Abstract: The present invention provides a planar reference electrode (10) comprising a substrate (20); an electrode connecting part (24); an electrode (22); an internal reference layer (23); a micro porous polymer membrane (38) which functions as both a junction and a protection membrane layer made of micro-porous organic polymeric membrane, characterised in that the internal reference layer further comprises; a hydrophilic bulk (26) made of bulk hydrogel layer containing organic internal electrolytes; a photo hydrophilic (28) layer conditioned with internal electrolyte; a hybrid solgel (30) which functions as junction between internal electrolyte and a bridge salts; a hydrophobic bulk (32) which is bulk hydrogel containing the bridge salts; a photo hydrophobic (34) which is conditioned with the bridge salts; a hybrid solgel (36) which functions as junction between external analyte and the bridge salts, wherein the at least two layers of hydrophilic hydrogel consisting of an electrolyte preferably ionic liquid selected from the group of hydrophilic chloride.
FIELD OF THE INVENTION

The present invention relates to a planar reference electrode and method of fabrication thereof.

BACKGROUND OF THE INVENTION

Electrochemical methods such as potentiometry or voltammetry have been commonly applied in the analytical laboratories to determine specific chemical components contained in clinical, environmental or industrial samples. However, the samples collected from remote sampling sites often require special pretreatment to prevent them from contamination or degradation during transportation to the laboratories. The laboratory-based instruments is expensive to operate, and should be managed by the technicians specially trained in the area.

Chemical sensor is current used for rapid in-situ analysis of soil macronutrients in the field of precision agriculture and environmental monitoring of ionic pollutants. Ion selective sensor membrane coupled to a reference electrode, gives voltage closed to the ideal Nernstian value. The flow of current in the circuit is complete and electrons flow in the conductive wires to form passage of current in the analyte. In this approach stability and lifetime of the reference electrode is as important as the sensitivity and selectivity of the sensing membrane. Conventional double-junction reference electrode is too bulky for field deployment and contains liquids electrolyte that gradually leaks and prone to contamination from the external ion. Weekly maintenance that includes thorough cleaning, drying and replacement of electrolyte is necessary to ensure reliable measurement and this is not possible for in-situ measurement in soils and fluids.
Several types of miniaturized reference electrodes have been proposed previously. US 6,964,734 disclosed a method for fabricating planar reference electrodes. Particularly, the present invention relates to the planar reference electrode comprising plate; electrode connection; electrode; insulating membrane; inner reference solution; junction; and protecting membrane, and processes for fabrication thereof, in which the junction is composed of porous material such as cotton thread, glass fiber, cellulose nitrate, cellulose acetate, filter paper and any material that can produce capillary action; porous polymer membrane; or a capillary either directly printed on the substrate or inserted with a thin film.

Another publication WO 05119235 disclosed a planar electrochemical sensor with membrane coatings used to perform chemical analyses. The object of this invention is to provide unit-use disposable sensors of very simple and inexpensive construction, preferably with only a single membrane coating on an electrode. The invented devices are potentiometric salt-bridge reference electrodes and dissolved gas sensors constructed with a heterogeneous membrane coating of a conductor. The heterogeneous membrane, which is an intimate admixture of a hydrophobic and a hydrophilic compartment, concurrently supports constrained transport of non-volatile species through its hydrophilic compartment and rapid gas and water vapor transport through its hydrophobic compartment.

Known double junction liquid-electrolyte reference electrode is too bulky and prone to leaking and contamination, and therefore not suitable for field deployment.

Hence, planar durable maintenance-free reference electrode is needed for integrated chemical sensor array to be used in harsh environment. The present invention proposes miniaturized planar reference electrode based of moisture-retaining hydrogels whereby the internal reference electrolytes are novel hydrophilic organic chlorides. Our results show that the proposed planar reference electrode can be manufactured in a few fabrication steps,
and it can be employed in potentiometric chemical sensors with good
stability and prolonged lifetime.

