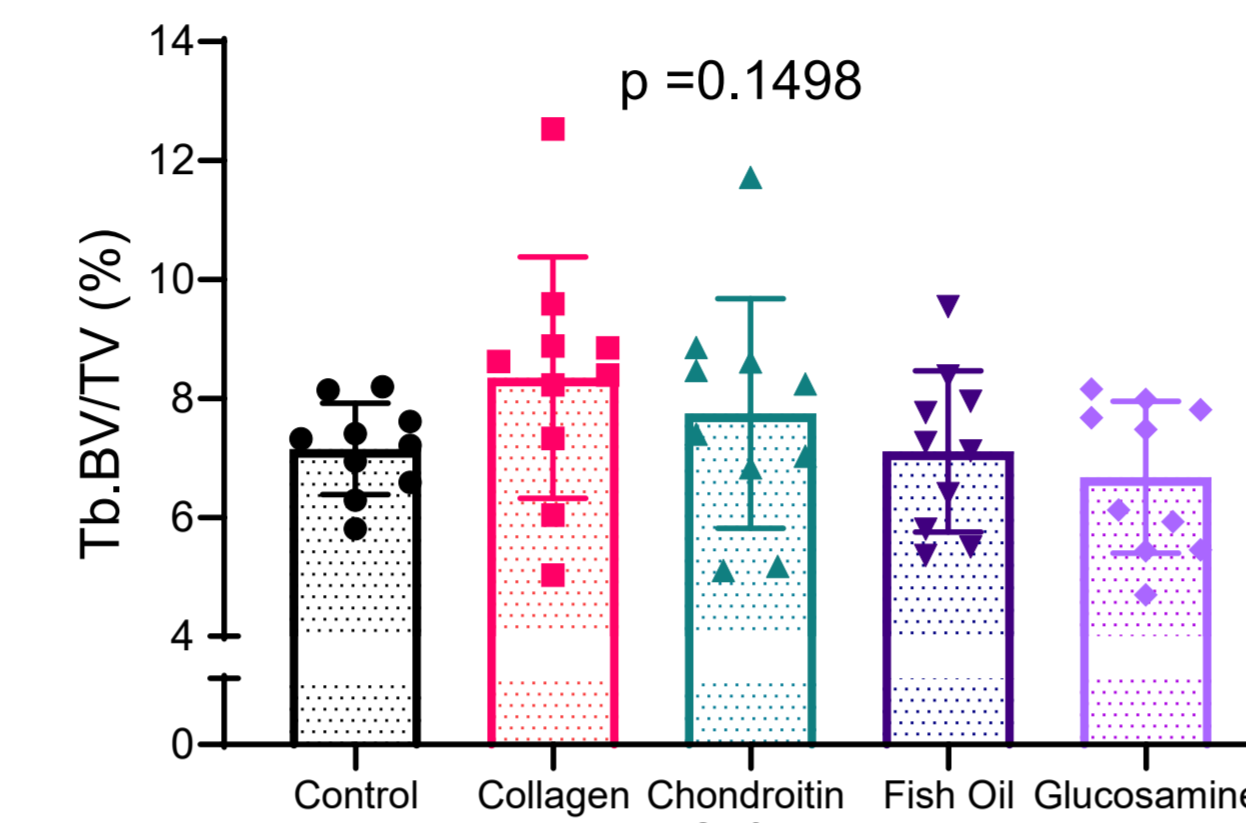


Bending test

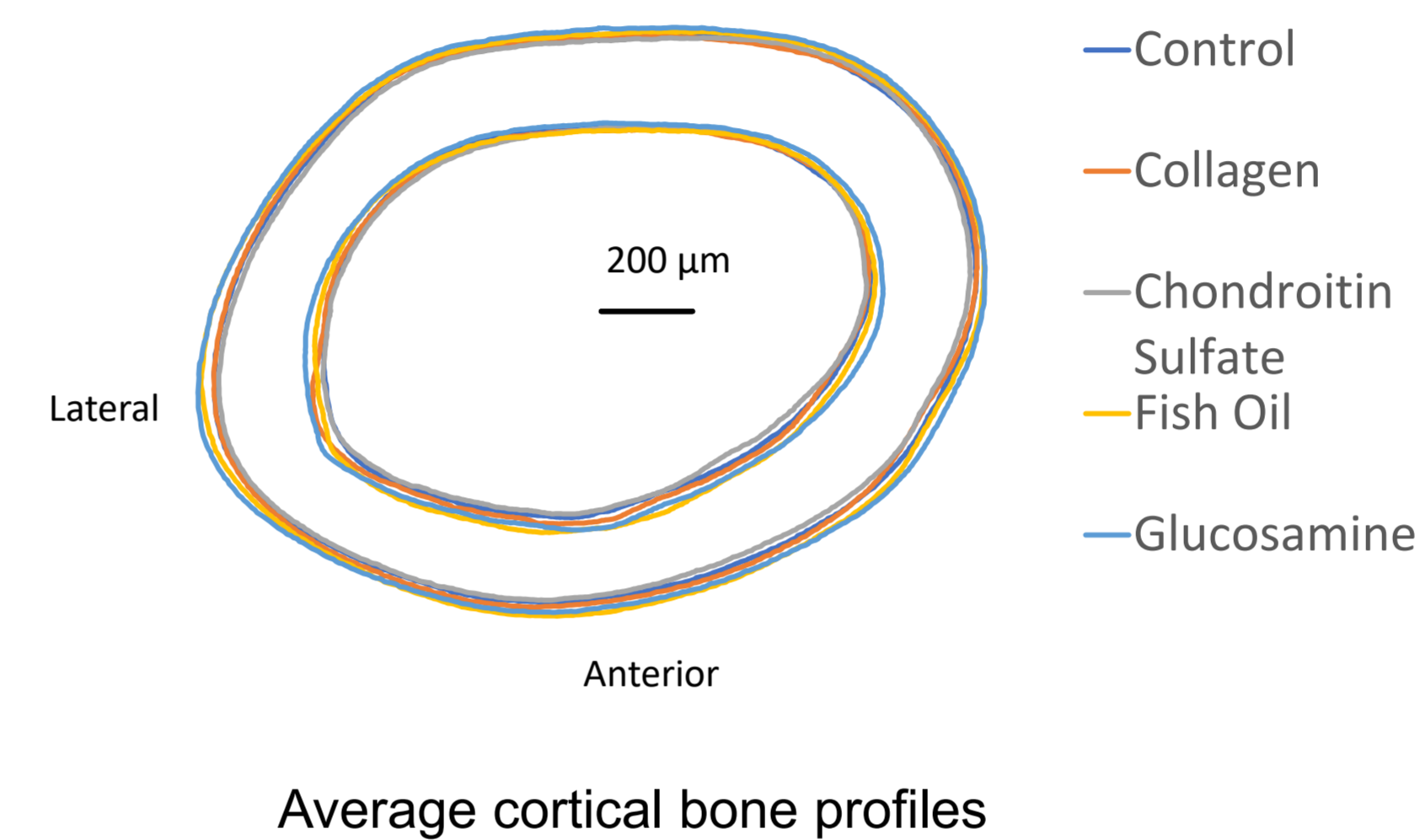


