**Appendix 2: Comparison of Animal Trial Methods**

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| **Study** | **Transection Method** | **Repair Surgery** | **Biomechanical Evaluation** | **Histology** | **ASC Source** | **ASC Preparation** | **Immobilization After Treatment** |
| Barco et al. 2015 [28] | Supraspinatus was transected with a blade. Subjects were randomized after transection | Group 1: Intraosseous tunnel was drilled into the humerus using 5/0 monofilament suture. Group 2: same repair with addition of fibrin sealant. Group 3: Same repair with 2 × 106 ASC embedded in fibrin sealant. | There was no preconditioning. Electrohydraulic load cell was preloaded to 0.1N and loaded until failure at a rate of 14 µm/s | Samples were decalcified, fixed, embedded, and sectioned in 5 um sections in the coronal plane then stained. | Epiploon and subcutaneous fat from 2 rats. | After harvesting, fat was mechanically disaggregated and enzymatically digested. RBCs were lysed using ammonium chlorhydrate. ASCs were then isolated and their lineage was confirmed. ASCs were then cultured for multiplication before injection. | No |
| Chen et al. 2015 [29] | Collagenase was injected around the supraspinatus tendon |  | JSVH1000 machine used. Tissue preloaded to 5N and pulled tendons at rate of 10 mm/min. | Samples were fixed, embedded, and sectioned in 2 mm then stained. | 3 g of ASCs were obtained from the abdominal fat of a 46 year-old Asian male | ASCs were exposed to trypsin and 3 × 108 cells/ml in PBS were injected at collagenase site. | No |
| Kaizawa et al. 2019 [30] | Supraspinatus and surrounding scar tissue was transected from the humerus. | All groups underwent repair with 5-0 Prolene through a bone tunnel and were then randomized. | A 5944 materials testing system was used. Opposite ends of specimens were secured in the machine or a vise. | Samples were fixed, embedded, and sectioned in 5‐μm‐thick coronal sections then stained. | ASCs were extracted from the flexor tendons of human cadavers | Flexor tendons were decellularized, lyophilized, and reduced to a powder that was digested and then neutralized. Cells were then incubated. | No |
| Lee et al. 2020 [31] | Supraspinatus, infraspinatus, and suprascapular nerve were all transected. |  | None | Samples were frozen with isopentane that underwent cooling with liquid nitrogen | Adipose tissue from UCP-1 reporter mice | No ASCs underwent any alterations before being injected. | No |
| Lu et al. 2020 [33] | Supraspinatus was transected with a blade. | Supraspinatus was sutured to greater trochanter using mattress method. | MTS858 testing machine was used. | Samples were fixed, embedded, sectioned, and stained. | Inguinal yellow fat from rabbits | ASCSs were washed with PBS, digested with type I collagenase, and centrifuged to yield SVF-FG | Yes, immobilization with plaster for 3 weeks |
| Lu et al. 2018 [32] | Supraspinatus was transected with a blade. | Supraspinatus was sutured to greater trochanter using mattress method. | MTS858 testing machine was used. | Samples were fixed, embedded, sectioned, and stained. | Inguinal yellow fat from rabbits | ASCSs were washed with PBS, digested with type I collagenase, and centrifuged to yield SVF-FG | Yes, immobilization with plaster for 3 weeks |
| Oh et al. 2014 [34] | Supraspinatus was severed at its insertion on the humerus. It was wrapped in a Penrose drain to prevent adhesions | At 6 weeks, Penrose drain was removed and the supraspinatus tendon was reattached with 2 Mini QUICKANCHOR Plus anchored 2-0 Ethibond sutures that were tied using mattress technique. | Intron 5565A machine. Preconditioning. Rate of elongation was 1 mm/s preloaded with 5N for maximum load testing | Samples were fixed, embedded, and sectioned transversely, then stained. There were 2 evaluators for each slide who were blinded. | Inguinal fat from 2 male rabbits not involved in the study. | Fat tissue was digested with type I collagenase, centrifuged, exposed to RBC lysis buffer, filtered with a mesh, and centrifuged once again to create SVF. ASCs were 'detached' and washed with PBS. ASCs (1 × 107) in 500 μL Hank's balanced salt solution was injected | No |
