Exercise Improves Structural Properties in Load Bearing Bones of Female but not in Male SPARC\textsuperscript{-/-} mice

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Introduction: Secreted protein acidic and rich in cysteine (SPARC) is the most abundant glycoprotein in bone and is considered a "matricellular protein" as it mediate interactions between cells and the extracellular matrix (ECM). In bone, SPARC is expressed by osteoblasts and is thought to play a critical role in bone remodeling and homeostasis, although the exact mechanism is not clear. Sex as a variable effecting health, illness and treatment efficacy has not been well studied and is not well understood. The purpose of this study was to quantify differences in the structural and biomechanical properties between the calvarial and femoral bone from male (M) and female (F) wild-type (WT) and SPARC null (SPARC\textsuperscript{-/-}) mice (Fig. 2B-C). Conversely, F-SPARC\textsuperscript{-/-}-SED had significantly greater bone volume (BV) and bone volume ratio (BS/BV) compared to SPARC\textsuperscript{-/-} mice (Fig. 2B-C). The only significant difference in BV among females was between SPARC\textsuperscript{-/-}-SED and WT-SED. BS/BV was significantly greater in F-SPARC\textsuperscript{-/-}-SED compared to all other female groups (Fig. 2B-C). Conversely, F-SPARC\textsuperscript{-/-}-RUN had significantly greater connective density compared to M-WT-RUN and SED females. BV was significantly greater for WT compared to SED males with M-SPARC\textsuperscript{-/-}-RUN femurs having greatest reduction (Fig. 2C). The only significant difference in BS/BV was between SPARC\textsuperscript{-/-}-SED and SED-SED (M-SPARC\textsuperscript{-/-}-RUN and SED) WT-SED. BS/BV was significantly greater in M-SPARC\textsuperscript{-/-}-SED compared to M-WT-SED. Furthermore, exercise had little to no effect on rescuing structural and biomechanical properties, with only stiffness being affected. Similar reductions in structural and biomechanical strength were observed in both male and female SPARC\textsuperscript{-/-} calvaria, besides BMD which was elevated in M-SPARC\textsuperscript{-/-} mice. Most measures were not significantly affected by exercise, however, exercise did increase the compromised stiffness observed in male and female SPARC\textsuperscript{-/-} calvaria. This suggests that the effect of SPARC\textsuperscript{-/-} was greater than exercise on calvaria bone structure and function. In femurs, the effect of SPARC\textsuperscript{-/-} was detected in the structural and biomechanical parameters of both sexes. It was, however, more noticeable in females with bone quality rescued with exercise. Further research of the mechanisms that cause these differences is needed.

Methods: 7-9 month old male and female WT (n = 40) and transgenic SPARC\textsuperscript{-/-} (n=43) mice were randomly assigned to exercise or sedentary group (n=12). Exercise groups were given a running wheel while in the sedentary groups the wheel was fixed. After 6 months, the mice were sacrificed and the calvaria and femur harvested. Bone structural parameters were quantified ex-vivo using micro computed tomography (µCT). Biomechanical properties were evaluated using push out testing (calvaria) and three-point flexural testing (femurs). Morphological features of the bone were examined via histological staining.

Results: Calvaria: µCT found that M-SPARC\textsuperscript{-/-} had significantly greater bone mineral density (BMD) compared to M-WT (Fig. 2A). WT mice of both sexes had significantly greater bone volume (BV) and bone volume ratio (BS/BV) compared to SPARC\textsuperscript{-/-} mice (Fig. 2B-C). Conversely, F-SPARC\textsuperscript{-/-}-SED had significantly greater connective density compared to M-WT-RUN and SED females. BV was significantly greater for WT compared to SED males with M-SPARC\textsuperscript{-/-}-RUN femurs having greatest reduction (Fig. 2B-C). The only significant difference in BS/BV was between SPARC\textsuperscript{-/-}-SED and SED-SED (M-SPARC\textsuperscript{-/-}-RUN and SED) WT-SED. BS/BV was significantly greater in M-SPARC\textsuperscript{-/-}-SED compared to M-WT-SED. Further research of the mechanisms that cause these differences is needed.

Discussion: This study emphasizes significant differences in the structural, biomechanical, and morphological properties of bone in regards to the gender and exercise in SPARC\textsuperscript{-/-} and WT mice. In males, exercise had little to no effect on rescuing structural and biomechanical properties, with only stiffness being affected. Similar reductions in structural and biomechanical strength were observed in both male and female SPARC\textsuperscript{-/-} calvaria, besides BMD which was elevated in M-SPARC\textsuperscript{-/-} mice. Most measures were not significantly affected by exercise, however, exercise did increase the compromised stiffness observed in male and female SPARC\textsuperscript{-/-} calvaria. This suggests that the effect of SPARC\textsuperscript{-/-} was greater than exercise on calvaria bone structure and function. In femurs, the effect of SPARC\textsuperscript{-/-} was detected in the structural and biomechanical parameters of both sexes. It was, however, more noticeable in females with bone quality rescued with exercise. Further research of the mechanisms that cause these differences is needed.

Significance / Clinical Relevance: Research into gender differences can help understand, inform, and personalize treatment options for patients in order to treat patients more accurately.

ORS 2024 Annual Meeting Paper No. 518