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EFFECT OF AC VOLTAGE ON THE PERFORMANCE OF A DISPOSABLE pH SENSOR FOR WOUND CARE

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ABSTRACT

Moisture and pH content in a wound have exhibited to play a significant role in determining the degree and rate of healing of the wound. This enhances wound care by helping the clinicians make treatment decisions and ultimately towards a targeted therapeutic approach to wound management.

A recent breakthrough in this area of healthcare is development of a moisture sensor that measures the moisture content in a wound at different stages during the healing process. The moisture sensor works on the principle of measuring the impedance levels from the exudate which indicates the condition of the wound as being wet, moist, dry etc. This moisture sensor utilizes AC voltages to give the moisture readings.

Another significant sensor has been designed that would measure the pH of the wound. This sensor along with the moisture sensor would provide the content of the two significant parameters that determine the wound conditions as mentioned earlier. The pH sensor is a potentiometric sensor and it might lose its voltage stability due to influence of AC voltages when measuring moisture content in the wound.

The main task of the project is to study the behavior and stability of the pH sensor upon application of AC voltages. This study would determine the efficiency of the pH sensor upon integrating with the moisture sensor and if successful, the combination of both the sensors would greatly enhance the area of wound management. The results obtained from this project provide essential data to proceed with the integration of the two sensors or not.

DISCLAIMER

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CONTENTS

Chapter 1: Introduction

This chapter details the necessary background on the medical condition of wounds and their management.

1.1: General Overview

A wound can be defined as a defect or a rupture in the skin that may occur due to physical, mechanical or thermal damage or also it may be a consequence of underlying medical and physiological disorder [1]. Since the wounds inflict the skin which is called the first line of defence in the human body, it becomes an imperative to discuss the structure and the function of the skin in brief before discussing the impact the wounds have and the extent of damage it causes on a human body.

1.2: Structure and function of the skin

In order to understand the occurrence of wounds, it is essential to know the structure of the skin so that the severity and also the healing of the wounds can be studied effectively.

The skin comprises the mainly of two layers namely the outer epidermis and the inner dermis [2]. The other layers are subdivisions of either of the above two layers and have their own functions to protect the skin from external inflictions. A brief description of the structure and the related function is given below to understand the first line of defence better.

1.2.1: Epidermis

The epidermis forms the outermost layer of the skin and comprises of stratified epithelium [2] [3]. The stratified epithelium further comprises of one to many layers. The epidermis functions mostly as a producer of keratin which is a tough and water insoluble protein. The epidermis is further divided into:

- <u>Stratum corneum (horny layer)</u> – It is the outermost layer of the skin and comprises keratin which as mentioned earlier is a tough and insoluble protein.

This layer is the first point of contact among the other skin layers and it helps in inhibiting effects of variation in external factors such as pH, temperature etc.

- Stratum Lucidum – This is a transparent layer which is mostly found in the areas where the skin is not thin such as the soles of the feet and palms of the hands. It is a layer that provides extra defence since their presence is in those area where maximum wear and tear occur.
- Stratum granulosum – This area is mostly concerned with the process of keratinization.
- Stratum spinosum – This layer is concerned with maintenance of the integrity of the epidermis and comprises of desmosomes.
- Stratum Basale – It forms a well defined layer differentiating the epidermis from the dermis and is mostly a controls blood supply to the epidermis from the dermis because the epidermis is devoid of blood supply.

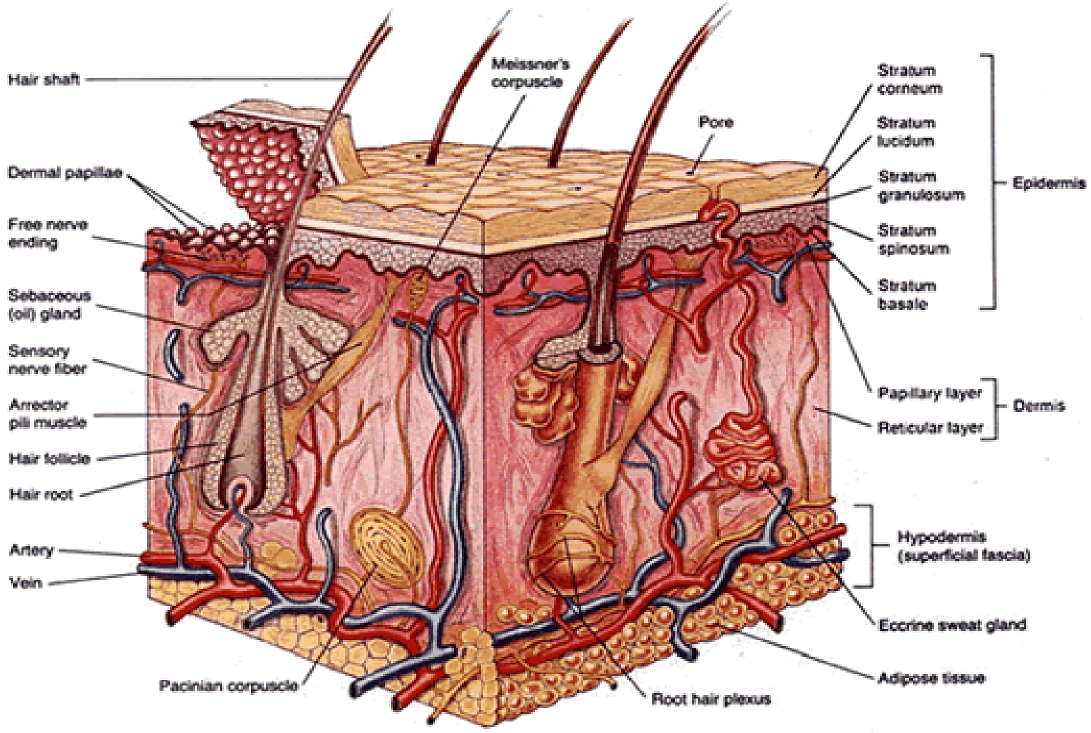

Figure 1.1a: The figure above depicts the detailed structure of the skin with its underlying layers that mostly form the initial points of contact to cause a wound

and the severity of the wound depends on the extent of damaging the underlying layers of the skin.(Source: http://www.lhsc.on.ca/Health_Professionals/Wound_Care/intro/structur.htm)[4]

1.2.2: Dermis

The dermis is the immediate underlying layer to the epidermis[2]. Since the epidermis is devoid of blood supply and nourishment on its own, the dermis helps in providing these essential components to the epidermis [2] [3]. The dermis can be subdivided into two layers namely:

- The papillary layer – also called the stratum papillare. It has a wave-like structure which enables maximum surface contact to the epidermis to provide the essential nourishment. The papillary layer comprises of loose connective tissue, capillaries, elastic fibres and also collagen bundles that are layered.
- The reticular layer – It is comparatively a thicker layer comprising of the layers similar to that of the papillary layer but being much more denser.

The dermis contains the elastic connective tissue which plays a crucial role in making the dermis highly tensile and elastic leading to sustenance of everyday stretching and also protection from mechanical damage.

1.3: Classification of Wounds

The wounds are mainly classified based on their tendency to heal [3]. In this section the wounds are classifies according to their tendency to heal along with the mode of infliction as detailed in the following contents.

1.3.1: Mechanical injuries

They include [2]-

- Abrasions (grazes) generally caused by friction due to rubbing or a tangential contact between the skin and another hard surface. These wounds are normally superficial.
- Lacerations (tears) are more damaging than abrasions and involve both the skin and tissues beneath the skin.
- Wounds caused by intense rupture of skin with sharp or pointed objects which may have a deeper penetrating effect. In such wounds the external appearance may look minor but the damage would be considerably high.
- Acute wounds include burns which may occur as a result of thermal, chemical and also which may occur due to radiation but on the whole thermal injuries are most common. They may be classified into[1]:
    - Superficial burns involve only epidermis and superficial dermis as a consequence of low intensity heat for a long time.
    - Deeper dermal burns in which majority of the epidermis and considerable layers of dermis are damaged.
    - Burns covering the entire skin as a result of extreme thermal impact.

Figure 1.2a: The above picture shows a type of acute wound resulting from an abrasion of a finger.

(Source: http://www.burnsurgery.org/Modules/BurnWound/rationale/acute_wounds/traumatic_wounds.htm)[5]

1.3.2: Chronic Wounds:

Ulcers constitute this type of wounds and are further divided into different types keeping the cause as a factor [2].

They are:

- Decubitus ulcers are usually caused by the prolonged application of surface pressure over a bony presence that blocks the capillary blood flow to the skin and tissues beneath. If the pressure is not removed, it will finally result in cell death and necrosis would follow.
- Leg ulcers, that can be venous, ischaemic or traumatic originally.
- Ulcers linked with certain systemic infections.
- Ulcers initiated from radiotherapy treatment.
- Ulcers due to malignant disease.

Figure 1.3a: A chronic leg ulcer in woman who is suffering from lower leg edema due to decreased mobility[6].

1.4: Wound healing process

Wound healing is a very complex process owing to the large number of biochemical and cellular processes involved [3]. The main mechanisms of healing are briefly discussed here to give an idea of the approach a body takes to repair itself on the occurrence of a wound and also the wound management techniques are mentioned here in brief.

The wound healing mechanism can be classified into:

1.4.1: Primary closure (healing by first intention):

When a wound occurs due to surgical reasons or trauma, they are managed by the primary closure mechanism [3]. The surgeon brings the edges of the wound close enough to individually suture the different layers of tissue together. This causes least granulation of the tissue in the wound and upon successful healing, the process culminates in formation of a scar which eventually becomes diminished as it matures. A brief explanation of the healing process is given below:

Healing is initiated by an acute inflammatory response [2] [3] that begins within a few minutes of the injury and persists up to three days. The platelets exit from the injured damages blood vessels to the wound and come in contact with the mature collagen to become activated and aggregate. During this process the granules within the cells release many agents and factors to further enhance platelet aggregation. At this time, thromboplastin is also released from the injured cells in proximity to the wound and initiating the coagulation process and eventually breaking the fibrinogen into fibrin monomers, which further polymerises to produce a fibrin network. The above mechanisms lead to haemostasis and strengthening of the injured tissue.

Agents responsible for vasodilation such as histamine and serotonin, released upon occurrence of original injury, help increase the permeability of the local capillary bed and allowing the serum and the WBCs into the area surrounding the wound. It is these accumulation of fluid that gives rise to characteristic swelling and sensations of pain and warmth that are experienced by the patient.

Within hours, the neutrophils start to be seen in the wound followed by macrophages after a while. Both the neutrophils and macrophages play a crucial role in initiating the destructive process of healing characterized by removal of debris and engulfing of bacteria. During this process the unnecessary fibrin and dead cells are also broken down by the activity of enzymes.

Later after about 24 hours, the epithelial cells try to pervade the defected area for about 2-3 days and at this time there is a slight evidence of organization within the wound itself as fibroblasts start to lay down the strands of collagen which is a crucial constituent of the skin and also one which strengthens the wound. The collagen production is maximum around the 5th – 7th day in spite of this process characterised by proliferation lasts about 3 weeks. The re-modeling or the maturation phase is subsequent to the proliferation phase and can take up to a year to complete.

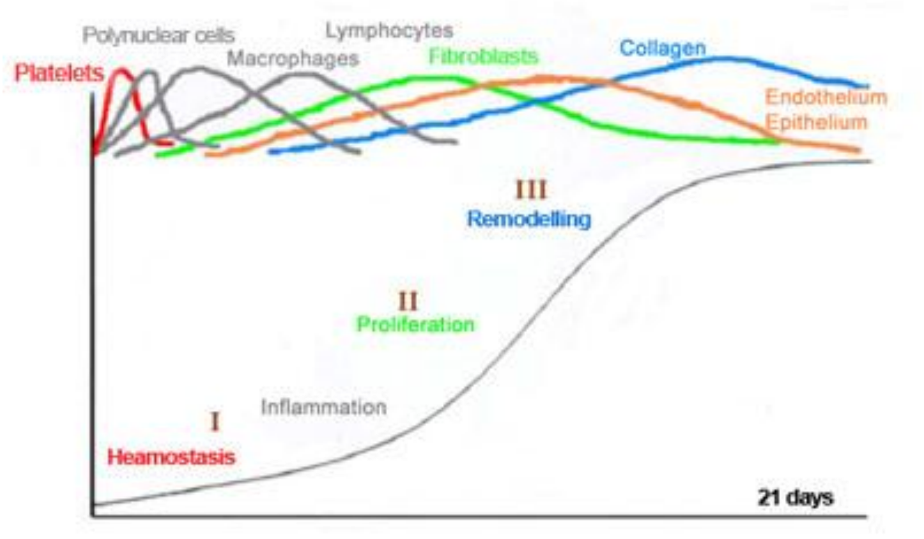

Figure 1.4a: The graph above depicts all the important phases along with their start times involved in the entire process of wound healing.(Source: Urgomedical)[7]

1.4.2: Secondary intention:

When the wounds occur as a result of considerable tissue loss as a result of surgery or trauma, it becomes quite difficult to apply the method of primary closure and the surgeon may opt to leave the wound open to heal by the process of secondary intention[3]. The mechanism is briefly explained in the following lines. The secondary intention alike the primary closure mechanism initiates with an inflammatory response and results in the exposed tissue becoming covered or filled with a layer of blood or serous fluid.

Consequently, the increased capillary and venous permeability release erythrocytes, leucocytes and platelets into the wound. The neutrophils predominate during the first few days and eventually macrophages replace neutrophils as the neutrophils reduce in number. Further by the complex actions by the macrophages, the fibroblasts appear in the base of the wound and are responsible for the production of intracellular factors for collagen which are later converted to collagen fibrils extracellularly.

