

higher concentrations will be used where local infusion of heparin into the blood is desired to inhibit thrombosis downstream of the graft, as described in Chen et al., *Boundary layer infusion of heparin prevents thrombosis and reduces neointimal hyperplasia in venous polytetrafluoroethylene grafts without systemic anticoagulation*, *J. Vascular Surgery*, v. 22, pp., 237-247 (1995).

[0040] The hydrogel supports the proliferation of eukaryotic cell cultures. Vascular cells such as endothelial cells, smooth muscle cells, and fibroblasts and other connective tissue cells, can thus be incorporated into the hydrogel. Human aortic endothelial cells and human dermal fibroblasts are also compatible with the hydrogels of the present invention. Hydrogels modified by such cell lines are, in turn, especially well adapted for implantation into the human body, and for use as soft tissue replacement parts in the human body. Indeed, replacement parts modified by such cell lines are better able to adapt and adjust to changing physical and physiological conditions in the body, and thereby to prevent any failure of the hydrogel which might otherwise occur. Hydrogels modified by such cell lines are, in sum, especially well adapted for implantation in the human body, and for use as replacement parts in the human body. These cellular lines can be incorporated into the hydrogel, after it has been produced, via standard cell culture protocol generally known in the art. It is especially effective to culture human aortic endothelial cells and human dermal fibroblasts using direct topical seeding and incubation in cell culture medium.

[0041] Besides the soft tissue replacement uses set forth for the poly(vinyl alcohol) hydrogel, discussed above, the hydrogels of the present invention can be used in any application in which poly(vinyl alcohol) hydrogels are generally suitable, including as an MR (magnetic resonance) quality control phantom, as an ultrasound or radio frequency thermal therapy transmission pad, as a substitute for an ice bag, as a denture base, and in other medical applications.

[0042] Although the following examples set out specific parameters for constructing a PVA hydrogel in accordance with the present invention, the ordinarily skilled artisan will understand that mechanical properties of the PVA hydrogel may be affected by one of four factors. Those factors include: (1) weight percentage of the respective constituents within the hydrogel (e.g. PVA polymer and water); (2) the molecular weight of the PVA starting material; (3) the number of freeze/thaw cycles; and (4) the duration of a freeze cycle. It is also important to note that the freeze/thaw cycle promotes an interlocking mesh or entanglement between molecules of PVA to create the mechanical strength. This is different than the traditional cross link accomplished by the above-referenced cross linking agents which inevitably introduces a toxic agent into the biomaterial, thus decreasing biocompatibility of materials which utilize those cross linking agents.

EXAMPLE 1

[0043] A 15% by weight poly(vinyl alcohol) solution was prepared by mixing 17.6 grams of poly(vinyl alcohol) polymer (124,000-186,000 Av. MW), 99+% saponification, in 100 ml of deionized, sterile water. The mixture was placed in a loosely capped container, heated and sterilized at 121° C. and 17 p.s.i. in an autoclave for about 15 minutes. The

container was then sealed removed from the autoclave and placed under a sterile ventilation hood. The mixture was then stirred to ensure a homogenous solution. The mixture was poured into sterile syringes, being careful not to generate air bubbles. The poly(vinyl alcohol) solution was then injected upwardly into stainless steel annular molds having stainless steel mandrels. The outer tube of the annulus had an inner diameter of 8 mm which surrounded a 5 mm diameter mandrel. The time that the solution was exposed to air was minimized in order to prevent evaporation of water. The mold was designed to create a poly(vinyl alcohol) hydrogel with approximately a 1.5 mm wall thickness, 10 cm long, having a 5 mm inside diameter. The mold was sealed at both ends using O-rings and rubber caps. Air space, equaling about 8% of the volume of the mold was deliberately maintained in order to allow for expansion while the aqueous solution froze.

[0044] The tube was then subjected to three (3) cycles of freezing and thawing. In each of the cycles the tube was frozen by placing it upright in a commercial freezer regulated at about -20° C., and allowing it to air cool for about 12 hours. The tube was then thawed by removing the tube from the freezer and setting it upright under ambient conditions. The tube was allowed to thaw for about 12 hours before being returned to the freezer for another cycle.

[0045] After the mixture had been frozen and thawed three times, it was removed from the tube (under a sterile vacuum hood) and immersed in a 50 ml, centrifuge vial containing 35 ml of deionized, sterile water. There was obtained a translucent to clear, gummy, weak material which was substantially unable to maintain its shape outside of water or other liquid. The material was handled carefully with forceps and immersed in water as quickly as possible. The inner diameter of the material was preserved by keeping the inner mandrel in place. The container was then sealed and placed in a freezer at about -20° C. The mixture was kept in the freezer for about 12 hours, and then removed and allowed to stand at room temperature for about 12 hours. The freezing and thawing process was repeated once, thus considering the three previous cycles within the mold, the mixture was subjected to a total of five (5) cycles of freezing and thawing.

[0046] The material obtained was opaque, elastic, and non-sticky, with mechanical properties very similar to a native artery tissue. The material was tested for mechanical strength according to standards of the Association for the Advancement of Medical Instrumentation and the American National Standards Institute, published in *Cardiovascular implants—Vascular Prosthesis*, ANSI/AAMI VP20-1994, section 8.3.3.3 (pressurized burst strength), and Section 8.8 (suture retention strength). The material had a burst pressure of about 540 mm Hg. Specifically, a 6-0 suture was placed 2 mm from the edge of the graft and pulled at a rate of 150 mm/min until it pulled through the graft. The average peak pullout load for the material a suture test was about 289 grams, which is greater than the pullout loads reported in the literature for human artery and vein. Finally, the tensile modulus of elasticity of the material was measured to be approximately 4.0×10^5 Pa.

EXAMPLE 2

[0047] A 25.9% by weight poly(vinyl alcohol) solution was prepared by mixing poly(vinyl alcohol) polymer (124,