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(54) Title: SENSOR DEVICES

(57) Abstract

Sensor devices for examining fluid samples having, between the sample under examination and a detector, a membrane made of polyvinyl chloride in un-plasticised form. This membrane material acts as a barrier to paracetamol and sugars but is permeable to hydrogen peroxide and to oxalate, so these species can be determined after passing through the membrane. When they are not present as such in the sample, they may be formed from analytes which are sought (e.g. by enzymic action, which is especially applicable to the determination of glucose, using an oxidase enzyme). The membranes may be made by solvent casting, conveniently to a thickness of 10 to 40 μm, and used in electrolytic systems with a platinum anode. The membrane material can be formed into multi-layer membranes, which may incorporate layers of material to protect it or modify the permeability, or added materials for example enzymes.

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Sensor Devices.

This invention relates to sensor devices such as are used in the determination of a component or components which may be present in a fluid sample, such as a physiological fluid (e.g. blood) or other fluids of biological origin (e.g. fruit), process fluids or effluents.

Many forms of sensor have been proposed, and commonly these rely on some form of membrane to control the extent to which the components present in a sample under examination can gain access to an electrode, at which they can then be detected and determined. Especially, it is well known to make sensors using membranes to separate the media being analysed from the active electrode itself. The main function of the membrane is to separate, as far as possible, those components which are desirable (i.e. can take part in the reactions at the electrode on which the desired determination depends) from interferents (i.e. compounds which may be present but are undesirable because they either interfere with the progress of the desired determination reactions or take part in reactions of their own which compete with those of the component sought and distort or overwhelm the signals which are to be measured. The forms of construction have much in common with each other, and mainly differ in the nature of the membrane or media within it or combined with it in some way.

Some forms of sensor rely on the components used to make the membrane, while others rely on the mode of fabrication of the membrane, selecting its physical properties (for example its porosity) or treatments given to it, as these factors can control its effectiveness and selectivity in use.

Other forms of sensors incorporate an enzyme, which converts one substrate compound or analyte into another which may then be more easily measured. Especially, it is known to use oxidase enzymes, which generate hydrogen peroxide -- a substance which can be measured very conveniently and very accurately by electrolytic methods, especially amperometrically. An example is

European Patent No. 216577 (of ICI PLC) which specifies an enzyme electrode sensor with membrane barrier of low (>5%) porosity.

Materials which have been proposed for the fabrication of membranes for sensors include polyvinyl chloride (PVC), but only in plasticised form, as described in European Patent Application No. 92302131.5, where the plasticiser performs a necessary part of the function of the membrane.

Such known sensors, utilising various membrane materials, are very valuable in analytical techniques and the analysis of biological fluids (e.g. blood) for the presence of substances considered critical for medical reasons -- e.g. glucose and other materials which may be oxidisable or reducable, or have toxic properties.

We have now found that the selectivity of such a membrane-enclosed sensor can be significantly and surprisingly modified and improved by making the membrane of polyvinyl chloride (PVC) itself, in un-plasticised form.

Thus according to our invention we provide an improved sensor device comprising means for detecting components present in fluid samples and providing an output representative of the content of said component comprising a detecting means and a membrane barrier between the detecting means and the sample to be analysed, characterised in that the membrane barrier is composed of polyvinyl chloride (PVC) itself, in un-plasticised form.

The detecting means is most conveniently of one of an electrochemical nature, but other types (e.g. spectrophotometric or optical detecting systems) may be used if desired. The detecting means will usually comprise an electrode system and a liquid or gel phase electrolyte-containing medium. In most applications the electrolyte will be aqueous (i.e. aqueous or aqueous-based) but the use of non-aqueous electrolyte media (for example organic-based media) is not excluded.

According to our invention we also provide a method for determining a component in a fluid sample, which comprises contacting the sample with a sensor device as defined above.

Especially, in the devices and method of our invention, the sensor device comprises a detecting means in contact with an electrolyte medium and both associated with a membrane of polyvinyl chloride (PVC) itself, in un-plasticised form, which provides an interface for contact with a sample to be analysed and interposed between the active electrode (anode) of the cell used as detector. Alternative forms of construction may have (a) both the electrodes of the detecting means (cell) enclosed within the PVC membrane (so that both are separated from the sample) or (b) only the anode of the detecting means (cell) enclosed within the PVC membrane (so that the PVC membrane is between the electrodes and the cathode is not separated from the sample). Of these, the former is very convenient and compact, but the latter is more simple and is made practicable by the fact that PVC in un-plasticised form is less insulating in its properties, and H^+ ions can pass through it.

