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(54) **Title:** SENSOR DEVICE AND METHOD OF PREPARING THEREOF

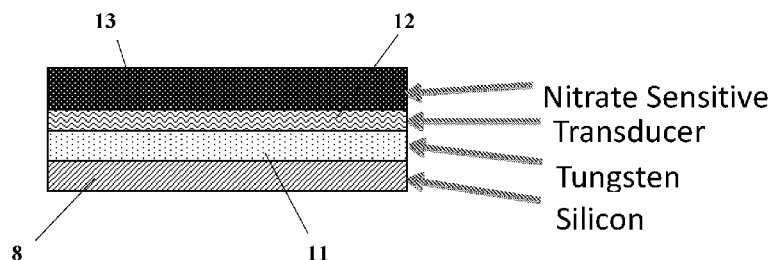


Figure 1

(57) **Abstract:** The present invention relates to relates to a sensor device and a method of preparing thereof, more particularly the present embodiment relates to a device for sensing nitrate comprising a substrate (8), a first layer (11) for transferring signals, a second layer (12) for converting energy into signals, and a nitrate sensitive material (13) formed on the second layer.

SENSOR DEVICE AND METHOD OF PREPARING THEREOF**TECHNICAL FIELD**

- 5 The present invention generally relates to a sensor device and a method of preparing thereof, more particularly the present invention relates to a device for sensing nitrate and a method of preparing thereof.

BACKGROUND OF INVENTION

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Applications using Nitrates have been widely implemented in various industries such as agriculture, chemical industry, medicine and food industry. However, uncontrolled use of nitrate can be hazardous to human and the environment.

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Detection of nitrates and the determination of the concentration of the nitrates are therefore important for controlling the levels of nitrates that can be introduced to humans and the environment. Kjeldahl method is commonly used to determine the concentration of nitrate in a sample. This method however involves long procedure and that the method has been tedious in determining nitrate levels.

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Alternatively, ion selective sensor can be used to determine nitrate levels via in situ measurement, and that the ion selective sensor uses membrane as an active supporter of the sensor for sensing nitrate. The use of membranes as a supporting matrix however has several drawbacks of delamination and leaching, such that the membrane will be broken off

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from the electrode surface when continuously and intensively used.

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US 2003/0232322 A1 discloses a biosensor method and apparatus for detecting and measuring nitrate, and the biosensor is based on the fluorescence properties of a receptor molecule fragment. The biosensor apparatus as disclosed in the prior art contains the active-site fragment of the receptor molecule for detecting nitrate.

WO 2005/119237 A1 discloses a biosensor for detecting nitrate ions and a measuring method using the same are provided, and the biosensor includes a working electrode having a nitrate reductase (NaR) immobilized thereon, a counter electrode and a reference

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electrode. The method as disclosed in the prior art is used to measure the concentration of nitrate in a sample using the biosensor.

US 5,776,715 discloses a biosensor for detecting nitrate or nitrite ions, and that the biosensor comprises a biochemical capable of producing a biochemical reaction in the presence of nitrate or nitrite ions and a transducer arranged to detect a transducible output agent produced directly or indirectly by the reaction produced by the biochemical in the presence of nitrate or nitrite ions.

However, these prior art documents do not provide solutions to overcome the problems of delamination and leaching associated with membranes in nitrate sensors.

SUMMARY OF INVENTION

The present invention pursues a sensor device and a method of preparing thereof, and that the sensor device is used for sensing nitrate levels. The sensor device of the preferred embodiment addresses the problems of delamination and leaching.

An object of the present invention is to provide a sensor device comprising a substrate, a first layer for transferring signals, a second layer for converting energy into signals and a nitrate sensitive material formed on the second layer.

Another object of the present invention is to provide a sensor device constructed from the second layer comprising a transducer layer grown on the first layer comprising a conductive layer, and that the nitrate sensitive material is grown on the second layer of the transducer layer.

