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"Using the Articularis Genu to Test Peri-Articular Muscle Health During Knee Osteoarthritis"

Knee osteoarthritis (OA) involves atrophy and alterations to myofiber distribution in the quadriceps. The *Articularis genu* (AG), an intra-articular muscle that retracts the suprapatellar bursa during extension, originates at the distal quadriceps, with similar mechanism, concurrent innervation, and composition. The AG is removed during total knee arthroplasty (TKA) and understanding of its sensitivity to OA is limited. We predict that the sensitivity of AG fibers to OA will be similar to published data on the vastus medialis (VM) and vastus lateralis (VL) during OA and that changes in fiber phenotype will associate with total range of motion (ROM).

OA patient AGs (n=40) collected during TKA were fixed, processed for paraffin sectioning, and grouped by ROM as poor (≤85º; n=11), fair (90º to 115º; n=19), and good (≥115º; n=10). Using quantitative immunofluorescence against slow and fast myosin heavy chains, we differentially co-detected type-I (T1) and type-II (T2) myofibers to compare mean distribution and cross-sectional areas (CSA) between groups by ANOVA. Cryopreserved AGs were homogenized, RNA isolated, cDNA synthesized, and qPCR executed per manufacturer instructions, and assayed. Primer pairs for *MYH1*, *MYH7*, *FBXO32*, and *TRIM63* were used against the housekeeping *hHPRT1* using the comparative CT method and log scaled. Non-parametric, unpaired t-tests were used between groups and RNA data. Linear regression with Pearson's coefficient (r) was used to test associations. Alpha was set to 0.05.

The T1/T2 ratio in fair and poor ROM groups was lower than those published on healthy AGs and quadriceps, but consistent with OA quadriceps. Decreasing T1 percentages (mean±SEM) from good (56.69±3.72), to fair (49.04±1.36), and poor (39.90±2.50) ROM groups was significant between poor versus fair ($p=0.018$) and good ($p<0.0001$), supported by negative *MYH7* (T1) and positive *MYH1* (T2) expression trends from good to poor ROM groups. We tested significant associations between ROM versus T1, and conversely T2, counts ($r=\pm0.729$; $p<0.0001$) and CSA changes in T1 ($r=0.5854$; $p<0.001$) and T2 ($r=0.5200$; $p=0.0006$). Increasingly positive trends in expression of atrophy-specific genes *TRIM63* and *FBXO32* from good to poor ROM groups support a ROM-dependent fiber atrophy.

By integrating phenotypic changes to the OA AG with clinical parameters, we generated pilot data indicating that the AG undergoes alterations in fiber distribution and atrophy, associated with knee disuse, consistent with findings from similar analyses of the VL and VM. Results suggest that OA-related disuse may act in synergism with aging-related muscle wasting to more severely alter the AG. This study warrants further investigation to overcome its limitations, such as a larger sample size and healthy AG controls. Yet, our data suggest that the AG has the potential to surrogate the OA quadriceps in a diagnostic platform, which could potentially guide peri-operative knee pain management and strength rehabilitation strategies post-TKA.