5 SUMMARY OF THE INVENTION

The present invention relates to a durable planar reference electrode based
on hydrophilic organic chloride. A planar reference electrode comprising: a
substrate; an electrode connecting part; an electrode; an insulating
membrane; an internal reference layer; a micro porous polymer membrane
which functions as both a junction and a protection membrane, wherein
porous polymer is dissolved in the organic solvent to form porous polymer
solution and the porous polymer solution is dispensed onto the internal
reference layer to form the porous polymer membrane, wherein the
substrate is formed of a material partially soluble in the organic solvent to
make the porous polymer membrane directly fixed to the substrate,
characterised in that the internal reference layer is at least two layers of
hydrophilic hydrogel consisting of an electrolyte preferably ionic liquids
selected from the group of hydrophilic organic chloride.

BRIEF DESCRIPTION OF THE DRAWINGS

Fig.1 illustrates layers of a preferred embodiment of planar electrode.
Fig.2 illustrates exemplified test on Ag/AgCl screen printed electrode (SPE)
response Cl⁻ test with standard of Ag/AgCl double junction reference
electrode based on Agar gel.
Fig.3 illustrates exemplified test on Ag/AgCl screen printed electrode (SPE)
with Potassium Ion Selective commercially available from Nico 2000.
Fig.4 illustrates exemplified test on Ag/AgCl screen printed electrode (SPE)
with Nitrate Ion Selective commercially available from Nico 2000.
Fig.5 illustrates exemplified test on Ag/AgCl Disc Electrode response Cl⁻ test with standard of Ag/AgCl double junction reference electrode based on Agar gel.

Fig.6 illustrates exemplified test on Ag/AgCl Disc Electrode with Potassium Ion Selective commercially available from Nico 2000.

Fig.7 illustrates exemplified test on Ag/AgCl Disc Electrode with Nitrate Ion Selective commercially available from Nico 2000.

DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENT

The proposed durable reference electrode is fabricated using reproducible planar process on silver-silver chloride electrode. The electrode layer can be fabricated by chlorination of silver foil, thick film screen printing and composite materials.

The silver-silver chloride layer can be deposited on many common substrates such as polyester, polycarbonate, ceramic, FR4 and silicon.

The polymeric media can be polyHEMA, acrylamide gel, polyvinyl alcohol, polyvinyl pyrrolidone and agar. There are two hydrophilic gel layers; internal layer contains low concentration of hydrophilic organic chloride reference electrolyte and external layer contains salts containing pation and anion of similar size and mobility such as lithium acetate or potassium chloride.

The polymeric gel layers are protected against swelling and delamination by a thin hydrophobic membrane such as Nafion17, TEOS sol-gel and blends of cellulose nitrate and cellulose acetate.

The internal reference electrolyte contains low concentration of novel hydrophilic organic chlorides of formula 1, 2, 3 and 4.
The construction of layers of the durable planar reference electrode 10 is as illustrated in Fig. 1. Substrate 20 may be selected from polyester, polycarbonate, FR4 PCB material, flex, ceramic or silicon. Layer 22 is silver-silver chloride electrode from chemical or electrochemical chlorination of metallic or screen printed silver. Electrical connection 24 provides interface to sensor readout circuit. Hydrophilic bulk 26 is bulk hydrogel layer containing proposed organic chloride internal electrolytes. Photo Hydrophilic 28 is polymerized acrylate monomer conditioned with internal electrolyte. Hybrid Solgel 30 is nano-porous channel functions as junction between internal electrolyte and bridge salts. Hydrophilic bulk 32 is bulk hydrogel containing bridge salts. Photo Hydrophilic 34 is polymerized acrylate monomer conditioned with bridge salts. Hybrid Solgel 36 is nano-porous channel functions as junction between external analyte and bridge salts. The top most layer 38 is physical protection layer made of micro-porous organic polymeric membrane.
Example 1

Preparation of Planar Reference Electrode based on Organic Chloride Reference Electrolyte

i. Reference Electrode on Screen Printed Silver

Silver paste in mixture of organic solvents was printed through fine mesh to form round-shaped electrodes with diameters of 2mm or 3mm and 0.5mm wire and space. The substrate can be polyester, polycarbonate, flex material, printed circuit board, ceramic or silicon wafer. Polyester sheet with 0.3mm thickness and standard A4 size is preferred.

