Multi-Omic Profiling Reveals Dose-Dependent Immune Response to BMP-2 in Preclinical Critically Sized Defect Fracture Model

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Purpose: Bone morphogenetic protein-2 (BMP-2) is a potent osteoinductive agent, but it carries significant risk because of side effects, including inflammation. In this study, we delivered BMP-2 using an injectable alginate system in a rat critically sized defect fracture model to investigate the impact of BMP-2 dose on bone regeneration and associated immune response.

Methods: Segmental bone defects (6 mm) were surgically created in femurs of female Wistar rats. Defects were internally stabilized and treated with 0 µg (control), 3 µg (minimal effective), or 30 µg (supraphysiologic) BMP- 2 (n = 7–8/group) (see figure). Bone regeneration was analyzed using radiographs and micro-CT. Longitudinal blood samples were collected to evaluate immune cells, cytokines, and metabolites. Additional subjects were euthanized at days 7 and 14 for cellular analysis of bone marrow, defect tissues, and quadriceps.

Results: A minimal effective BMP-2 dose regenerated significantly less ectopic bone ($p<0.04$) while inducing robust healing within the defect without significantly modulating immune cell dynamics or the cytokine profile. Contrastingly, in addition to inducing ectopic bone, a supraphysiologic dose significantly modulated immune cells systemically and locally ($p<0.05$) and upregulated immunomodulatory cytokines ($p<0.05$), deviating from an endogenous healing response. Concurrently, metabolite expression exhibited dose dependency.

Conclusion: BMP-2 dose potently affects bone formation, circulating and local immune cells, cytokines, and metabolites, reinforcing the need for further preclinical work tuning dose-response, enabling lower clinical doses.