The Effect of Exogenous Administration of rhBMP-7 In Distraction Osteogenesis; A Dose-Response Curve and An Alternative Pathway for Bone Formation

Ali Izadpanah, MD, CM, MSc, Loai Alsalmi, MD, FRCSC, Reggie Hamdy, MD, MSc, FRCSC, Lucie Lessard, MD, FRCSC, FRCSC

Introduction

Distraction osteogenesis (DO), a widely used technique for bone and soft tissue formation in numerous craniofacial and limb defects such as traumatic bone loss, congenital anomalies, infection, and malignancies, is not short of limitations. Long consolidation required with multiple associated morbidities is the main limitation related to DO. Thus, we sought to determine the effect of exogenous administration of rhBMP-7 (OP-1) at multiple doses during wild type mice's tibial DO to ascertain an optimal dose if existing.

Materials

Eighty wild type mice in eight randomized groups underwent unilateral tibial DO with 5 days of latency, followed by 12 days of distraction at a rate of 0.2 mm/12hr, and 34 days of consolidation. On day 6, first day of distraction, animals underwent 4 doses of rhBMP-7 local administration to the distracted site; 0µg/kg (controls), 2µg/kg, 4µg/kg and 20µg/kg. Animals were sacrificed and samples collected at day-34 (mid-consolidation) and day-51 (end of consolidation). Specimens were analyzed using faxitron, micro-CT, static histomorphometry, biomechanical testing, and immunohistochemical analysis.

Results

All study groups had greater values compared to the controls (0µg/kg). We observed a gradual and step-wise decrease in bone formation upon increasing the administered dose of rhBMP-7 for faxitron, micro-CT, static histomorphometry, and biomechanical testing. Interestingly, at the end of consolidation, the mid-dose group of 4µg/kg had the greatest and strongest bone formed (graphs 1-4). Our Immunohistochemical analysis results demonstrated an inverse relation between the administered dose of rhBMP-7, and the expression of endogenous inhibitors such as Inhibin, Noggin, and Chordin and bone formation at mid-consolidation. Similarly, end of consolidation demonstrated the lowest expression of these inhibitors at 4µg/kg dose.

Conclusion

There appears to exist an optimal dose of exogenous rh-BMP7 during DO. Increasing administered doses can have an inhibitory effect due to up-regulation of endogenous inhibitors. Thus, manipulation of these inhibitors could be an alternative pathway in up-regulation of osteogenesis in DO.

Fig. 1. Bone histomorphometrical analysis of distracted segments at mid-consolidation

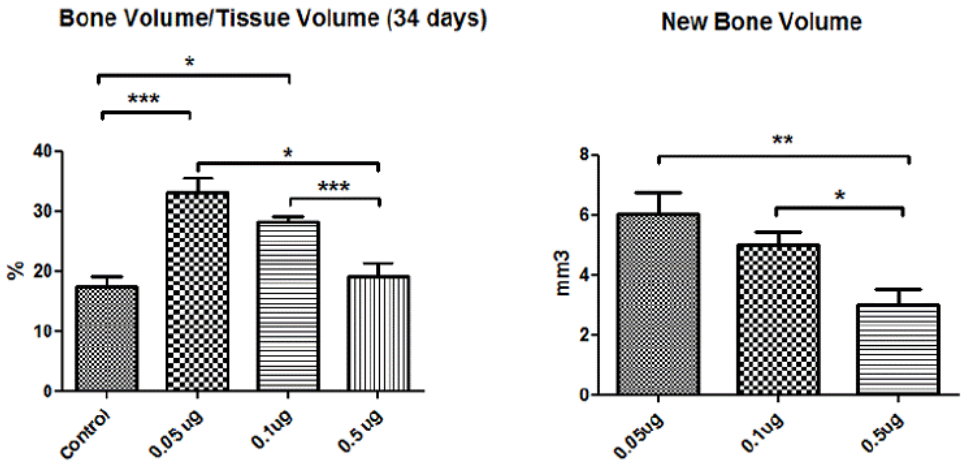

Table 2. Immunohistochemical Results of *Chondrocytes* staining positive for BMP signaling pathway members

	Concentration 0.05		Concentration 0.10		Concentration 0.50	
Gene	34 days	51 days	34 days	51 days	34 days	51 days
Ligands						
BMP-2	+	++	++	+	+++	++
BMP-4	+	+++	++	+	+++	++
Inhibitors						
Noggin	++	++	++	+	++	+
Chordin	+	++	+	+	+++	+

Staining of chondrocytes in the distracted zone, -: no positive staining, +: less than ¼ of cells stained positive, ++: ¼ to ½ of cells stained positive, +++: ½ to ¾ of cells stained positive, and ++++: more than ¾ of cells stained positive.