A Wet Route to Nanofiber-based Chitosan Sponges

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A simple wet route for the fabrication of nanostructured chitosan sponges has been developed on the base of a novel phase-separation technique, thus providing a new method for the preparation of chitosan nanomaterials.

Polymer nanofibers, with diameters in the nanometer range, possess larger surface areas per unit mass and permit easier addition of surface functionalities compared with polymer microfibers. Hence, polymer nanofiber-based mats or sponges are being considered for use as filters, scaffolds for tissue engineering, protective clothing, reinforcement in composite materials and sensors.[1] In biomaterial research, tissue engineering requires the design of ideal scaffolds from synthetic or natural materials that provide temporary templates for cell seeding, invasion, proliferation, and differentiation, resulting in regeneration of biologically functional tissue. Scaffolds made of nanofibers from biodegradable polymers may be helpful in adjusting the degradation rate of a specialized biomaterial in the in vivo environment. In addition, it has been proved that diameters of microfibers do affect the degradation features[2,3] and related mechanical properties of the materials used.[4] Furthermore, it has been theorized that cells attach and organize well around fibers with diameters smaller than the diameter of the cells.[5] Hence, researchers have tried to convert biopolymers into nanofiber mats that mimic biological structures. Up to now, nanofibers from various synthetic polymers have been reported, as well as those from natural polymers, including proteins,[6,7] nucleic acids,[8] and polysaccharides.[9,10] Among these biopolymers, chitosan is more attractive because of its abundant production in nature, excellent biocompatibility, extensive versatilities and potential applications in biomaterial areas, and thus the fabrication of chitosan nanofibers have attracted widely attention in recent years. Currently, the fabrication of chitosan nanofibers is mainly depended on electrospinning technique.[11–13] Although electrospinning has been recognized as an efficient method for the fabrication of submicron-sized fibers and various macromolecules have been electrospun into ultrafine fibers as thin as several nanometers, obviously, this technique was greatly limited by the electrospinning apparatus. There are few previous work related to the fabrication chitosan nanofibers using wet chemical route.[14] To meet this challenge, a facile and low-cost method is more desired. In this letter, we demonstrated the fabrication of chitosan nanofiber-based sponges via a wet chemistry way.

The principle of our method is schematically shown in Figure 1. Our method is based on an interface diffusion and precipitation process. When oil-phase (OP) substance containing triethylamine (TEA) (or triethanolamine, TEOA) was introduced to chitosan solution (water phase, WP), they would form an oil–water interface between the two phase. TEA would diffuse from OP to WP and hydrolyze into ammonia. This ammonia would neutralize the H^+ of the chitosan solution at the interface. As a result, chitosan would precipitate out at the oil–water interface. Obviously, this controlled process can be utilized to carry out the controlled precipitation of chitosan. Here, we demonstrate the fabrication of chitosan nanofibers based on the above mechanism. This can be easily accomplished via an emulsion technique. The detailed method is described as follows.

(1). Water-phase solutions were prepared as following. Stock chitosan solutions with different concentrations were prepared. The chitosan was stirred in 1% v/v aqueous acetic acid. After resting for 24 h, the undissolved chitosan was separated by filtration through a medium porosity filter. In a typical process, 0.5 g of chitosan with deacetylation degree (DD) of 82% was dissolved in 100 mL of 0.5% acetic acid solution.

(2). Oil-phase solutions were prepared as following. TEA was dropped to hexane under gentle stirring. In a typical process, 0.5 g of TEA were dissolved in 10 mL of hexane (to stabilize TEA in hexane, 0.1 g of Span-80 was introduced to the oil-phase solution).

(3). The oil-phase solutions were introduced to the water-phase solutions under violent stirring. As a natural glucosamine, chitosan can be used as polymer cationic emulsifiers in acidic aqueous solutions (see Figure 2). The oil-phase drops were emulsified and stabilized by the chitosan cationic emulsifiers. In this emulsion system, chitosan solution was the continuous phase and the oil-phase drops were the dispersed phase. The oil beads were suspended and stabilized by the chitosan cationic emulsifier micelles.

With the diffusing and hydrolyzing of TEA to ammonia, chitosan would precipitate out at the oil–water interface and grow into nanofibers. To improve the intensity of the chitosan nanofibers, crosslinking agent (e.g., glutaraldehyde) can be introduced into the chitosan solutions. The final chitosan nano-

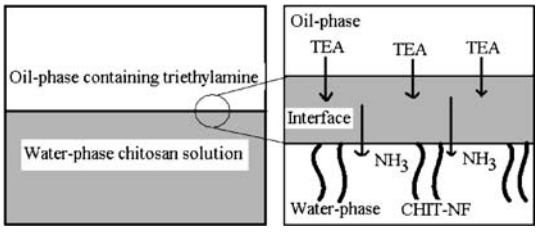

Figure 1. The interface precipitation process for the formation of chitsoan nanofiber-based sponge.

