Dynamic Loading Enhances Integrative Meniscal Repair in the Presence of Interleukin-1

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 INTRODUCTION: Meniscal tears are a common knee injury, and in addition to the pain and loss of function associated with the initial injury, damage or loss of meniscal tissue is associated with degenerative changes in the joint that ultimately lead to osteoarthritis (OA). Inflammatory cytokines, particularly interleukin-1 (IL-1), are upregulated in injured and osteoarthritic joints, inducing many degradative and pro-inflammatory pathways. Our previous studies using an in vitro repair model have shown that IL-1 diminishes repair by decreasing the integrative shear strength, meniscal cell accumulation, and tissue formation at the interface of meniscal lesions (1,2). In addition to biochemical factors, biomechanical factors also influence cell metabolism and matrix turnover in the meniscus (3). Mechanical stress and IL-1 modulate meniscal biosynthetic activity and degradation, and dynamic loading can induce anabolic and anti-inflammatory responses that overcome some of the effects of IL-1 (4,5,6). However, the effects of dynamic compression on the repair of meniscal lesions are not known. We hypothesized that mechanical compression at lower magnitudes (<10%) would enhance meniscal repair under both normal and inflammatory conditions, and that higher magnitudes (>10%) of dynamic compression would inhibit repair.

METHODS: Explants (5 mm diameter) were harvested from skeletally mature female porcine medial menisci. To simulate a full-thickness defect, a 3 mm diameter core was removed and reinserted. Explants (n > 9 per treatment group) were loaded in a custom-built closed-loop mechanical compression bioreactor for 4 hours per day at 1 Hz at various levels of strain, ranging from 0 – 26% for 14 days in the presence of 0 or 100 pg/mL of porcine IL-1 alpha (R & D Systems). Media was assessed for matrix metalloproteinase (MMP) activity with a fluorescence based assay (7), sulfated glycosaminoglycan (S-GAG) release using the dimethylmethylene blue (DMB) assay (8), and nitric oxide (NO) production using the Greiss reagent to measure both nitrite and nitrate (9). At the end of the culture period, a push-out test was performed to determine the interfacial shear strength between the outer ring and inner core (1). Statistical analyses were performed using Statistica 7.0 (StatSoft Inc.). A two-factor ANOVA and the Newman-Keuls post hoc test were performed to determine significant differences and the interactive effect of strain and IL-1.

RESULTS: The MMP activity in the media of meniscal repair model explants was increased by IL-1 treatment in the absence of strain (Figure 1; p < 0.0005). However, dynamic compression at strains of 1%, 10%, and 26% blocked the increase in MMP activity due to IL-1 (p < 0.03). At both 1% and 10% strains, there was an interactive effect of strain and IL-1 on MMP activity (p < 0.001). In the absence of IL-1, there was no change in MMP activity at any of the strains tested.

In the absence of strain, S-GAG release was enhanced ~5-fold by IL-1 treatment (Figure 1; p < 0.0005). Strains of both 1% and 10% were effective in reducing the IL-1-mediated release of S-GAG (p < 0.005). However, 26% strain had no effect on S-GAG release in the presence of IL-1. At both 1% and 10% strains, there was an interactive effect of strain and IL-1 (p < 0.03). None of the strains tested altered the S-GAG release into the media in the absence of IL-1.

NO release into the media was increased by IL-1 treatment in the absence of strain (Figure 2; p < 0.0005). However, the application of all tested strains reduced the IL-1-mediated NO release (p < 0.002). At all strains, there was an interactive effect of strain and IL-1 that contributed to NO release into the media (p < 0.02). In the absence of IL-1, none of the strain magnitudes altered NO release from meniscal repair explants.

In the absence of strain, IL-1 decreased the integrative shear strength of repair in meniscal repair model explants (Figure 2; p < 0.03). The administration of 1%, 10% and 26% strain increased the shear strength of repair of IL-1 treated explants to strengths that were comparable to that measured in unstrained explants (p > 0.01). Additionally, there was a positive correlation between increasing shear strength in the presence of IL-1 and actual applied strain (determined from the thickness of the individual explants) (r² = 0.25; p < 0.0005). In the absence of IL-1 at 26% strain, there was a 2-fold increase in the shear strength of repair compared to unstrained samples (p = 0.003), but there was no effect of either 1% or 10% strains alone. Furthermore, there was a positive correlation between increasing shear strength of repair and the applied strain in the absence of IL-1 (r² = 0.14, p < 0.005).

DISCUSSION: We demonstrate here that dynamic loading blocks the IL-1 inhibition of integrative repair of meniscal tissue in an in vitro repair model. This finding is consistent with previous studies showing that mechanical stimulation can induce an anti-inflammatory response in meniscal cells (4,5,6), as well as in articular chondrocytes (10). The mechanisms for the interaction between mechanical factors and IL-1 are not fully understood but appear to involve mechanical inhibition of the NF-kB pathway, a pathway involved in IL-1 signaling. Taken together, our data indicate that dynamic loading is beneficial after meniscal injury, and that it promotes repair of meniscal lesions in an inflammatory microenvironment, such as those noted following meniscal injury, repair, or degeneration.

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