It is yet another object of the present invention to provide a method for preparing the sensor device comprising the steps of fabricating a conductive layer, growing a transducer layer on the conductive layer, preparing a mixed composition from materials sensitive to nitrate, dispensing the mixed composition on the transducer layer, curing a layer of the mixed composition on the transducer layer, and forming a layer of nitrate sensitive layer responsive to nitrate.

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The present invention therefore aims to provide a sensor device for sensing nitrate without the use of lipophilic membrane or polymer matrix.

BRIEF DESCRIPTION OF DRAWINGS

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Figure 1 illustrates a graphical representation of a cross-sectional view showing compositional layers of the preferred embodiment.

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Figure 2 illustrates a flow chart representing the process for preparing the device of the preferred embodiment.

Figure 3 illustrates a graphical representation of a cross-sectional view showing compositional layers of an alternative embodiment.

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Figure 4 illustrates chemical structures of examples of conductive polymers of an alternative embodiment, wherein (a) is a polypyrrole and (b) is a polythiophene.

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Figure 5(a) illustrates a graph obtained from the table of Figure 5(b) showing performance of the preferred embodiment.

DETAILED DESCRIPTION OF EMBODIMENTS

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Described below are preferred embodiments of the present invention with reference to the accompanying drawings. Each of the following preferred embodiments describes an example not limiting in any aspect.

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Referring to Figure 1, there is illustrated a graphical representation of a cross-sectional view showing compositional layers of the preferred embodiment, wherein the preferred embodiment is a sensor device without membrane for sensing nitrate.

The sensor device for sensing nitrate comprising a substrate (8), a first layer (11) for transferring signals, a second layer (12) for converting energy, and a nitrate sensitive material (13) formed on the second layer (12). The sensor device is characterized in that the second layer (12) comprising a transducer layer which is grown on the first layer (11)

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comprising a conductive layer, and that the nitrate sensitive material (13) is grown on the second layer (12) of the transducer layer. The transfer of signals comprises the transfer of electrons and ions.

- 5 The substrate (8) is preferably but not limited to silicon, and that the substrate facilitates seeding for the first layer (11). The first layer (11) which is defined as the conductive layer of the preferred embodiment, is preferably a layer of microelectrode arrays, such that the microelectrode arrays contain multiple connectivity means for obtaining and delivering signals through electrons and ions transfer.

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Preferably, the microelectrode arrays are made of tungsten and that wherein the microelectrode array is at least 0.2 micrometer in width with 0.2 micrometer spacing between the said microelectrodes.

- 15 The second layer (12) which is defined as the transducer layer is preferably but not limited to an electrochemical transducer layer comprising at least one or in a combination of doped polypyrrole and doped polythiophene, and comprising dopants of at least one or in any combination of an inorganic chloride, organic chloride tetrafluoroborate, iodide, para-toluene sulfonate, trifluoromethane sulfonate, camphor sulfonate, poly styrene sulfonate, 20 nafion, hexafluorophosphate.

- The nitrate sensitive material (13), which is grown on the second layer (12) of the transducer layer comprises a conductive polymer with at least 0.5% weight of dopant, and the conductive polymer comprises at least one or in any combination of polypyrrole, 25 polythiophene, polyaniline, polyacetylene, polyphenylene, poly(phenylene sulphide) and poly (phenyl-quinoline).

- Referring to Figure 1 and Figure 2, Figure 2 illustrates a flow chart representing the process for preparing the device of the preferred embodiment, which is the sensor device as shown 30 in Figure 1.

The process for preparing the sensor device comprises the steps of initially fabricating a conductive layer on a substrate, and then growing a transducer layer on the conductive layer. Subsequently, a mixed composition from materials sensitive to nitrate is prepared and

- 5 -

dispensed on the transducer layer, and that a layer of the mixed composition is cured on the transducer layer. Then, a nitrate sensitive layer (13) responsive to nitrate is formed on the transducer layer.

- 5 The nitrate sensitive material (13) is preferably prepared using electrochemical deposition in a monomer solution having at least 0.001M of the said nitrate salt.