As the wound progresses in to the 2nd or 3rd day, endothelial cells start to appear in the developing inflammatory tissue as capillary buds. Further, the macrophages are responsible for clearing away the portions of the fibrous clot and release of growth factors that eventually pave way for the production of the capillary network. These capillaries are still permeable and thus provide cells and fluid for the developing tissue constantly.

In the process of healing, the wound gets filled by granulation tissue comprising of collagen and proteoglycans that produce a gel-like matrix present within the fibrous collagen network [3]. The granulation tissue pervades until the base of the original cavity is almost level with the surrounding skin. At this phase, the epithelium starts to grow over the surface of the wound and revives the integrity of the epidermis.

Another crucial part of the healing process is contraction a phase wherein the boundaries of the wound are pulled towards the centre. Wound contraction is initiated about the end of the first week and persists till the wound is completely covered. The myofibroblasts are the key components responsible for this phase.

1.5: Wound diagnostic instrumentation

The objective analysis of the wound bed is imperative since this enhances the management of the wound in a more efficient way rather than a subjective analysis[8].The surface pH and the moisture content in a wound provide useful information for determining the healing rates of the wound and also whether the wound is properly healing or not before the complete wound closure[8][9].This would lead to efficient wound management since it paves way for determining the response of the wound to treatment[8] and also the wound healing does not get disturbed by unnecessary dressings[9].The nursing time of dressings consumes considerable costs which may be minimized by keeping track of the two crucial parameters.

To develop such diagnosis approach, it is necessary to develop sensors for monitoring moisture and pH so that it can be easy to determine the crucial parameters involved in wound healing and also a suitable instrumentation is required. Hence, to proceed with such diagnostic approach, two sensors have been developed each for a different parameter and they are discussed in detail in the following pages.

Figure 1.5a: The picture above shows the currently applied wound diagnostic instrument designed by Ohmedics company named 'WoundSense' to measure the levels of moisture in a wound and determine the healing rates without disturbing the dressing.(Source: Ohmedics)[10]

1.6: Factors influencing the healing process of the wound

There are many factors which determine the healing rates of a wound but the main factors which have been found to play a crucial role in the wound healing process are:

1) Moisture content in the wound

2) pH levels in the wound

1.6.1: Moisture content in the wound

Natural healing occurs in moist wound environment, this is due to the ease of migration of cellular and molecular elements in these conditions [9].Healing is more efficient in moist environment because it enhances the proliferation of fibroblasts which produces the elements of the new dermal ECM(collagen and elastin fibre)enabling the healing of the wound.

Achieving a good moist environment depends on the ability of good clinical judgement for finding the accurate therapeutic levels since too little moisture would dry the wound and too much of moisture would make the wound bed and also the surrounding soft by maceration [11].

1.6.2: pH levels in the wound

The healing of both acute and chronic wounds was observed to be delayed when the pH levels of the wound were alkaline as compared to healing wounds wherein the pH was found to be close to neutral[12][13][14].It has been observed that when a wound heals, the pH of the wound shifts to neutral and then to acidic[12][13].The pH levels have been found to play a vital role in the regulation of the wound healing process such as speeding the enzymatic factors[14], promotion of angiogenesis, increased macrophage etc.[14-18].

The pH levels in a wound can also influence the release of oxygen into the tissues and it has been observed that oxygen levels in tissues enhance the process of wound healing[19].

1.7: Sensors developed for wound management

In order to determine the wound healing process comprehensively two sensors capable of measuring the moisture and the pH levels each have been developed which would greatly enable the efficiency of wound management techniques without aggravating the healing process by unnecessary dressings.

1.7.1: Moisture sensor

The moisture sensor has been developed based on the principle of impedance[9].The sensor utilizes AC voltages and provides a feedback based on the impedance levels in the wound bed depending on the exudate levels in the wound. The moisture content in a wound can be categorized as:

i) Dry -High impedance

ii) Dry to moist-Fall of impedance from high level to midrange

iii) Moist - Mid range impedance

iv) Moist to wet - Impedance tending to low

v) Wet – Low impedance

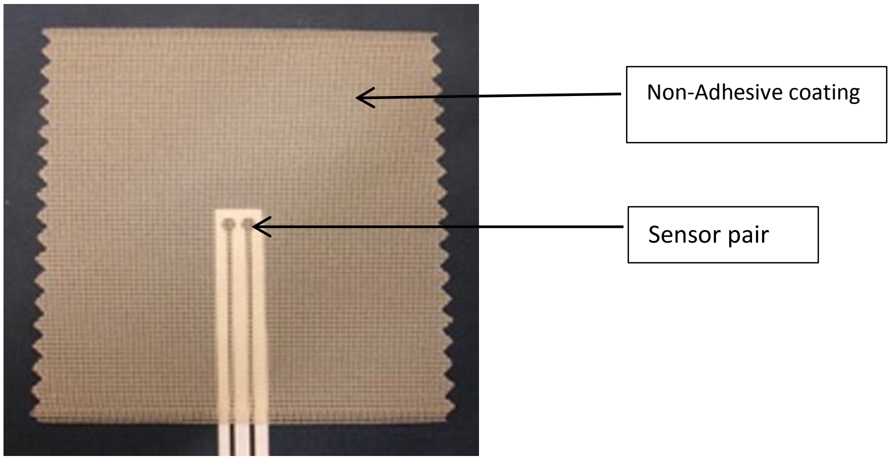

Figure 1.6a: A sample moisture sensor designed in the Department of Bioengineering at the University of Strathclyde, UK which works on the principle of bio-impedance and measures the moisture concentration in a wound [9].

1.7.2: pH sensor

The pH sensor works on the principle of potentiometry [20] wherein the voltage developed by the sensor upon immersing it in a test solution is directly proportional to the levels of pH in it. The pH sensor shares similar working properties as with the potentiometric sensor developed by Arnaud Emmanuel Musa et al.,(2011)[20].

The pH readings obtained were such that pH of 7 was close to null potential and high pH indicated negative values of potential while low pH indicated positive values of potential all recorded with respect to the standard value of the reference electrode.

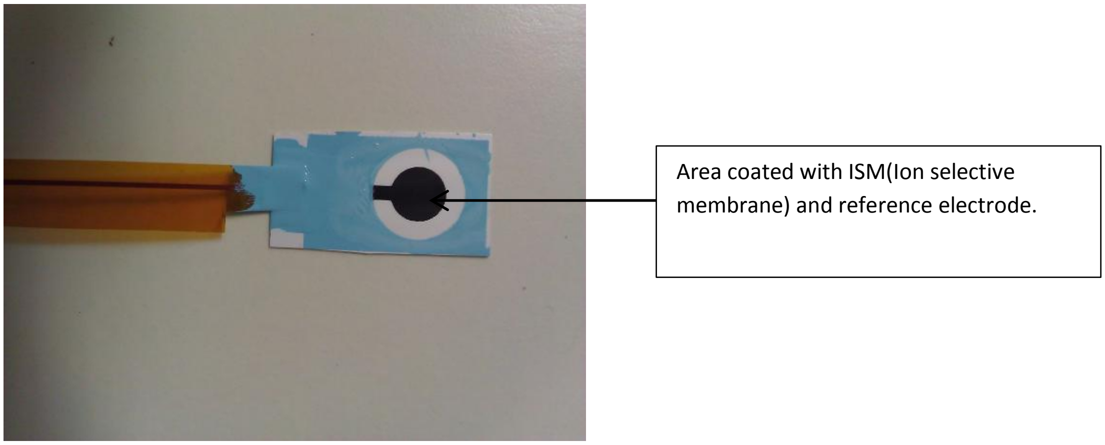

Figure 1.7a: A sample disposable pH sensor designed in the Department of Bioengineering at University of Strathclyde, UK which works on the principle of potentiometry to measure the pH concentration in a solution.

1.8: Integration of the moisture sensor and the pH sensor

For an efficient objective analysis of the wound bed and determining the wound healing rates, it becomes imperative to keep monitoring the above mentioned two crucial parameters namely the moisture content and the pH content in the wound bed[8][9]. This yields efficient wound management since unnecessary dressings are avoided and associated unnecessary expenses are minimized. Thus by keeping track of the two parameters and finding out if their levels in the wound bed are optimal, wound management can become more patient friendly and yield quicker healing results.

For determining the levels of the two parameters it is necessary to use the respective detecting sensors and integrating both the sensors in one device would greatly enhance the measurement protocols and also cost effective wound management can be achieved.

Since the two sensor’s working principles are not similar as mentioned earlier, experiments are necessary to be conducted so that a study of the influence on the readings given by both the sensors and the accuracy of the readings obtained can be examined.

Chapter 2: Experimental theory

This chapter details all the related theoretical aspects that are essential prior to conducting the experiment of studying the AC interference on the pH sensor.

2.1: Alternating Current Voltages (AC voltages)

The theory of the alternating currents is very essential in conducting the experiments in this project due to the utilisation of this voltage by the moisture sensor as discussed in the earlier chapter. The AC voltages play a very crucial role in measurement and analytical studies such as electrochemical impedance spectroscopy (EIS) [21]. It becomes an imperative to discuss the theory of AC voltages in brief to comprehend the experimental results.

2.1.1: Introduction

The term AC is an acronym for Alternating Current and implies that the current is constantly changing its direction in an alternate manner [22]. The other parameters associated with it alternate too such as voltage and phase etc. Since the AC is a signal, it is a waveform in nature and hence can be also called as AC waves.

2.1.2: Sine Wave

The sine wave is the simplest of the AC waves depicting a smooth and simple waveform. It is very basic waveform owing to its single frequency and the regularity in the time period in alternating above and below the mid value [23]. The sine wave is easy to measure and there are a few important measurement parameters to define the AC voltage. These parameters are discussed in brief in the following contents.

2.1.3: Measurement of the Sine wave

As mentioned in the previous section, there are parameters required to characterise or define an AC sine wave [23]. These parameters enable to quantify the signal and study its effects on the other systems. The experiment involved in this project requires only certain specific parameters to define the AC waves and only those are discussed here. Since in this project, the voltage aspect of the AC signal was only taken into account the parameters described below are only in terms of the voltage and not current.

The parameters are discussed here in brief:

- Peak to peak value - It is the distance between the top part of the wave to the bottom part of the wave and it is measured in volts. It may be labeled as Vp-p.
- Amplitude – The maximum height achieved by the wave from the center in either direction (top or bottom) is the amplitude of the wave and is expressed in volts.
- Frequency - The frequency is defined as the number of times the waveform repeating itself over a time period of 1 second. It is measured in Hertz (Hz). Thus if an AC signal is said to have 10Hz, it implies that the waveform is repeating 10 times in 1 second.

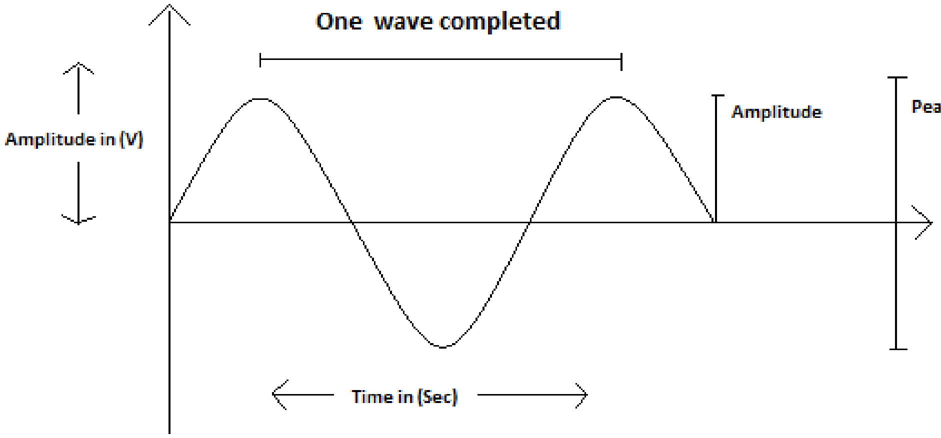

Figure 2.1a: The above diagram illustrates a simple sine AC wave along with the parameters important in understanding the working of the moisture sensor and the project.

2.2: Potentiometry

As discussed earlier in Chapter 1 under 1.7.2, the pH sensors work on the principle of potentiometry and in order to understand their working and the laws governing them, it is essential to discuss the theoretical aspects and hence this section contains the relevant theory to explain the pH sensor working.

2.2.1: Introduction

Potentiometry is a measurement technique used to measure the concentration of a specific ion in a solution [24]. The technique utilizes the relation which states that the potential developed being directly proportional to the concentration of that specific ion in the solution, thus higher the potential developed then higher is the concentration of that ion in the solution [25].

The experiment with which the project deals here is mostly concerning the potentiometric measurements of the unknown pH concentration in a pH solution. The pH sensor which has been designed to measure the pH in the wound bed as detailed in earlier chapter is used.

2.2.2: Development of potential

The pH sensor which is used in the experiments of this project is able to measure the pH concentration because of the attraction of the specific ions onto its surface with the help of an ionophore [26] and thus leading to an accumulation of that specific ion on the sensor which later gives rise to a potential due to excessive charge formation.

The above process starts as soon as the pH sensor is immersed in the solution. It must be noted that the potentials in sensors develop whenever there is a change in the immediate material around it due to the variation in the charges in the different materials.

2.2.3: Electrochemical cell and the potential measurement

In order to measure the concentration of a specific ion with the help of a sensor (ion-selective electrode) in a solution (electrolyte), a reference electrode is essential [27]. The reference electrode provides a standard constant potential throughout the measurement process and the potential difference between the sensor and the reference electrode gives the concentration of the pH in the solution.

