A Novel Oxysterol Analogue Promotes *in Vitro* and *in Vivo* Bone Regeneration

Luis Andres Segovia, MD, Plastic Surgery, UCLA, Los Angeles, CA
Angeles, CA; Akishige Hokugo, DDS PhD, Plastic and Reconstructive Surgery, University of California, Los Angeles, Los Angeles, CA; Andrew Li, MD, Plastic and Reconstructive Surgery, University of California, Los Angeles, Los Angeles, CA; Reza Jarrahy, MD, Plastic and Reconstructive Surgery, University of California, Los Angeles, Los

Title

A Novel Oxysterol Analogue Promotes *In Vitro* and *In Vivo* Bone Regeneration

Introduction

The repair of complex craniofacial defects poses significant reconstructive challenges. Current methods employing autologous bone grafts or alloplastic implants are fraught with complications. Tissue engineering approaches using bone morphogenetic proteins (BMPs) are associated with adverse side effects and exorbitant costs. Here we investigate a novel molecule with significant osteoinductive capacity for its potential use in clinical applications. We examine the impact of Oxy133, a novel oxysterol analogue, on *in vitro* and *in vivo* osteogenic differentiation of rabbit bone marrow stromal cells (BMSCs).

Methods

Rabbit BMSCs were isolated, cultured, and treated with control media and varying concentrations of Oxy133 or BMP-2. *In vitro* osteogenic differentiation was assessed via alkaline phosphatase (ALP) assay, quantitative real-time PCR of osteogenic genes, and mineralization assays. *In vivo* activity was measured by healing of critical-sized rabbit calvarial defects that were treated with a collagen sponge carrying an inert control vehicle, collagen sponge carrying Oxy133, collagen sponge carrying BMP-2, or left untreated. The calvarium was harvested after seven weeks for histologic and radiographic analysis.

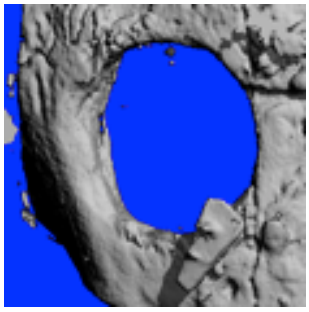
Results

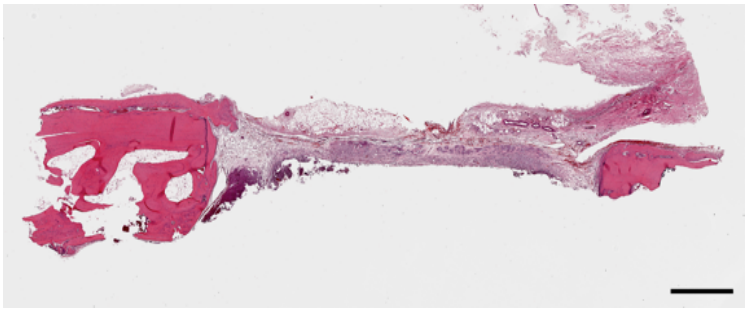
Rabbit BMSCs treated with Oxy133 demonstrated increased ALP activity, up-regulation of osteogenic gene expression, and increased mineralization of cultures compared to controls. Oxy133-treated cells demonstrated *in vitro* osteogenic differentiation with an efficacy similar to that of cells treated with BMP-2. Critical-sized rabbit calvarial defects showed complete bone regeneration when treated with collagen sponges carrying Oxy133, with an efficacy similar to that seen in animals treated with BMP-2. (Figure 1)

Conclusions

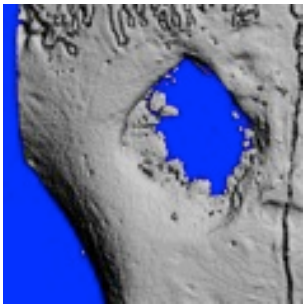
Oxy133 induces osteogenic differentiation in rabbit BMSCs as effectively as BMP-2 in both *in vitro* and *in vivo* models. Oxysterols, which are relatively inert and less costly to scale for mass production, may therefore represent a viable alternative to BMP-2 in bone tissue engineering models. Its application to the design of a clinically viable, safe, and cost effective bone graft substitute warrants further study.

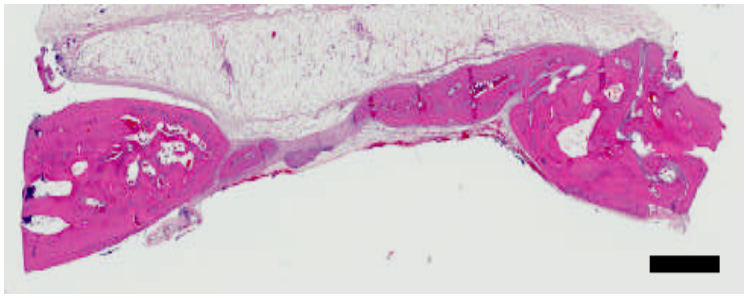
NO TREATMENT



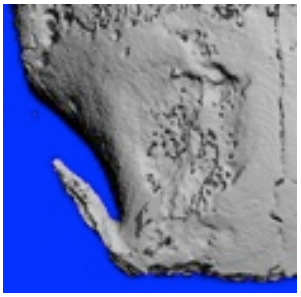


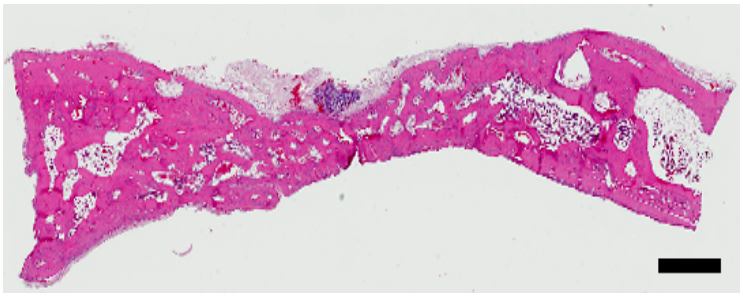
COLLAGEN SPONGE





COLLAGEN SPONGE + BMP2





COLLAGEN SPONGE + OXY133