

## Injectable Hyaluronan Gels Forms Bone In-vivo

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### Introduction

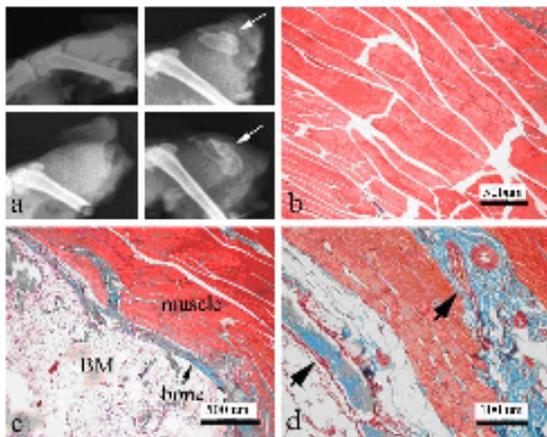
Autologous bone grafts are routinely used to heal large bone defects but has the disadvantages limited graft quantity, donor site morbidity, pain, and cost of harvesting. A promising alternative to this autografting is the use of a growth factor loaded biomaterial where endogenous cells are recruited *in-vivo* by loading the gel with bone morphogenetic protein-2 (BMP-2).

### Materials and Methods

To tailor a non-toxic injectable system with rapid *in vivo* gel formation in water at 37°C, chemical modification of hyaluronic acid (HA) was carried out in aqueous conditions following (1). Polyvinylalcohol (PVA) with 5% hydrazide functionality (PVAH) was synthesized following (2). The ability of the gels to induce bone formation was assessed in Sprague Dawley rats by co-injecting either HAA and PVAH with BMP-2 into quadriceps muscles. Critical size skull defects 4 x 2 cm were prepared in minipigs and filled with the same material.

### Results

The two multi-functionalized polymers react spontaneously with each other in aqueous solutions to form a cross-linked network. Examination after 4 weeks show the formation of ectopic bone at the site where gels loaded with rhBMP-2 were injected (figure 1).

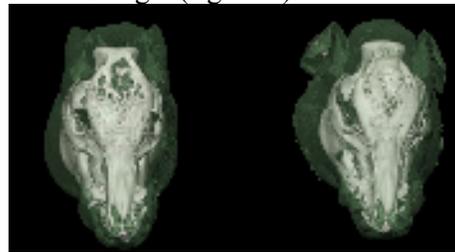


**Fig.1.** a: Ectopic bone formation at the injection site (right) in the BMP-group, gel

without the growth factor had no effect (left pictures). Active bone formation with osteoid and surrounding osteoblasts (c & d) and blood vessel formation (d). Control, no residues of the gel was found with complete absence of inflammatory reaction (b).

Bone formation, including mineralized tissue surrounding a bone marrow cavity, was visualized by histology. Arteries and veins were found in adjacent soft tissues in BMP-2-hydrogel specimens, but not in the controls indicating the concurrent induction of vascular tissue.

In similarity to bone induction the rat model, critical sized defects in the minipig were completely closed after 12 weeks using the BMP-2 loaded gel (figure 2).



**Fig.2.** The mini pig model showed no bone formation with the gel alone (left) while BMP-2 loaded gel gave full closure of the 4 x 2 cm cranial defect after 12 weeks (right).

### Discussion and Conclusions

The benefit of not having to rely on transplantation of cells, in combination with the non invasive procedure i.e. injection, makes this approach towards guided bone repair particularly attractive in clinical use for the treatment of slow- or non-healing closed fractures.

### References

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2. Ossipov, D.A., Brännvall, K., Forsberg-Nilsson, K. & Hilborn, J., *J. Appl. Polym. Sci.* **106**, 60-70 (2007).