Vascular endothelial growth factor expression in posterolateral rabbit fusion: An evaluation of bone graft materials

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Statement of Purpose: Large bone defects and non-unions caused by trauma, revision surgery, inflammation, tumor surgery and developmental deformity remains a major challenge for orthopaedic surgeons. In addition to the osteoconductive, osteoinductive and osteogenic requirements for an "ideal bone graft", rapid vascularization of bone graft materials is becoming recognized as an important process for long-term clinical performance[1].

Rabbit posterolateral fusion is a widely used model to assess the performance of synthetic bone graft materials[2]. This is a challenging model for bone substitutes because the transverse processes are extremely thin, and the space to be filled with bone can be considered a critical gap defect and the tissue space is avascular [3].

The extent of immunohistochemical staining specific for VEGF was measured for synthetic bone grafts mixed with both bone marrow aspirate and autograft relative to a control group (autograft plus BMA). Two types of synthetic bone graft materials were compared, nanOss Bioactive and nanOss Bioactive 3D (Pioneer Surgical, Marquette, MI).

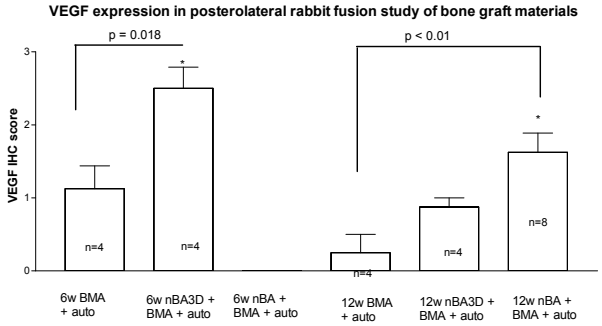
Methods: Single-level posterolateral lumbar fusion was performed in 69 adult New Zealand white rabbits as reported and developed by Dr. Boden et al. [4] following institutional ethical approval. The study design is outlined in Table 1. Animals were anesthetized for surgery using isoflurane/oxygen inhalation. A midline incision was made in the skin, and the intermuscular plane between the multifidus and longissimus muscles was bluntly incised to expose the L5 and L6 transverse processes. A high-speed burr (Midas Rex, Medtronic, Memphis, TN) was used to decorticate the transverse processes in a uniform and reproducible manner in all animals, 10 mm from the base of the transverse processes.

Table 1 Study design

Group	6 weeks (*n*)	12 weeks (*n*)
Autograft + BMA	6	12
nBA3D + BMA + Autograft	6	12
nBA + BMA + Autograft	N/A	12

For animals receiving synthetic bone graft materials plus BMA with autograft, a total of 3 mL of bone marrow harvested with a bone marrow biopsy needle and 3 cc of morcelized corticocancellous bone harvested from the iliac crests were mixed prior to implantation on each side (1.5 cc per side) over the decorticated transverse processes.

Animals were sacrificed at 6 and 12 weeks, and tissue specimens were fixed in cold phosphate-buffered formalin solution for a minimum of 48 h and decalcified in 10% of formic acid–phosphate buffered formalin solution for routine paraffin histology. Immunohistochemical staining for VEGF was used to identify vascular structures adjacent to bone graft implants. A semi-quantitative grading scale was used to assess overall VEGF expression & distribution.



Results: VEGF staining was found adjacent to all implant materials at both time points. Surprisingly, autograft implants showed statistically less VEGF staining than some synthetic bone graft materials. Putty-like bone graft materials showed the highest levels of VEGF expression.

Conclusions: VEGF staining provides an additional metric to assess the performance of synthetic bone grafts and may guide the development of improved biomaterials for bone regeneration.

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