The silver paste was cured at 120 °C under continuous flow of nitrogen blanket. Uniform and reproducible dry thickness of 30 micrometer silver can be manufactured using this method. Solder mask isolation material was then printed repeatedly and oven cured over five times to isolate neighboring electrode wells and passivate silver wires.

The printed electrodes were separated in groups of silver wells in one row, wherein the wells are 4mm apart. Only the wells and the wire pins are exposed for further processing. The silver wells were exposed in 0.1 M Ferric Chloride for 2 minutes under continuous flow of nitrogen blanket. Reproducible quality of silver-silver chloride electrode can be manufactured using this method. The quality of silver chloride is tested based on chloride response in potassium chloride solutions versus conventional reference electrode.

Hydrophilic gel such as poly hydroxyethyl methacrylate (polyHEMA), poly vinylalcohol or polyvinyl pyrrolidone (5 µL) containing 0.1 M to 3M of hydrophilic organic chloride was coated onto screen printed silver-silver chloride electrode surface and dried under vigorous flow of nitrogen for 30 minutes. Additional photocured layer of HEMA monomer conditioned with 0.1 M of potassium chloride on top of the hydrophilic gel is an option.
Porous glass layer formed by solgel cocktail of tetraethoxysilicon separates the reference electrolyte and the external electrolyte. Hydrophilic gel such as poly hydroxyethyl methacrylate (polyHEMA), poly vinylalcohol or polyvinyl pyrrolidone (5 µL) containing 0.1 M of salts containing cation and anion of similar sizes and mobility such as lithium acetate or potassium chloride was coated onto the porous separator layer and dried for 30 minutes. Additional photocured layer of HEMA monomer conditioned with 0.1 M of lithium acetate on top of the second hydrophilic gel is an option.

The internal, porous and external layers are protected against physical damage by micro porous organic membrane. Cellulose acetate or cellulose nitrate 5-20% by weight in dioxane-water or tetrahydrofuran (5 µL) was coated on the external layer and dried under vigorous flow of nitrogen blanket. The planar reference electrode was conditioned in deionized water for one hour and subjected to performance tests.

**ii. Reference Electrode of Silver Metal Disc**

Commercially available silver foil, 99.99% and 0.25mm thickness was cut by high power laser into round shape with tail for soldering to electrical wiring. The discs are 2mm or 3mm in diameter and upon wire soldering, was glued to printed circuit board. Then black epoxy was coated to form circular dam of 0.3mm high and oven cured at 120 0C.

The silver surface was washed with detergent and rinsed with deionized water. Then soft sand paper was used to remove remaining oxide on the silver surface, followed by rinsing with deionized water and drying with vigorous blow of nitrogen.

Ferric chloride (0.1 M, 1 drop) was dropped into the clean silver surface and left for chlorination of silver or 20 seconds. Reproducible silver-silver chloride electrode can be manufactured in this manner. Coating of
internal, porous, external and top-most protection layer can be repeated in the same manner described earlier for screen printed electrode. The planar reference electrode was conditioned in deionized water for at least one hour and subjected to performance tests.