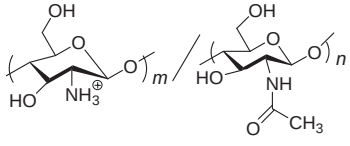

Figure 2. Chitosan molecular section in acidic aqueous solution.

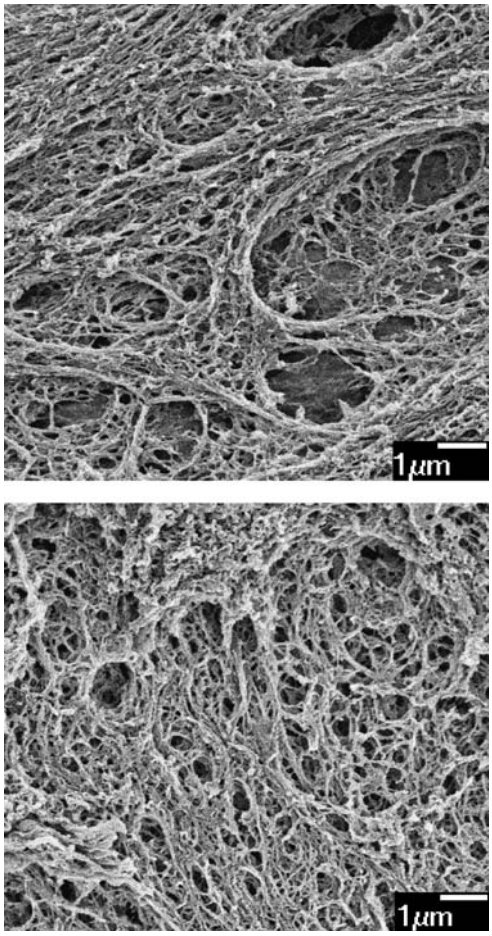

Figure 3. SEM images of the chitosan nanofiber-based sponges.

structured products were collected by centrifugation and washed with water and ethanol for several times. The chitosan products were dried at 60 °C under vacuum and then their morphology was analyzed using scanning electron microscopy (FE-SEM, JEOL JSM-6335F).

Figure 3 shows the SEM images of the chitosan nanofiber-based sponges. It reveals that well-constructed chitosn nanofiber-based sponges were fabricated after using the controlled diffusion and precipitation process. By changing the concentration of chitosan in water-phase solutions, the microstructures of the sponges can be controlled. Figure 4 presents the SEM images of the chitosan nanofibers. It shows that these nanofibers have an average diameter about 100 nm. Figure 4 also shows that those chitosan nanofibers have considerable surface roughness, and this rough surface structure will be very useful for the combination of cells in tissue engineering.

In summary, a simple wet-chemistry route for the fabrication of chitosan nanofiber-based sponges has been developed on the basis of a controlled interface diffusion and precipitation process. Chitosan nanofibers and nanofiber-based sponges were obtained, thereby providing an efficient and simple wet-chemistry way for the fabrication of chitosan nanomaterials. The approach was also cost effective with the simple procedure and inexpensive apparatus. These chitosan nanomaterials will have potential applications in many area of science.

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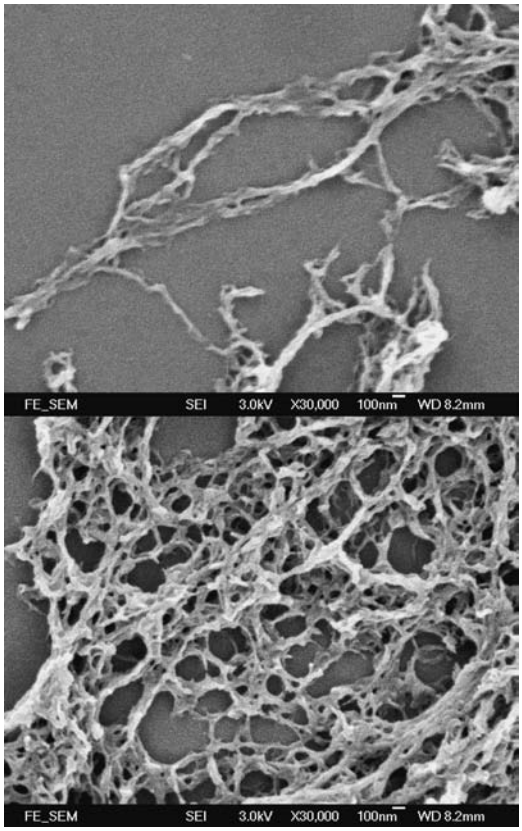

Figure 4. SEM images of the chitosan nanofibers.

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