The sensor device of our invention can have a single membrane or, if desired, multiple layers of membrane material. When multiple layers of membrane are used, these may be the same or different. The preference for the position to be occupied by the un-plasticised PVC membrane differs to some degree according to the particular use to which the sensor is being applied and what substrate compound is to be detected by the sensor.

The governing factor is mainly the fact that un-plasticised PVC is permeable to hydrogen peroxide and some low molecular weight species (for example oxalate) but impermeable to sugars, for example glucose. Thus, when the sensor is to be used for detecting a species to which the PVC is impermeable, a sensor assembly is best made with an enzyme (with which the desired substrate can interact and generate a species which can pass through the un-plasticised PVC) situated outside the un-plasticised PVC membrane, so that the desired species in the sample under examination can make contact with the enzyme -- either directly or indirectly, through another selective membrane layer -- and thereby generate a species to which the un-plasticised PVC membrane is permeable. Especially, when glucose is the species sought, an oxidase enzyme can be used and the hydrogen peroxide thus generated can pass through the un-plasticised membrane and be determined at the detector means.

When multiple membrane layers are used, any membrane layer or layers other than any comprised of un-plasticised PVC may be made of any of the wide variety of materials known in the art. Examples of these include dialysis membranes, and in general are preferably non-diffusion limiting membranes, at least to the extent that they do not limit diffusion and passage of desired species towards the detecting means. When un-plasticised PVC is used as in inner membrane (i.e. a layer which is not the outermost), then one or more outer layers may be used which are of material which protects the sensor assembly in a mechanical manner (e.g. from mechanical damage) or in a chemical or any other manner considered appropriate for the use to which it is to be applied. Thus for example, there may be used an outer layer comprising a polycarbonate (especially in a porous form).

The active electrode may be any of those known in the art, for example a metal electrode, but especially a platinum anode. This is most conveniently made in combination with a silver/silver chloride counter electrode, as for example in the so-called Clark electrode, which comprises a platinum electrode surrounded by a silver/silver chloride ring.

The PVC (polyvinyl chloride) may be any polymer of vinyl chloride, as for example those made and available commercially, but should be free from any added plasticiser (an ingredient which is often present in some commercial products intended for uses such as moulding). Such "un-plasticised" PVC polymers are readily obtainable in commerce, however, and it is necessary only for the quality and purity of any polymer to be checked, whether by its specification or labelling. The molecular weight of the PVC is relatively non-critical, and most commercial grades will be satisfactory in use. A typical molecular weight is in the range 10,000 to 200,000, but others may be used if desired.

The material (i.e. the un-plasticised polyvinyl chloride) may be made into membranes by any conventional method. Most conveniently, this can be done by solution casting techniques, using solvents to dissolve the polymer and then spreading the solution on a plate or flat surface and allowing the solvent to evaporate. A convenient solvent is tetrahydrofuran (THF), but other solvents or mixtures of solvents which are known to be able to dissolve PVC be used if desired.

The thickness of the membranes can be of the order already used conventionally in the art, but may be varied as found most appropriate having regard for the particular mixed polymer composition being used and the conditions under which it is to be used. Thus a convenient thickness is in the range 10 to 40 μm, though larger or smaller thicknesses can be used if desired.

There are two principal forms of construction which may be used to secure the advantages of the new membrane material we now propose. In one, the un-plasticised PVC is the outer membrane, and in the other it is the inner membrane.

The forms using an enzyme will have basically the construction sequences:-

(A)	OR	(B)
Sample		Sample
---------		---------
Membrane (e.g. Polycarbonate)		Membrane (Un-plasticised PVC)
---------		---------
enzyme		enzyme
---------		---------
Inner (Un-plasticised PVC) membrane		Inner (e.g. polycarbonate) membrane
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Electrode.		Electrode.

For these, the components (apart from the un-plasticised PVC membrane) are mainly the conventional ones, and the many variants known in the art may be used.

Un-plasticised PVC is not permeable to glucose, so direct electrochemical detection of glucose cannot be achieved through this material as membrane. However, it is permeable to hydrogen peroxide, so enabling an alternative glucose-permeable membrane and an oxidase enzyme to be used to produce hydrogen peroxide, which then can selectively permeate the un-plasticised PVC membrane.

This un-plasticised PVC membrane has a particular advantage over plasticised PVC in that it is (unlike the plasticised form) impermeable to paracetamol -- so that this compound is excluded from interfering with hydrogen peroxide detection, as well as ascorbate/urate. This selectivity against paracetamol and the like while in favour of hydrogen peroxide is a valuable property which is not easily found and it very useful in clinical and related analytical, diagnostic and monitoring work.

