

Measuring Denatured Collagen Debridement After *in vivo* ACL Cyclic Loading in Mice

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Background/Objective:

Anterior cruciate ligament (ACL) injuries are one of the most common and debilitating injuries in sports. Once thought to be caused due to acute stress events, recent research has demonstrated that this could be from chronic overuse and fatigue. We hypothesized that the estimated time that denatured collagen removal occurs is around 6 days to start ACL repair and there will be no changes between right and left knee mechanical parameters.

Methods:

Forty B6 female 10-wk old mice were used in a custom setup that cyclically loads to 60% of the ACL's max force for 500 cycles. All right legs were tested, and the specimens were randomly separated into four equal cohorts for rest times in 3-day increments. Following each cohort's rest time, the left knee was loaded in the same manner (n=7) and the remaining from each group (n=3) were used as untested contralateral controls. Immediately after each cohort was tested, they were euthanized, and legs were harvested, fixated, and decalcified for paraffin infiltration. Tissue was sectioned and deparaffinized for staining with R-CHP for immunofluorescence, followed by Raman spectroscopy to examine proteoglycan activity.

Results:

Current results demonstrate the mechanical data of all tested ACLs via measures of hysteresis and stiffness. No statistical differences were found, except for the hysteresis of the left ACLs between cohorts 2 and 3 and upper stiffness of the right ACLs in all mice compared to the left ACLs in cohort 3. The results from CHP staining will be analyzed in future work.

Conclusion and Implications:

Ultimately, this study will help narrow down when denatured ACL collagen from fatigue begins as an indication of the repair process taking place. This knowledge may be used in athletes with strained ACLs to know how long to adequately rest before continuing sport activity.