The substrate (8) is preferably but not limited to silicon, and that the substrate facilitates seeding for the conductive layer of the preferred embodiment. The conductive layer is
10 preferably a layer of microelectrode arrays, such that the microelectrode arrays contain multiple connectivity means for obtaining and delivering signals through electrons and ions transfer.

Preferably, the microelectrode arrays are made of tungsten and that wherein the
15 microelectrode array is at least 0.2 micrometer in width with 0.2 micrometer spacing between the said microelectrodes.

The transducer layer is preferably but not limited to an electrochemical transducer layer comprising at least one or in a combination of doped polypyrrole and doped polythiophene,
20 and comprising dopants of at least one or in any combination of an inorganic chloride, organic chloride tetrafluoroborate, iodide, para-toluene sulfonate, trifluoromethane sulfonate, camphor sulfonate, poly styrene sulfonate, nafion, hexafluorophosphate.

The nitrate sensitive material (13), which is grown on the transducer layer that comprises a
25 conductive polymer with at least 0.5% weight of dopant, and the conductive polymer comprises at least one or in any combination of polypyrrole, polythiophene, polyaniline, polyacetylene, polyphenylene, poly(phenylene sulphide) and poly (phenyl-quinoline).

Referring to Figure 3 and Figure 4, Figure 3 illustrates a graphical representation of a cross-sectional view showing compositional layers of an alternative embodiment, with examples
30 of conductive polymers shown in Figure 4.

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The sensor device as shown in Figure 3 preferably comprising a substrate (18), a conductor layer (21), a carbon layer (22), an electrochemical transducer layer (23), and a nitrate sensing layer (24).

- 5 The transducer layer (23) is preferably but not limited to an electrochemical transducer layer comprising at least one or in a combination of doped polypyrrole and doped polythiophene, and that any one of the doped polypyrrole and doped polythiophene is at least one or in any combination of an inorganic chloride, organic chloride tetrafluoroborate, iodide, para-toluene sulfonate, trifluoromethane sulfonate, camphor sulfonate, poly styrene
10 sulfonate, nafion, hexafluorophosphate.

The transducer layer (23), which is generally used as a conductive polymer layer or layers of electrochemical sensors, the transducer layer (23) must necessarily first doped using a dopant to increase conductivity value. Dopant used is usually a salt or materials that have a
15 charge such as inorganic salts or organic materials charged.

The nitrate sensing layer (24) preferably comprises a conductive polymer, and that the conductive polymer comprises at least one or in any combination of polypyrrole, polythiophene, polyaniline, polyacetylene, polyphenylene, poly(phenylene sulphide) and
20 poly (phenyl-quinoline), and comprising dopants of inorganic nitrate salts and organic nitrate. Also, the nitrate sensing layer (24) comprising solvents of water, ethanol, methanol, isopropanol, n-propanol and n-butanol.

The nitrate sensing layer (24) used is preferably but not limited to conductive polymer
25 doped with nitrate salts such as KNO_3 or NaNO_3 , and that the purpose of the use of nitrate salts in addition serves to increase the conductivity of conductive polymers, but also nitrate ions contained in this conductive polymer will serve to catch and hold the nitrate ions contained in the sample above the surface of the electrode to detect. Interaction between two similar ions would be more easily to occur when compared to different species, and
30 that the conductive polymer doped with nitrate-based salts can therefore function as a nitrate sensor and also has positive levels of selectivity against other ion versus nitrate ion.

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The sensor device can be developed with two layers of conductive polymers, and that the conductive polymers are conjugated polymers that show changes in single and double bonds between carbon atoms in the polymer main chain. The carbon double bonds have four valence electrons, but in the conjugated molecules, the carbon bonds have only two or three other atoms. The remaining electrons form a π bond, and that the electrons are delocalized on the whole molecule. A substance can be conductive polymer if it has a conjugated double bond. Examples of the conductive polymer include polypyrrole and polythiophene as shown in Figure 4.

