TS035
Superhydrophobic patterned chips for the combinatorial and rapid study of 3D biomaterials-cells interactions and protein delivery systems

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The development of optimized products in the tissue engineering (TE) field is a time and resource consuming process due to the unpredictable influence of the combination of variables such as biomaterials, cells and soluble factors. As multiple combinations can be considered high-throughput (HT) methods were suggested as a way to master complexity in TE. Usually, HT systems test cell-2D biomaterials interactions and, more recently, cell-3D hydrogels interactions. Using a chip consisting of superhydrophobic surfaces patterned with wettable regions we tested cells-hydrogels interactions in three-dimensional environment [1]. The versatility of the chip allowed its use for the first time on-chip combinatorial study of 3D miniaturized porous scaffolds. Arrays of biomaterials were dispensed and processed in situ as porous scaffolds with distinct composition, surface characteristics, porosity/pore size and mechanical properties. Those characteristics were assessed by adapting microcomputed tomography equipment and a dynamic mechanical analyzer. The interactions between cell types of two distinct origins – osteoblast-like and fibroblasts - and the scaffolds modified with distinct amounts of fibronectin were studied by image-based methods and validated by comparison with conventional destructive methods. Physical and biological on-chip results were coherent with conventional measures, and conclusions about the most favorable media for the growth of both cell types were taken. Growth factors (GF) proved to play an important role in TE approaches, mainly for determining cell fate in applications containing stem cells. We developed a chip based on wettability contrast with torus-shaped hydrophilic transparent regions disposed in an array matrix. Concentrically to these wettable regions a superhydrophobic circle was maintained, so the hydrogels could be processed as protein-loaded spheres with minimum protein loss [2] and fixed with an indentation. A combinatorial system of BSA-FITC – a commonly used GF model – encapsulated in alginate hydrogels was designed. The protein release from the hydrogels could be studied by image analysis, avoiding manipulation and protein loss. The results were compared with conventional protein release tests and similar tendencies were observed. We believe that the proposed innovative uses for the superhydrophobic chip and their upgrade in future applications may constitute a promising breakthrough in integrated technologies for the rapid development of TE systems.

References:

1. Salgado CL, Oliveira MB, Mano JF. Combinatorial cell-3D biomaterials cytocompatibility screening for tissue engineering using bioinspired superhydrophobic substrates. Integrative Biology. 2012;4:318–27.
2. Song WL, Lima AC, Mano JF. Bioinspired methodology to fabricate hydrogel spheres for multi-applications using superhydrophobic substrates. Soft Matter. 2010;6:5868–71.

TS036
Nanostructured Multilayer compartments: towards multifunctionality and "cell-like" hierarchical complexity

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In living organisms, there are phenomena that require the presence of specific biomolecules with distinct function and in variable concentrations at a given time, such as the healing and regeneration of tissue and organ lesions. In this work, we propose the use of a compartmented drug delivery device for the multiple release of bioactive agents. It consists of nanostructured microcapsules confined within a millimetric container that can be easily handled, mimicking the concept of cells which possess organelles with specialized functions. Each hierarchical structure was conceived using the layer-by-layer (LbL) method to form micro and macrocapsules that could individually carry either molecules and release them with distinct kinetics or magnetic nanoparticles (MNPs) to be used in targeted therapies. Furthermore, the internal microcontainers were constructed with a temperature-responsive elastin-like recombinamer (ELR) to further add smart properties to the proposed system. Sacrificial $CaCO_3$ microparticles empty or entrapping either rhodamine or Fe_3O_4 MNPs were incubated in chitosan and ELR solutions using LbL for the conception of the microcapsules. Then, the microcapsules were suspended in alginate which was ionically cross-linked in $CaCl_2$ drop-wise. Rhodamine could be encapsulated at this point in the alginate. The bead was coated with chitosan and alginate to build the external macrocapsule compartment. All structures were coated with 3 bilayers. The $CaCO_3$ cores were chelated and the alginate beads liquefied using EDTA. Fluorescence microscopy using FITC and rhodamine markers showed a uniform distribution of the microcapsules within the macroreservoir. The release of rhodamine from either in the micro or macrocapsule was assessed at 25 and 37 °C in PBS. While the release from the macrocapsule follows a profile similar to that of traditional drug delivery systems, it is more sustained and delayed when released from the internal compartments. Such retention is more pronounced at 37 °C (65% of release in comparison to 90%). This is due to the temperature responsive behavior of ELRs, which undergo a phase transition and make the LbL shell less permeable. For the magnetic response, the incorporation of the MNPs was observed by transmitted light microscopy. The attraction of the devices was observed by applying an external magnetic field along a defined trajectory. The results let foresee the use of such multilayer devices as compartmented structures to encapsulate growth factors, MNPs and stem cells for their controlled differentiation and maintenance or for guided regeneration of tissues and organs.