Design, Development and Characterisation of Smart Synthetic Bone Graft Materials for Advanced Therapies

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The common occurrence of bone defects and bone destruction caused by disease (osteoporosis, bacterial infections, osteoarthritis and tumour) or accidental factors (car accidents and trauma) have a huge impact on a patient's quality of life, and demand suitable grafting remediations [1]. Because of the limited availability of biological bone substitutes, several tissue engineering strategies have been widely considered in the reconstruction of vascularized bone tissue and in the treatment of bone defects, namely, the combination of cells, biological molecules and biomaterials [2]. The synthetic bone grafts might comprise different types of materials, including metals, ceramics, bioactive glasses, polymers and composites. They have to be judiciously selected according to the specific requirements of the bone substitutes in terms of the relevant physical, and functional properties. The close resemblance between the chemical composition of the inorganic part of the bone and hydroxyapatite (HA-$Ca_{10}(PO_4)_6(OH)_2$, almost bioinert) and tricalcium phosphate (TCP-$Ca_3(PO_4)_2$, resorbable) make them attractive to develop bone grafts based on calcium phosphates (HA, TCP, or biphasic compositions, pure and doped with therapeutic ions [3]. Moreover, bone tissue engineering strategies often involve the use of porous three-dimensional (3D) scaffolds that act as temporary supports and provide a suitable environment and architecture for bone regeneration and development [4]. A high and interconnected porosity with adequate pore sizes for allowing cell adhesion and proliferation, ensuring the diffusion of oxygen and nutrients to the cells and the removal of waste products are essential requirements. These requirements can be commonly achieved by using additive manufacturing [5], or by biomimetic approaches, such as transforming the aragonitic cuttlefish bone in the desired calcium phosphate composition, while preserving the highly porous and interconnected structure [6]. The first part of this presentation discloses the overall composition and, in particular, the surface chemistry of such scaffolds can be modified with special coatings for endowing them with specific functional properties such as enhanced bioactivity, drug storage and controlled *in situ* release. Bioactive glasses are other competitor materials for bone repair and regeneration [7]. Although the discovery of the first 45S5 Bioglass® has been regarded as a great breakthrough, its high alkali content causes serious shortcomings. In contrast, Alkali-free bioactive glasses [8] possess a set of high-performing biological key features [7,9]. Moreover, Alkali-free bioactive glass compositions inherently exhibit stable/metastable thermodynamic driven liquid-liquid phase separation (LLPS) that facilitates the sintering of the glass powders, being well suited for developing biomedical devices that depend on powder processing techniques. The first glass transition temperature is significantly lower than the crystallization onset temperature, providing a wide temperature window for controlling liquid state sintering [10]. On the other hand, LLPS negatively affects the fabrication of devices that involves glass shaping from the melt, such as fibre drawing [11]. This is because the second

glass transition temperature is close to that of the onset of crystallization, thus leaving only a narrow temperature window for glass shaping.

The second part of this presentation is focused on the effects of $NaBO_2$ addition to a phase-separated Alkali-free bioactive glass composition (38.49 SiO_2 • 36.07 CaO • 19.24 MgO • 5.61 P_2O_5 • 0.59 CaF_2). Scanning electron microscopy reveals binodal phase separation involving two Si microphases with a droplet size of ~200 μm. The local environments and spatial distribution of silicate, phosphate, and fluoride ions in this phase-separated system were studied, using ^{29}Si, ^{31}P, ^{11}B, ^{19}F, ^{25}Mg, and ^{23}Na nuclear magnetic resonance (NMR) and infrared spectroscopy. The silicate units are dominantly of the metasilicate (Si^2 or $Q_{2(Si)}$) type. The phosphate units exist mostly as orthophosphate (P^0 or $Q_{0(P)}$) while the borate is present in the form of pyroborate (B^1 or $T_{1(B)}$). Multinuclear dipolar re-coupling experiments indicate that the minority components F, P, and Na all occur within a common phase. Thus, atomic distribution scenarios involving the separation of these components into separate phases can be excluded. Based on the ^{31}P spin echo decay (SED) method along with Monte Carlo simulations analysis, the phosphate component forms clusters of sizes 1-4 nm, which are randomly embedded in an environment more dilute in phosphate. While ^{19}F SED results indicate that the fluoride ions do not form clusters and are close to randomly distributed, dipolar recoupling with ^{31}P, using $^{19}F\{^{31}P\}$ REDOR experiments, suggests a local environment resembling that of fluorapatite. Such local environment might be the reason behind the fast biomineralization rate of this type of bioactive glasses. All SBG compositions were subjected to cytocompatibility tests using human mesenchymal stem cell cultures (hMSCs). It was found that all SBGs elicited good biocompatibility, with some SBG formulations promoting hMSCs proliferation and differentiation, while others sustaining the stem cell phenotype. These findings prove that SBGs are a class of materials with immense potential for tailored bone grafting applications in future medicine.

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