

IONICALLY COVALENTLY CROSSLINKED AND CROSSLINKABLE BIOCOMPATIBLE ENCAPSULATION COMPOSITIONS AND METHODS

BACKGROUND OF THE INVENTION

The present invention relates to compositions and methods for encapsulating biologics such as biologically active materials or diagnostic markers. More specifically, the present invention relates to an ionically and covalently crosslinked biocompatible material which provides a non-cytotoxic, immunoprotective barrier for a biologic, including a xenotransplanted biologic. In another aspect, the present invention relates to a mixture of an ionically crosslinkable component and a covalently crosslinkable component, suitable for use in encapsulating a biologic. The present invention also relates to processes for encapsulating biologics with the crosslinked biocompatible material.

Many biocompatible materials such as lipids, polycations, and polysaccharides, such as hyaluronic acid, have been used to encapsulate materials such as living cells and tissue. For instance, the polysaccharide alginate has been used for the encapsulation, immunoisolation, and immobilization of cells. The combination of alginate with multivalent cations, such as calcium, can form mechanically stable, ionically crosslinked microcapsules. However, when used in a physiological environment, such as in the transplantation of microencapsulated islets or cells, the mechanical stability of the ionically crosslinked alginate microcapsules will erode since the extracellular concentration of monovalent cations exceeds the concentration of the divalent calcium cations, resulting in the exchange by diffusion of the monovalent and divalent cations.

Attempts to chemically modify alginate microcapsules or gels by utilizing covalent crosslinking rather than ionic crosslinking have been made in order to improve mechanical stability. However, the reagents and reaction conditions involved with these techniques often prove toxic and even fatal to the encapsulated materials. In order to further improve mechanical stability, covalently modified alginates have been photopolymerized to produce covalently crosslinked alginate gels. The rapid photopolymerization or photocrosslinking of the alginate avoids the exposure of the encapsulated materials to the toxic species that are present during the above-described processes.

It has also been found that implanted alginate gels having higher fractions of α -L-guluronic acid (G blocks) are more biocompatible than those containing a larger fraction of β -D-mannuronic acid (M blocks), since they do not induce a cytokine response from monocytes. Alginates having a higher fraction of M blocks on the other hand, induce a cytokine response when implanted in a physiological environment. This M block induced cytokine response results in fibrous overgrowth of the implanted alginate encapsulation material, thereby restricting the supply of nutrients and preventing the encapsulated material from permeating the encapsulation barrier for delivery to the physiological environment.

Polylysine has been used as an additional layer with microcapsules using high G-block alginates in order to improve chemical and mechanical stability. Nevertheless, although high G block alginate-polylysine gels have been demonstrated to successfully reverse diabetes

in spontaneous diabetic dogs when used to encapsulate transplanted islets of Langerhans, the long term function of these ionically crosslinked gels have been hampered by chemical and/or mechanical disruption of the alginate-polylysine membrane, thereby resulting in rejection and fibrous overgrowth.

Water-insoluble polymers, such as acrylate polymers or copolymers, and methacrylate polymers or copolymers, have been used in efforts to improve the stability of encapsulation materials. Photopolymerized polyacrylamides have also been in this regard. Likewise, however, these materials, or the organic solvents associated with them, often prove cytotoxic to the encapsulated living cells. Moreover, these water insoluble polymers tend to be bioincompatible in vivo in the long-term.

Crosslinked polyethylene glycol (PEG) gels have been used to immobilize enzymes and microbial cells. Polymerizable derivatives of PEG, such as PEG dimethacrylate, have been photocrosslinked using ultraviolet light in the presence of a suitable initiator to form a gel. This technique is desirable for the encapsulation of not only enzymes, but also for cells and organelles, due to the absence of heating, the avoidance of extreme pH values, and the absence of toxic chemicals in the photopolymerization process.

Polyethylene glycol is generally suitable as a biocompatible, protein repulsive, non-inflammatory, and nonimmunogenic modifier for drugs, proteins, enzymes, and the surfaces of implanted material. These characteristics of polyethylene glycol are attributable to its non-ionic character, its water solubility, the flexibility of its backbone, and its volume exclusion effect in solution or when immobilized at a surface. Further, surfaces modified with PEG have been found to be extremely non-thrombogenic, resistant to fibrous overgrowth in vivo, and resistant to bacterial adhesion. PEG bound to bovine serum albumin has been shown to have reduced immunogenicity and increased circulation times in a rabbit.

PEG has been covalently bound to poly-L-lysine (PLL) to enhance the biocompatibility of alginate-PLL (PLL) microcapsules for encapsulation of cells. PEG has also been covalently bound to polysaccharides such as alginates in order to make them soluble in organic media. However, this material requires the additional steps necessary to chemically modify the alginate before it can be crosslinked.

Polyethylene glycol has also been utilized as a component of interpenetrating polymer networks (or IPNs), which are a combination of two or more different polymer systems, at least one of which is synthesized or crosslinked individually in the presence of the other. In particular, polyethylene glycol has been used as a component in IPNs with several polymers and monomers such as N-acryloylpyrrolidine, polysiloxane, epoxy resins, and poly acrylic acid. Nevertheless, the IPNs known in the art have not utilized alginates as one of the crosslinked components, nor are IPNs known in which one of the components is ionically crosslinked.

Although many of the polyethylene glycol gels known in the art elicit a very low fibrotic response in physiological environments, such gels often require an essential emulsion or co-extrusion step with an oil or water immiscible phase for the formation of droplets or microcapsules. This may result in some toxicity due to