Consequently, any other sensor device or system using an oxidase enzyme to generate hydrogen peroxide can, with advantage, incorporate an un-plasticised PVC membrane as part of its construction, especially as an inner membrane near to the active electrode surface.

Another advantage is that sensor devices of the electrolytic (amperometric) type are often used for examination of biological fluids for example blood, urine, and the like. It happens that urine contains a number of interferents which -- even though their identity has not yet been established -- have the ability to permeate many of the known conventional membrane materials, for example plasticised PVC, and so interfere severely with the accurate estimation of components such as hydrogen peroxide and of compounds which can be used to generate it. The un-plasticised form of PVC does not allow these unidentified components to pass through, is it constitutes an effective barrier to them and facilitates the wider applicability and accuracy of measurement. Thus, for many applications the un-plasticised PVC membrane will be preferable, though it will not always be so in applications in which it is desired to have a phenolic compound permeating the membrane.

Another use for the un-plasticised PVC membrane is in the construction of a sensor device for the determination of oxalate. It acts in this in two ways, both of which are valuable. One is a form of sensor in which the un-plasticised PVC membrane is used as an inner membrane beneath a layer or membrane comprising an oxalate oxidase system, so that hydrogen peroxide is generated from the sample and the resulting hydrogen peroxide passes through the un-plasticised PVC membrane; this relies on its action as a membrane selectively permeable to hydrogen peroxide.

Alternatively, the un-plasticised PVC membrane can be used as an outer layer with the means for oxalate detection beneath it; this relies on its unexpected ability to act as a membrane selectively permeable to oxalate. Un-plasticised PVC is the only material that we know of at present which is oxalate-selective, and the mechanism by which it performs in this way is still not clear to us. In this mode or embodiment of our invention, the means for detecting the oxalate which has passed through the un-plasticised PVC membrane may be any of those known and may be either an enzyme one (oxalate oxidase based) or one which detects and measures the oxalate directly by electrolytic action. This may be amperometric, and the pH of the medium around the active electrode may be adjusted as may be found most appropriate by simple trial.

Whatever the mechanism by which the permeability to oxalate takes place, the ability of an un-plasticised PVC membrane to act so as to exclude serum and urine interferents from the active electrode region of a sensor is true and highly effective and a valuable feature of this invention.

An example of a basic construction sequence utilising this un-plasticised PVC membrane can be represented as follows:-

(1) SAMPLE
(2) ========== un-plasticised PVC membrane
(3) ENZYME (Oxidase)
(4) ========== inner membrane
(5) ELECTRODE.

In this, oxalate or oxalic acid from sample (1) passes through the un-plasticised PVC membrane (2) and then, by contact with the enzyme (3), generates hydrogen peroxide and carbon dioxide. The hydrogen peroxide then passes through the inner membrane (4) to the electrode (5), at which it is determined electrolytically.

The inner membrane (4) may be of any convenient material -- for example a dialysis membrane or a polycarbonate membrane -- intended to perform such actions as regulation of the flow of components to electrode (5) (should this be considered desirable) or to provide mechanical protection and add to the robustness of the sensor assembly and minimise risk of damage to the innermost electrode elements.

Thus according to our invention we also provide, as useful new products, membranes comprising un-plasticised polyvinyl chloride. Especially, we provide these in the form of multi-layer membrane products, in which at least one layer is formed of un-plasticised polyvinyl chloride and is combined with one or more layers of other materials of appropriate properties to enhance the properties of the un-plasticised polyvinyl chloride itself. Such materials may be of appropriate permeability to regulate the access of components before or after passage through the un-plasticised polyvinyl chloride, and/or of a physical form or strength which protects the un-plasticised polyvinyl chloride from damage or provides it with any desired degree of stability of shape or positioning in use.

Especially, we provide membranes of un-plasticised polyvinyl chloride incorporating an enzyme, for example an oxidase. In these, the enzyme may be immobilised by the chemical means known in the art, or they may be held between the layers of a multi-layer structure.

CLAIMS.