Example 2
Method of fabrication

i. Using Ag/AgCl Screen Printed Electrode (SPE)

5 µL Hydrophilic gel contains of hydrophilic organic chloride coated onto Ag/AgCl screen printed electrode surface and dried for 30 min. 5 µL Hydrophilic gel contains of salts containing cation and anion of similar size and mobility such as lithium acetate or potassium chloride coated onto hydrophilic organic chloride layer and dried for 30 min. 5 µL protection layer coated onto hydrophilic gel contains of salt such as lithium acetate or potassium chloride and dried for 10 min.

ii. Using Ag Disc Electrode

Ag Disc Electrode was chlorinated with ferric chloride (FeCl₃) 1 M for 20 seconds. After chlorination, rinsing with deionized water and drying with nitrogen flow, silver-silver chloride electrode is ready for membrane application. Hydrophilic gel (5 µL) containing hydrophilic organic chloride was coated onto the silver-silver chloride layer of disc electrode surface and dried under copious flow of nitrogen for 30 min. Hydrophilic gel (5 µL) containing bridge salts of similar cation and anion size and mobility such as lithium acetate or potassium chloride was coated onto hydrophilic organic chloride layer and dried with flow of nitrogen for 30 minutes. 5 µL Cocktail of cellulose acetate or cellulose nitrate in tetrahydrofuran or dioxane-water protection layer was coated onto the dried hydrophilic gel containing lithium acetate or potassium chloride bridge salt.
Example 3

Test on the performance of the reference electrode
Using Ag/AgCl screen printed electrode (SPE)

The performance and stability of planar reference electrode was tested for the response of chloride ion (Cl⁻) in a wide range of potassium chloride solution versus conventional double-junction reference electrode.

Table 1 shows that the planar reference electrode in the current invention shows good stability over a wide range of concentrations. The stability of chloride response from $10^{-1}$ to $10^{-5}$ for the intended application in Precision Agriculture is very good.

<table>
<thead>
<tr>
<th>Log [Cl⁻]</th>
<th>mV</th>
</tr>
</thead>
<tbody>
<tr>
<td>-1</td>
<td>20.6</td>
</tr>
<tr>
<td>-2</td>
<td>18.7</td>
</tr>
<tr>
<td>-3</td>
<td>22.3</td>
</tr>
<tr>
<td>-4</td>
<td>26.3</td>
</tr>
<tr>
<td>-5</td>
<td>12.4</td>
</tr>
<tr>
<td>ΔmV</td>
<td>13.9</td>
</tr>
</tbody>
</table>

Table 1

Example 4

The stability of the fabricated planar reference electrode was also tested with commercial ion-selective electrode (ISE) chemical sensors. The response of potassium ion was studied using Nico 2000 potassium sensor versus the fabricated planar reference electrode.

<table>
<thead>
<tr>
<th>Log [K⁺]</th>
<th>mV</th>
</tr>
</thead>
<tbody>
<tr>
<td>-1</td>
<td>411.1</td>
</tr>
<tr>
<td>-2</td>
<td>364.5</td>
</tr>
<tr>
<td>-3</td>
<td>309.6</td>
</tr>
<tr>
<td>-4</td>
<td>248.7</td>
</tr>
<tr>
<td>-5</td>
<td>197.3</td>
</tr>
</tbody>
</table>

Table 2
Fig. 3 illustrates exemplified test on silver-silver chloride screen printed electrode (SPE) with Potassium Ion Selective Electrode, commercially available from Nico 2000. A comparison test was conducted for potassium response of commercial potassium sensor (Nico 2000) versus planar reference electrode based on organic chloride electrolyte in potassium chloride solutions. The planar reference electrode containing hydrophilic chloride reference electrolyte has been used as reference electrode in potentiometric measurement of potassium ion. Using commercial potassium sensor with known potentiometric response based on conventional reference electrode, the planar reference electrode has shown similar potassium response, reproducing the expected slope and linearity.

Example 5

The stability of the fabricated planar reference electrode was also tested with anion commercial ion-selective electrode (ISE) chemical sensor. The response of nitrate ion was studied using Nico 2000 nitrate sensor versus the fabricated planar reference electrode.

