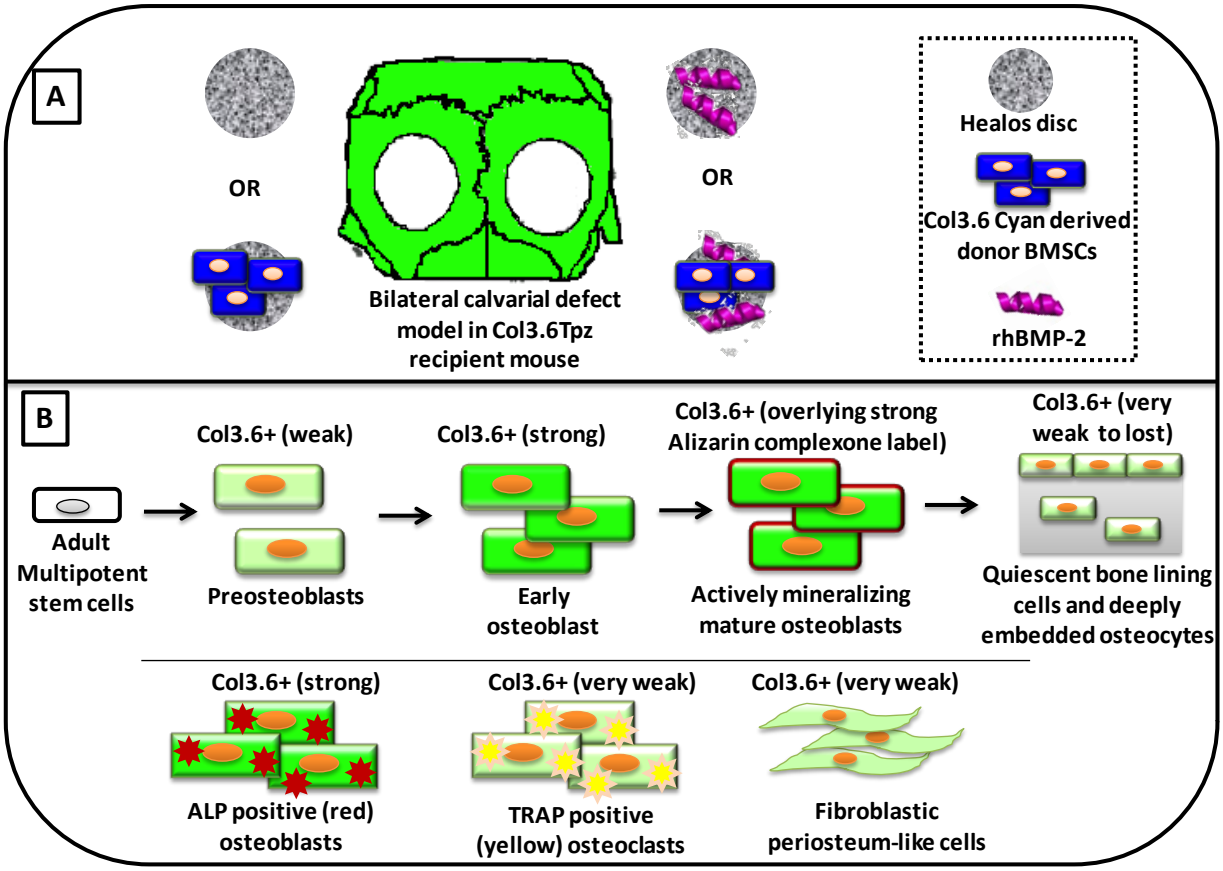
Supplementary data:

Supplementary figure S1.



A) A bilateral calvarial bone defect model was used where Col3.6Tpz transgenic reporter mice were used as recipient and Col3.6Cyan transgenic reporter derived BMSCs were used as donor cells for studying BMP-2 mediated bone healing; B) Use of GFP reporter expression and histological staining to identify the cellular response during bone formation.

Supplementary figure S2.