Preparation of conductive polymers can be suggestively made by electrochemical processes or by chemical processes. Electrochemical process is usually done by using a three electrode cell of namely the reference electrode, working electrode and counter electrode.

The three electrodes are inserted into a solution of monomers that contain adequate dopant and given the current and certain polymerization time or by using cyclic voltammetry on scanning area and a certain number of cycles, depending on the type of conductive polymer, dopant, concentration, electrode surface area and also the desired thickness. The preparation by using a chemical process is usually done by adding oxidising agents. A response slope of close to -59 millivolt per decade can be achieved with correlation coefficient of greater than 0.99 according to the preferred embodiment.

The method of preparing the membraneless nitrate sensor with cast doped polythiophene nanocomposite transducer and polypyrrole doped nitrate comprising the steps of initially depositing carbon on electrode, then preparing electrochemical transducer materials, depositing electrochemical transducer material on carbon electrode surface, and preparing nitrate sensing material, depositing nitrate sensing material on electrochemical transducer layer, and characterizing membraneless nitrate sensor response.

The method of preparing the membraneless nitrate, wherein the step of preparing the electrochemical transducer layer is by electrochemical process and chemical process, the step of preparing the nitrate sensing layer is by electrochemical process and chemical process, and that the membranes nitrate sensor is used in precision agriculture, food sensors and medical sensors.

The present invention is further described but not limited to the following examples.

Example 1

5 Preparation of Screen Printed Carbon Electrode

Carbon paste is type of screen printed on copper-gold or prefabricated screen printed silver. The optimized dry thickness of both screen printed carbon and silver are about 100 micrometer.

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The circular shaped electrodes with 3mm diameter is printed on polyester or printed circuit board substrate and separated by 2 mm spacing from each other for integrated multi-sensor application, and the printed paste is cured at 120 °C to give the desired dry thickness. Solder mask insulating layer was also screen printed to define the electrode window, separate the wells and to protect the printed conducting wires.

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Example 2

Membraneless Nitrate Sensor

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Membraneless nitrate sensor composition was prepared by mixing 1 M potassium nitrate (KNO_3) and 0.5 M monomer pyrrole (Py), the Screen printed electrodes (SPE) with 4 mm diameter were cleaned ultrasonically with deionised water for 1 min, and 2 μL Polythiophes cocktail was coated on SPE and dried under N_2 flow at 30 minutes.

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Homogeneous solution of monomers in electropolymerization of pyrrole doped nitrate on polythiophene transducer surface using a method of cyclic voltammetry, where a platinum electrode (Pt) as counter electrode, the electrode double-junction Ag / AgCl as reference electrode and transducer polythiophene as the working electrode.

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Electropolymerization run with a scan rate 0.1 V / sec in the area of -1 V to 1 V in 5 cycles, and this membraneless nitrate sensor was tested using commercial Ag/AgCl double junction reference electrode with 0.1M LiOAc as outer solution. The results are shown in

