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A hybrid network of synthetic polymer mesh and collagen sponge

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A novel hybrid network of synthetic polymer mesh and collagen sponge was prepared by forming collagen sponges with interconnected microporous structures in the interstices of the synthetic polymer mesh.

Biodegradable synthetic poly(α-hydroxy acids), such as poly(lactic acid) (PLA), poly(glycolic acid) (PGA) and their copolymer of poly(DL-lactic-co-glycolic acid) (PLGA), and collagen have been widely used for tissue engineering as temporary scaffolds to accommodate transplanted cell masses, and as materials for assisting in surgeries in clinical applications.[1–6] The synthetic polymer meshes demonstrate good biocompatibility, good mechanical properties, and are easy to handle. Their rate of degradation can also be controlled to meet the rate of new tissue formation in tissue repair. However, these synthetic polymers are relatively hydrophobic. These hydrophobic properties, together with the large mesh interstices, hinder smooth cell seeding. In contrast, collagen offers the advantage of specific cell interactions and hydrophilicity, but scaffolds constructed entirely of collagen have poor mechanical strength. Therefore, synthetic biodegradable polymers and collagen have been hybridized to combine their advantages.[7–13] In the present study, a novel hybrid biomaterial was prepared by combining synthetic biodegradable polymer mesh with collagen sponge.

The hybridization of synthetic poly(α-hydroxy acids) and collagen was achieved by forming collagen sponges between the interstices of poly(α-hydroxy acids) mesh. A Vicryl knitted mesh made of polylactin 910 (a 90 : 10 copolymer of glycolic acid and lactic acid), as shown in Fig. 1a, was immersed in a bovine collagen acidic solution (type I, pH 3.2, 0.5 wt%), and frozen at −80 °C for 12 h. It was then freeze dried under a vacuum of 0.2 Torr for 24 h to allow the formation of collagen sponge. The collagen sponge was further cross-linked by treatment with glutaraldehyde vapor saturated with 25% glutaraldehyde aqueous solution at 37 °C for 4 h. After the cross-linking, the sponge was treated with 0.1 M glycine aqueous solution to block unreacted aldehyde groups. After being washed with deionized water and freeze dried, the polymer–collagen hybrid mesh was prepared.

The hybrid mesh was coated with gold and observed by scanning electron microscopy (SEM). The SEM photomicrographs of the hybrid mesh are shown in Fig. 1. Collagen sponges with interconnected microporous structures were formed in the interstices of the synthetic polymer mesh. The polymer mesh was embedded in the collagen sponge sheet so that the fiber bundles of polymer mesh and the collagen sponges were alternately chained. The thickness of the collagen sponge sheet surrounding the polymer mesh could be manipulated by adjusting the volume of the collagen solution. A greater volume of collagen solution resulted in a thicker layer of collagen sponge. The position of the polymer mesh in the hybrid network could also be manipulated. It could be a sandwich type with the polymer mesh embedded in the middle of the collagen sponge layer, or be overlapped. The hybrid mesh could be prepared in the shape of a sheet or a cylinder by using a polymer mesh sheet or tube.

20-mM HEPES buffer-soaked hybrid mesh, polymer mesh and collagen sponge were used for static tensile mechanical tests. They were pulled to failure at a rate of 0.5 mm min^{-1}. Load–deformation curves were obtained from a chart recorder. The moduli of elasticity of the hybrid mesh, polymer mesh and collagen sponge determined from the load–deformation curves and the dimensions of each sample were 35.42 ± 1.42, 35.15 ± 1.00 and 0.02 ± 0.00 MPa, respectively. The hybrid mesh possessed almost the same mechanical property as that of the polymer mesh, much higher than that of the collagen sponge alone.

Human skin fibroblasts were subcultured in 106S serum medium supplemented with 2 (v/v)% fetal bovine serum (FBS), 10 ng mL^{-1} recombinant epidermal growth factor (rEGF), and