| Rothrauff et al. 2018 [35] | Supraspinatus and infraspinatus tendons were transected with number 11 blade. Remaining fibrocartilage was taken out with a burr. | Adhesions were lysed and an intraosseous tunnel was created with a 22-gauge needle. Supraspinatus and infraspinatus tendons were sutured with 5-0 Prolene using the Mason-Allen stitch and passed through the intraosseous tunnel and "partially" tightened. | Instron 5965 tester was used. Tissues preloaded and preconditioned Load-to-failure assessed with a rate of 5 mm/min and determined with a load-elongation curve. | Samples were decalcified, fixed, embedded, and sectioned in 6 um sections in the coronal plane then stained. | Inguinal fat from rats | Fat was rinsed with PBS, digested with collagenase type I, filtered, and centrifuged. The pellet was washed, filtered and centrifuged again and subsequently underwent RBC lysing. This was centrifuged again, and the resulting pellet was exposed to growth medium that contained fibroblast growth factor 2. Colony-forming ASCs were used for injection | No |
| Valencia et al. 2014 [67] | Supraspinatus was transected with a blade. | Intraosseous tunnel was drilled into the humerus. The supraspinatus tendon was sutured using 5/0 suture and a modified Masson stitch, then it was passed through the bone tunnel and tied off. Following surgery, subjects were randomized. | Linear encoder preloaded to 0.10N with a rate of 14 µm/s until failure was reached. | Samples were fixed, embedded, and sectioned in 4 um coronally then stained. | Subcutaneous fat from 2 rats | Fat was rinsed using PBS, digested with collagenase type I, neutralized with fetal bovine serum, and centrifuged. The pellet was treated to lyse RBCs and was then washed with DMEM with 10% FBS. SVF created. The ASC group received 2×106 ASCs in a collagen carrier | No |
| Wang et al. 2019 [37] | Supraspinatus and infraspinatus were transected near the humeral greater tuberosity. Penrose drain used to prevent adhesions | Holes in humerus were drilled for transosseous RCR using two No. 2 orthocord sutures. Following repair, rabbits were injected with 1011 ASC-Exos with 20 uL of saline. | Preloading and preconditioning performed. Maximum load to failure found with load-elongation curve | Samples were fixed, embedded, and sectioned in 5 um sections then stained. | Human subcutaneous fat obtained via liposuction | Extracted fat was conditioned and centrifuged to get rid of unwanted material. The samples were then centrifuged again to separate ASC-Exos into a pellet. This pellet was treated with PBS and centrifuged for a final time. Afterwards, ASC-Exos was suspended in saline for injection. | No |
| Wang et al. 2020 [38] | Supraspinatus and infraspinatus were transected near the humeral greater tuberosity. | No surgery | Instron 5568. Preconditioning Testing elongated specimens to 3% strain with a rate of 50%/s and a subsequent 300 s relaxation period. Load-to-failure was determined at a rate of 0.3%/s. | Samples were fixed, embedded, and sectioned in 7um sections then stained. | Human subcutaneous fat obtained via liposuction from 7 women (19-63 years). | After several rounds of passage to reach 70%-80% confluency, ASCs were washed with PBS, centrifuged, and ultracentrifuged. ASC-Exos in centrifuge tube were again suspended in PBS and centrifuged once more to extract ASC-Exos pellets. | No |

ASC, adipose stem cell; DMEM, Dulbecco's modified Eagle's medium; Exos, Exosomes; FBS, Fetal Bovine Serum; FG, Fibrin Glue; min, minutes; PBS, Phosphate Buffered Saline; RBC, red blood cells; s, seconds; SVF, Stromal Vascular Fraction.