Fibroblast Proliferation and Collagen Secretion Are Required For Myofascial Release-Induced Wound Healing In Three Dimensional Bioengineered Tendons

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BACKGROUND: Myofascial release (MFR) is one of the most common manual medicine treatments and we recently reported that modeled MFR hastens wound healing in vitro. However, little is known about the mechanisms underlying the efficacy of MFR. Fibroblasts are the principle cell type in fascia, secrete collagen critical to the extracellular matrix and are prime targets for the mechanical strains that MFR imparts. We have previously shown that relatively long duration and low magnitude modeled MFR results in optimal wound healing rates in bioengineered tendons (BETs). The objective of this study is to elucidate the role of fibroblast proliferation and collagen secretion in MFR-induced wound healing.
METHODS: BETs were cultured on deformable matrices and then wounded using a steel cutting tip. Using vacuum pressure, BETs were uniaxially strained with a modeled MFR stretch magnitude of 3% beyond resting length for 5 minutes using previously published loading and unloading parameters. BETs were then incubated in either 2% fetal bovine serum (FBS; required concentration for fibroblast proliferation) or 0.2% FBS (required concentration to arrest fibroblasts in the G_o phase of the cell cycle). Daily measurements of BETs' width and wound area were recorded over 6 days and quantified microscopically. Human fibroblast collagen staining and nuclei deposition were assessed using colorimetric and immunofluorescence techniques.
RESULTS: Although significant reductions in wound size were observed in nonstrained (NS) and MFR groups cultured with 2% FBS compared to Day 0 (day of wounding), the MFR group displayed significantly improved wound healing versus the NS group (Figure). Both NS and MFR groups showed about 60% reduction in BETs' width at Day 6 compared to Day 0 (n=11, $P<0.05$). MFR revealed a 61% increase in collagen staining and more nuclei deposition compared to the NS group at Day 6 (n=3, $P\leq0.05$). All measured differences between NS and MFR were completely abrogated when fibroblast proliferative responses were blocked (n=6, $P<0.05$).
CONCLUSION: Modeled MFR greatly enhances wound healing rates in BETs. In the absence of fibroblast proliferation, wound healing rates were slowed and collagen secretion was reduced in MFR-strained BETs. If these results are clinically translatable, they support a likely mechanism by which MFR improves wound healing.
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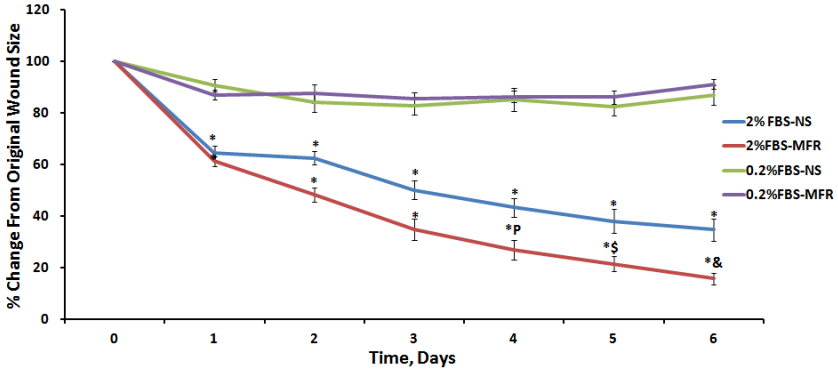

Figure: Wounded bioengineered tendons (BETs) treated with 3% myofascial release (MFR) for 5 minutes. Change in wound size was measured, as a percent change from Day 0 (100%), daily for six days after MFR treatment in BETs cultured with 2% FBS (n=11) and 0.2% FBS (n=6). Data are given as a (SEM). *Statistically significant decrease ($P<0.05$) in wound size compared with Day 0. (P, $ and &) Statistically significant decrease ($P<0.05$) in wound size compared to 2% FBS cultured Non strain (NS) BETs at Days 4, 5 and 6 respectively.