CAPSULES notes and applications from BASi

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Comparison of Two Biosensors Used for the Determination of Glucose Concentrations

Purpose

One method that has been widely investigated for the determination of glucose is based on its reaction with glucose oxidase; that is, the sensitivity of amperometric detection is coupled with the selectivity of enzymes. A variety of designs for such glucose biosensors have been proposed, and two of these are discussed in this Capsule.

Reference

Critical Comparison of Metallized and Mediator-Based Carbon Paste Glucose Biosensors, J. Wang, L. Chen, and J. Liu, Electroanalysis 9 (1997) 298-301.

Method

The enyzmatic scheme on which glucose oxidase biosensors is based is shown in ***F1***. Glucose is

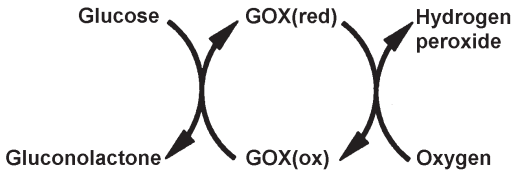

Figure 1. Scheme for the oxidation of glucose by glucose oxidase.

oxidized to gluconolactone by the flavin adenine nucleotide active site of glucose oxidase (GOX). The reduced (red) active site is then reoxidized by oxygen. The oxidation of glucose therefore involves consumption of oxygen and production of hydrogen peroxide, and ideally it should be possible to measure glucose concentrations by monitoring changes in the concentrations of oxygen or hydrogen peroxide using amperometric methods. However, there are problems associated with each of these measurements. Measurement of oxygen concentrations can be difficult due to natural variations in oxygen concentrations, and a relatively high potential (+0.6 V vs. Ag/AgCl) is required for the oxidation of hydrogen peroxide at a platinum electrode (which makes such measurements susceptible to interferences by e.g., ascorbic acid).

The high potential required for detection of hydrogen peroxide can be decreased by taking advantage of the catalytic reduction of hydrogen peroxide at various transition metals. In this study, rhodium was used as the catalytic material for one of the biosensors characterized in this study, and a potential of 0.05 - -0.1 V was used. Hydrogen peroxide can also be detected at moderate potentials using a biosensor based on peroxidase (1).

An alternative design for oxidase-based biosensors is based on the reoxidation of the active site electrochemically. Direct electron transfer between the electrode surface and the active site of the enzyme is generally not possible, since the active site may be buried inside the enzyme, so indirect (or mediated) electron transfer is used. Indirect electron transfer uses small redox-active molecules (mediators - Med) to carry electrons between the electrode and the active site of the enzyme (***F2***). Typical mediators include

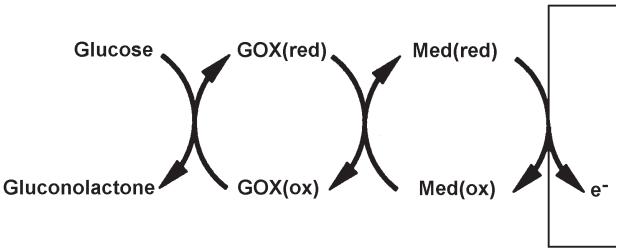

Figure 2. Scheme for the oxidation of glucose by glucose oxidase using mediated electron transfer between the electrode and the enzyme.

ferrocene, ferricyanide, quinones, and osmium and ruthenium complexes. The second biosensor characterized in this study was based on indirect electron transfer, using dimethylferrocene as the mediator.

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To summarize, one biosensor consisted of glucose oxidase, rhodium particles, and an enzyme stabilizer (polyethylenimine) in a carbon paste matrix. The second biosensor was made from glucose oxidase, dimethylferrocene, polyethylenimine in a carbon paste matrix.

Results

The first step in characterizing these sensors was the optimization of the fixed potential for the amperometric measurements based on hydrodynamic voltammograms. Potentials of 0.05 - -0.1 V (vs. Ag/AgCl) were used for the metallized biosenor, whereas a slightly more positive potential (+0.1 V) was used for the mediated biosensor.

The performance of each biosensor was examined by monitoring the current response to a series of standard additions of glucose under hydrodynamic conditions (i.e., stirred solution). A typical plot of current vs. time is shown in ***F3***. The following properties were examined: the response time, the sensitivity, the detection limit, the effects of varying the proportion of polyethylenimine, the effects of interferants, and the oxygen dependence.

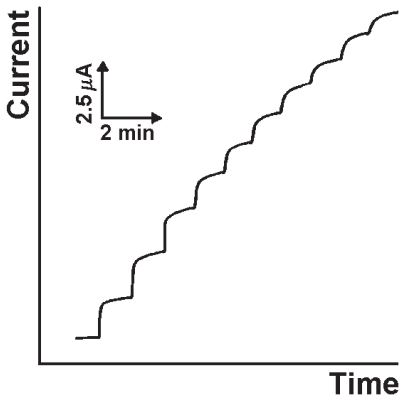

Figure 3. Typical current-time plot for the oxidation of glucose. Each current step corresponds to an addition of glucose. Adapted with permission from primary reference. Copyright 1997 VCH.

It was found that the detection limits of the biosensors were similar, whereas a higher sensitivity and slower response time was observed for the mediated biosensor. The addition of three common physiological interferants (ascorbic acid, uric acid, and acetaminophen) had little effect on the current response of the metallized biosensor, whereas the addition of ascorbic acid caused a significant increase in the current response of the mediated biosensor. Under anaerobic conditions, there was a 10 - 15 % decrease in the current response of the mediated biosensor, whereas the decrease for the metallized biosensor was much larger (this is to be expected, since this biosensor uses oxygen, rather than mediators, to reoxidize the active site of the enzyme).

It was concluded by the authors that there was no clear advantage in using one of these biosensors over the other. It was also noted that there are a variety of mediators and metal catalysts that can be used, as well as different electrode materials, all of which could affect the performance of the biosensor.

References

1. A.W. Bott, Curr. Seps. 17 (1998) 25 - 31 and references therein.