The potential developed is a resultant of three individual potentials developed across the different phase contacts namely the Ion selective electrode/electrolyte potential $P1_{(ISE/EL)}$,Reference Electrode/Electrolyte potential $P2_{(REF/EL)}$, and Ion selective electrode/Reference electrode $P3_{(ISE/REF)}$[28].

The sum of the potentials is given by equation below:

$$E = P1_{(ISE/EL)} + P2_{(REF/EL)} + P3_{(ISE/REF)} \text{ ....} \mathbf{2.1a}$$

A simple illustration of the arrangement mentioned above is shown below.

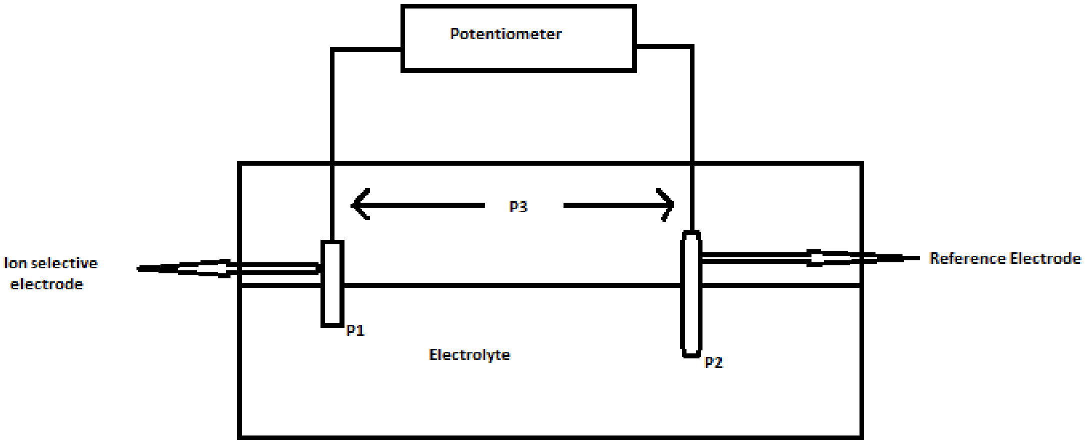

Figure 2.2a: A simple illustration of a cell potential arrangement depicting the regions constituting the individual interfacial potentials which sums up to give out the actual potential.

With regards to the experiment involved in the project, $P2_{(REF/EL)}$ and $P3_{(ISE/REF)}$ is a constant. It is because $P2_{(REF/EL)}$ is the potential difference between the reference electrode and the electrolyte would always remain a standard potential and thus a constant. The $P3_{(ISE/REF)}$ is the potential difference between the ion selective electrode (in this case the pH sensor) and the reference electrode (in this case the Ag/AgCl electrode) is also a constant because the potential difference between the two is independent of the electrolyte.

Hence in the equation terms,

$$E= P3_{(ISE/REF)} - P1_{(ISE/EL)} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \mathbf{2.2a}$$

If the above equation is re-written in terms of the function of the sensor detecting the changes in the pH of the solution(electrolyte) then the cell potential would be altered with the variation of the pH in the solution as

$$\Delta E = \Delta P3_{(ISE/REF)} - \Delta P1_{(ISE/EL)} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \mathbf{2.3a}$$

Therefore any change in the pH of the concentration would be indicated as a relative to the standard potential of the reference electrode.

2.2.4: Nernst Equation

The potential developed by the pH sensor while conducting the potentiometric measurements as detailed above can be theoretically quantified by the use of the Nernst equation[29] which is given as

$$E = E_0 + \frac{RT}{nF} \text{In (Ion conc.)} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \mathbf{2.4a}$$

where E = Potential developed in (V)

E_0= Potential of the sensor under standard conditions in (V).

R = Universal Gas constant (8.314 J/deg-mole).

T = Temperature (degree K).

n = Number of electrons released/atom which is reduced or oxidized.

F = Faraday constant

Ion Conc. = Ion concentration in moles/liter of electrolyte.

Under standard conditions at 25^0C, the potential developed on the sensor should be less than or equal to **0.059V.**

2.2.5: Helmholtz double layer/Double Layer

The electrodes when immersed in an electrolyte solution undergo various changes prior to actually giving a stable potential [28]. One of the most significant change occurring is the accumulation of the opposite charge from that of the charge of the electrode on the surface of the electrode which are present in the electrolyte.

This change and event taking place in proximity to the surface of the electrode (in this case the pH sensor) has a considerable influence on the measurement abilities of the pH sensor. The reason is due to the development of a capacitor type of region wherein one surface is of one particular charge and the other layer formed on the surface of the sensor possesses the opposite charge along with the separation between the two surfaces by a dielectric [28]. These changes mainly occur as result of difference in the potential between the sensor and the electrolyte.

The entire region in proximity to the sensor can be said to have developed a double layer or the Helmholtz layer. This double layer can be divided into:

i) Inner Helmholtz plane (IHP) – This layer is the closest to the surface of the sensor and it mostly accumulates the specific ions onto the surface which is the potential determining factor.

ii) Outer Helmholtz plane (OHP) – Immediate to the IHP, comes the Outer Helmholtz plane which is a result of the accumulation of the non-specific ions close to the surface of the IHP due to the Coulomb forces. Only the opposite non-specific ions are attracted towards the IHP while the same charges are repelled. The non-specific ions are distributed from the OHP to the bulk solution.

iii) Diffuse layer – The region of the electrolyte starting from the OHP and away from the sensor to the bulk solution forms the diffuse layer it has a charge density of µC/cm^2.

A detailed illustration of the double layer is given below:

The illustration given below shows schematically the changes that occur near the surface of an electrode after immersion in an electrolyte.

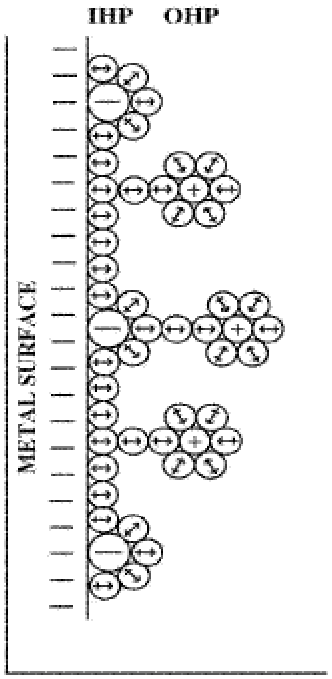

Figure 2.3a: The illustration above depicts the water dipole model of the double layer extracted from Bockris, Devanathan and Muller[30]. As evident from the above illustration the region in proximity to the electrode forms a double layer which eventually becomes like a capacitor.

It can be concluded that the total charge density of the double layer σ^{Total} is the sum of the IHP charge density σ^{IHP} and the diffuse layer charge density $\sigma^{Diffuse}$ because the diffuse layer and the OHP comprise mostly of the similar charges developed by the electrode and thus compensate for the negative charge of the sensor [31]. An equation can be written relating the above 3 charge densities as

$$\sigma^{Total} = \sigma^{IHP} + \sigma^{Diffuse} = -\sigma^{Sensor} \ldots\ldots\ldots\ldots\ldots\ldots \mathbf{2.5a}$$

wherein σ^{Sensor} is the charge density of the sensor. When a graph of potential against the distance of the layers from the surface of the electrode is plotted, it becomes evident that the potential keeps decreasing with increase in distance from the surface of the sensor (assuming that the sensor possesses the maximum negative charge). A term called Zeta potential has been given to the potential value between the OHP and the diffuse layer.

The model described above is the simplest model to characterise the properties of the double layer that forms a capacitor because of the accumulation of two oppositely charged ions as two layers separated by a dielectric in between. To determine the effects produced by the sensor due to the double layer which acts as a capacitor, an equation relating the charge density and the voltage drop across the plates is given and is mentioned below [31]:

$$\sigma = \frac{\varepsilon\varepsilon_0}{d} V \ldots\ldots\ldots\ldots\ldots\ldots \mathbf{2.6a}$$

where V = Voltage drop across the plates of the double layer.

ε = dielectric constant of the electrolyte.

ε_0= permittivity of the free space.

d = distance in between the two plates.

Since the capacitance of a capacitor is given by the ratio of charge developed to the potential applied the above equation in terms of capacitance can be re-written as

$$C = \frac{\sigma}{V} = \frac{\varepsilon\varepsilon_0}{d} \ldots\ldots\ldots\ldots\ldots\ldots \mathbf{2.7a}$$

2.2.6: Polarisable and Non-polarisable electrode-electrolyte interfaces

The concept of polarity of the interface between the electrode-electrolyte is a major factor for measurement purposes. A polarisable interface behaves as a capacitor due to the charge alteration as a result of a potential applied but with no current flow between the double layers. A non-polarisable interface on the other hand allows current flow freely owing to the invariable charge across the layers and thus resulting in a constant potential [32]. But according to Bockris and Reddy [28], no electrode is fully polarisable or non-polarisable but would be closer to either of the two.

The above properties of electrodes enables sensor applications because an electrochemical cell requires two electrodes wherein one electrode (sensor) due to its function should preferably made polarisable while the other electrode (reference electrode) should preferably made non-polarisable. This helps in the measurement systems in electrochemical systems possible and efficient.

2.2.7: Membrane potential

This section deals with the actual process that occurs in a sensor internally which leads to the development of the potential and measurement of the concentration of the specific ion. The main component responsible for the measurement of concentration is the ionophore comprising ion selective membrane [26].

It is the ion selective membrane which acts as a medium to attract the specific ions and pass it over to the rest of the conduction circuit in an electrode. The ionophore is the crucial element in differentiating the specific ion from all other ions in the electrolyte.

The Nernst equation as detailed in section 2.2.4 can be re-written in another form to determine the activity of ions between the electrolyte and in the membrane. The equation is

$$E = E_0 + \frac{RT}{nF} \text{In} \frac{i_{sol}}{i_m} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \mathbf{2.8a}$$

Wherein all the terms denote the same parameters as stated in equation 2.4a and additionally i_{sol} refers to the activity of the ions in electrolyte whereas i_m refers to the activity of the ions in the membrane.

As the capacity to accommodate the ions by the ionophore is constant the activity denoted by i_m is constant and can be set as 1. Hence the potential is only determined by the i_{sol} term. According to the Denye-Huckel equation, the i_m term can be expressed in terms of the concentration of the ion in the solution and hence when the i_{sol} term is replaced by the concentration term c_{sol}, the above equation can be re-written as

$$E = E_0 + \frac{RT}{nF} \text{In } c_{sol} \text{ ……………………………..} \mathbf{2.9a}$$

Diffusion plays a major role in determining the response rates of the sensor once it has been immersed into the electrolyte for the first time. Since initially the membrane is devoid of any ion accumulation by the ionophores, the sensor may take some time to get into action. It has also been observed by Bakker et al.[33], in 1997 that the ion is slow in movement in a solid state membrane and hence requires a longer time to stabilise. After this process, the sensor gets conditioned such that in the future the sensor would take negligible time in stabilising.

2.2.8: Ion selective electrode working

A best example of an ion selective electrode is that of a liquid pH electrode. The working of a typical ion selective electrode (ISE) is detailed here. As discussed earlier the ion selective membrane plays the crucial role in the absorption of the specific ions from the electrolyte to the sensor. In order to explain the working of the ISE, the total process needs to be classified into the respective interfaces [30]. The potential obtained is a result of the summation of all the individual potentials developed in the respective interfaces. In order to efficiently measure the concentration of the specific ion only the concerned interface should be kept variable while all the potentials arising from the rest of the interfaces need to be a constant. The equation along with the explanation of the terms is given below:

$$E = E_{EL/M} + E_M + E_{M/IS} + E_{IS/REF} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \mathbf{2.1b}$$

wherein $E_{EL/M}$ denotes the potential difference between the electrolyte and the membrane of the ISE, E_M denotes the membrane potential, $E_{M/IS}$ is the potential difference between the ISE membrane and the internal solution of the electrode and lastly $E_{IS/REF}$ is the potential difference between the internal solution and the reference electrode fixed inside the ISE[34].

In the equation stated above, E_M is a constant due to the fixed diffusion of ions as stated earlier, $E_{M/IS}$ is also a constant due to the saturation activity throughout the measurement process, $E_{IS/REF}$ is also a constant as stated earlier that a reference electrode would always give a standard potential owing to its non-polarisability property and lastly only the term $E_{EL/M}$ remains which according to the function of the electrode is the crucial term in determining the ion concentration in the electrolyte. Therefore, during the measurement process only potential between the electrolyte and the ISE membrane varies with respect to the standard potential reference electrode and thus the concentration of a specific ion is obtained.

2.2.9: Ion selective electrode in a solid state (in this case the pH sensor)

The sensor which has been designed for monitoring the pH levels in the wound and that which would be used in this project is a type of the solid state ISE. The solid state ISE does not have a liquid phase internal filling solution instead is attached by a solid contact[20].

The entire process of measurement is the same as that of the normal ISE. In this type of electrode the term $E_{M/IS}$ is absent due to the absence of liquid phase internal filling solution and thus the total potential equation is given by

$$E = E_{EL/M} + E_M + E_{M/CON} \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \mathbf{2.2b}$$

Similar to the reason given in the earlier section, the only potential determining term is the $E_{EL/M}$. In this case the term $E_{M/CON}$ is the potential difference between the ISE membrane and the ISE conductor [35]. This term is also a constant because there is no transfer of the absorbed taking place across the interface.

There is one issue with using such an electrode wherein the interface between the membrane and the conductor has a polarising property and hence any minute fluctuations in the double layer in the electrolyte may lead to variation of $E_{M/CON}$. This property may influence the measurement [36].