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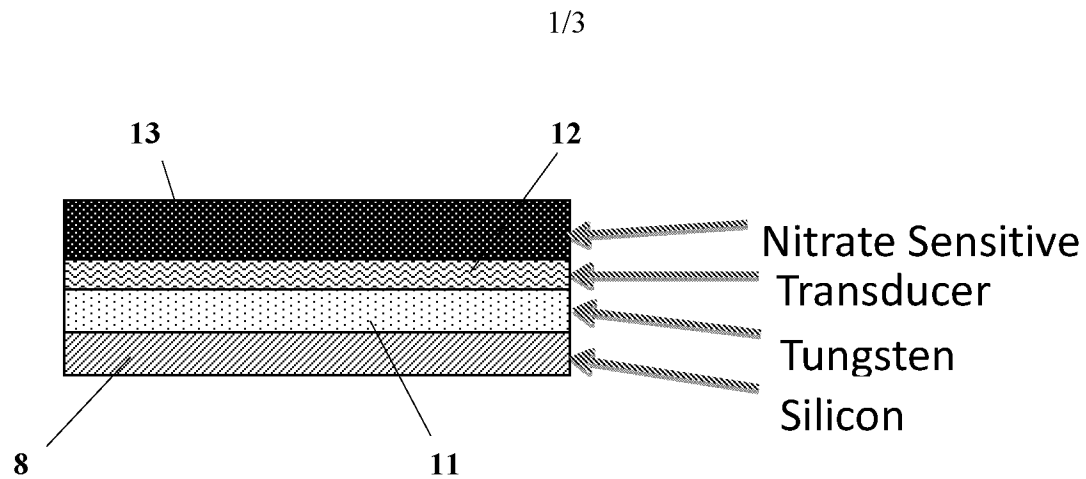
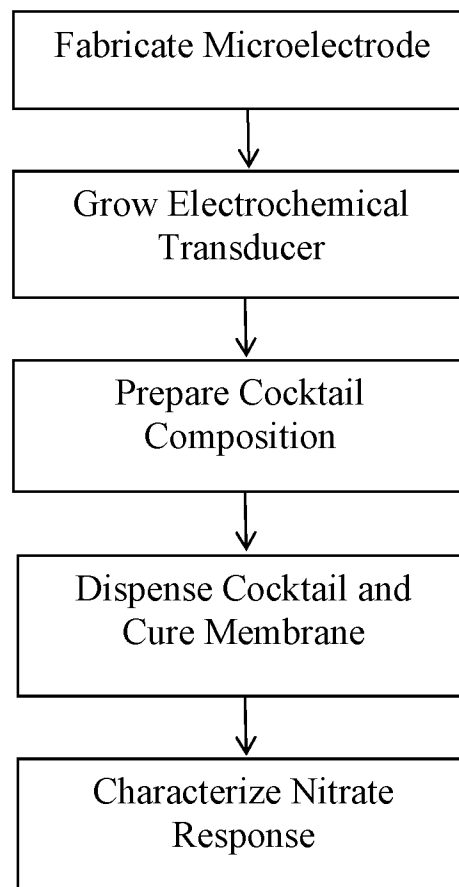
Table 1 of Figure 5(b) and plotted in Figure 5(a), and that the plot of emf response versus activity of nitrate ion shows good Nernstian response and good linearity.

5 In as much as the present invention is subject to many variations, modifications and changes in detail, it is intended that all matter contained in the foregoing description or shown in the accompanying drawings shall be interpreted as illustrative and not in a limiting sense.

CLAIMS

1. A sensor device comprising:
 - a substrate (8);
 - 5 a first layer (11) for transferring signals;
 - a second layer (12) for converting energy into signals; and
 - a nitrate sensitive material (13) formed on the second layer;
 - characterized in that the second layer (12) comprising a transducer layer which
 - is grown on the first layer (11) comprising a conductive layer, and that the nitrate
 - 10 sensitive material (13) is grown on the second layer (12) of the transducer layer.
2. A sensor device according to claim 1, wherein the substrate (8) is a silicon substrate for seeding the first layer (11).
- 15 3. A sensor device according to claim 1, wherein the conductive layer is a layer of microelectrode array.
4. A sensor device according to claim 3, wherein the microelectrode array is a layer of tungsten material.
- 20 5. A sensor device according to claim 1, wherein the transducer layer is an electrochemical transducer layer comprising at least one or in a combination of doped polypyrrole and doped polythiophene.
- 25 6. A sensor device according to claim 5, wherein the transducer layer comprising dopants of at least one or in any combination of an inorganic chloride, organic chloride tetrafluoroborate, iodide, para-toluene sulfonate, trifluoromethane sulfonate, camphor sulfonate, poly styrene sulfonate, nafion, hexafluorophosphate.
- 30 7. A sensor device according to claim 1, wherein the nitrate sensitive material (13) comprises a conductive polymer with at least 0.05% weight of dopant.
8. A sensor device according to claim 7, wherein the conductive polymer comprises at least one or in any combination of polypyrrole, polythiophene, polyaniline, polyacetylene, polyphenylene, poly(phenylene sulphide) and poly (phenyl-quinoline).
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9. A method for preparing the sensor device of claim 1 comprising the steps of:
- fabricating a conductive layer;
 - growing a transducer layer on the conductive layer;
 - 5 preparing a mixed composition from materials sensitive to nitrate;
 - dispensing the mixed composition on the transducer layer;
 - curing a layer of the mixed composition on the transducer layer; and
 - forming a layer of nitrate sensitive material (13) responsive to nitrate;
- 10 10. A method according to claim 9, wherein the mixed composition comprises conductive polymer with at least 0.05% weight of dopant, and that the conductive polymer comprises at least one or combination of polypyrrole, polythiophene, polyaniline, polyacetylene, polyphenylene, poly(phenylene sulphide) and poly(phenyl-quinoline).
- 15 11. A method according to claim 9, wherein the nitrate sensitive material (13) is prepared using electrochemical deposition in a monomer solution having at least 0.001M of the said nitrate salt.