1. A sensor device comprising means for detecting components present in fluid samples and providing an output representative of the content of said component, comprising a detecting means and a membrane barrier between the detecting means and the sample to be analysed, characterised in that the membrane barrier is composed of polyvinyl chloride (PVC) itself, in un-plasticised form.
2. A sensor device as claimed in Claim 1 wherein the detecting means is of an electrochemical nature.
3. A sensor device as claimed in Claim 1 or Claim 2 which comprises a detecting means in contact with an electrolyte medium and both associated with a membrane of polyvinyl chloride (PVC) itself, in un-plasticised form, which provides an interface for contact with a sample to be analysed and interposed between the active electrode (anode) of the cell used as detector.
4. A sensor device as claimed in Claim 3 wherein the detecting means comprises an electrode system and a liquid or gel phase electrolyte-containing medium, which is preferably aqueous or aqueous-based.
5. A sensor device as claimed in any of Claims 1 to 4, wherein the polyvinyl chloride has a molecular weight in the range 10,000 to 200,000.
6. A sensor device as claimed in any of Claims 1 to 5, wherein the polyvinyl chloride membrane has a thickness in the range 10 to 40 µm.
7. A sensor device as claimed in any of Claims 1 to 6, wherein the polyvinyl chloride is made by solution casting.
8. A sensor device as claimed in any of Claims 1 to 7, wherein the active electrode (usually the anode) of the detecting means is made of platinum.
9. A sensor device as claimed in any of Claims 1 to 8 wherein the membrane of un-plasticised polyvinyl chloride is used as in inner membrane in conjunction with one or more outer layers of

material, for example a polycarbonate (especially in a porous form) which protects the sensor assembly.

10. A sensor device as claimed in any of Claims 1 to 9 wherein an enzyme is used to convert a component or analyte to be determined into another compound which is then detected and measured by the detecting means.

11. A sensor device as claimed in Claim 10 adapted for detecting a species to which the un-plasticised polyvinyl chloride is impermeable, wherein the enzyme (with which the desired substrate species can interact and generate another species to which the un-plasticised polyvinyl chloride is permeable) is situated outside the un-plasticised polyvinyl chloride membrane, so that the desired species in the sample under examination can make contact with the enzyme and thereby react to generate a species which can pass through the un-plasticised polyvinyl chloride membrane to the detecting means.

12. A sensor device as claimed in Claim 11 wherein the enzyme is an oxidase, which is used to interact with an oxidisable component, for example glucose, to generate hydrogen peroxide -- to which un-plasticised polyvinyl chloride is permeable.

13. A sensor device as claimed in Claim 10 adapted for detecting a species to which the un-plasticised polyvinyl chloride is permeable, wherein the enzyme is situated within the un-plasticised polyvinyl chloride membrane, so that the desired species in the sample under examination first pass through the un-plasticised polyvinyl chloride membrane and then make contact with the enzyme and thereby generate a species to which the detecting means is responsive.

14. Sensor devices incorporating un-plasticised polyvinyl chloride, substantially as described.

15. Method for determining a component in a fluid sample, which comprises using a sensor device as claimed in any of Claims 1 to 14.

16. Method as claimed in Claim 15 wherein the component to be determined is hydrogen peroxide (to which the un-plasticised polyvinyl chloride membrane is permeable) and this is

determined either directly by being present as such or indirectly by being generated from another component or analyte (and so able to serve as a measure thereof), for example by enzymic action.

17. Method as claimed in Claim 15 wherein the component to be determined is oxalate (to which the un-plasticised polyvinyl chloride membrane is permeable) either directly as such or as another compound generated therefrom, for example as hydrogen peroxide generated therefrom for example by enzymic action (for example using an oxalate oxidase).

18. Method as claimed in Claim 17 wherein the means for detecting the oxalate which has passed through the un-plasticised polyvinyl chloride membrane is either an enzymic one (oxalate oxidase based) or one which detects and measures the oxalate directly by electrolytic action.

19. Method as claimed in any of Claims 15 to 18, employed for the study of biological fluids.

20. Method for determining a component in a fluid sample, using a membrane of un-plasticised polyvinyl chloride, substantially as described.

21. A multiple-layer membrane product, comprising at least one membrane layer of un-plasticised polyvinyl chloride and also an enzyme (for example an oxidase) for converting one analyte component into another which is more readily determined at a detector means.

22. Un-plasticised polyvinyl chloride membranes for use in a sensor device or method, substantially as described.

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INTERNATIONAL SEARCH REPORT

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A. CLASSIFICATION OF SUBJECT MATTER
IPC 5 C12M1/40 G01N27/40 G01N27/30

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 5 C12Q G01N C12M

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	EP,A,0 216 577 (IMPERIAL CHEMICAL INDUSTRIES PLC) 1 April 1987 cited in the application see the whole document ---	1-22
P,A	EP,A,0 503 943 (IMPERIAL CHEMICAL INDUSTRIES PLC) 16 September 1992 cited in the application see the whole document -----	1-22

☐ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

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Information on patent family members

Inte. ɔnal Application No
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Patent document cited in search report	Publication date	Patent family member(s)	Publication date
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