<table>
<thead>
<tr>
<th>Log [NO₃⁻]</th>
<th>mV</th>
</tr>
</thead>
<tbody>
<tr>
<td>-1</td>
<td>147.7</td>
</tr>
<tr>
<td>-2</td>
<td>195.8</td>
</tr>
<tr>
<td>-3</td>
<td>250.3</td>
</tr>
<tr>
<td>-4</td>
<td>318.1</td>
</tr>
<tr>
<td>-5</td>
<td>361.7</td>
</tr>
</tbody>
</table>

Table 3

Fig. 4 illustrates exemplified test on silver-silver chloride screen printed electrode (SPE) with Nitrate Ion Selective Electrode commercially available from Nico 2000. Nitrate response of commercial nitrate (Nico) sensor versus planar reference electrode based on organic chloride electrolyte in potassium nitrate solutions. The planar reference electrode containing hydrophilic chloride reference electrolyte has been used as reference.
electrode in potentiometric measurement of nitrate ion as well. Using commercial nitrate sensor with known potentiometric response based on conventional reference electrode, the planar reference electrode has shown similar nitrate response, reproducing the expected slope and linearity.

Example 6
Using Silver-Silver Chloride (Ag/AgCl) Disc Electrode

Response of chloride ion (Cl\(^-\)) with conventional double-junction reference electrode as illustrated in Fig. 5.

<table>
<thead>
<tr>
<th>Log [Cl(^-)]</th>
<th>mV</th>
</tr>
</thead>
<tbody>
<tr>
<td>-1</td>
<td>31.6</td>
</tr>
<tr>
<td>-2</td>
<td>30</td>
</tr>
<tr>
<td>-3</td>
<td>22.4</td>
</tr>
<tr>
<td>-4</td>
<td>33.4</td>
</tr>
<tr>
<td>-5</td>
<td>44.6</td>
</tr>
<tr>
<td>(\Delta mV)</td>
<td>22.2</td>
</tr>
</tbody>
</table>

Table 4

Example 7

Response of potassium ion using commercially available potassium Ion Selective Electrode from Nico 2000 versus the fabricated planar reference electrode as illustrated in Fig. 6.

<table>
<thead>
<tr>
<th>Log [K(^+)]</th>
<th>mV</th>
</tr>
</thead>
<tbody>
<tr>
<td>-1</td>
<td>436.1</td>
</tr>
<tr>
<td>-2</td>
<td>380.3</td>
</tr>
<tr>
<td>-3</td>
<td>333.0</td>
</tr>
<tr>
<td>-4</td>
<td>260.5</td>
</tr>
<tr>
<td>-5</td>
<td>239.7</td>
</tr>
</tbody>
</table>

Table 5
Example 8

Response of nitrate ion using commercially available nitrate ion selective electrode from Nico 2000 versus the fabricated planar reference electrode as illustrated in Fig. 7.

<table>
<thead>
<tr>
<th>Log [NO₃⁻]</th>
<th>mV</th>
</tr>
</thead>
<tbody>
<tr>
<td>-1</td>
<td>215.0</td>
</tr>
<tr>
<td>-2</td>
<td>269.2</td>
</tr>
<tr>
<td>-3</td>
<td>334.7</td>
</tr>
<tr>
<td>-4</td>
<td>398.1</td>
</tr>
</tbody>
</table>

Table 6

Example 9

Preparation of Hydrophilic Organic Chlorides Reference Electrolytes
i. Preparation of Hydrophilic Polyhydroxyl Ammonium Chloride 1

\[
\text{H}_3\text{N} - \text{OH} \quad \text{Cl} - \text{OH} \quad \text{H}_3\text{N} - \text{OH} \quad \text{Cl}^-
\]

A 100-mL three-necked, round-bottomed flask, equipped with thermometer, nitrogen inlet, addition funnel, magnetic stirrer and reflux condenser was flushed with argon and charged with 40 ml of anhydrous dichloromethane. Freshly distilled tris(hydromethyl)aminomethane (i, 0.01 mol) was added into the flask. The flask was chilled with ice-water bath and 3-chloro-1-propanol (0.011 mol) in 30 mL of anhydrous dichloromethane was added cautiously into the flask with continuous stirring over 40 minutes. The solution was heated under reflux for 20
hours and then allowed to cool to room temperature. The dichloromethane solvent was removed under vacuum distillation to afford 81\% of polyhydroxyl ammonium chloride 1.

