Δ Facilitated Fracture Repair via Noninvasive Localized Cold Therapy

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Purpose: The ability of fractured bone to regenerate and undergo repair is often compromised. Successful fracture healing involves an inflammatory cascade leading to bone repair. Cold treatment is commonly used to prevent inflammation after musculoskeletal injuries and earlier work on using cold stimuli for critical cortical defect healing has been shown to enhance bone growth. However, no study has looked at its effect or mechanism on fractures. Here, we hypothesized that enhanced bone healing demonstrated using cold therapy in cortical defects will be replicated in a clinically relevant fracture model and cold therapy stimulates angiogenesis, leading to improved vascularity.

Methods: Bilateral femoral fractures using retrograde nailing were formed in C3H strain mice. Initially, a guidewire was inserted followed by usage of a home-made 3-point impact device to produce a closed midshaft femur fracture. A 24-gauge stainless steel needle tip was then implanted over the guidewire to stabilize the fracture and the guidewire removed. Experimental legs were immersed in a cold-water bath reaching an internal bone temperature of 19°C for 15 minutes daily. Core-body and control leg temperatures were maintained with a heating pad. Femurs were harvested at days 7, 14, and 28 and underwent micro-CT analysis. Staining of ALP (alkaline phosphatase), TRAP (tartrate-resistant acid phosphatase), and VEGF (vascular endothelial growth factor) followed.

Results: Day 28 time-point analysis revealed daily exposure of fractured femurs to cold therapy increased bone volume / tissue volume (BV / TV) by 15% when compared to untreated controls ($P<0.001$). Simultaneously, a 1.42% increase in channel volume / tissue volume within cold-treated femora illustrates the prevalence of an enhanced vascular network ($P<0.05$). Biomarkers ALP and TRAP demonstrated expressions consistent with physiological bone remodeling within cold-treated femora at day 28, with staining revealing a significant 5.3% reduction in ALP ($P = 0.028$) detection without significant alteration of TRAP expression ($P = 0.44$). Bony callus formation is visibly enhanced at the 14-day time point from daily exposure of fractured femurs to cold therapy, reflecting a 6.61% increase in BV / TV when compared to untreated controls ($P<0.001$).

Conclusion: Findings confirm that localized cold treatment accelerates bone growth in a clinically relevant closed femoral fracture model. A strong increase in BV / TV coupled with decreased ALP staining of osteoblasts without significantly altered TRAP staining of osteoclasts indicates osteoclastic driven bone remodeling is attained faster within cold-treated femora. Simultaneously, improved vascularity indicates enhanced angiogenesis. Currently, potential fluctuations in genetic expression, vasomotor tone, pH, and hemoglobin oxygen saturation are being investigated to uncover the mechanistic propensity of therapeutic hypothermia.

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