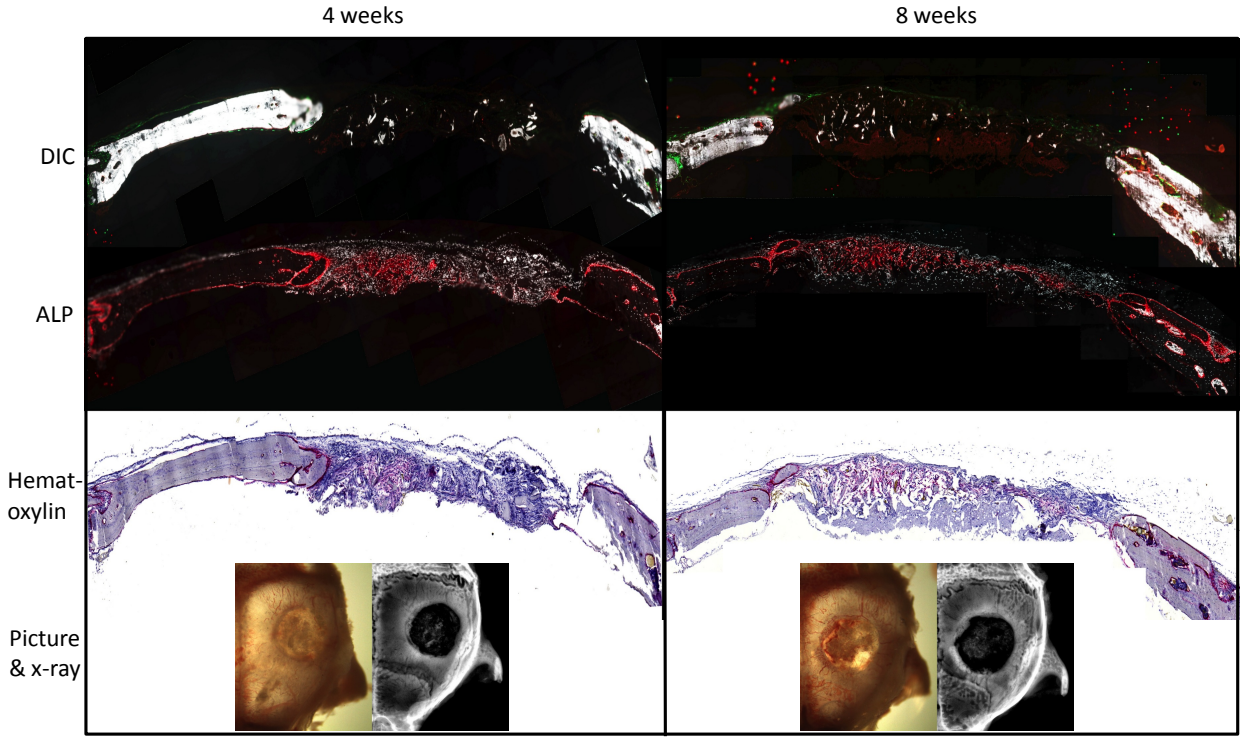


Figure S2. DIC, ALP and hematoxylin stained sections along with photo and x-ray of the critical sized defect implanted with Healos where the neighboring defect did not have any rhBMP-2, after 4 and 8 weeks of surgery. Healos alone, in absence of rhBMP-2 in the neighboring defect, did not show significant bone formation.

Methodology details:

Tape transfer process: Cryosections (5 μm) through the nondecalcified calvaria were obtained on a Leica CM3050S cryostat (Leica, Wetzlar, Germany) using a disposable MX35 PREMIER+ microtome blade (Thermo Scientific, Waltham, MA) and then transferred on to a Superfrost® Plus microscope slide using tape transfer process [Cryofilm Type2C (10), Section-lab, Hiroshima, Japan]. The Cryofilm (tape) is designed to capture a frozen cryostat section on a special cold adhesive tape to assist in transferring the section to a Superfrost® Plus microscope slide. The tape is color coded for adhering either to the slide (silver) or to the cryosection (gold). The tissue sections, which remain adherent to the tape through all of the subsequent steps, are placed sample/cryosection side (gold) up on a glass slide and stored at -20^{o}C until use.

The adhesive properties of the tape are however lost during TRAP staining due to use of acidic solutions in the staining process. Hence the method was modified where one day prior to staining; the tape along with the cryosection was transferred, sample/cryosection side (gold) up, onto a microscopic glass slide using 1.6% chitosan (Cat no 3646-500G, Sigma) solution in 0.25% acetic acid, as an adhesive. The chitosan solution keeps the tape adhered to the slide during multiple staining procedures and does not show any auto-fluorescence. After this step, the slides were again transferred to -20^{o}C for at least 24h to ensure complete drying of the adhesive followed by staining.