Chapter 8

Specific Applications

Learning Objectives

- To see the application of a number of the sensor techniques presented in this text to five different analyses.
- To appreciate the structure and performance of the Medisense 'ExacTech' biosensor for the determination of glucose in blood.
- To see how complexation and differential pulse polarography are combined to measure low concentrations of copper(I) in water.
- To understand the developments towards a 'laboratory on a chip' – using arrays of sensors to determine mixtures of analytes.
- To see specifically the use of ISFET sensors for the simultaneous determination of four different metal ions in blood.
- To understand how the combination of an immunoassay linked via enzyme processes to a bioluminescent assay can determine pmol amounts of explosive materials.
- To appreciate the use of a broad-spectrum enzyme available in plant tissues in the amperometric analysis of flavanols in beers.

DQ 8.1

What are the main factors to consider when devising a new sensor?

Answer

(a) *Any special criteria for the application.*
(b) *Decide on the selective element.*
(c) *Select the transducer.*
(d) *Decide on the method of immobilization.*
(e) *The performance factors required.*

(f) *Construction of the sensor.*
(g) *Operation of the sensor*
(h) *Testing of the sensor.*

8.1 Determination of Glucose in Blood – Amperometric Biosensor

8.1.1 Survey of Biosensor Methods for the Determination of Glucose

It is said that about half of the research papers published on biosensors are concerned with glucose. In addition to its metabolic and medical importance, this material provides a good standard compound on which to try out possible new biosensor techniques. There are, in fact, a number of different ways of determining glucose just by using electrochemical transducers. Reference to this has already been made under other transducer modes in Chapters 5 and 6 earlier.

Figure 5.18 shown above illustrates the overall pattern for assays which use these transducers. It can be seen from this figure that there are several different ways in which glucose may be determined, although glucose oxidase (GOD) lies at the centre of them all.

There are of course many other systems that can be used to determine glucose, employing different enzymes, such as glucose dehydrogenase (GDH), or different transducers, including thermal and photometric devices.

8.1.2 Aim – to provide a simple, portable, sensor for use at home by diabetics for regular monitoring of the level of glucose in their own blood

8.1.2.1 Special Criteria for this Application

For use by patients at home, it must be simple, reliable and cheap.

8.1.2.2 Decide on the Selective Element

Glucose oxidase is an inexpensive, readily available enzyme obtained from *Aspergillis niger*. It is stable over a long period of time, especially when sealed in foil.

8.1.2.3 Select the Transducer

An electrochemical transducer, particularly an amperometric type, is cheap, reliable and will give a direct read-out to a liquid crystal display (LCD). Most commercial glucose sensors use glucose oxidase coupled to a mediator (usually a ferrocene derivative) as the electron donor, rather than oxygen.

8.1.2.4 Decide on the Method of Immobilization

For 'long life', a covalent-bonding method is best. The original biosensors on which the Medisense 'ExacTech' sensor was based used a graphite foil coated with dimethylferrocene as the mediator. The oxidase was immobilized by reaction with 1-cyclohexyl-3-(2-morpholinoethyl)carbodiimide *p*-methyltoluenesulfonate. The Medisense 'ExacTech' device appears to use a screen-printed technique.

8.1.2.5 The Performance Factors Required

- Blood glucose range of 1.1–33.3 mM
- Precision of ±3–8%
- Test time of −30 s
- Lifetime of sensor – test-electrode strip to last at least six months in sealed foil

8.1.2.6 Construction of the Sensor

The device shown in Figure 8.1 has dimensions of 53 × 90 mm and weighs 40 g. It uses a non-replaceable 3 V battery that lasts for about 4000 tests. It is marketed at about £35[†] and is replaced free when the battery runs out.

The electrode strip is shown in more detail in Figure 8.2. This is used just once to ensure optimum reproducibility. These strips cost about 40 p each.[†] Miniaturization is achieved by immobilizing the enzyme and mediator on to a screen-printed strip, such as the one shown in Figure 8.1. One electrode contains the immobilized enzyme–mediator, while the other is a silver–silver chloride reference electrode. (Silver–silver chloride 'ink' can be screen-printed on to the appropriate electrode.) The electrode strip has two metal contacts at one end which are connected to the measuring device.

8.1.2.7 Operation of the Sensor

A drop of blood is placed on the strip across the two electrodes, making contact with both and thus obviating the need for a cell container. No supporting electrolyte is needed, and no de-gassing is involved.

The switch on the measuring device is then pressed. The timing is automatic and is usually displayed on the LCD read-out. After 30 s, the current is read and the reading is then converted into a measure of glucose in the blood and displayed.

The transducer is an electrode that is set to a potential (versus a reference electrode) at which the reduced ferrocene derivative may be re-oxidized, i.e. it is used in the amperometric mode. The oxidation current is measured for a fixed time (usually 30 s) and is then recorded and displayed on the LCD.

[†] Price as at September 2001.

Figure 8.1 The Medisense 'ExacTech' biosensor device. Reproduced by permission of Medisense, Birmingham, UK.

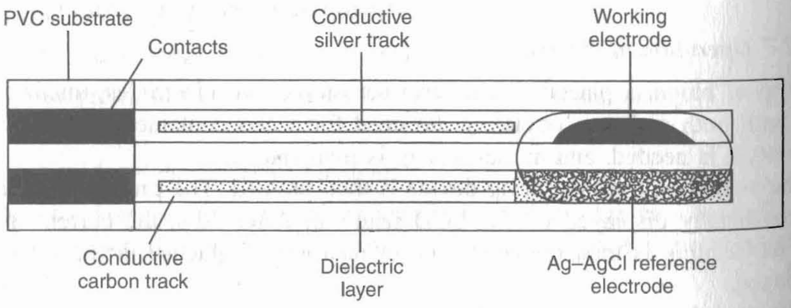

Figure 8.2 Schematic of the 'ExacTech' biosensor disposable electrode strip. From Hilditch, P. I. and Green, M. J., *Analyst*, **116**, 1217–1220 (1991). Reproduced with permission of The Royal Society of Chemistry.

8.1.2.8 Testing of the Sensor

The following shows test data provided by Medisense.

Precision Three blood samples with three different glucose levels were tested, with 20 readings being taken using electrode test strips from the same batch. The results obtained are shown in the table below.

Parameter	Sample		
	1	2	3
Number of readings	20	20	20
Mean concentration (mM)	3.0	4.8	15.5
Standard deviation	0.24	0.19	0.52
Coefficient of variation (%)	8.1	3.9	3.3

Accuracy A clinical study was carried out, comparing the 'ExacTech' sensor with the blood glucose-monitoring device for laboratory use from Yellow Springs Instruments (YSI Model, No. 23AM). The correlation over 200 readings was given by the following linear regression:

$$y = 1.041x + 0.271 \text{ mM}$$

with a correlation coefficient of 0.985.

Patient use A study compared the results obtained from a group of nurses and medical technicians with those obtained from a number of patients. The similarity in the findings showed that the 'ExacTech' device performed equally well in the hands of patients as it did with trained professionals.

SAQ 8.1

How could a glucose analyser be used for continuous monitoring of a patient's blood glucose levels?

8.2 Determination of Nanogram Levels of Copper(I) in Water Using Anodic Stripping Voltammetry, Employing an Electrode Modified with a Complexing Agent

8.2.1 Background to Stripping Voltammetry – Anodic and Cathodic

During cyclic voltammetry, the concentration of the reduced species builds up at the electrode surface. This process can be improved if the material is adsorbed