Hydrogels with Fully Interconnected and Tunable Pores Prepared from Co-Continuous Polymer Blends.

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Introduction. Porous hydrogels, also called microfluidic hydrogels, are gels that possess a network of interconnected pores.[1] They are of particular interest in the fields of biomedical and tissue engineering for applications targeting the repair and/or regeneration of vascularized tissues.[2,3] Gas-foaming, solvent casting/porogen leaching, cryogelation, soft-lithography and rapid-prototyping are some of the methods that are currently being developed and used to prepare these materials. Some of the challenges associated with the preparation of porous gels include control over the pore volume fraction, pore interconnection, the average pore diameter and size distribution, and compatible gel chemistries. Furthermore, there is a need to develop techniques allowing the quantitative characterization of their microstructural characteristics. All of these challenges are complicated by their 3-D microstructure. The objectives of this work were to (1) develop a new method, using co-continuous polymer blends,[4] to prepare hydrogels comprising networks of fully interconnected pores and (2) to develop techniques to characterize their 3-D microstructure.

Methods. Co-continuous polymer blends of polystyrene (PS) and polylactide (PLA) (50/50 %vol.) were melt-processed in an internal mixer at 190°C during 5 min at 50 RPM, then quenched in cold-water to freeze-in the morphology. Quiescent annealing (QA) of the blends at 190°C during 10, 30, 60 or 90 min allowed coarsening of the microstructure. A number of samples were subsequently cryogenically microtomed, while others were shaped in 1 cm^3 cubes. The PS phase was then selectively extracted using cyclohexane to obtain porous PLA materials. Their microstructure was characterized by scanning electron microscopy (SEM) and image analysis (IA). Solutions of 2% sodium alginate (colored in blue) or 3% agar (colored in red) were then injected into the porous cubic PLA materials. The extent of solution penetration was assessed by visual inspection and by optical microscopy. After *in situ* gelling, the PLA was selectively extracted using chloroform to obtain porous hydrogels. Their microstructure was characterized by X-ray microtomography (μCT) and IA.

Results. After melt-processing and quiescent annealing of the PS/PLA polymer blends, the amount of extracted PS with cyclohexane, as measured by gravimetric analysis, indicated the level of continuity of the PS phase - nearly 100% for all QA times. SEM inspection of the cryomicrotomed and PS-extracted samples revealed the microstructure of the blends (Figure 1a).

IA has demonstrated that the specific PS/PLA interfacial area (which subsequently corresponded to the PLA specific surface once the PS was extracted) decreased from 5800 cm^{-1} (right after melt-processing) to 45 cm^{-1} (after 90 min of QA). From these data, it was calculated that the average pore size increased from 3 μm to 450 μm.

Injections of the agar or alginate solutions within the porous PLA materials were realized with a system using a manually applied pressure. Figure 1b shows a PLA mold completely filled with the agar solution.
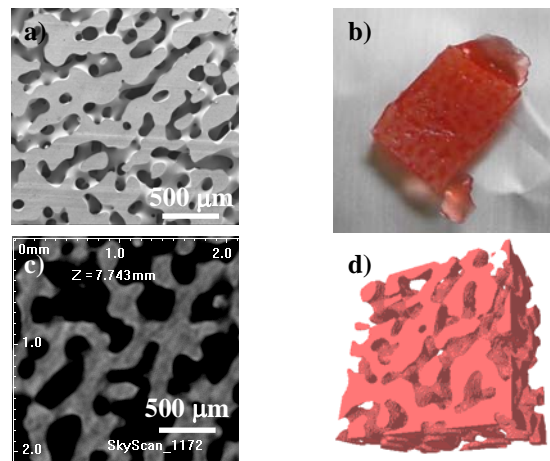

Figure 1. a) Porous PLA obtained after 60 min of quiescent annealing (QA); b) Injected agar solution in a PLA mold annealed during 60 min; c) 2-D μCT image of a porous agar gel (grey = agar, black = pores) (QA = 60 min); d) 3-D reconstruction of porous agar gel in (c).

Porous hydrogels were obtained after the selective extraction of the PLA phase. μ-CT, a non-destructive 3-D imaging technique, was employed to visualize the microstructure. Figure 1c shows a 2-D section of a porous agar gel prepared with a PLA mold initially annealed during 60 min. Figure 1d shows the 3-D reconstruction. The average pore sizes of agar and alginate gels prepared with PLA molds annealed during 60 min were respectively of about 275 μm and 260 μm, as compared to 200 μm for the PLA molds. The differences' causes remain to be clearly identified.

Conclusions. A new methodology, based on using co-continuous polymer blends, has been developed to prepare hydrogels with fully interconnected pores. The average pore size can potentially be tuned from ~ 1 μm to near 1000 μm by control of the porous PLA molds. A variety of chemistries could be used – in this work, demonstrations are shown for thermoreversible (agar) and ionic (sodium alginate) gels. A future work will look at the applicability of this approach for other types of gels.

References.
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