**Figure 1****Figure 2**

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Membraneless Nitrate Sensor

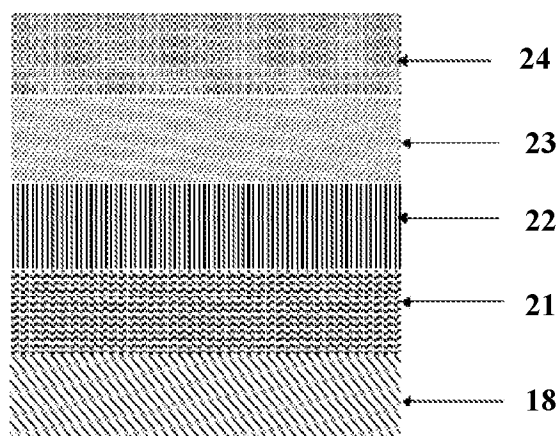
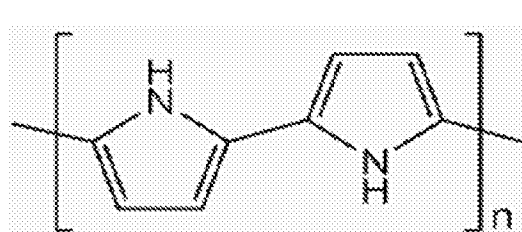
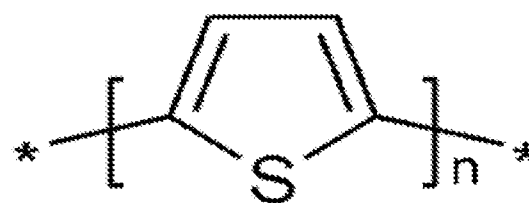


Figure 3



(a)



(b)

Figure 4

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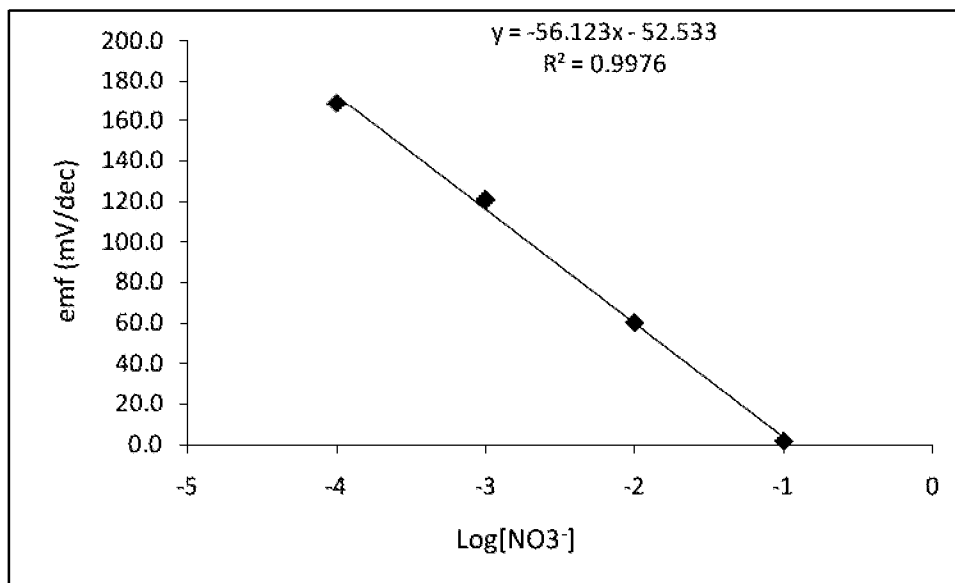


