INTRODUCTION
Approximately 5 to 10% of the fractures that occur annually in the United States exhibit some degree of impaired healing. Several compounds have been approved for the management of these conditions including BMP-2 and OP-1 (BMP-7). While these compounds show promising results, their applicability is limited due to the need for surgical implantation at the fracture site. Several studies have recently reported on the enhanced healing of fractures treated with systemically administered PTH (1,2). However, the molecular mechanisms by which either enhanced healing or regain of bone mass in osteoporotic conditions is achieved remain elusive (3). The majority of research to date has focused on the role of PTH in modulating osteoblast function in the context of coupled remodeling. While this may be an important component of PTH mediated enhanced bone formation, alternate mechanisms may exist in fracture repair that occurs through an endochondral process dependent on early chondrogenic events. In order to define the stages of fracture repair enhanced by PTH, we analyzed the tissue, cellular, and molecular effects of PTH treatment (40 µg/Kg, PTH 1-34) during bone healing in a murine femoral fracture model.

METHODS
Standardized closed femoral fractures were produced in C57Bl/6 mice and treated with PTH (40µg/Kg) or vehicle alone (sterile saline) via daily subcutaneous injections over the first 14 days of the 28 day standard repair time course. Calluses were harvested at days 5, 7, 10, 14, 21 and 28 post fracture for faxitron radiographic analyses, histologic analyses and isolation of RNA for molecular analyses using ribonuclease protection assay (RPA). These times points represent early chondrogenic recruitment (day 5), chondrogenic maturation (days 7 and 10), replacement of cartilaginous callus (day 14) and bone remodeling (days 21 and 28).

RESULTS
Radiographically, we observe that PTH treatment lead to increased mineralization of the fracture callus by day 10 and an earlier bridging of the fracture site (Figure 1). Histological analysis demonstrated that PTH increased the rate of cartilage recruitment, the total quantity of cartilage and its rate of hypertrophic maturation. TRAP staining and histomorphometric assessment demonstrated no evidence of increased bone resorption within PTH treated fractures. Molecular analysis confirmed these observations showing that PTH treated fractures expressed Col2A1 and Col10A1 on average two days earlier than in vehicle treated animals (Figure 2).

DISCUSSION
As opposed to the bone resorptive effects seen with continual exposure to PTH, intermittent administration leads to an increase in bone formation. Clinically this has been demonstrated in postmenopausal osteoporotic women where intermittent PTH injections resulted in a decrease in the risk of vertebral and non-vertebral fractures, as well as increased vertebral, femoral and total-body bone mineral density (3). In fracture repair, our results demonstrate that this anabolic enhancement of PTH is in part due to the induction of nascent mesenchymal cell recruitment into the chondrogenic lineage and by stimulating the subsequent maturation and expansion of these cells through a Wnt mediated mechanism. A systemically administered drug that enhances bone repair could have widespread clinical applications in the use of treating fractures, enhancing osteointegration of porous implants and promoting joint and spinal arthrodeses. PTH is the first bone formation agent that has been approved for the management of bone healing.

REFERENCES