# Biocompatible phosphate glass fibre scaffolds Engineering of the hard/ soft tissue interface

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**INTRODUCTION:** Phosphate based glasses maintain the survival, phenotype and morphology of adherent human bone and ligament cells (1) and may prove ideal, as scaffolding material, for tissue engineering dealing with ligament-tendon/ bone attachment defects. This work assesses the biocompatibility of fibre constructs of both, ternary ($CaO–Na_2O–P_2O_5$) and Quaternary (iron containing) compositions as candidates for a contiguous osteoblast/ fibroblast seeded in vitro systems. This is achieved by firstly, identifying the optimum fibre diameter and secondly, comparing various glass compositions for biocompatibility through evaluating cell adhesion, proliferation and morphology.

**MATERIALS AND METHODS:**

**Cells:** Primary Human osteoblasts (HOB), oral (HOF) and Tendon (HTF) fibroblasts were obtained from explants derived from alveolar bone, buccal mucosa and the flexor tendon of the hand respectively.

**Glass fibre production:** Ternary glass compositions containing 46 and 48 mol% calcium together with quaternary forms ($CaO–Na_2O–Fe_2O_3–P_2O_5$) of 1, 2 and 3 mol% iron were utilised to generate fibres of various diameters. By keeping $P_2O_5$ fixed at 50 mol% Solubility of the glass was controlled as such where higher calcium or iron content resulted in lower degradation rates (2). Glass fibres where arranged in mesh and parallel monolayer constructs where a distance of 5 to 10 µm was maintained between alternating fibres. Cells were seeded at 3.2 $x10^3$ /$cm^2$ density.

**Cell adhesion, proliferation and morphology:** Cell numbers at various time points, and on various glass compositions and fibre diameters, were measured by direct counting of propidium iodide (PI) stained nuclei. Cell morphology was visualized by fluorescent immunolabeling of the intermediate filament protein vimentin. Cell count and imaging were conducted using Leica DM IRB fluorescent microscope.

**RESULTS:**

Fibres less than 20-25 µm in diameter were excluded from further studies due to the lack of cell attachment, faster solubility and scaffold degradation. Whilst cell survival on ternary glass compositions ceased at day 7, a clear proliferation pattern, for both cell types, was observed on quaternary glasses of 2-3 mol% iron up to 14 days in culture (figure 1). Adherent cells exhibited a well spread morphology aligned to the fibres orientation (figure 2).

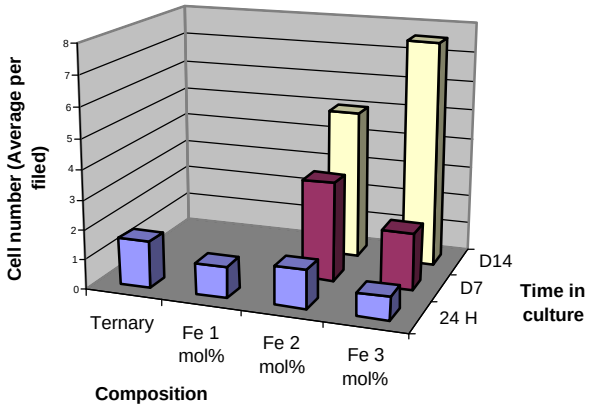


***Figure 1HOF*** *proliferation pattern on various glass compositions at different time points. (n=3).*

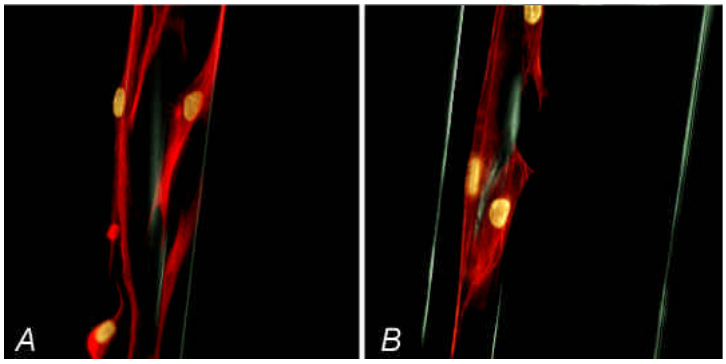


***Figure 2, X20 Leica fluorescent microscope image of A, HOF cells and B, HOB cells on 3 mol% iron containing glass fibres.***

**Conclusion:**

Phosphate based glass fibres, of quaternary glass form and containing between 2 and 3 mol% of iron are to be considered as scaffolding material for tissue engineering. These scaffolds are to be seeded with co-cultures, of both cell types, and treated with the appropriate intrinsic factors attempting to mimic the hard/ soft tissue interface in vitro.

**References:**

(1) *Bitar et al.* Biomaterials 2004; 25(12):2283-2292.

(2) *Ahmed et al.* Biomaterials 2004; 25(16):3223-3232.