Zwitterionic poly(carboxybetaine) hydrogels for glucose biosensors in complex media

Wei Yang, Hong Xue, Louisa R. Carr, and Shaoyi Jiang

Department of Chemical Engineering, University of Washington, Seattle, WA 98195, US.

Statement of Purpose: Nonspecific protein adsorption is a crucial issue for many biomedical applications, but at present there are few effective nonfouling biomaterials available to meet the challenges of practical applications. Recently, zwitterionic materials such as carboxybetaine methacrylate (polyCBMA) have been shown to exhibit ultra-low protein adsorption. Amperometric glucose sensors based on glucose oxidase (GOx) have played a leading role in the move towards continuous monitoring of blood glucose. However, undesirable interactions between the surface of sensors and the biological medium have been proved to be the major barrier to the development of reliable *in vivo* glucose sensors. Hydrogel coatings based on PEG or poly(2-hydroxyethyl methacrylate) (polyHEMA) have been applied to minimize nonspecific protein adsorption. However, they dramatically decrease the electrochemical signal of the sensor. Herein, polyCBMA hydrogels were developed to protect implantable electrochemical glucose biosensors from biofouling in complex media.

Methods: Glucose sensors were designed based on the coil-type implantable sensor previously described.[1] The enzyme layer over the sensor was firstly formed by coating it with aqueous solution of GOx. To enhance the linearity and sensitivity of the sensing profile, both physical and chemical adsorption methods were used to prepare hydrogel-coated sensors via photopolymerization. Next, a novel CBMA-based crosslinker (CBMAX) was introduced to prepare polyCBMA hydrogels, and GOx was reacted with activated carboxylate groups of the polyCBMA hydrogels via sulfo-NHS and EDC chemistry. The *in vitro* performance of the sensors was examined with glucose solution dispersed in PBS, 10%, 50%, and 100% human blood serum. Amperometric measurements were performed at room temperature at +0.75 V vs. Ag/AgCl along with a platinum wire counter electrode. Mechanical and COS-7 cell adhesion properties of the hydrogels were also evaluated.

Results: After polymerization, all the hydrogels were transparent and colorless in appearance and had a uniform and smooth surface. Cell adhesion tests showed that polyCBMA hydrogels were highly resistant to cell attachment compared to polyHEMA hydrogel. The bare sensors without any hydrogel coating displayed very high sensitivity to glucose in PBS but failed when exposed to 10% blood serum. The polyHEMA hydrogel coating did not help to increase the linearity and decreased the sensitivity in 10% blood serum. The polyCBMA hydrogel coating via physical adsorption improved the sensor response linearity in 10% blood serum but failed when exposed to 50% blood serum. To improve the adhesion of the hydrogels, (trimethoxysilyl)propyl methacrylate was used to modify the sensor surface.[2] Results show the sensors coated with polyCBMA hydrogel via chemical attachment retained excellent linearity and high sensitivity in 10%, 50%, and 100% blood serum over 12 days. Then,

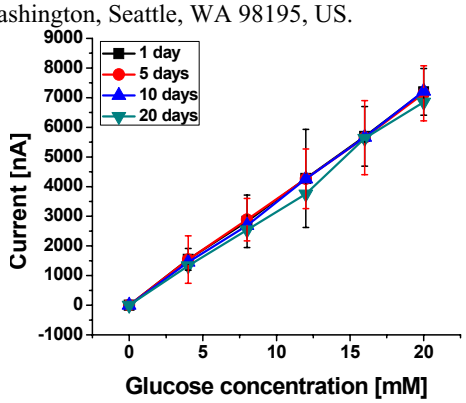

Figure 1. Current response of glucose sensors coated with polyCBMA hydrogels as a function of glucose concentration in 100% blood serum.

the novel CBMAX crosslinker and GOx immobilization method was introduced to reduce the loss of immobilized GOx. The current response of these CBMA-treated sensors to glucose in PBS had a 100-fold increase compared to the previous results.[1] Moreover, as illustrated in Figure 1, the sensors coated with 0.1% CBMAX molar ratio polyCBMA hydrogels did not show a decline in either sensitivity or linearity after exposure to blood samples over 10 days.

Conclusions: In this work, glucose sensor coating based on zwitterionic poly(carboxybetaine) hydrogels is demonstrated to effectively minimize blood–materials interactions and related adverse effects for long-term applications in complex biological media such as undiluted blood serum. The polyCBMA hydrogel layer via physical adsorption method improved the sensor response linearity in 10% blood serum but failed when exposed to 50% blood serum. Pt/GOx electrochemical biosensors chemically coated with polyCBMA hydrogels possess very high sensitivity and good linearity to glucose for over 12 days in PBS, 10%, 50%, and 100% blood serum. Furthermore, after introduction of the CBMAX crosslinker and GOx immobilization method, all the polyCBMA hydrogels are highly resistant to cell attachment before and after GOx immobilization. All the sensors displayed very fast current response with excellent linearity over the 4-20 mM range and the current response to glucose in PBS had a 100-fold increase. Moreover, the sensors coated with 0.1% CBMAX molar ratio polyCBMA hydrogels retained excellent sensitivity and linearity after exposure to undiluted blood samples over 10 days. With a single polyCBMA hydrogels coating, which could serve as both the diffusion-limiting layer and biocompatible outermost coatings, significant improvements of the *in vivo* performance of implanted glucose sensors are anticipated.

References: 1 Yu BZ, Biosens. Bioeletron. 2008; 23: 1278-1284; 2 Jimenez C. Anal. Chim. Acta 1997; 351:169-176.