#1586 - Experimental Study / Free Papers

Cold Delivered To Bone Causes Bone Growth

General Topics / Basic Sciences

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Background
The effect of cold temperature on bone/fracture healing has not been investigated directly. There are conflicting studies regarding the effect of hypothermia on osteoblast activity. Further there have been no in-vivo studies of the effect of cold therapy on the bone healing response. Despite several case studies showing cold causes bone growth no research has been explored to investigate if this is true after fracture.
Objectives
The purpose of this study is to investigate the effect of local cold therapy on bone healing. The hypothesis is that the group exposed to cold therapy will show more signs of healing in both microCT and histologic analysis when compared to the control group.
Study Design & Methods
Twelve C3H wild-type mice aged two and three months were used in the study. One-mm burr was used to create a one × three mm unicortical rectangular bone window in the lateral aspect of the femoral diaphysis. The right side was designated as the experimental side in all mice. A temperature sensor implanted on the quadriceps of the index mouse was able to confirm that using a six degree Celsius cold bath, the legs were cooled down to 19°C within three minutes. The experimental side was immersed daily in a six degree Celsius cold bath for a total of 15 minutes. A total of 12 mice underwent the abovementioned protocol without complications. MicroCT analysis was performed using a Skyscan 1172 microCT (Bruker Corp). A region of interest (ROI) was defined using a one-mm fixed diameter circle centered on the medullary canal. A constant total volume (TV) was created by extrapolating the ROI over a distance of 2.3 mm, centered at the middle of the cortical defect. Using a threshold of 55 Hounsfield units, the total bone volume (BV) was extrapolated from the ROI. For histologic analysis, the femora were fixed and cut in 5-µm sections. Staining for alkaline phosphatase (ALP), CD34, and tartrate-resistant acid phosphatase (TRAP) was then performed.
Results
The average bone window length was 2.68 ± 0.16 mm. The percent bone volume (BV/TV) in the experimental group was 34.1 ± 5.0, which was significantly higher than that of the control group 26.9 ± 7.1 ($P < 0.001$). Histological analysis revealed a significant decrease ($P < 0.001$) in the percentage of ALP stained cells in the experimental group (0.44 ± 0.2) when compared to the control group (1.2 ± 0.4). There was also a significant decrease ($P = 0.03$) in the percentage of CD34 stained cells in the experimental group (0.22 ± 0.08) when compared to the control group (1.58 ± 0.6). Finally, there was no significant difference ($P = 0.4$) in the percentage of TRAP stained cells between both groups ($P = 0.4$).
Conclusions

The results of our experiments show that daily treatments with cold therapy stimulate the bone growth/healing process in a murine model. Furthermore, the histological analysis reveals that the mechanism by which cold therapy stimulates growth is not necessarily linked to osteoblast activity since osteoblast activity was not increased in the experimental group despite increased bone formation. Postulation of the pathway and possible translation of the technique are discussed.

Mechanistic treatments to cause bone healing are desired over drug pathways. Innovation along these lines has been difficult in the past but is much easier in todays biomedtech milieu.