**m. Preparation of Hydrophilic Ammonium Chloride of formula 2**

![Chemical Structure]

A 50-mL three-necked, round-bottomed flask, equipped with thermometer, nitrogen inlet, addition funnel, magnetic stirrer and reflux condenser was flushed with argon and charged with 30 mL of anhydrous dichloromethane. Freshly distilled triethanolamine (Mf 1.49g, 0.01 mol) was added into the flask. The flask was chilled with ice-water bath and 3-chloro-1-propanol (1.04g, 0.011 mol) in 10 mL of anhydrous dichloromethane was added cautiously into the flask with continuous stirring over 20 minutes. The solution was heated under reflux for 14 hours and then allowed to cool to room temperature. The dichloromethane solvent was removed under vacuum distillation through Vigreaux column to 87\% of polyhydroxyl ammonium, chloride of formula 2.
iii. Preparation of Hydrophilic 1-Methyl-3-Propanol Imidazolium Chloride of formula 3

A 100-mL three-necked, round-bottomed flask, equipped with thermometer, nitrogen inlet, addition funnel, magnetic stirrer and reflux condenser was flushed with argon and charged with 40 mL of anhydrous dichloromethane. Freshly distilled 1-methyl imidazole (iii, 0.01 mol) was added into the flask. The flask was chilled with ice-water bath and 3-chloro-1-propanol (1.04g, 0.011 mol) in 30 mL of anhydrous dichloromethane was added cautiously into the flask with continuous stirring over 40 minutes. The solution was heated under reflux for 20 hours and then allowed to cool to room temperature. The dichloromethane solvent was removed under vacuum distillation to afford 81% of imidazolium chloride 3.

iv. Preparation of Hydrophilic Imidazolium Chloride of formula 4
A 100-mL three-necked, round-bottomed flask, equipped with thermometer, nitrogen inlet, addition funnel, magnetic stirrer and reflux condenser was flushed with argon and charged with 40 mL of anhydrous dichloromethane. Freshly distilled 4-methyl-5-imidazolemethanol (iv, 0.01 mol) was added into the flask. The flask was chilled with ice-water bath and 3-chloro-1-propanol (1.04g, 0.01 mol) in 30 mL of anhydrous dichloromethane was added cautiously into the flask with continuous stirring over 40 minutes. The solution was heated under reflux for 20 hours and then allowed to cool to room temperature. The dichloromethane solvent was removed under vacuum distillation to afford 81% of imidazolium chloride 4.

Although the preferred embodiments of the present invention have been described herein, the above descriptions are merely illustrative. Further modification of the invention herein disclosed will occur to those skilled in the respective arts and all such modifications are deemed to be within the scope of the invention as defined by the appended claims.
CLAIMS

1. A planar reference electrode 10 comprising:
   a substrate 20;
   an electrode connecting part 24;
   an electrode 22;
   an internal reference layer 23;
   a micro porous polymer membrane 38 which functions as both a junction and a protection membrane layer made of micro-porous organic polymeric membrane, characterised in that the internal reference layer further comprises; a hydrophilic bulk 26 made of bulk hydrogel layer containing organic internal electrolytes; a photo hydrophilic 28 layer conditioned with internal electrolyte; a hybrid solgel 30 which functions as junction between internal electrolyte and a bridge salts; a hydrophilic bulk 32 which is bulk hydrogel containing the bridge salts; a photo hydrophilic 34 which is conditioned with the bridge salts; a hybrid solgel 36 which functions as junction between external analyte and the bridge salts, wherein the at least two layers of hydrophilic hydrogel consisting of an electrolyte preferably ionic liquids selected from the group of hydrophilic chloride.