2.2.10: Ion to electron conduction

As mentioned in the earlier section, the solid phase is used instead of the liquid junction and this interface is fabricated as a good redox couple so that the ions carried over by the ionophores is converted into electrons on the conductor. This interface is made non polarisable by increasing the exchange current density and thus inhibiting the charge accumulation.

There are three conditions [36] that are required to decrease the inefficiency of a solid state sensor:

1. The ionic to electronic conversion must be reversible and stable.

2. High exchange current than the current developed by the measuring devices.

3. Minimal interference response to gases and ions.

Chapter 3: Materials and Methodology

This section details the materials used and methods adopted to conduct the experiments concerning the AC voltage interference on the pH sensor. The AC voltages have two important parameters namely its frequency and amplitude. In order to study the effects of AC voltage interference on the pH sensor, it is essential to study the effects of the above two parameters on the pH sensor individually. Thus, in this chapter all the details of individual analysis and also additional analysis such as cumulative effects etc. which were necessary to study the effect on the sensor comprehensively have been included.

3.1: Materials

The materials or components involved for conducting the experiments are detailed in the following contents:

3.1.1: Universal Buffer solution and pH solutions

The sensor needs to be tested in pH solutions ranging from pH4-pH10.These range of pH were prepared from universal buffer solution. For preparing this universal buffer solution, a bottle is taken and filled with 900 ml of distilled water. In this bottle containing the distilled water, 2.3ml of Acetic acid, 2.74ml of Phosphoric acid and 2.4732g of Boric acid is added and dissolved by proper mixing. Now the bottle is filled with more distilled water to make it to 1 litre. Now in another fresh bottle 0.2M of NaOH solution is prepared by adding 7g of NaOH crystals to 875ml of distilled water in the bottle and well mixed. To prepare the different pH solutions, certain standard volume of NaOH solution is added to the 1 litre universal buffer solution each time to obtain individual pH solutions. The detailed procedure for preparing the universal buffer solution and corresponding pH solutions are given in the appendix later in the thesis. The pH solutions were filled in a beaker when performing the experiments each time and the pH and the moisture sensor were immersed along with the reference electrode and the outputs were recorded.

Mostly the use of pH 4 and pH 7 solutions was made to study the drift per decade.

3.1.2: Reference electrode

The reference electrode as discussed in the previous sections is a standard electrode which provides a constant potential throughout the measurement [33]. The reference electrode used in this project is a Ag/AgCl (Silver/Silver Chloride) electrode which is the most widely used electrode due to several advantages. It can be easily prepared in laboratories. The Ag(Silver) metal is coated with an ionic compound AgCl(Silver Chloride).The AgCl is negligibly soluble in water which makes it stable. This ion compound coated Ag metal is now immersed in an electrolyte bath containing the Cl^- (Chloride) as the anion. Thus the reactions involved are:

1. $Ag \longleftrightarrow Ag^+ + e^-$

The above equation denotes the first reaction involving the process of oxidation of Ag to become Ag^+ ions in solution at the interface.

2. $Ag^+ + Cl^- \longleftrightarrow AgCl$

The above equation denotes the second reaction involving the precipitation of the AgCl.

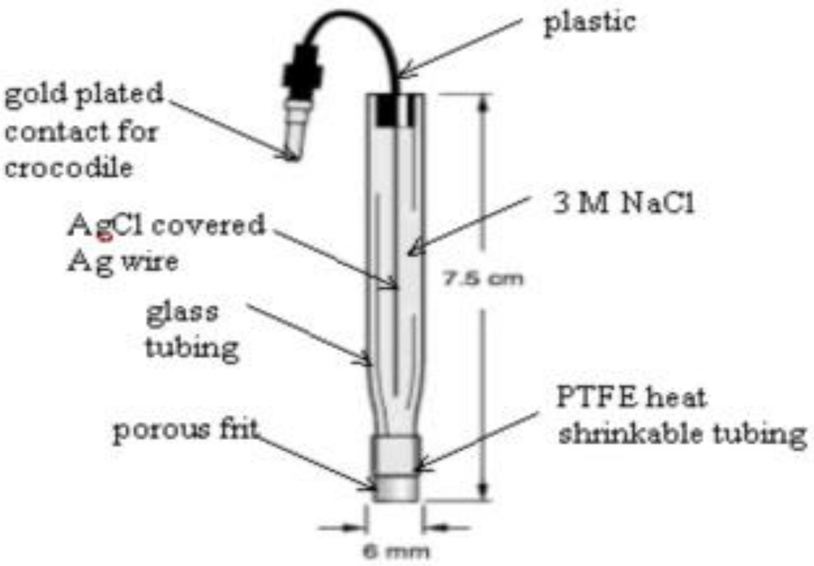

Figure 3.1a: A typical Ag/AgCl liquid junction reference electrode used commonly in electrochemical cells to act as a standard potential electrode. The reference electrode provides standard potential irrespective of the pH concentration variation thus enabling accurate pH measurement by the sensors. (Source: http://www.chm.bris.ac.uk/pt/diamond/mattthesis/chapter5.htm)[36]

3.1.3: Moisture sensor

The moisture sensor is used as a source to introduce the AC voltage interference to the pH sensor in the solution or electrolyte. The moisture sensor is connected to the function generator to generate the characteristic AC sine waveforms. The detailed information of the moisture sensor is given in chapter 1 under section 1.7.1.

3.1.4: pH sensor

The pH sensor is the subject of study in this entire project. It is connected to the Solartron 1286A Electrochemical Interface instrument so that the output obtained from the sensor can be recorded and analysed. A detailed information of the pH sensor is given in chapter 1 under 1.7.2.

3.2: Methodology

The methods and procedures adopted for the experimental study are detailed in the following contents.

3.2.1: Voltage potential analysis:

The voltage potential analysis was conducted to verify the working of the pH sensor and its stability in giving the output of the voltage developed depending upon the pH of the solution. This analysis would help in selecting a good sample of sensors without any defects for the main experimental procedures. It would also help in determining the ideal potential values and the ideal voltage/pH measured by the sensor.

The schematic diagram of conducting the above analysis is depicted below:

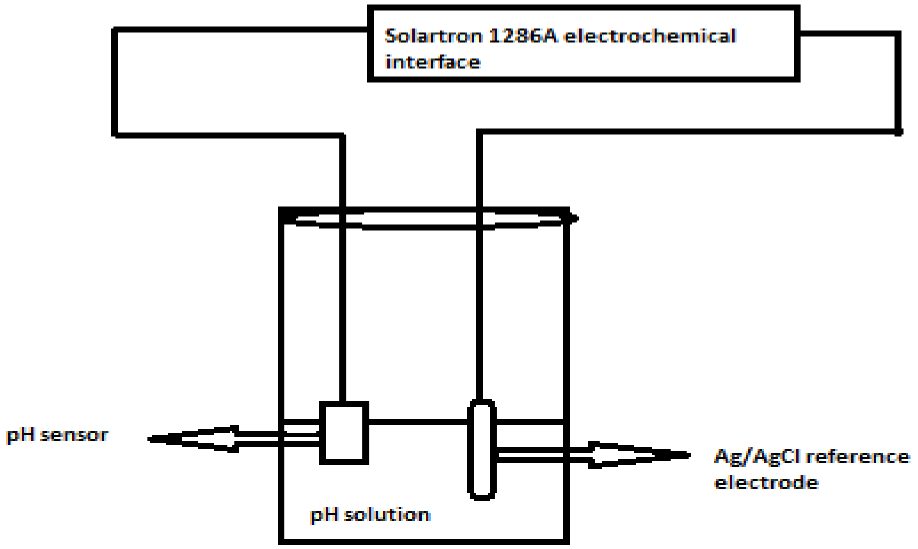

Figure 3.2b: The diagram above depicts the arrangement of the components for conducting the voltage potential analysis experiment. This arrangement does not introduce any AC interference and hence the moisture sensor is not included in the diagram.

The procedure for conducting the above experiment is as follows:

1) A pH sensor illustrated in figure 1.7a was connected to the Solartron 1286A electrochemical interface.

2) An Ag/AgCl reference electrode illustrated in figure 3.1a was used to complete the circuit and also to give a standard potential difference between the pH sensor and itself throughout the experiment.

3) A medium sized beaker was used to fill the pH 4 solution as obtained from the procedure in section 3.1.1 and was placed under the clamp holding the pH sensor and the reference electrode.

4) Both the pH sensor and the reference electrode were immersed into the pH 4 solution and the measurement of the developed potential on the pH sensor was recorded with the help of CorrWare software provided with the equipment measuring the potential.

5) The above test was repeated on each of the other sample of small and large sensors on different pH solutions ranging from 4 to 10.

3.2.2: Schematic diagram for all the following analysis:

All the analytical experiments in the following sections are arranged in the way depicted below except for the changes in the individual procedures of conducting the experiments and the timings at which the AC interference is introduced and the corresponding recording of the sensor behavior.

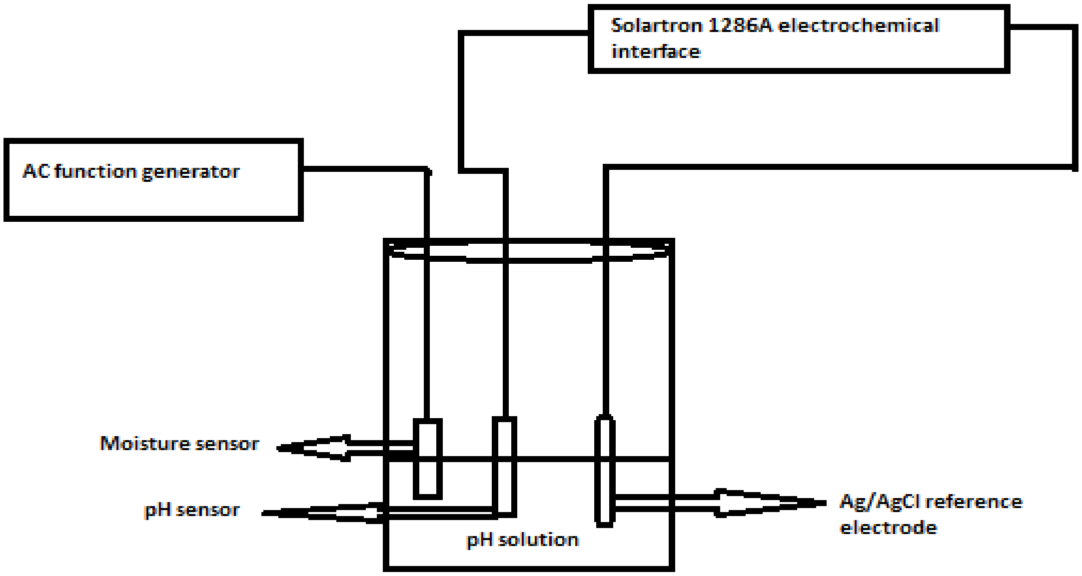

Figure 3.3a: The above diagram shows the arrangement for performing the main study experiments by introducing the AC interference on the pH sensor. The AC voltage is introduced by the function generator with the desired values of the parameters defining the interference characteristics. Though the analytical experiments have the similar setup as illustrated above, different analysis require different immersion timings of the sensors.

3.2.3: Frequency testing

The frequency analysis was conducted to learn the variation of the potential developed on the pH sensor with different frequencies and a constant amplitude introduced via the wound moisture sensor. The experiment was conducted in a pH 7 solution with a constant amplitude of 0.25V

The procedure for conducting the above experiment is as follows:

1) A pH sensor illustrated in figure 1.7a was connected to the Solartron 1286A electrochemical interface.

2) An Ag/AgCl reference electrode illustrated in figure 3.1a was connected to the Solartron 1287 electrochemical interface and is used to complete the circuit and also to give a standard potential difference between the pH sensor and itself throughout the experiment.

3) A medium sized beaker was used to fill the pH 7 solution obtained by preparation as detailed in section 3.1.1.

4)The Ohmedics moisture sensor as illustrated in figure 1.6a was connected to a function generator.

5) The pH sensor and the reference electrode were immersed in the pH 7 solution and the potential was recorded till 120 seconds approx. The moisture sensor is set at 10Hz frequency and is introduced after 120 seconds and the response of the pH sensor is recorded till 240 seconds approx. Now the AC interference is removed and the response is recorded till 300 seconds approx. and the experiment is stopped. The graphs with all the timing points are depicted in the next chapter.

6) The above steps are repeated for 3 randomly chosen sensors labeled as S4, S7 and S11 from the small sensor group and L2,L3 and L10 from the large sensor group by varying the voltage levels as 10Hz and 1kHZ.

3.2.4: Amplitude testing

The amplitude testing was conducted to learn the variation of the potential developed on a pH sensor by varying the amplitude of AC interference voltage with a constant frequency of 1kHz introduced via the moisture sensor. The amplitude of AC voltage as told earlier is another important parameter determining the characteristic of the AC interference . The variation of amplitudes was set as 0.1V, 0.25V and 0.5V. The schematic diagram illustrating the arrangement for this analysis is depicted in 3.3a.

The procedure for conducting the above experiment is as follows:

1) A pH sensor illustrated in figure 1.7a was connected to the Solartron 1286A electrochemical interface.

2) An Ag/AgCl reference electrode illustrated in figure 1 was connected to the Solartron 1286A electrochemical interface and is used to complete the circuit and also to give a standard potential difference between the pH sensor and itself throughout the experiment.

3) A medium sized beaker was used to fill the pH 7 solution which is prepared as mentioned under 3.1.1.

4) The Ohmedics moisture sensor illustrated in figure 1.6a was connected to a function generator.

