

Onset of obesity late in life increases bone matrix strength in a sexually dimorphic manner

Erika L. Cyphert^{1,2}, Chongshan Liu², Emily Blackford², Angie L. Morales², Jacob C. Nixon², Christopher J. Hernandez^{1,3}
¹University of California, San Francisco, CA, ²Cornell University, Ithaca, NY, ³Chan Zuckerberg Biohub, San Francisco, CA
Email of presenting author: elc98@cornell.edu

Disclosures: None to disclose

INTRODUCTION: Aged adults are at an elevated risk for musculoskeletal injuries that contribute to loss of mobility and subsequent weight gain. The relationship between late-onset obesity and postmenopausal osteoporosis is multi-faceted and not fully understood [1]. In mice, the effect of obesity on bone strength and geometry has been studied primarily in young animals [2-3], however, it remains unclear how the onset of obesity in aged mice influences bone health when mice are at an increased risk for osteoporosis. The effect of the onset of obesity late in life causes changes to muscle and immune function and behavior that are not observed in young animals [4-5] but has not been studied in bone. Here we evaluated the effect of the onset of obesity after 18 months of age on bone strength and femoral geometry in both female and male mice. We hypothesized that the onset of obesity late in life would increase loading forces and bone formation, increasing the size and strength of the femora in both males and females.

METHODS: Animal procedures were approved by the local Institutional Animal Care and Use Committee. Eighteen month old male and female C57Bl/6 mice bred in our facility were divided into two experimental groups (n=9-12/group/sex; 40 total): 1) high-fat diet 18-20 months of age (60% Kcal fat, D12492, Research Diets) and 2) standard lab chow. Eighteen months of age in a mouse is considered analogous to 56-60 years of age in humans. Animals were euthanized at 20 months of age due to rapid premature death of animals from weight gain. Femora were scanned using micro-computed X-ray tomography (10 μ m voxel) using a mineral calibration phantom. Geometric measurements of the cortical diaphysis were processed by applying a Gaussian blur and a threshold according to Otsu's method and analyzed using BoneJ. Cortical-cross-sectional area, cortical thickness, moment of inertia, distance from the neutral axis to the edge of the bone surface, and section modulus were measured. Femora were tested to failure using three-point bending in the anterior-to-posterior direction.

RESULTS: Females on the high fat diet had a greater whole bone strength ($p = 0.006$) and tissue strength ($p = 0.005$) relative to mice on the chow diet, whereas no differences in bone strength were associated with the high fat diet in males. Females in the high fat group also had greater cortical cross-sectional area ($p = 0.121$) and section modulus ($p = 0.105$) relative to mice on the chow diet. During the two-month period of high fat diet, females and males nearly doubled their body weight ($p < 0.001$) whereas mice on chow lost weight. At euthanasia, females on the high fat diet had an average body mass of 44.2 ± 8.2 g (chow = 25.6 ± 3.4 g) and males on the high fat diet weighed 50.0 ± 6.5 g (chow = 30.1 ± 2.3 g). When normalized by body weight, the difference in whole bone strength between high fat diet and chow was no longer significant in females ($p = 0.143$). Femoral geometry (cross-sectional area, moment of inertia, section modulus, and femur length) was positively correlated with body weight and fat pad mass in females, but not males.

DISCUSSION: High fat diet applied late in life resulted in rapid and profound changes in adiposity across both sexes and increases in femur size and strength in females but not males. Accounting for body weight removed the effects of diet on mechanical properties and femur geometry in females, suggesting that changes in bone strength and femoral geometry were secondary to increased body weight from the high fat diet. Despite similar increases in adiposity in both females and males on the high fat diet, adiposity did not influence bone strength and geometry in males. While it is unclear why sexual differences in the effect of adiposity on bone strength and geometry were present, it is possible that changes in hormonal levels in the aged females rendered them more susceptible to perturbations in bone modeling. Further studies are necessary to measure aspects of bone quality (mineral-to-matrix ratio, crystallinity, trabecular microarchitecture) and serum estrogen and bone formation/resorption markers (P1NP, TRAP5b) to provide mechanistic insight towards observed sex-dependent differences. In summary, we found that dietary changes late-in-life can influence bone biomechanics in a sexually dependent manner, providing insight into the relationship between adiposity and bone health in an aged murine model.

SIGNIFICANCE/CLINICAL RELEVANCE: The development of obesity late in life frequently occurs following musculoskeletal injuries in aged adults and can exert secondary effects on bone health. Here we examine the effect of late onset obesity on bone strength in an aged model where there is an increased risk of osteoporosis.

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ACKNOWLEDGEMENTS: This work was supported by NIH R01AG067997 (C.J.H.) and NIH F32AG076244 (E.L.C.)

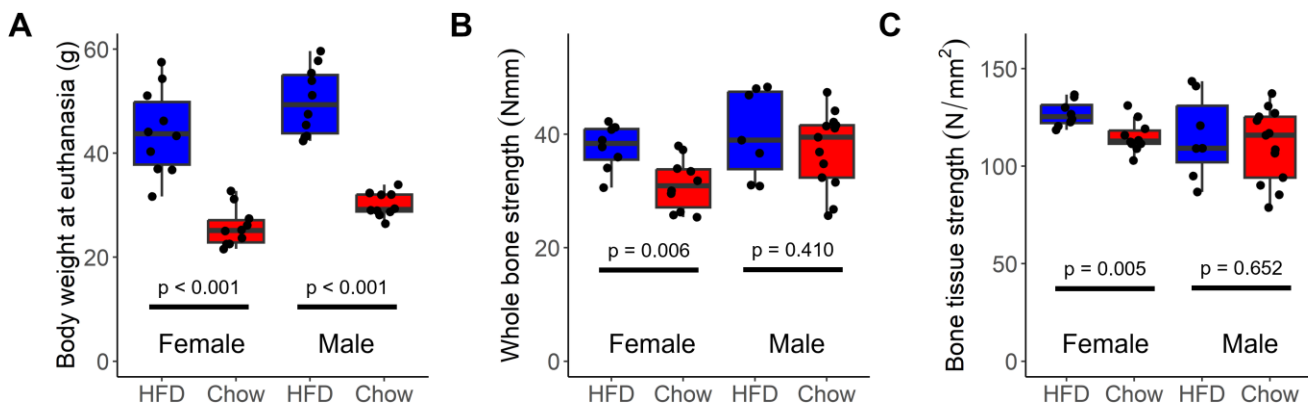


Figure 1: (A) Both sexes gained considerable weight on the high fat diet. (B-C) Whole bone strength and tissue strength was greater in females on the high fat diet relative to age-matched on the chow diet, whereas no changes were detected in males.