2. The planar reference electrode 10 according to claim 1, characterised in that the internal reference layer include an electrolyte which is hydrophilic poly hydroxyl organic chloride of formula 1.
3. The planar reference electrode 10 according to claim 1, characterised in that the internal reference layer include an electrolyte which is hydrophilic poly hydroxyl organic chloride of formula 2.

\[ \text{Cl}^- \text{OH} \]
\[ \text{HO-} \text{N}^+ \text{OH} \]
\[ \text{OH} \]

5. The planar reference electrode 10 according to claim 1, characterised in that the internal reference layer include an electrolyte which is hydrophilic imidazolium chloride of formula 3.

\[ \text{Cl}^- \text{OH} \]
\[ \text{H}_3\text{C} \]

10

5. The planar reference electrode 10 according to claim 1, characterised in that the internal reference layer include an electrolyte which hydrophilic imidazolium chloride of formula 4.

\[ \text{H}_3\text{C} \text{Cl}^- \text{OH} \]
\[ \text{HO} \]
\[ \text{H} \]

15

6. The planar reference electrode 10 according to claim 1, characterised in that the micro porous polymer membrane is formed of cellulose nitrate.
7. The planar reference electrode 10 according to claim 1, wherein the substrate is selected from polyester, polycarbonate, FR4 PCB material, flex, ceramic or silicon.

8. The planar reference electrode 10 according to claim 1, characterised in that the electrode is selected from the group consisting of Ag, Pd, Cu, Pt, Au, Ag-AgCl, coated with Nation.

9. A method for fabricating the screen printed electrode (SPE) planar reference electrode of claim 1 which comprises:
   (a) forming the electrode connecting part on the substrate;
   (b) forming the electrode on the plate by using a screen printing method;
   (c) forming the insulating layer by screening printing on the electrode, to provide a well around the electrode;
   (d) placing the internal reference layer within the well; and
   (e) forming the porous protection membrane to cover the internal reference layer, characterised in that the step of forming the porous protection is provided by placing hydrophilic gel containing salts having cation and anion of similar size and mobility.

10. A method for fabricating the silver disc planar reference electrode of claim 1 which comprises:
   (a) forming the electrode connecting part on the substrate;
   (b) forming the electrode on the plate by using chlorination of silver disc method;
   (c) forming the insulating layer by depositing black epoxy surrounding round-shape electrode wells;
   (d) placing the internal reference layer within the well; and
   (e) forming the porous protection membrane to cover the internal reference layer, characterised in that the step of forming the porous
protection is provided by placing hydrophilic gel containing salts having cation and anion of similar size and mobility.

11. The formula 1 in claim 2 characterised in that the polyhydroxyl ammonium chloride 1 is prepared from tris(hydromethyl)aminomethane and 3-chloro-1-propanol in refluxing dichloromethane.

12. The formula 2 in claim 3 characterised in that the polyhydroxyl ammonium chloride 2 is prepared from triethanolamine and 3-chloro-1-propanol in refluxing dichloromethane.

13. The formula 3 in claim 4 characterised in that the 1-methyl-3-propanol imidazolium chloride 3 is prepared from 1-methyl imidazole and 3-chloro-1-propanol in refluxing dichloromethane.

14. The formula 4 in claim 5 characterised in that the imidazolium chloride 4 is prepared from 4-methyl-5-methanol imidazole and 3-chloro-1-propanol in refluxing dichloromethane.
Figure 1

Cl⁻ response SPE vs Ag/AgCl Double junction reference electrode based on Agar gel

Figure 2
**Figure 5**

**Figure 6**

**Figure 6**

\[ y = 49.26x + 481.7 \]

\[ R^2 = 0.9976 \]
Figure 7

\[ y = -61.48x + 150.55 \]

\[ R^2 = 0.9984 \]