5) The pH sensor and the reference electrode were immersed in the pH 4 solution and the potential was recorded till 120 seconds approx. The moisture sensor is set at 0.1V amplitude and is introduced after 120 seconds and the response of the pH sensor is recorded till 240 seconds approx. Now the AC interference is removed and the response is recorded till 300 seconds approx. and the experiment is stopped. A schematic graph to explain this is illustrated in the results chapter.

6) The above steps are repeated for 3 randomly chosen sensors labeled as S4, S7 and S11 from the small sensor group and L2, L3 and L10 from the large sensor group by varying the amplitudes of AC voltage as 0.1V, 0.25V and 0.5V.

7) The above experiments are conducted in both pH 4 and pH 7 solution to study the Voltage/decade change in hydrogen ion concentration.

3.2.5: Cumulative effect testing:

This testing was done to study the change in the drifting levels of the pH sensor upon consecutive introduction and removal of AC interference which would help to study phenomena occurring on the surface of the sensor.

This testing was imperative owing to the sensor's drifting voltage levels each time AC interference was introduced and this testing allows to find the trend line to the changes of drift levels between the consecutive interferences.

The following procedure was adopted to examine the cumulative effects of the sensor:

1) A pH sensor as illustrated in figure 3 was randomly chosen and the voltage developed on it was recorded with the help of the Solartron 1286A electrochemical interface. This constitutes the start or the initial voltage with no interference at all.

2) Next, an AC interference is introduced with 1kHz and 0.1V as its parameters. Later the interference is removed after the sensor reaches a stable potential and the newly drifted voltage values are recorded by repeating step 1 as stated above. This step would constitute the voltage upon 1st interference effect.

3) The step 2 is done repeatedly till the 10th interference effect of the sensor is recorded and the values obtained are analysed.

3.2.6: Calibration testing

This test was conducted to determine if the sensor would exhibit constant drift levels upon AC interference between the pH 4 and pH7 solutions and also to determine if the sensor lost sensitivity after interference. The test was conducted multiple times with other sensors to verify the results obtained.

The procedure adopted for this testing is detailed below:

1) Initially the pH sensor as illustrated in figure 1.7a was tested for its potential developed in the pH 4 solution and later in pH 7 solution and the readings are recorded.

2) Next, the AC interference is applied via the moisture sensor with 1 kHZ and 0.1V as parameters. This would lead to a drift in the sensor and the potential developed after the drift is recorded by executing step 1.

3) The above step 2 is repeatedly executed with 3 different small and 3 large sensors to verify the obtained results

Chapter 4: Results

This chapter details the results obtained upon conducting the experiments detailed in the previous chapter. As already mentioned in the previous chapter the sensor was subjected to the two important parameters that define the AC interference and the effects were recorded. To study the sensor's characteristics further, the cumulative and the calibration analysis were done which showed useful results.

The details of the results obtained are given in the following contents.

4.1: Voltage potential analysis

As detailed in the previous chapter under 3.2.1 the voltage potential analysis was mainly conducted to enable proper selection of sensors with no fault in measurement for the main study experiments. The potential stability developed by the sensors under each pH solution further gave the ideal values in the pH solutions with no external interference.

Both small and large sensors constituted 22 in number with 11 in each group. The test was conducted on all the 22 sensors and an average potential value for each pH concentration was recorded.

Both the tabulation and the corresponding graphs have been depicted to show the potential developed by the sensors over the pH range from 4 to 10.

The sensors have shown quite stable behavior with minimal or no variations.

pH value	Small sensors (V)	Large sensors (V)	Difference between the two sensors(V)
4	0.261	0.254	0.007
5	0.216	0.214	0.002
6	0.159	0.159	0.000
7	0.108	0.112	0.004
8	0.057	0.056	0.001
9	0.004	0.003	0.001
10	-0.049	-0.05	0.001

Table 4.1a: The table above shows the average potential output obtained by the sensors for each pH solution along with the difference between the small and large sensors.

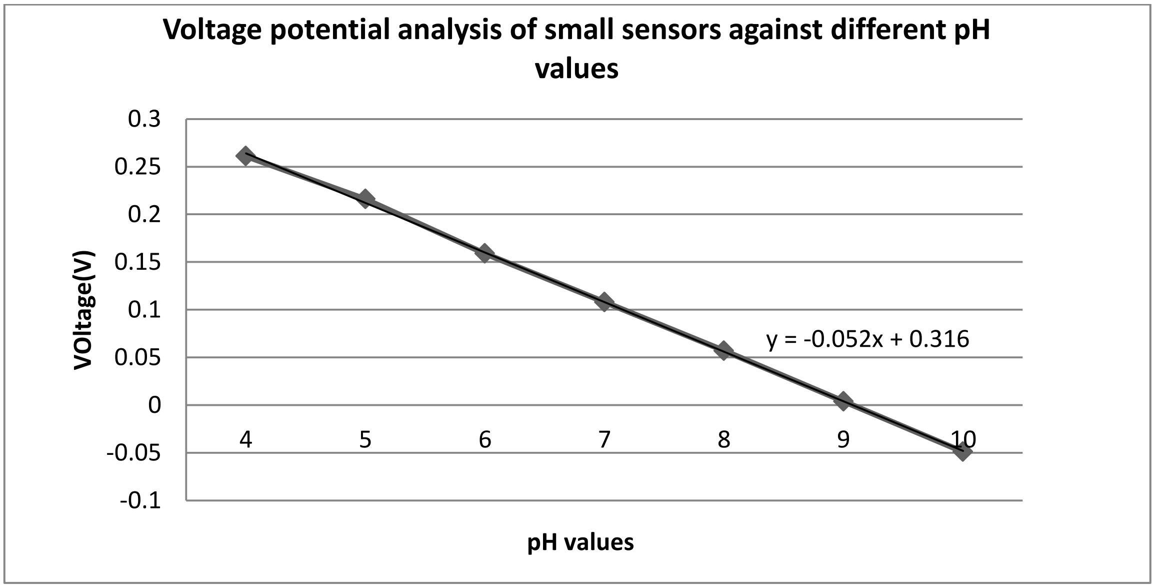

Figure 4.1a: The graph above depicts the potential developed by small sensors over a pH range. The slope from the graph is found to be -0.052 V/decade change in hydrogen ion concentration.

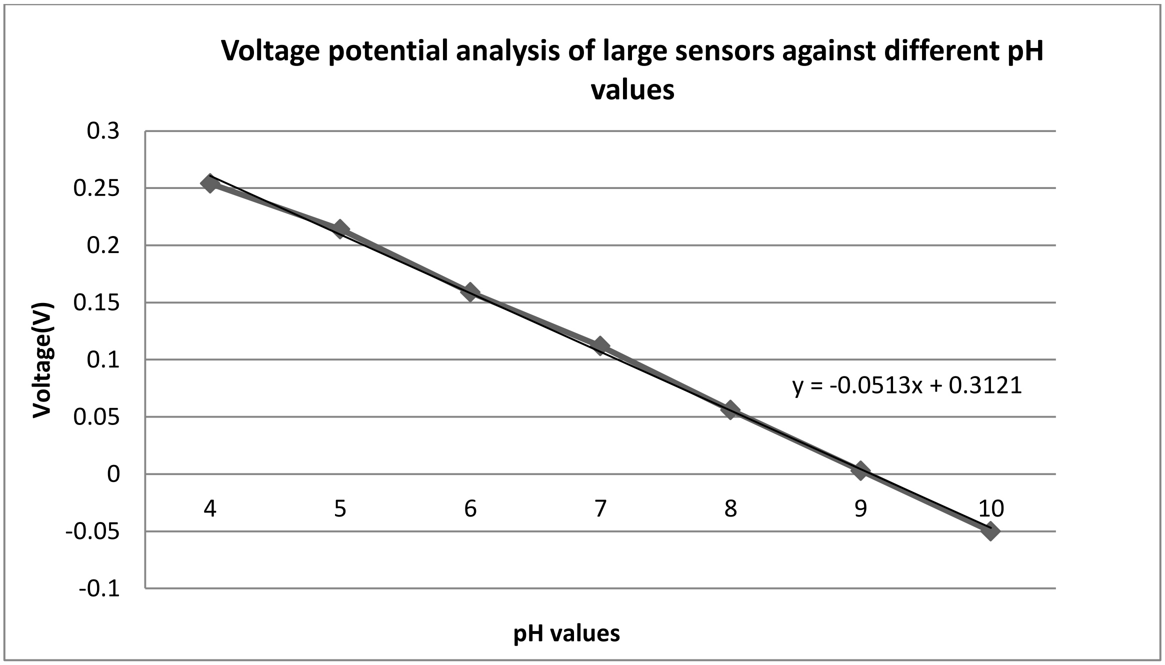

Figure 4.2a: The graph above depicts the potential developed by large sensors over a pH range. The slope from the graph is found to be -0.0513 V/decade change in hydrogen ion concentration.

From the above results obtained, it is very clear that the sensors are very stable with minimal or no variations between them and the graph depicted makes it evident that the voltage values drop in a linear manner with similar slope values between the large and the small sensors.

The slope values found are -0.052V/decade change in hydrogen ion concentration and -0.0513V/decade change in hydrogen ion concentration for small and large sensors respectively.

The values obtained from the above experiment are kept as a base with which the next experiments could be conducted and analysed in an effective manner by looking into the drifting potential of the sensors.

4.2: Frequency testing analysis

This analysis helps study the behavior of the sensor with variations in one of the two main parameters of AC interference i.e frequency by keeping the amplitude constant at a value of 0.25V and varying the frequency as 10 Hz and 1kHz as already detailed in 3.2.3.

The analysis would pave way for determining the ideal values of frequency at which there is minimal drift in the sensors.

The related tables and graphs are shown below:

Peak-Peak value of sensors at 10Hz:

Small sensors in (V)	Large sensors in (V)
0.145	0.141

Table 4.2a: The table above show the average peak-peak voltage values upon AC interference of 10Hz frequency and it is clearly seen that the peak-peak value is quite large.

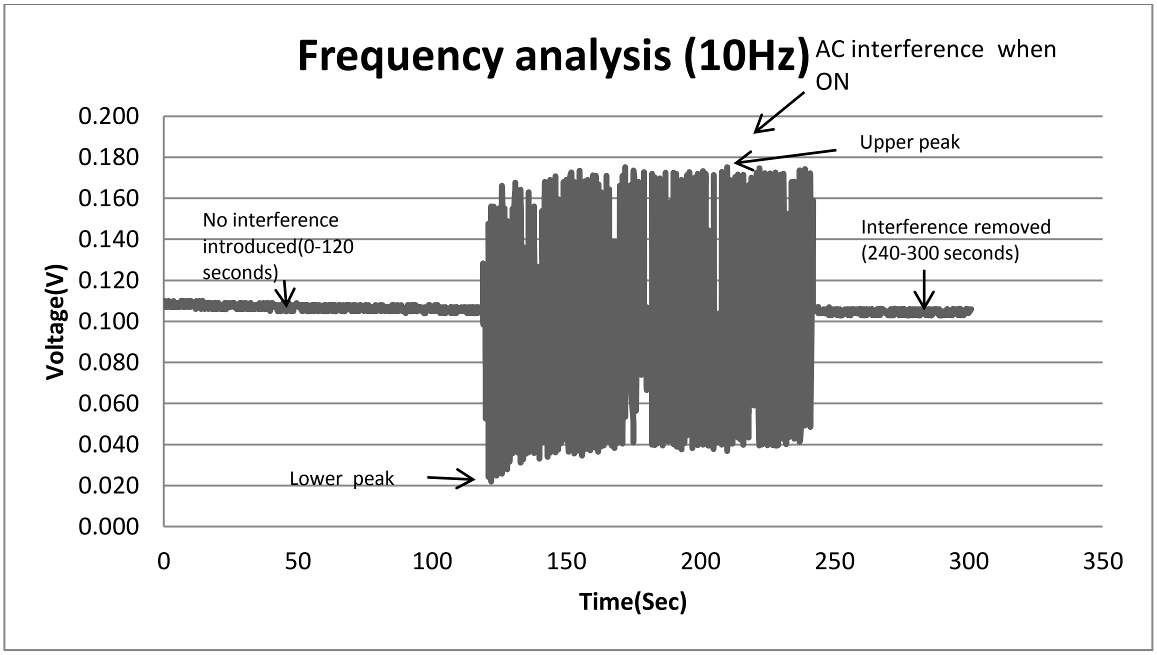

Figure 4.3a: The graph above depicts with indications the large fluctuations of the sensor upon low frequency interference. It can be seen that the drastic fluctuations are very clear indicating the intense effect a low frequency AC voltage has on the pH sensor.

Peak-Peak value of sensors at 1kHz:

Small sensors(V)	Large sensors(V)
0.0024	0.0041

Table 4.3a: The table above show the average peak-peak voltage values upon AC interference of 1kHz frequency and it is clearly seen that the peak-peak value is very minimal.