Compression test

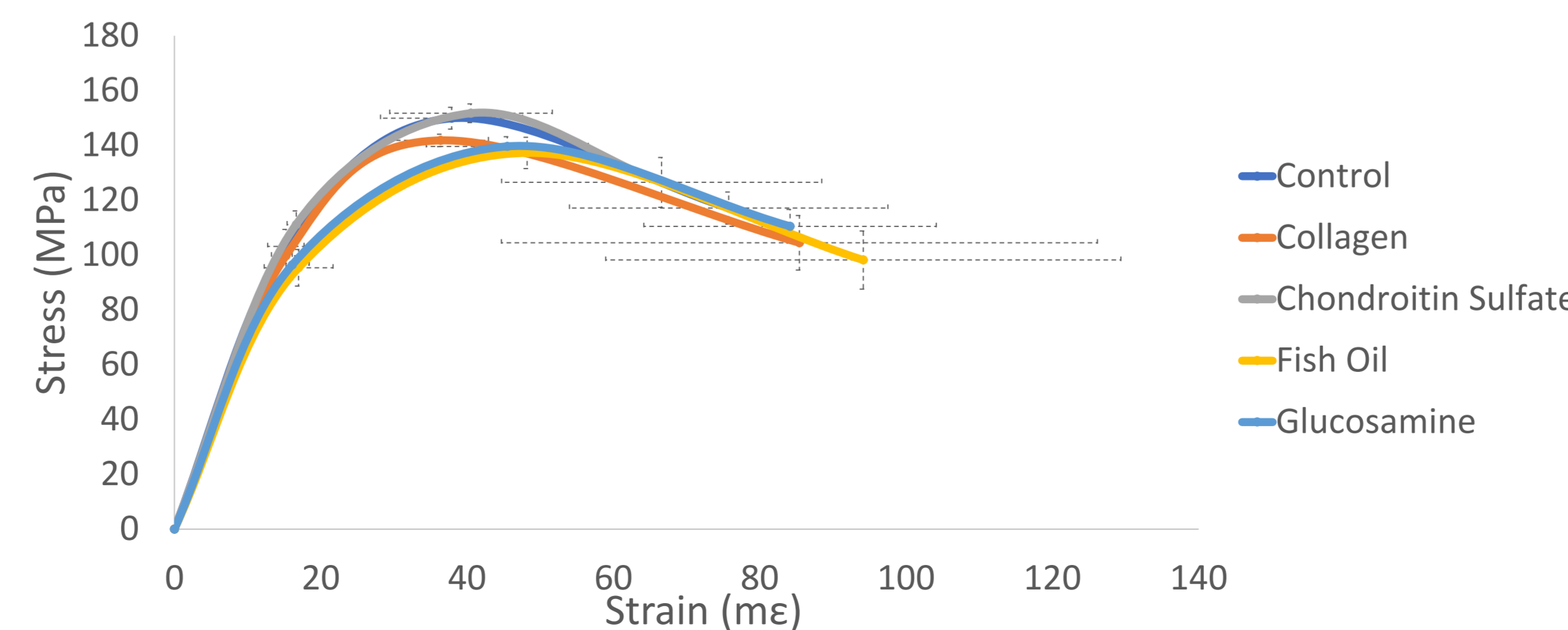
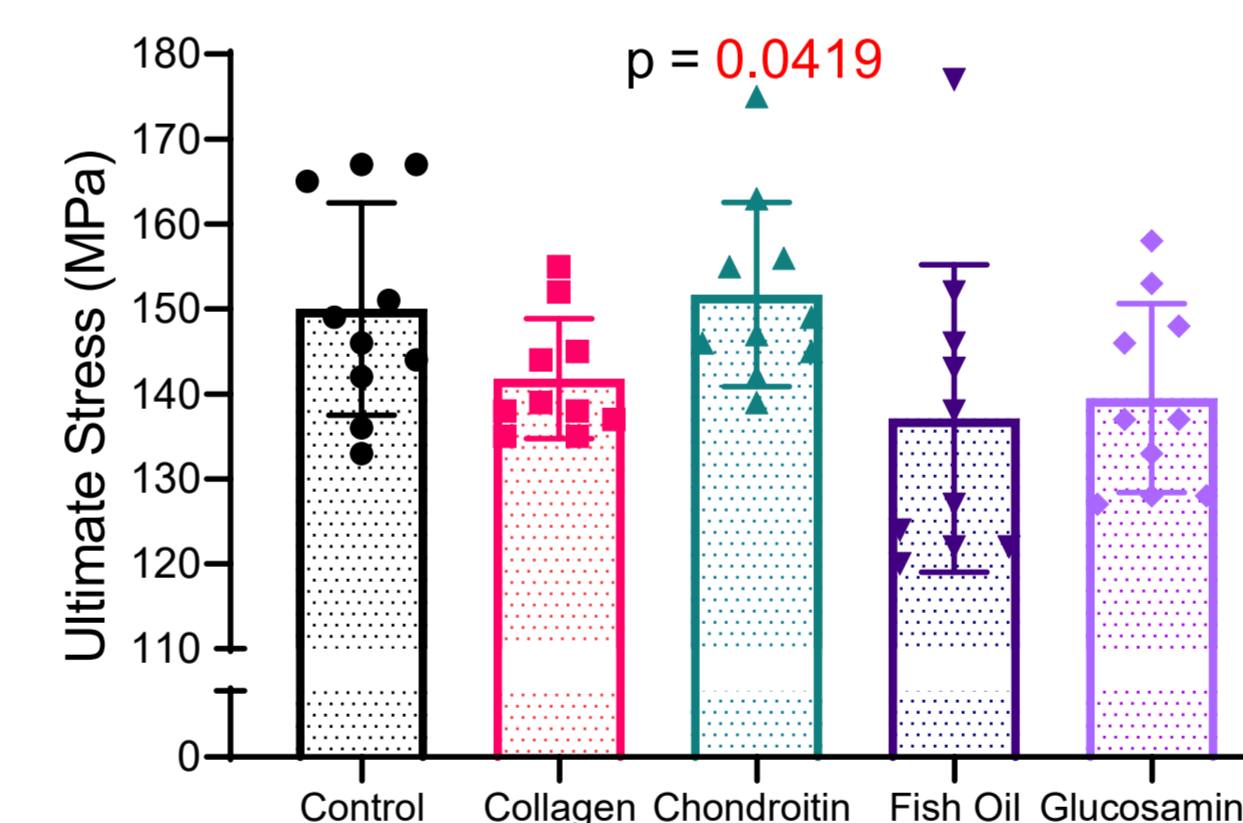
There were few structural and material level changes in femurs of treated mice



