Bioactive Glass Scaffold with Oriented Porous Structure for the Repair of Load Bearing Bones

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Introduction: Scaffolds fabricated by conventional methods from biodegradable polymers, bioactive ceramics, and bioactive glass often lack the combination of high strength and high porosity for skeletal substitution of load-bearing bones. Unidirectional freezing of aqueous suspensions has been shown to produce porous scaffolds with high strength (in the orientation direction) but the pore width is often too small to support tissue ingrowth[1]. In the work, a two-step process, unidirectional freezing of camphene-based suspensions followed by annealing of the frozen constructs, was investigated as a route for creating bioactive glass scaffolds for potential application in the repair of load-bearing bones.

Methods: Glass particles (1 ± 0.5 μm) with the 13-93 composition (wt %) ($53SiO_2$, $6Na_2O$, $12K_2O$, $5MgO$, $20CaO$, $4P_2O_5$) were dispersed in liquid camphene (2 wt% isostearic acid) by ball milling for 24 h at 55°C. The slurry was solidified unidirectionally by pouring into molds placed on cold Cu plates (3°C), after which the frozen constructs were annealed for 24 h at 34°C to grow the camphene crystals. After sublimation of the camphene, the constructs were heated for 1 h at 700°C to densify the glass particles in the pore walls. The fabricated scaffolds were characterized using SEM, X-ray tomography, and mechanical testing in compression (scaffolds 6 mm in diameter × 10 mm were deformed at a cross-head speed of 0.5 mm/min). In vitro culture was performed to determine the ability of the scaffolds to support cell proliferation and differentiated function. The scaffolds were seeded with 50,000 MLO-A5 cells (2 ml of a-MEM medium with 10% fetal bovine serum plus 100 U/ml penicillin and 100 μg/ml streptomycin sulfate) and incubated for 2, 4, and 6 days at 37°C. After incubation, the cell-seeded constructs were evaluated using SEM, MTT staining, and assays for protein and alkaline phosphatase activity. Scaffolds of 13-93 glass with a trabecular microstructure[1] (porosity = 85%; pore size = 100–500 μm) were used as controls.

Results and Discussion:

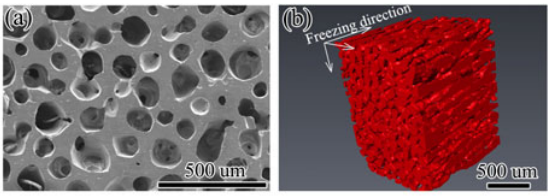

Fig. 1. (a) Cross-section of scaffold (perpendicular to the orientation direction; (b) microCT image of scaffold. .

Images of the cross-section perpendicular to the freezing direction of the sintered scaffolds (**Fig. 1a**) showed the circular shape of the pores (porosity = 50%; pore diameter = 50–150 μm). MicroCT showed that the pores were oriented along the freezing direction (**Fig. 1b**), with neighboring pores connected at several positions along their length. The scaffolds showed an elastic response, and a compressive strength of 34 ± 11 MPa.

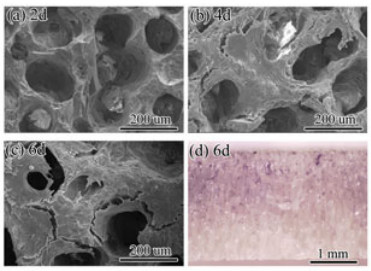

Fig.2. Cell proliferation on and into the scafoldsscaffolds

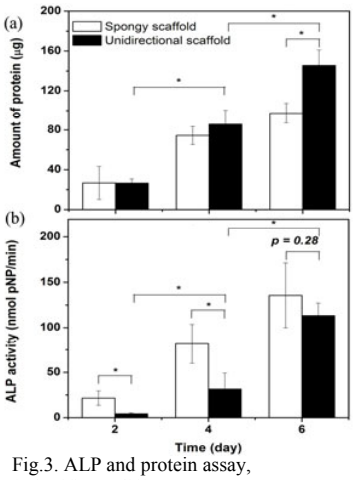

Fig.3. ALP and protein assay, * significant difference

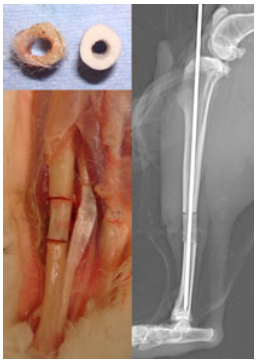

Fig.4. Oriented scaffold implanted in cadaver rabbit tibia

Cells proliferated on the surface of the scaffolds and into the interior pores (**Fig. 2a, b, c**). MTT staining of the longitudinal cross section (**Fig. 2d**) confirmed the ingrowth of live cells into the interior pores. Assays showed the ability of the oriented scaffolds to support cell proliferation and differentiated function (**Fig. 3**). After incubation for 6 days, cells on the oriented scaffolds had a higher density, despite having a lower porosity and smaller pore size than the trabecular scaffolds. Pilot experiments showed the ability of the scaffolds to survive implantation into cadaver rabbit tibia (**Fig. 4**).

Conclusions: Oriented bioactive glass (13-93) scaffolds (porosity = 50%; pore diameter = 50–150 μm) prepared by unidirectional freezing of camphene-based suspensions, showed promising results for application in the repair of load-bearing bones. The scaffolds had a compressive strength compressive strength = 34 ± 11 MPa, supported the proliferation and differentiated function of osteogenic MLO-A5 cells on the surface and into the interior pores, and survived implantation into segmental defects in cadaver rabbit tibia.

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References

[1]Deville et al., Biomaterials 2006;27:5480-9
[2]Fu et al., Acta Biomater 2008;4:1854-64