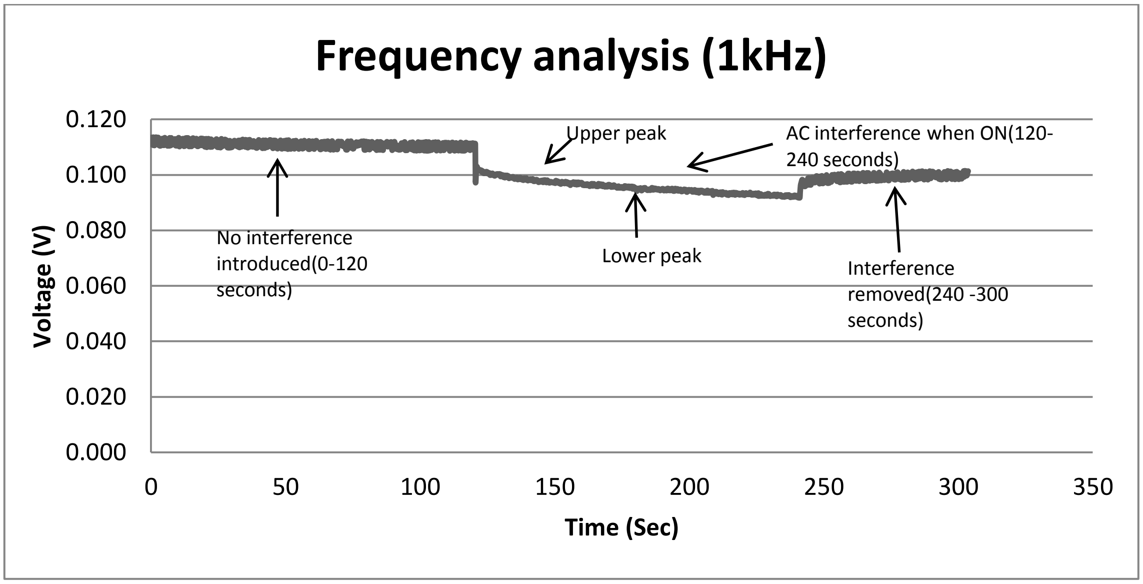

Figure 4.4a: The graph above depicts with indications the minimal fluctuations of the sensor upon high frequency interference.

The peak –peak values were calculated as difference between the highest peak and the lowest peak while the AC interference is ON.

From the results obtained above, it is clear that the sensors drastically fluctuate under 10Hz frequency while at 1kHz frequency, there is very minimal fluctuation. There was no much variation between the small and large sensors in low frequencies but at high frequencies the small sensors seem to fluctuate lower than the larger sensors with a difference of 0.0017V.

4.3 Amplitude testing analysis:

The other important parameter in AC interference is the amplitude of the signal. When the sensor behavior with regards to amplitude variation with constant frequency of the AC interference was studied, the results obtained were slightly inconsistent and there was large standard deviation between each sensor operated upon with the same AC voltage parameters. The detailed procedure is included in the previous chapter under 3.2.4.

But one conclusion derived from this experiment was that while performing the experiments, the large sensors were predominantly found to achieve the stable potential quickly than their counterparts.

Note: The graphs convey the average drift (upon AC interference) in voltage obtained from the sample of 3 sensors each from a small sensor group and a large sensor group. The standard deviation too is obtained from the values recorded by each of the group of small sensors and the large sensors.

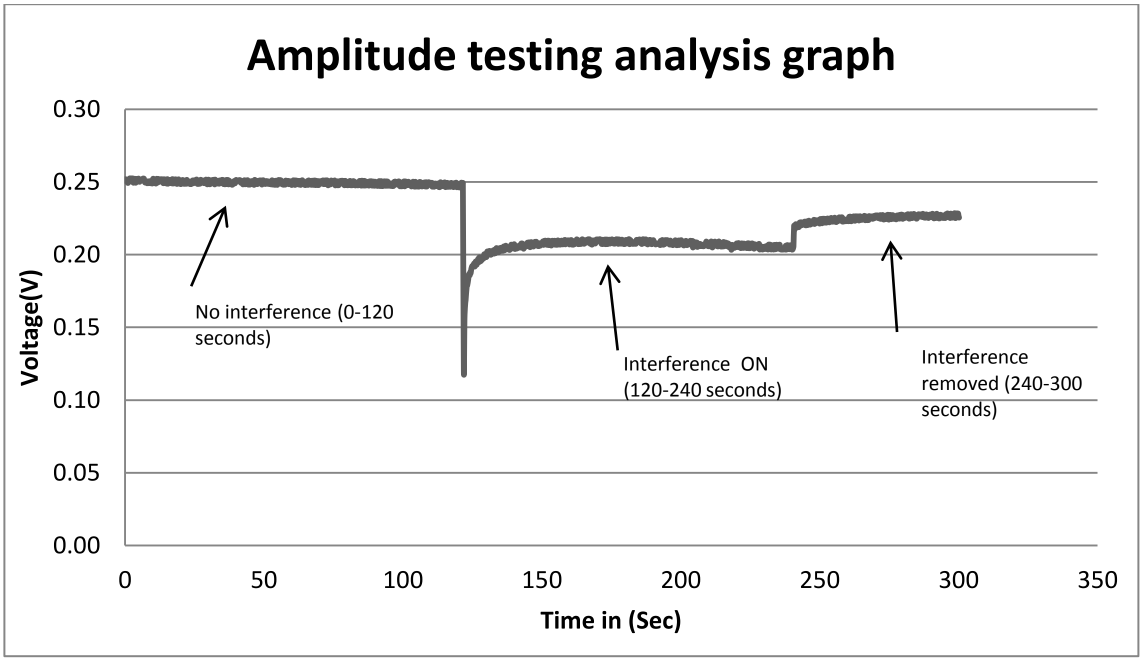

Figure 4.5a: A typical amplitude analysis testing graph wherein at 120 seconds approx.,AC interference is introduced and at 240 seconds approx.,AC interference is removed as indicated by the arrows in the graph.

4.3.1: Amplitude analysis in pH 4 solution

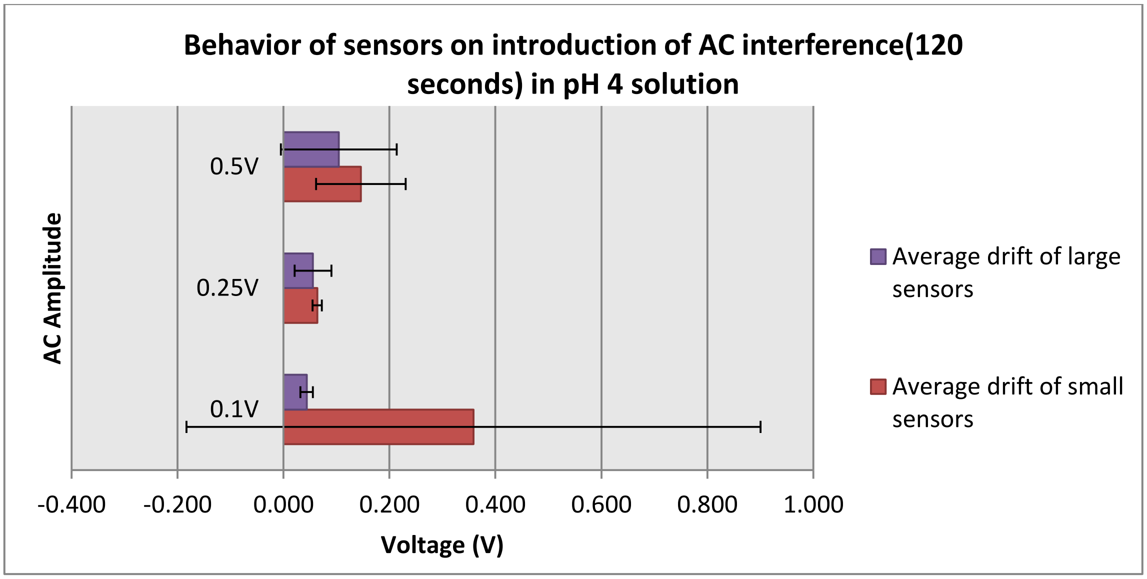

Figure 4.6a: The graph above shows the average drift in voltage and standard deviation of the sensors when AC interference at 0.1V, 0.25V and 0.5V amplitude and constant frequency of 1kHz is introduced at 120 seconds approx. in a pH4 solution.

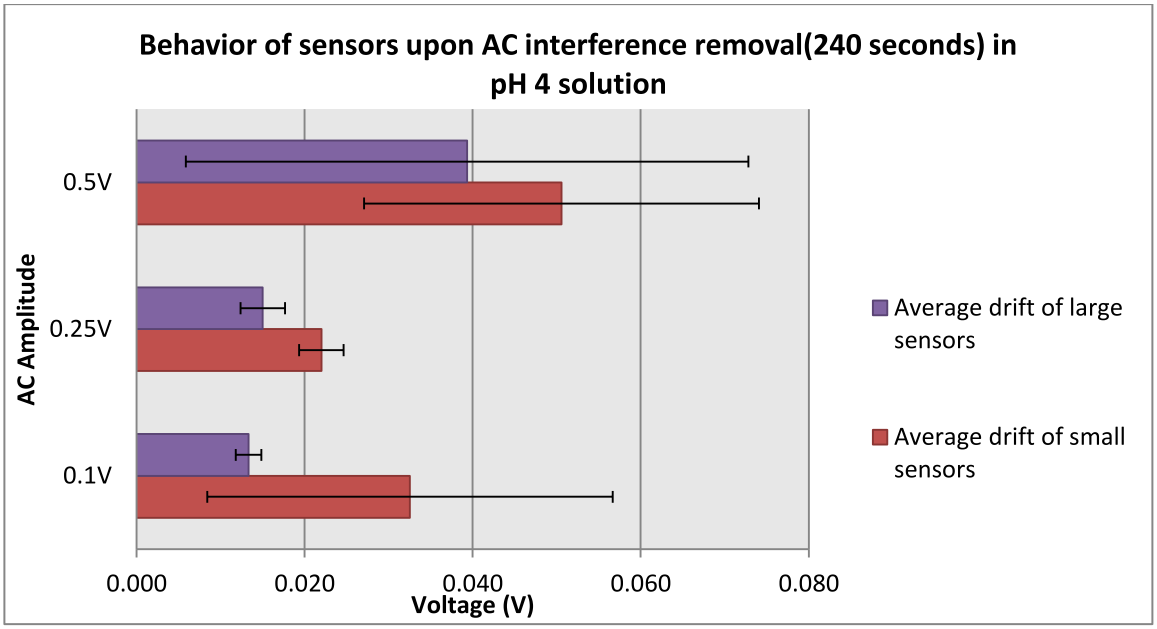

Figure 4.7a: The graph above shows the average drift in voltage and standard deviation of the sensors when AC interference at 0.1V,0.25V and 0.5V amplitude and constant frequency of 1kHz is removed at 240 seconds approx. in a pH4 solution.

4.3.2: Amplitude analysis in pH 7 solution:

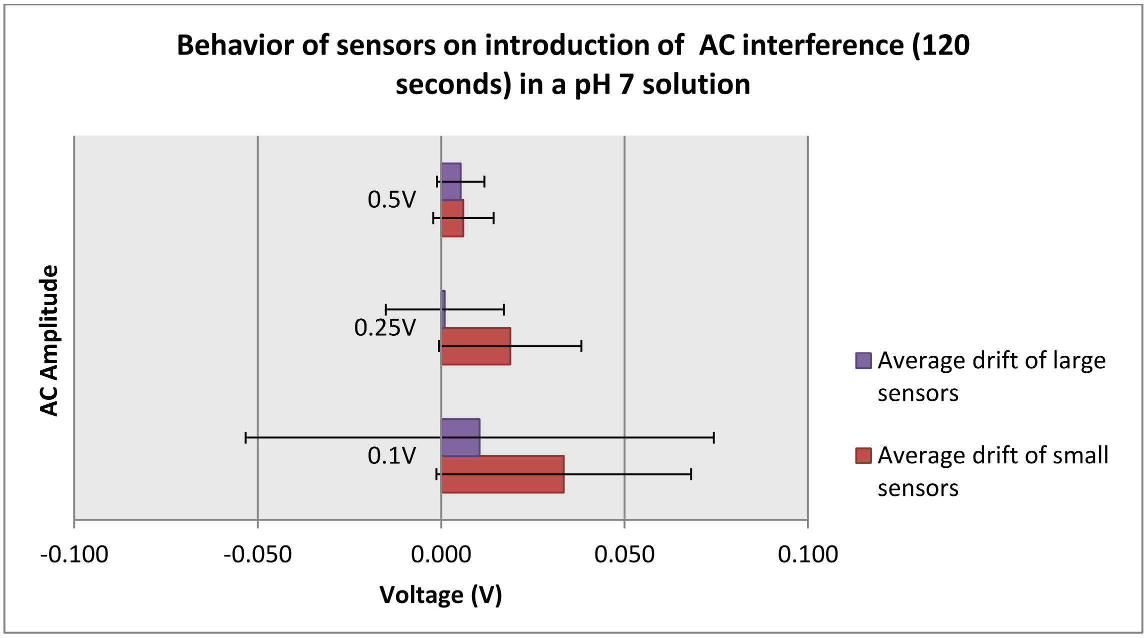

Figure 4.8a: The graph above shows the average drift in voltage and standard deviation of the sensors when AC interference at 0.1V, 0.25V and 0.5V amplitude and constant frequency of 1kHz is introduced at 120 seconds approx. in a pH7 solution.