All graphs show mean, standard deviation and individual data points. ANOVA p-values are listed above. * indicate a statistically significant difference to the control data.

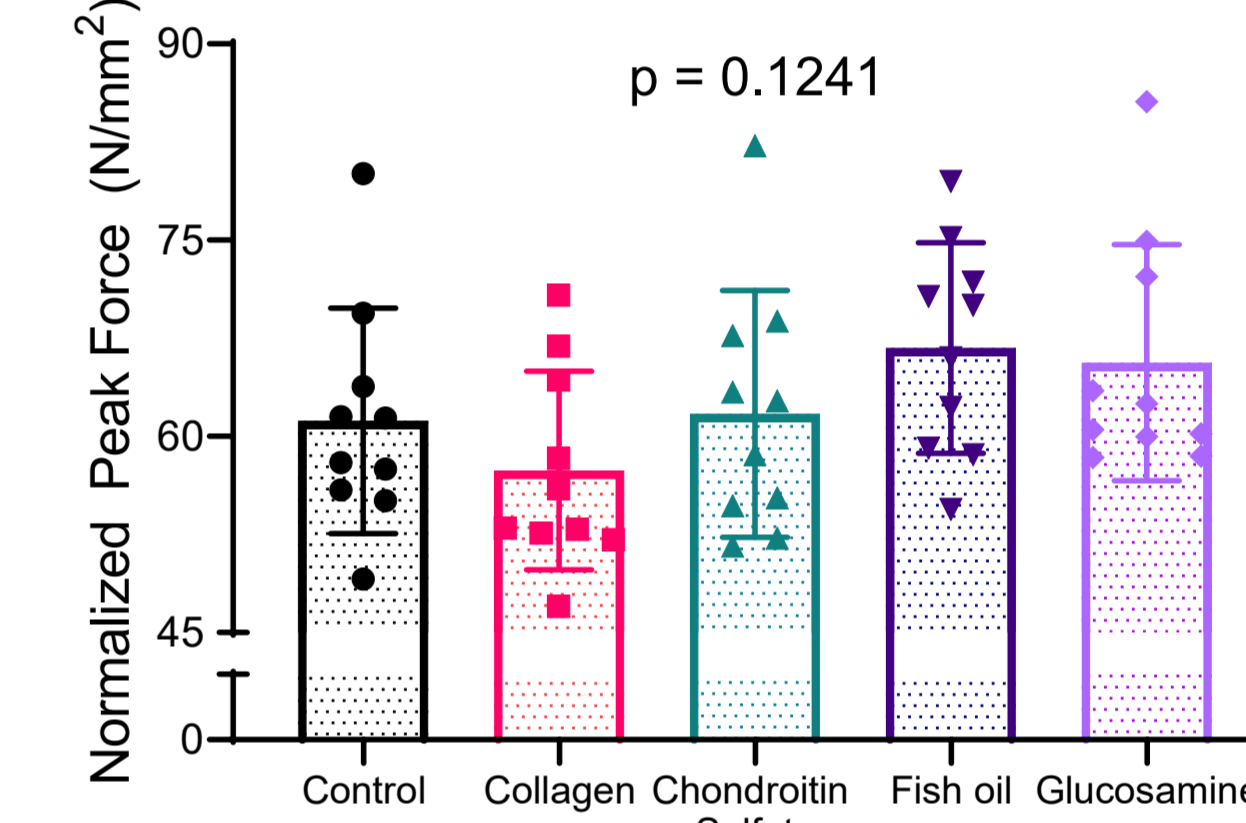
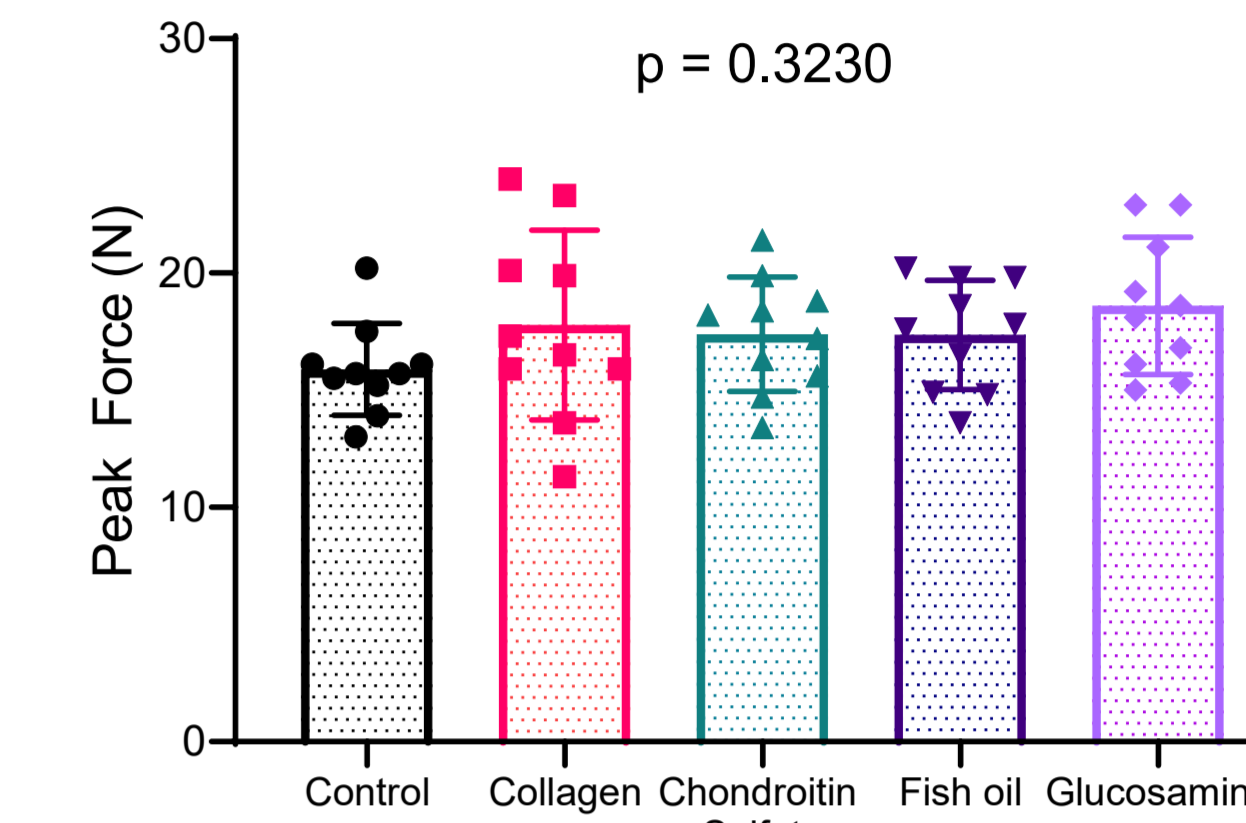
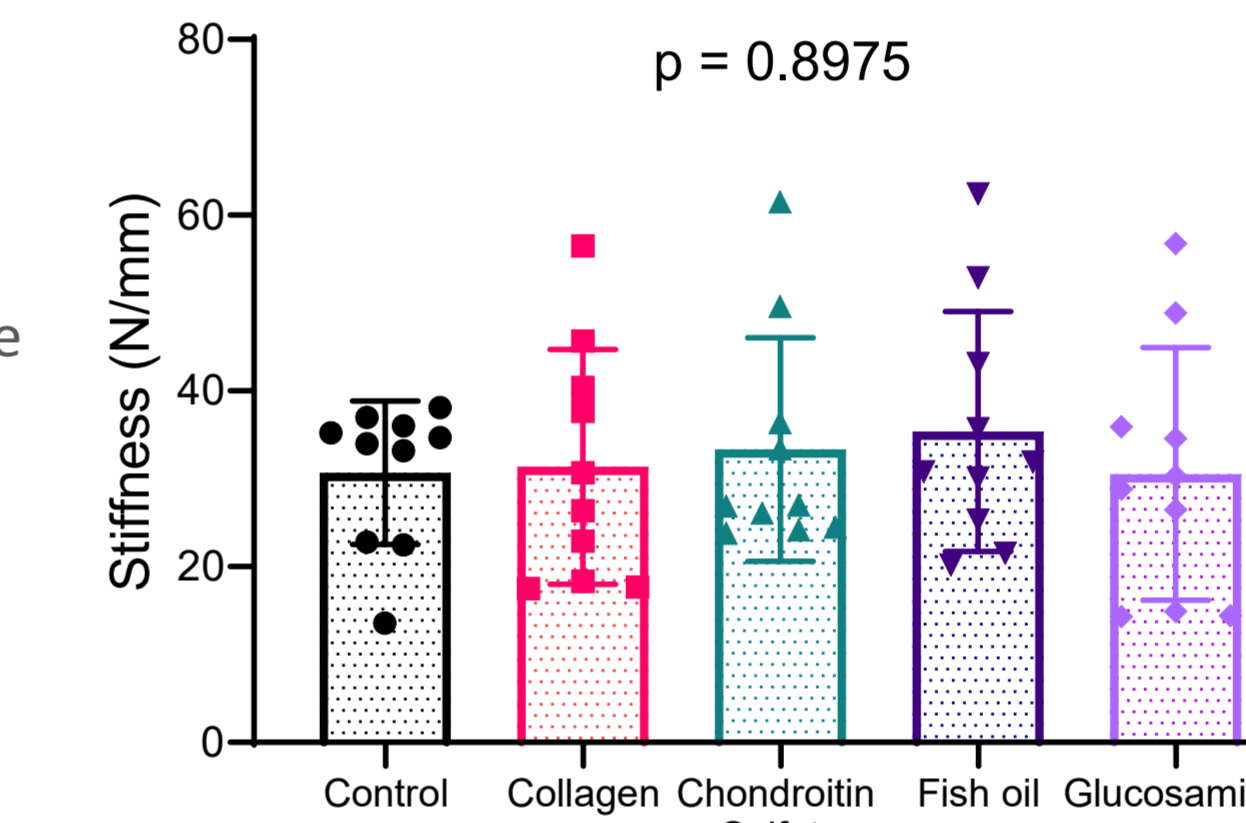
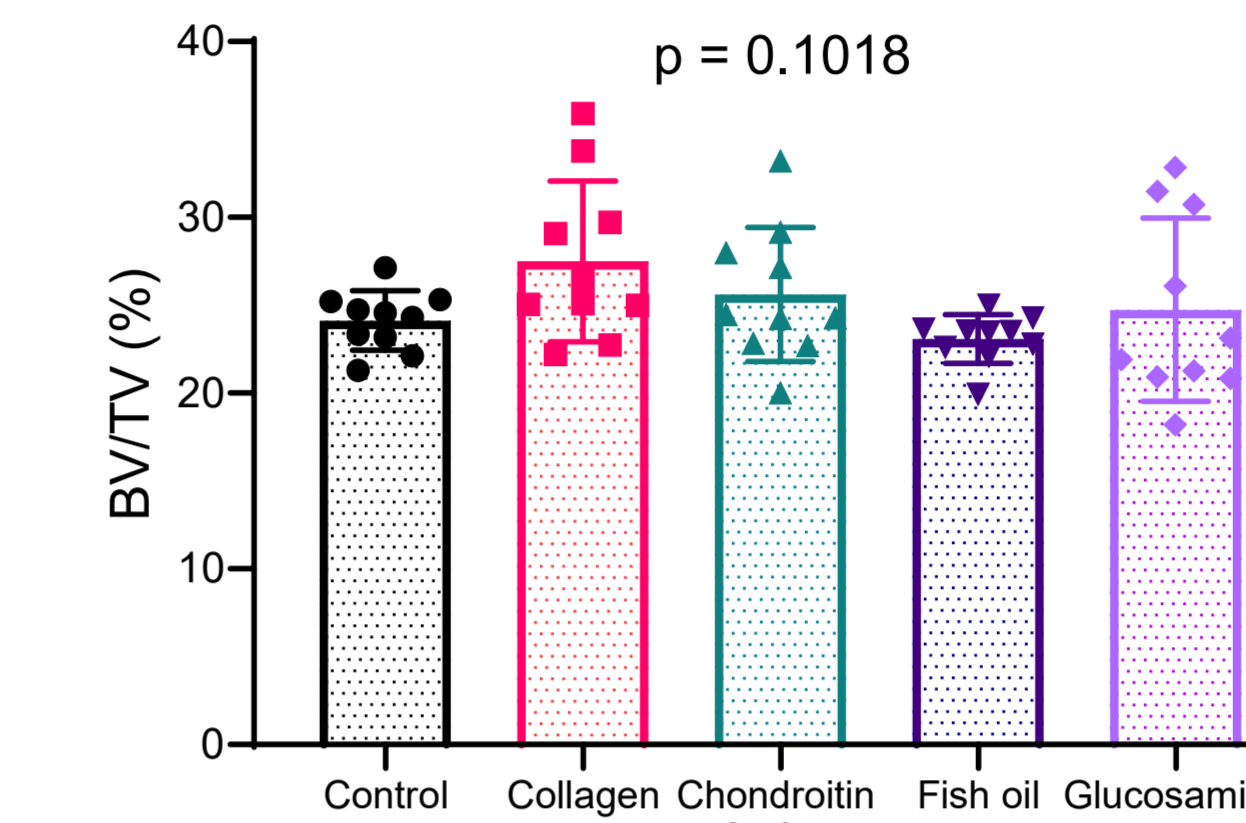


Average cortical bone profiles



Schematic stress-strain curve of average material properties with SEM

Bone volume and compressive strength did not alter in trabecular bone of vertebrae



There were no differences in bone volume fraction (BV/TV), peak force to compression, stiffness, or peak force to compression normalized by the bone area of trabecular bone in the vertebrae. ANOVA p values are given above graphs

Discussion

Overall, dietary supplements did not improve bone quality in long bones or in trabecular bone at either the femur or vertebrae indicating a probable lack of incorporation into the bone matrix

Some limitations of this study include that it is unsure if each mouse ingested the same amount of dietary supplement and only healthy females were treated

Dietary supplements may not be able to improve bone quality in treated mice relative to healthy younger adults in mice

Conclusion

Dietary supplements are not appropriate for improving bone quality in healthy, younger adults

References

- [1] Unal et al. *Curr. Osteoporos. Rep.* 2018. [2] Judex et al. *Calcif. Tissue Int.* 2000. [3] Jackix et al. *J. Med. Food.* 2010.

Acknowledgements

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