Biomimetic approaches to engineer bioactive glass-based nanosystems

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Statement of Purpose:
Despite the remarkable osteoconductive properties attributed to bioactive glass since its discovery, it is still a brittle material. Therefore, its applications are limited by a proper engineering of the material's structure, or by its combination with other materials, like polymers. In native mineralized tissues, the blend of organic with inorganic phases is frequently the key for the remarkable mechanical properties of this class of natural materials.[1]
The main goal of this work was to give a step further in producing *in vitro* materials able to mimic the structural and chemical environment necessary to bone growth. Micro and nanofabrication techniques were used to recapitulate the complex environment of mineralized tissues. Bioactive glass and chitosan were chosen as materials to be combined respectively as mineral and organic phase in order to mimic bone structure.
Methods: Bioactive glass nanoparticles (BG-NPs) were previously produced by sol-gel route[2]. Chitosan, a natural origin polymer, was combined with the bioactive glass nanoparticles to obtain nanocomposites. Two strategies were followed. BG-NPs were dispersed in a chitosan solution and then transformed in membranes through a traditional solvent casting procedure and a microcontact printing (mCP) technique was employed to print BG-NPs on the surface of the chitosan membranes.
Regarding the 3D level, a simple procedure of adding a drop of aqueous suspension of BG-NPs and then leave it to dry on a superhydrophobic surface was used to induce the self-assembly of the BG-NPs,
Results: SaOs-2 osteoblastic-like cells were seeded on the nanocomposites films based on a chitosan blend with bioactive glass nanoparticles with different formulations, namely SiO_2:CaO:P_2O_5(mol.%)=55:40:5 and SiO_2:CaO:P_2O_5:MgO(mol.%)=64:26:5:5. Their behavior was followed by SEM observations, citotoxicity assessments, DNA quantification and ALP analysis. The nanocomposite containing particles doped with Mg presented moderate bioactive character and higher hydrophilicity and was found to stimulate a better osteoblastic response towards cellular differentiation and mineralization. When a microcontact printing technique was employed to print BG-NPs on the surface of the chitosan membranes, mineralized patterns were obtained and cells showed a tendency to attach accordingly to the created pattern, while maintaining their viability. See Figure 1.
By allowing the evaporation induced self-assembly of the BG-NPs on a superhydrophobic surface, bioactive glass based aggregate comprising the nano, micro and macro levels were created. Besides having a hierarchical organization, known to give mineralized materials their great mechanical properties, we also proved that these systems allow for the inclusion of drugs by dispersing dyeing additives in the macrospheres. Their bioactive character was controlled by changing the evaporation ratio by adjusting the environment temperature.

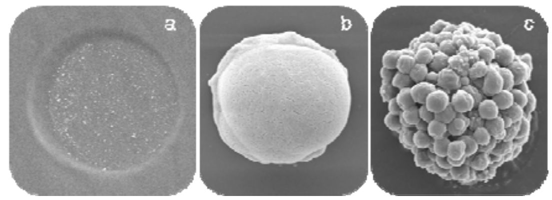

Figure 1. **a)** BG-NPs pattern on a chitosan membrane by mCP; **b)** Mineralized pattern; **c)** Cells cultured on the micropatterned membrane respect the BG-NPs pattern.

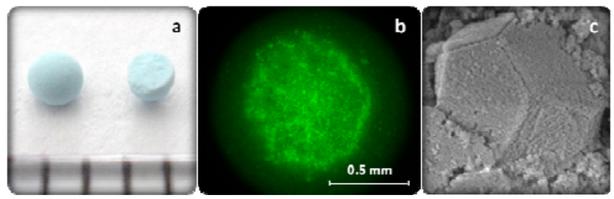

Figure 2. **a)** Methylene blue dyed whole sphere and a cross section; **b)** Fluorescence image of the cross-section of a calcein AM stained sphere; **c)** SEM image of a microstructure formed by the self-assembly of the BG-NPs.

Conclusions: Regarding 2D applications of the BG-NPs, bioactive nanocomposites were obtained by casting chitosan solutions containing the nanoparticles. BG-NPs were also doped with magnesium in order to enhance their biological influence. Results showed a positive effect on the osteoblastic response. In an extension to the previous successful results, mCP technique was used to control spatially the mineralization of chitosan membranes. A valuable and polyvalent platform was created to push even further the study of mineralization and cellular interactions.
At the 3D, a bottom-up approach to produce hierarchically organized BG-NPs based structures was used successfully. The results showed that BG-NPs are capable of self-assemble into organized multilevel structures. Therefore, self-assembly proved to be a powerful tool in mimicking mineralized structures.
The described research work is based on simple techniques highly competitive against other existing technologies for bone´s structure mimicking. Control over the BG-NPs distribution and was achieved at 2D. However, it would be interesting to develop modes of also extending the spatial control of mineralization to 3D structures, namely in hydrogels and scaffolds, in order to target the particles potential in a wider range of BTE applications.

References:
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