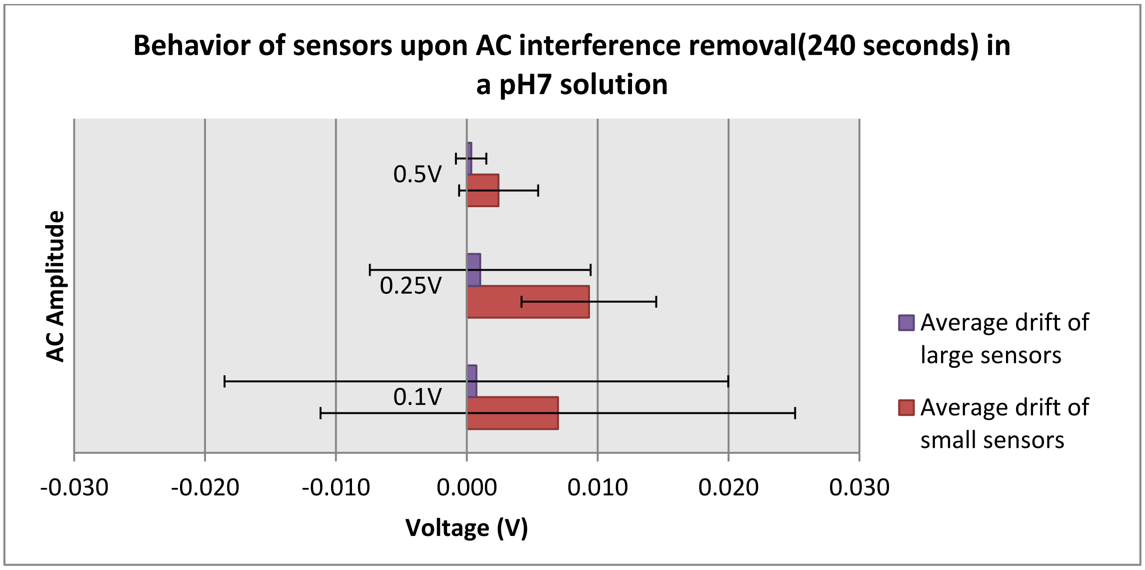

Figure 4.9a: The graph above shows the average drift in voltage and standard deviation of the sensors when AC interference at 0.1V, 0.25V and 0.5V amplitude and constant frequency of 1 kHz is removed at 240 seconds approx. in a pH7 solution.

Conclusion:

Upon careful observation of the above results, the following can be deduced:

- The large sensors are much more stable than the small sensors by basing it on the fact that the average drift values from the initial potential were lower as compared to the small sensors after the effect of AC interference.
- The standard deviation was greater in large sensors than in small sensors as shown in both the pH4 and pH7 solution graphs overall. The large and non-trending standard deviations observed from the above graphs indicates that the sensors have taken different values at the same specified points under the same specified influence of the AC voltage interference. This variation between the sensors has led to inconsistent readings which inhibits reasonable discussion.

NOTE: The conclusions mentioned above cannot be relied upon due to the slow recovery times of the sensor and other factors which played a crucial role in determining the output which are discussed in detail in the next chapter.

In the next chapter, all the probable reasons and a detailed review of the results are included.

4.4: Cumulative effect testing analysis

Since the readings obtained in the amplitude testing analysis were inconsistent, it was imperative to conduct further study on the sensor so that the activities occurring on the sensor interface could be found. This would be helpful in deciding if the sensor has some limiting factors in suffering AC interference progressively. The detailed methodology is given in the previous chapter under section 3.2.5.

The results are depicted below in the form of a graph:

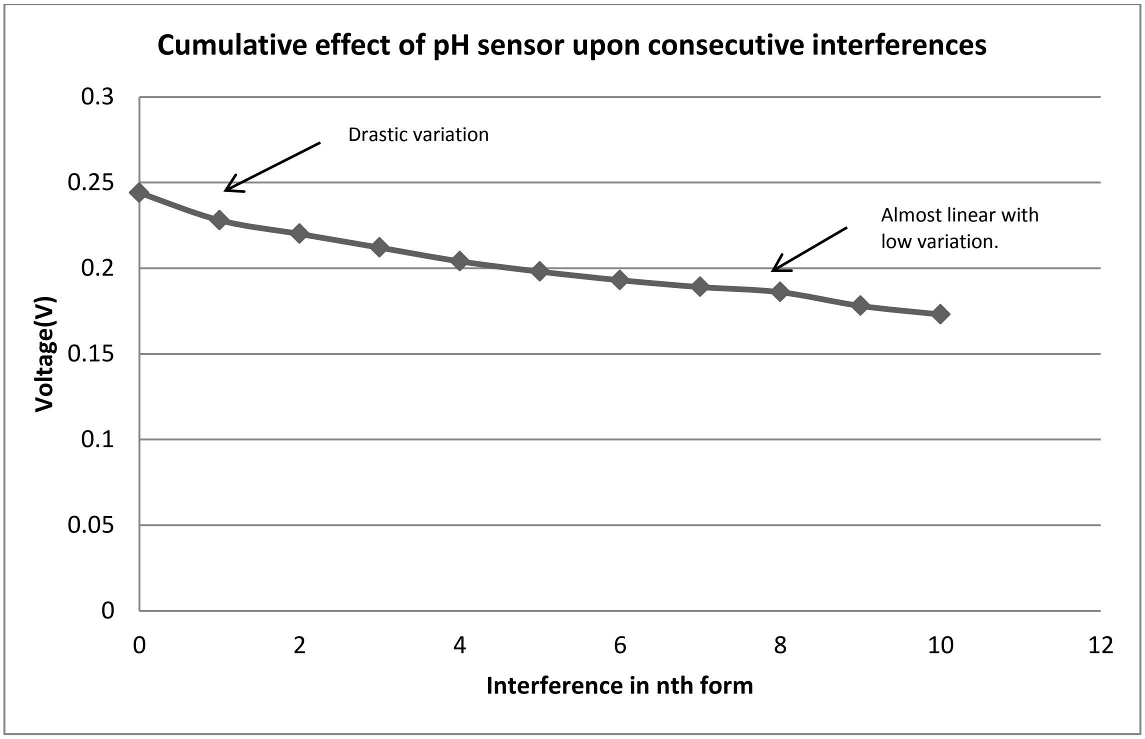

Figure 4.1b: The graph above depicts the falling voltage levels between consecutive introduction of AC interference. It is evident from the graph that the difference decreases as the sensor is subjected to consecutive AC interference.

The cumulative effect study showed interesting results wherein the sensor showed drastic variation in the initial consecutive interferences but as the number of number of interferences increased, the sensor almost became linear indicating no much voltage drop from the previous voltage values.

Conclusion:

It can be deduced from the results obtained that there is a limiting factor on the sensitivity of the sensor with increasing interference consecutively and probably it is due to the membrane changes in the sensor. A detailed discussion and review is included in the next chapter.

4.5: Calibration testing analysis

To obtain a solid understanding of the drifting characteristics of the sensor further, the calibration testing was conducted. This was mainly to determine the shift in the voltage levels in pH4 and pH7 solutions before and after AC interference. This would help in determining if the sensitivity of the sensor is lost or not after the first interference. The detailed procedure for conducting the experiment is included in the previous chapter under 3.2.6.

The calibration testing on the sensor exhibited positive results wherein the drifting of the sensor between the pH 4 and pH 7 before the interference and after the interference was constant.

The illustrations supporting the analysis are shown below:

Drift per pH before interference in (V)	Drift per pH after interference in (V)
0.044	0.044

Figure 4.4a: The table above shows the drift per pH before the interference and after the interference and it is evident that there is no variation and the drift is constant.

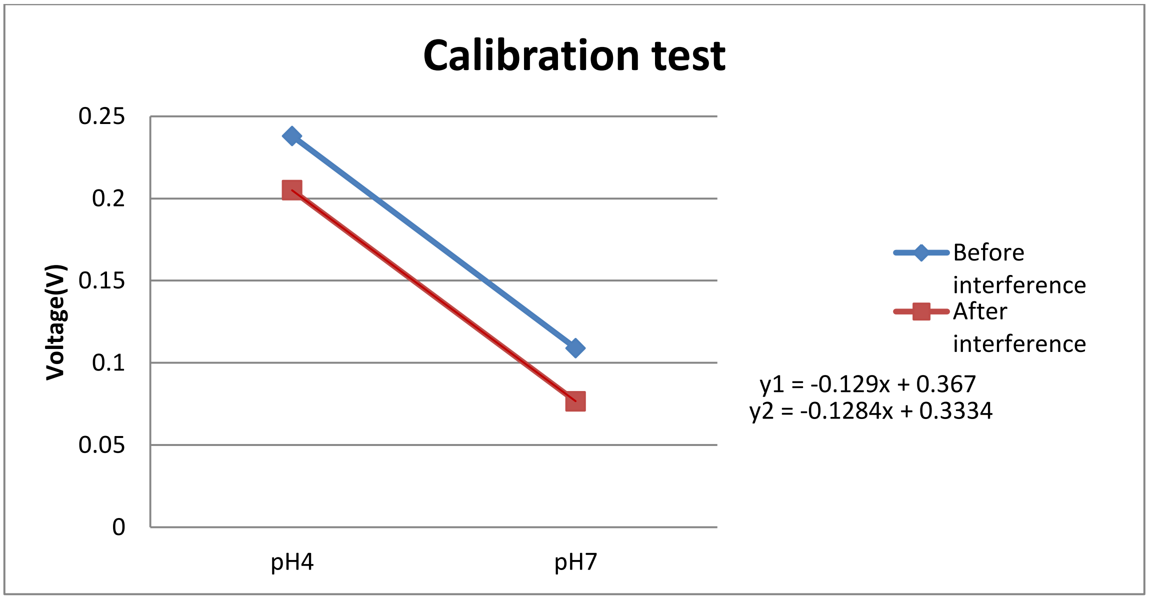

Figure 4.2b: The graph above depicts the voltage levels between the pH4 and pH7 solutions before interference and after interference. It is evident from the graph that the drift is constant and the slope values are also similar with a value close to -0.129V/ decade change in hydrogen ion concentration.

Conclusion:

It is evident from the above results that the sensor does not lose its sensitivity after the first interference and maintains its calibration characteristics. Further details and review of the results are discussed in the next chapter.

Chapter 5: Discussion

Having obtained the experimental results which help in determining the behavior of the sensor upon AC interference, it is now essential to discuss the results in a more elaborate way by utilizing the already existing theoretical concepts of the sensors and electrochemistry in general and bringing out the most probable reasons for such behavior as observed in the previous chapter.

There were five analysis done in total namely voltage potential analysis, frequency analysis, amplitude analysis, cumulative effects analysis and finally the calibration testing analysis. Each of the analysis are discussed and reviewed in detail in the following contents.

5.1: Tests analysis:

5.1.1: Voltage potential analysis:

This experiment was conducted to observe the voltage potential response of the sensors upon immersion in pH solutions each with a different pH concentration. This experiment also allows to sample all the ideal pH sensors and discard the defective sensors. This experiment was significant also in determining the voltage per decade change in hydrogen ion concentration because the potential developed on the sensor over a range of pH from 4 to 10 needs to follow a linear characteristic graph due to the linearly varying concentrations of hydrogen ion concentrations from pH 4 to pH 10.

Key observations:

1. The voltage levels obtained depending on the pH concentration in the solution showed a linear trend in the values measured and a negative slope of -0.052V/decade change in hydrogen ion concentration was recorded in small sensors and a -0.0513V/decade change in hydrogen ion concentration was recorded for large sensors.

2. The pH sensors attained quick stability while measuring the potentials of the pH solutions and did not show any major deviations in their measurements.

Discussion:

1. As observed from this experiment that the sensors were working well and there was no drastic variation in their measurement abilities, it can be said that the pH sensors were stable and the electrode –electrolyte interactions were taking place normally as illustrated in the chapter 2 under the ion-selective membrane section.

2. The theoretical slope of the same experiment mentioned above is 0.059V/decade change in hydrogen ion concentration at 25^0 C which has been obtained from the Nernst equation detailed in chapter 2 under 2.2.4.The equation is once again re-written below. The value obtained is close to the theoretical value and is slightly different due to the temperature variations in which the experiment was done.

$$E = E_0 + \frac{RT}{nF} \text{ In (Ion conc.)}$$

5.1.2: Frequency testing analysis

This experiment tests the behavior of the pH sensor upon with AC voltage interference whose frequency is varied at specific times keeping the amplitude constant. As discussed earlier in the chapter 2 the experiment allows to find the effect of different frequencies effect on the pH sensor working and thus would let conclude the ideal working frequency while using the sensor in real life applications.

Key observations:

1. At low frequency (10 Hz in this experiment), the sensor was drastically fluctuating from the instant when the AC interference was introduced at 120 seconds approx. till the removal of the AC interference at 240 seconds approx.

2. At high frequency (1000 Hz in this experiment), the sensor exhibited minimal fluctuations unlike the 10Hz frequency result from the time AC interference was introduced (120 seconds approx.) till the time the AC interference was removed(240 seconds approx.).

Discussion:

1. The behavior exhibited by the sensor upon conducting the above experiment is explainable when relating to the theory detailed in chapter 3.

It is known from chapter 2 under section 2.2 that a sensor when dipped in an electrolyte faces changes ahead of it such as the double layer formation and also the ion-electron conduction property needs to be ideal with large current densities to exhibit accuracy in measurement. The sensor needs to have a polarised double layer and a non polarised ion-electron conduction between the ion selective membrane and the solid contact(in this case the carbon contact). Relating that theory to the experiment it can be said that the sensor’s double layer formation and the ability of the sensor to polarize or not plays a very crucial role in reasoning such a behavior.

2. When the AC voltage interference is introduced at a low frequency as 10 Hz with the amplitude factor being fixed, the double layer in the sensor/electrolyte interface characteristics are altered by varying the capacitance due to charge variation constantly with change in polarity and external interference of AC field and this happens for a long duration since the AC waves take 0.1 seconds to complete one cycle in this case and this is a long time. This time suffices the sensor to suffer changes longer and thus exhibit such drastic variations.

3. In the case of the 1000Hz frequency interference, the double layer in the sensor/electrolyte interface characteristics are altered for a very short duration since the AC waves take 0.001 seconds to complete one cycle and this is a very short time. This short duration of alternating signal does not give sufficient time to the sensor to exhibit the large variations due to the decreased charging time and the same reactions mentioned above in point 2 occur rapidly.

5.1.3: Amplitude testing analysis:

Another important parameter which characterizes an AC voltage apart from its frequency is its amplitude. To determine the effect this amplitude parameter has on the pH sensor, it is essential to conduct an experiment with varying amplitudes and a fixed frequency. It must be noted that the moisture sensor utilising the AC voltages operates at a fixed frequency. Hence this experiment was conducted as detailed in the chapter 2.