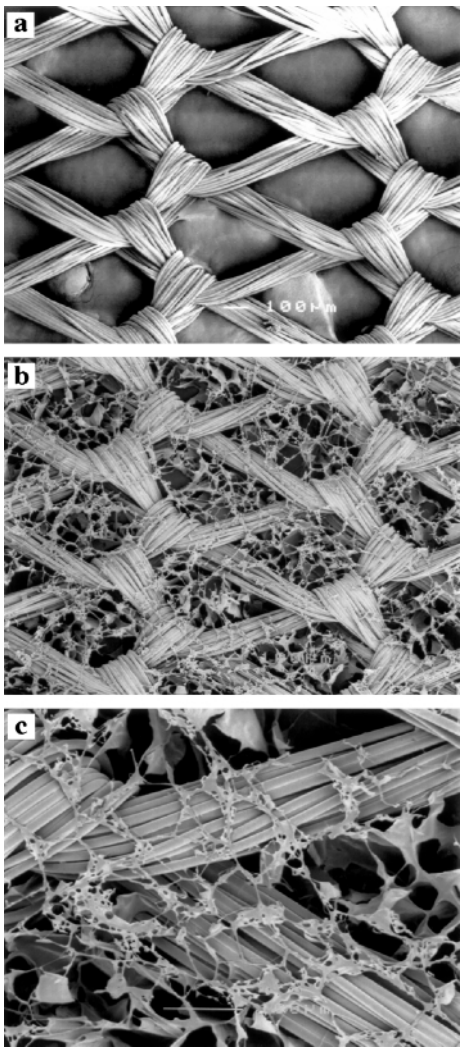

Fig. 1 SEM photomicrographs of polylactin 910 mesh (a) and its hybrid mesh with collagen at original magnification ×60 (b) and 200 (c).

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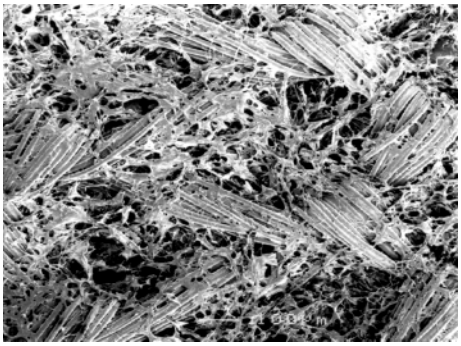

Fig. 2 SEM photomicrograph of human skin fibroblasts cultured on the hybrid mesh for 5 days at original magnification ×100.

3 ng mL^{-1} recombinant fibroblast growth factor-basic (rb-FGF). The subcultured fibroblasts were harvested and seeded on the hybrid and polymer meshes (7.64×10^4 cells/cm^2), and cultured in 106S serum medium under a 5% CO_2 atmosphere at 37 °C. The medium was replaced every 2 days. The cells adhering to the hybrid and polymer meshes after 24 h of culturing were $6.42 \pm 0.21 \times 10^4$ and $1.48 \pm 0.26 \times 10^4$ cells cm^{-2}, respectively. Many more cells adhered to the hybrid mesh than the polymer mesh. The cell morphology on the hybrid mesh was examined by SEM observation. Fig. 2 shows the appearance of the cells on the hybrid mesh after being cultured for 5 days. The fibroblasts adhered and spread well on the surfaces of the collagen sponge of the hybrid mesh after being cultured for 5 days. After 2 weeks, they proliferated to become completely connected in a layer structure. Over a longer culture period, the hybrid mesh degraded and eventually disappeared. Only cell sheet containing fibroblasts and extracellular matrices was left. These results suggest good cell interaction of the hybrid mesh.

Biodegradable synthetic poly(α-hydroxy acids) meshes have been used as temporary scaffolds for the tissue engineering of skin,[2] nerve,[9] esophagus,[10] ligament,[11] *etc*. However, the mesh interstices and their hydrophobicity hinder cell seeding. Cultured urothelial cells do not grow into a confluent layer on PGA or polyglactin mesh because of the large size of the mesh interstices.[14] To increase the cell seeding density on the PGA mesh, a method of surface hydrolysis has been used to improve the wettability of the mesh.[15] Hybridization of synthetic polymer with collagen has also been used to address these problems.[7–11] Polyglactin 910 mesh and PGA mesh have been coated with collagen solution, or embedded in collagen gels, to improve cell attachment and cell seeding. However, neither surface hydrolysis nor collagen coating changes the pore size of the interstices. The use of collagen gel produces a complete loss of pore structure. Compared to collagen gel, collagen sponge is porous enough to accommodate implanted cells, and its microporous structure facilitates cell seeding. The hybrid mesh of polylactin 910 mesh with collagen sponge exhibited a novel hybrid structure with interconnected microporous collagen sponges formed in the interstices of the synthetic polymer mesh. The polymer mesh, serving as a skeleton, reinforced the hybrid mesh and resulted in easy handling, while the collagen sponge provided the hybrid with a microporous structure and hydrophilicity, and, therefore, easy cell seeding. The hybrid mesh we developed could serve as a useful biomaterial for tissue engineering.

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