Parathyroid Hormone Suppressed Fatty Infiltration and Muscle Atrophy after Rotator Cuff Tear by Fibro-Adipogenic Progenitors Browning in a Rodent Model

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INTRODUCTION: Progressive fatty infiltration and muscle atrophy after rotator cuff tears cause increased retear rate after tendon repair and poor outcomes. Fibro-adipogenic progenitors (FAPs) are involved in fatty infiltration and skeletal muscle homeostasis. Inducing FAP differentiation into brown adipocyte-like "beige adipocytes" suppresses fatty infiltration. Moreover, their secreted factors are involved in muscle regeneration. This study aims to clarify that parathyroid hormone (PTH) inhibits fatty infiltration and muscle atrophy after rotator cuff tears in a rat model by browning of FAPs.

METHODS: PTH (Teriparatide, 30 ug/kg, three times a week) was administered subcutaneously for 4 or 8 weeks to a rat rotator cuff tear model. After treatment, fatty infiltration of supraspinatus muscles was evaluated by Oil Red O staining and muscle atrophy was evaluated by wet muscle weight and muscle fiber cross-sectional area. Co-staining of PDGFRα (FAPs marker) and UCP1 (Browning marker) was conducted to confirm the browning of FAPs by PTH. FAPs isolated from mice were cultured with PTH and assessed for browning-related gene expression by PCR and adipogenic differentiation by BODIPY staining. The expression of VEGF as secreted factors by browning FAPs for muscle regeneration was assessed by PCR and ELISA in vitro and immunostaining in vitro. Furthermore, myogenic differentiation of C2C12 myoblasts was assessed by co-culture of PTH-treated browning FAPs with C2C12. Myogenic differentiation and angiogenesis as an indirect effect of VEGF on muscle regeneration were also evaluated by immunostaining in vivo.

RESULTS: The wet muscle weight loss of the rotator cuff muscle of PTH-treated rats was suppressed at 4 and 8 weeks (Figure 1). PTH suppressed fatty infiltration after rotator cuff tear at 8 weeks (Figure 2). Moreover, the PTH-treated rats showed a larger myofiber cross-sectional area than untreated rats at 4 and 8 weeks, and PTH promoted the expression of UCP1, a browning marker (Figure 3). Co-staining indicated co-localization of PDGFRα and UCP1 and the promotion of the browning of FAPs by PTH (Figure 4). PTH increased the expression of browning-related genes in FAPs and inhibited the accumulation of fat droplets in vitro (Figure 5). PTH promoted the expression of VEGF, confirmed by PCR, ELISA, and immunohistochemistry (Figure 6). Co-culture with PTH-treated FAPs stimulated the differentiation of C2C12 cells into myotubes, and a VEGF receptor inhibitor partially inhibited the promotion of myogenic differentiation by PTH (Figure 7a). PTH also stimulated myogenic differentiation and angiogenesis in vivo (Figure 7b-c).

DISCUSSION AND CONCLUSION: PTH induced FAPs-derived beige adipocytes by promoting the browning-related gene expression, and the browning effect of PTH on FAPs suppressed fatty infiltration and muscle atrophy after rotator cuff tear in a rodent model. The results suggest that PTH might have the potential as a therapeutic agent for fatty infiltration and muscle atrophy after rotator cuff tears.