How electron microscopy can contributes to the elaboration of porous organogels

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Organogels are organic solvents or oils that have been gelled using an organic gelator. Organic gelators are low-molecular-weight compounds that promote non-covalent self-assembly of fiber-like structures to form a 3D network in solution that can gelate various types of oils. Among the low-molecular-mass organic gelators, 12-hydroxystearic acid (HSA), obtained from castor oil, has been largely investigated because it is a non toxic gelator able to gelify a large variety of organic solvents and oils.

The goal of the present study was the elaboration of porous organogels through the use of a particulate leaching method to introduce a controlled porosity into the material (Figure 1). We focused on a porous organogel made from renewable and biodegradable resources like soybean oil and HSA, which could be interesting for cell culture or as new adsorbent material.

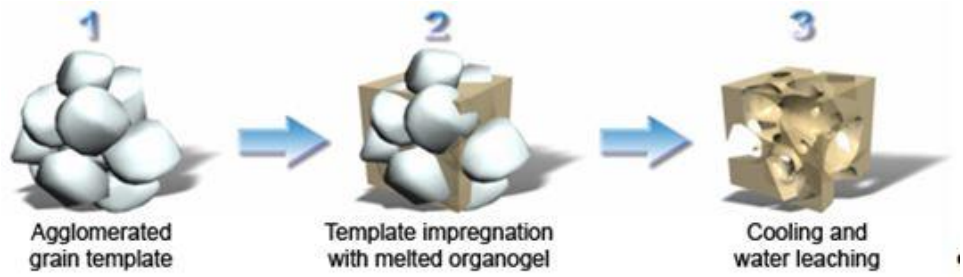

Figure 1: Schematic presentation of porous organogel scaffolds preparation by the particulate leaching technique.

Different microscopy observations techniques have been used at all the development steps of the porous material.

1- <u>Scanning electron microscopy (SEM) :</u>

In order to visualize the self-organization of HSA in soybean oil, SEM was carried out (Figure 2A), and clearly showed the nanofibrillar organization of the HSA. Sodium chloride and saccharose were chosen as porogens. Using SEM images analysis, the particle size and average were determined (Figure 2B and 2C)

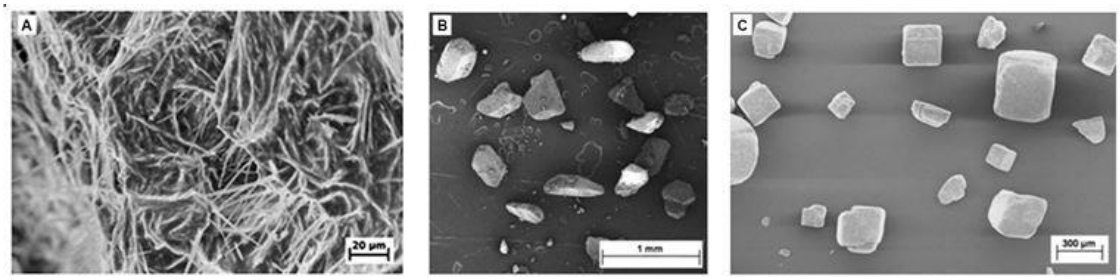

Figure 2: SEM micrographs of (A) Nanofibrillar organization of the organogelator ;

(B) sugar crystals as porogen particles ; (C) sodium chloride as porogen particles

2- <u>Low vacuum scanning electron microscopy (LV-SEM) :</u>

The microstructure and the morphology of the porous organogels were observed by LV-SEM (Figure 3A), in order to prevent a possible deformation of this soft material. Additionally microtomography reconstruction was applied (Figure 3B). By image analysis we determined the pore size average and the porosity of the material.

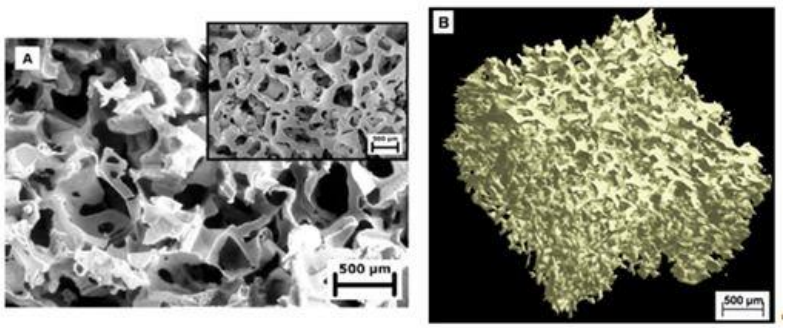

Figure 3: (A) LV-SEM images of the internal morphology ; (B) Microtomography reconstruction of the porous gelled soybean oil (15wt% of HSA: and sugar templated).

3- <u>Environmental scanning electron microscopy :</u>

The porous organogels were evaluated for cell culture viability and proliferation. After 21 days, environmental SEM images (Figure 4), clearly shows the cells attached on the 3D network of the material. Additionally, the results of histological staining and optical microscopy analysis indicate that these microporous organogels induces an appropriate environment that favors cell migration and collagen synthesis.

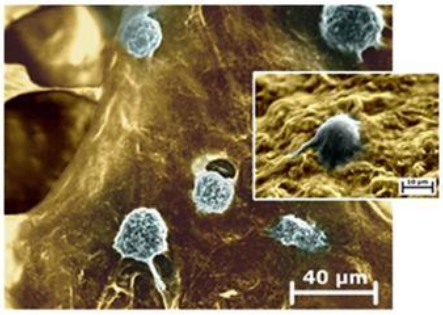

Figure 4: Colored environmental SEM micrograph of CHO fibroblast cells attached on the soybean oil based scaffold after 3 weeks of cell culture.

All of these results underline the usefulness and the importance of electron microscopy observations in the elaboration of such porous organogels. These microporous organogels can be potential candidates as artificial extracellular matrices for tissue engineering applications, and have also demonstrated interesting properties as adsorbent material.