Figure 5(a)

Log [NO ₃ ⁻]	mV 1	mV 2	mV 3	AVG
-1	1.6	1.7	1.8	1.7
-2	59.9	60.1	60.4	60.1
-3	120.6	120.6	120.8	120.7
-4	169.2	168.5	168.1	168.6
M				-56.123
C				-52.533
r²				0.9976

Figure 5(b)

INTERNATIONAL SEARCH REPORT

 International application No.
PCT/MY2015/050010

A. CLASSIFICATION OF SUBJECT MATTER

G01N 27/26 (2006.01)

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)

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 practicable, search terms used)

Databases: WPIAP, EPODOC, INSPEC & keywords: sensor, detector, monitor, measure, nitrate, nitrite, transducer, piezoelectric, electrochemical, electrode, contact, conducting-polymer and similar terms.

Espacenet: BERHAD, Mimos & keywords: nitrate, nitrite, sensor, detector, electrochemical and similar terms.

Google search & keywords: electro-chemical, sensor, detector, conducting polymer, nitrate, nitrite and similar terms.

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
	Documents are listed in the continuation of Box C	



Further documents are listed in the continuation of Box C



See patent family annex

* "A"	Special categories of cited documents: document defining the general state of the art which is not considered to be of particular relevance	"T"	later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"E"	earlier application or patent but published on or after the international filing date	"X"	document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"L"	document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)	"Y"	document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"O"	document referring to an oral disclosure, use, exhibition or other means	"&"	document member of the same patent family
"P"	document published prior to the international filing date but later than the priority date claimed		
Date of the actual completion of the international search 11 June 2015		Date of mailing of the international search report 11 June 2015	
Name and mailing address of the ISA/AU AUSTRALIAN PATENT OFFICE PO BOX 200, WODEN ACT 2606, AUSTRALIA Email address: pct@ipaustalia.gov.au		Authorised officer Jayati Ray AUSTRALIAN PATENT OFFICE (ISO 9001 Quality Certified Service) Telephone No. 0262223654	

INTERNATIONAL SEARCH REPORT		International application No.
C (Continuation). DOCUMENTS CONSIDERED TO BE RELEVANT		PCT/MY2015/050010
Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	WO 2012/067490 A1 (MIMOS BERHAD) 24 May 2012 Fig. 1 and the associated text	1-6, 9
X	Lu J. et al., "Polyaniline nanoparticle–carbon nanotube hybrid network vapour sensors with switchable chemo-electrical polarity", Nanotechnology, 21, 28 May 2010, 255501 pages 1-10. PaniNP-MWNT sensor, see abstract, item 2, Experimental details	1-11

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INTERNATIONAL SEARCH REPORT Information on patent family members		International application No. PCT/MY2015/050010	
This Annex lists known patent family members relating to the patent documents cited in the above-mentioned international search report. The Australian Patent Office is in no way liable for these particulars which are merely given for the purpose of information.			
Patent Document/s Cited in Search Report		Patent Family Member/s	
Publication Number	Publication Date	Publication Number	Publication Date
WO 2012/067490 A1	24 May 2012		
End of Annex			
Due to data integration issues this family listing may not include 10 digit Australian applications filed since May 2001. Form PCT/ISA/210 (Family Annex)(July 2009)			