CLINICAL ORAL IMPLANTS RESEARCH

Autologous human osteoblasts for maxillofacial bone tissue engineering: ex vivo good manufacturing practice (GMP)-level expansion and clinical evaluation

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Background: Bone tissue engineering (BTE) using osteogenic cells and biomaterial scaffolds can overcome the limitations of conventional grafting techniques and the need for extensive autogenous bone harvesting, especially in large bone defects. While autologous undifferentiated mesenchymal stem cells (MSC) have been used clinically for maxillofacial BTE with varying success, less is known about the regenerative potential of differentiated osteoblasts.

Aim/Hypothesis: The objectives of the study were: (1) to culture human bone marrow-derived osteoblasts ex vivo at the Good Manufacturing Practice (GMP) level, and (2) to evaluate the clinical bone regeneration potential of autologous osteoblasts in a fibrin sealant scaff.

Material and methods: Following informed consent, autologous cell-based reconstruction was planned in a 24-year-old female with extensive post-traumatic dentoalveolar loss in the anterior maxilla and mandible (esthetic zone). Preoperative radiography demonstrated horizontal bone defects, insufficient for implant placement. Bone marrow (8 ml) was harvested with a minimal-invasive technique from the patient's posterior iliac crest and transported to a GMP laboratory. MSC were isolated and differentiated into osteoblasts using specific induction media. Characterization of osteoblasts was performed by assessing alkaline phosphatase (ALP) activity and by alizarin red staining of extracellular calcium deposits after 14 and 21 days, respectively. Four weeks after bone marrow harvest, ex vivo culture-expanded autologous osteoblasts (~4.8 × 107 cells) mixed with a clinical-grade human-derived fibrin sealant (Tisseel, Baxter, USA) were implanted at the defect sites. The construct was allowed to polymerize and form a gel-like consistency, and was covered using a resorbable membrane for provision of guided bone regeneration (GBR). After 4 months new bone formation was evaluated via cone-beam computed tomography (CBCT) and dental implants of standard dimensions were placed in the regenerated bone (n = 7). Prosthetic rehabilitation with fixed partial dentures was completed after 4 months.

Results: Isolation of MSC from bone marrow and subsequent osteogenic differentiation was successfully performed. Cells displayed normal morphology when observed microscopically at different stages of culture. Positive ALP assay and alizarin red staining confirmed that the expanded cells possessed osteogenic capacity. Clinically significant numbers (>1 × 107) of GMP-level osteoblasts were generated within 4 weeks and cell viability was assessed to be >80% prior to implantation. No postoperative complications or adverse events were reported. Postoperative CBCT displayed considerable horizontal bone gain (~4–5 mm in each jaw) after 4 months. The tissue-engineered bone displayed a trabecular pattern and integration with adjacent pristine bone radiographically, and D2-D3 quality clinically at implant placement Adequate primary stability of all implants was achieved. At 1-year follow-up there was no radiographic evidence of loss of regenerated bone volume or remarkable peri-implant marginal bone loss.

Conclusions and clinical implications: Implantation of autologous ex-vivo expanded osteoblasts in a fibrin sealant scaffold was effective in terms of bone regeneration, allowing implant-supported prosthetic rehabilitation without the need for extensive autogenous bone harvesting.