Key Observations:

1. The readings recorded were inconsistent and no proper conclusion could be derived from it. The average drift in potential between the sensors and the specific amplitude variation did not follow any reasonable trend.

2. The standard deviation of the potential drift in the sensors was drastically fluctuating from being low to high again without following any trend in the behavior.

3. The sensor took quite a long time (may be around 4-5 hours) to recover back to its initial position after turning off the AC interference. When studying the sensors on the whole, it was found that the large sensors were much stable than the small sensors with ability to attain quick stability and show minimal fluctuations as detailed in the chapter 3 under 3.5.

Discussion:

1. First of all it is essential to explain the behavior of the sensor when it is subjected to the AC voltage interference. As mentioned in chapter 2 under section 2.2 which has detailed all the theoretical aspects of the experiment, the double layer and the polarizing ability of the region around the sensor play a crucial role in determining the potential developed for measuring the pH concentration in the solution.

Another most important point to be noted is that the reasons explained below cannot be supported or relied upon and are just probabilities that may be occurring in the sensor because of the event of the sensor not recovering back to its initial value after the influence of AC interference is turned off while conducting the experiments and the readings obtained in this way make the analysis incomplete and without any strong base. This may be a strong reason for obtaining the inconsistent readings.

2. Relating to the theory, it can be said that when the AC interference is introduced on the sensor with a fixed frequency (1kHz in this experiment) and only varying the amplitude, the ISM (Ion Selective Membrane) along with the ionophores may be getting disoriented due to the magnitude of the AC voltages. The ionophores are neutral in charge so the membrane is one component which may have been affected. Since in this case, only the amplitude is the varying factor, it implies that the magnitude of the AC interference field in the pH solution is varied and the ability of the magnitude to cause disorientation of the sensor surface and eventually alter its conduction depends on the value of the magnitude.

3. The attraction of the specific ions (hydrogen ions in this experiment) towards the sensor are disturbed due to the external higher electric field which pulls out the ions from entering the sensor region and since it is the AC interference, the polarity of the external field alternates at each time and this pushes the ions towards the sensor region and thus we obtain an alternating potential developed on the sensor depending on the frequency of the AC interference.

4. The pH sensors have showed a long time to recover to its initial state indicating the intense effect the AC interference has on the sensor. The long recovery time may be due to the disorientation of the molecular structure of the ISM and the ionophore as detailed in chapter 2 under on the sensor surface which takes such a long time to recover to its original molecular structure.

5. The delay in recovery time of the sensor is not a permanent change and is a temporary change as the sensors upon leaving it for a long time did recover to their normal values.

5.1.4: Cumulative effect testing analysis

The cumulative effects analysis was conducted as a part of studying the phenomena occurring in the region of the double layers and on the surface of the sensor and also to study the effects on the sensor upon AC interference further.

Key Observations:

1. Looking at the graph illustrated in chapter 4 under 4.4, it can be said that the first few AC interferences drift the sensor potential largely while the later interferences appear to give almost linear potential values indicating the proximity between the potential drifts after the AC interference introduction.

Discussion:

1. The behavior exhibited by the sensor can be explained with the help of the theory detailed in chapter 2 under. Initially the sensor behaves normally by developing the potential according to the pH concentration in the solution. Now the AC interference is introduced to the sensor and removed after 120 seconds. The potential developed by the sensor after this process is much lower than the initial value indicating a large drift. Upon repeating this process of introducing and removing the AC interference on the sensor, it is observed that the change in potential is reduced considerably wherein the line in the graph almost becomes horizontal as illustrated in chapter 4.

2. The reason for such a behavior exhibited by the sensor as stated above may be due to the persistency in the changes of the ISM after removal of the AC interference inside the sensor. These changes in the ISM are regarding the process of disorientation of the molecular arrangement and the ionophores[20] after the initial AC interference and eventually leading to diminished effects upon consecutive AC interferences. These changes and disturbances of the sensor are absent before the initial AC interference. Hence the initial voltage drift is large compared to the following interferences.

5.1.5: Calibration testing analysis:

To make the study of AC interference on the pH sensor more comprehensive, the above mentioned analysis was conducted. This analysis as stated earlier in previous chapters is to check if the sensitivity of the pH sensor is lost upon AC interference. It also helps in checking the re-calibration property of the sensor along with the effect of the variation of voltage potential on pH sensitivity.

Key Observations:

1. The sensor exhibited a drift condition wherein the potential developed on the sensor in both the pH 4 and pH 7 solutions detailed in chapter 3 under 3.1.1 before and after the AC interference was the same. This indicates that the sensitivity of the sensor is intact and the changes that occur on the sensor in a particular pH solution is carried over and reflected in the other pH solutions with a constant drift from the initial conditions of the sensor.

Discussion:

1. The explanation for the observations stated above may be due to the stability achieved by the sensor with the new voltage potential on its surface after the AC interference has been introduced. The main changes that occur on the sensor surface as detailed in the above contents regarding the disorientation of molecular structure of the conducting polymer and the charge distribution etc. may be taking a long time to recover due to the slow reactions and inertness of the material involved due to which the measurements taken follow a linear trend as illustrated in chapter 4 under 4.5.

5.2: The Moisture sensor

Since the project deals with the study of the effects of the AC voltage interference on a pH sensor to ultimately integrate with a moisture sensor, it is very essential to know the processes occurring in the moisture sensor as well. The moisture sensor may also be facing with the same phenomena occurring near its surface as that of the pH sensor but the difference is that the moisture sensor is an impedance based sensor which introduces small amplitude AC voltages and receives the resulting current from the test solution in real life applications while the pH sensor is a potentiometric sensor capable of only giving out the potential corresponding to the concentration of the specific ion in a solution.

In this experiment, the main role of the moisture sensor is to introduce the AC interference only without getting any feedback. The characteristics exhibited by the moisture sensor while conducting the experiments is detailed below.

Key Observations:

1. When the moisture sensor carrying the AC interference was immersed for the first time in the pH solution, it was observed that the pH sensor suffered a drop in potential as usual but the stabilizing time was prolonged and took a considerable time. Further, upon removal of the moisture sensor, a surface coating on one of the electrodes of the two was observed.

2. After the above processes took place and when then the moisture sensor was reused to resume with the experiment, the recording was normal as usually obtained in the rest of the experiments.

Discussion:

1. The reasons to explain for this coating are unknown and should be looked into further as this may have caused the prolonged stabilizing time of the pH sensor at the first instance of use.

2. Most probable reasons may include the high affinity of the hydronium ions [H_3O^+] to react with one of the materials in the moisture sensor or the oxidation of AgCl or the formation of molecules within the solution on the interface which takes some time to coat it fully.

An illustration to support the output obtained upon using a new moisture sensor at the first instance is given below as a graph:

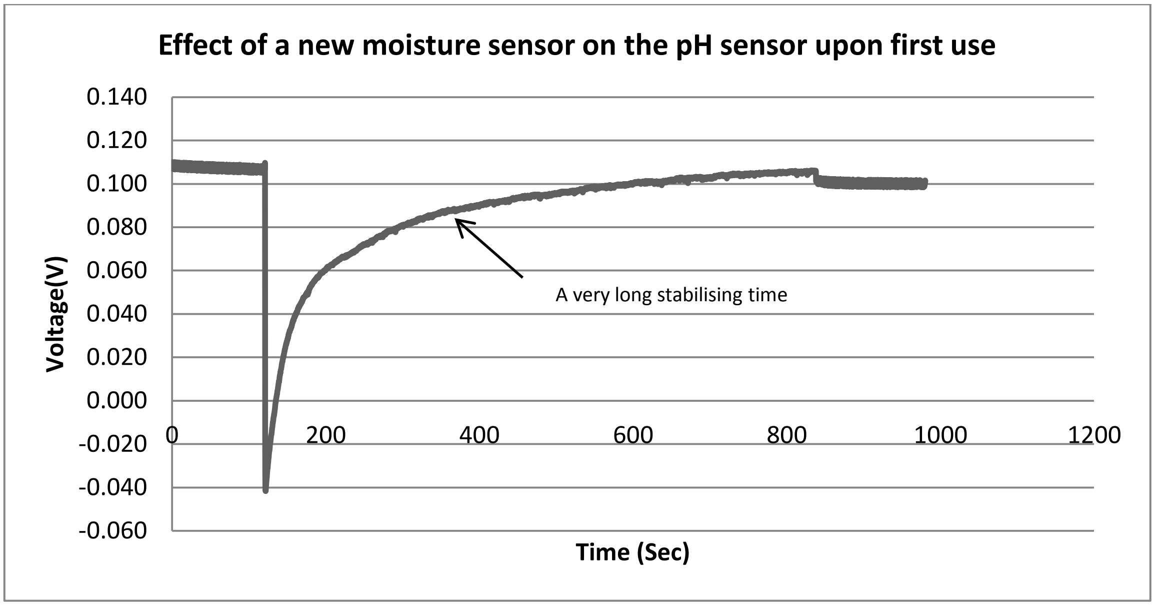

Figure 5.1a: The graph above illustrates the prolonging stabilising time upon the use of a new moisture sensor at the first instance.

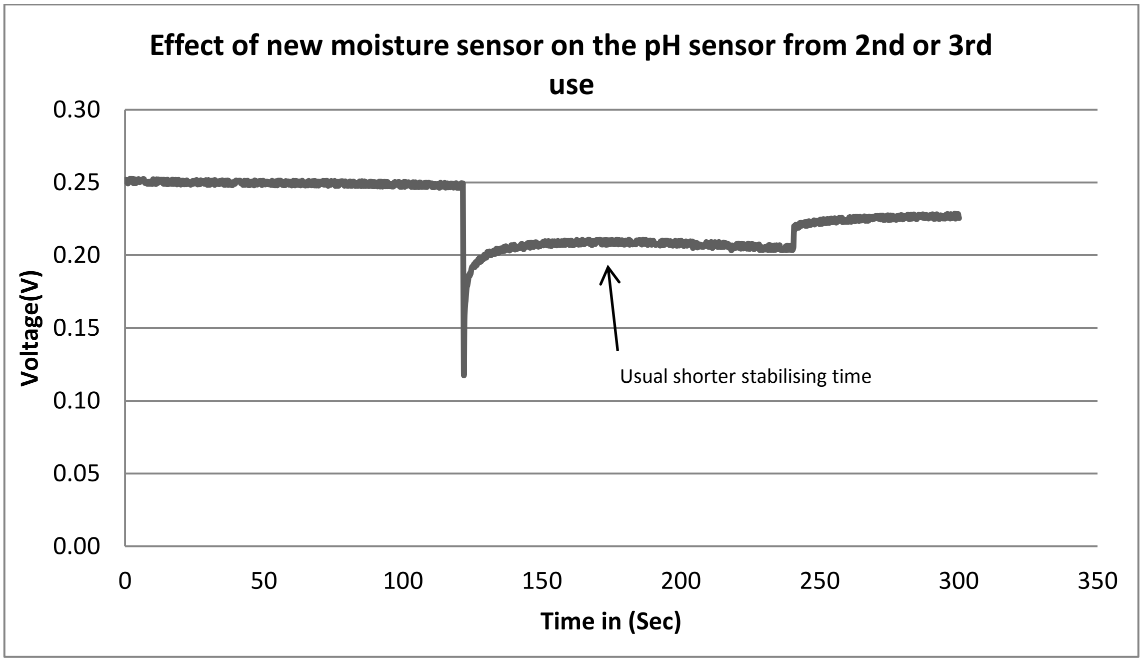

Figure 5.2a: The graph above illustrates the usual stabilising time of the pH sensor upon use of the new moisture sensor after the 2nd or 3rd time.

5.3: Conclusion

After having conducted the experiments to study the behavior of the pH sensors upon subjecting it to the AC voltage interference, the following conclusions have been made:

1. The sensor has shown results that suffice to prove the influence under the action of AC interference in most of the analytical experiments except for the amplitude testing analysis wherein the inconsistent readings were obtained with which no proper conclusions could be arrived at.

2. Based on the other results obtained, it can be concluded that the sensor would get effected lesser at higher frequencies rather than at low frequencies despite it getting effected by all the frequencies.

3. The voltage drift upon consecutive introduction of AC interferences on the pH sensor showed results that indicate the decreasing effect of the AC voltage on the sensor upon continuous subjection.

4. The sensitivity of the sensor is not lost after the first AC interference and remains intact. This also helps in concluding that the pH sensor can be re-calibrated easily to provide accurate response to the pH.

Since the pH sensor is a disposable sensor, the readings obtained favor the use of the sensor for real time wound care applications but further research needs to be done to overcome the inconsistency of readings obtained in the amplitude testing analysis.

5.4: Integration of the sensors based on the results obtained

As far as the integration of the pH sensor and the moisture sensor is concerned, the pH sensor does get influenced fairly under the action of AC interference but would exhibit effective sensing provided it gets re-calibrated. The results obtained from the cumulative effects analysis and the calibration test analysis favour the application of the sensor owing to its intact sensitivity and re-calibration properties.

5.5: Future Work

Since this experimental project is an introduction to study the effects of the AC interference on the pH sensor, it can be used to improvise the analysis by studying the reasons for the inconsistency in the amplitude analysis and the experiments in the future may be conducted by allowing the sensors to recover fully despite it taking a long time in order to study the effect of the crucial parameter which may give considerable support in integrating the moisture and the pH sensor for optimum objective analysis of the wound healing rates.

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Appendix:

Procedure for universal buffer solution

Revision	Author	Description	Date
3.0Final	Stephen Milne	Original document	26/10/09

1. Introduction

This details the method for producing 1 litre of universal buffer solution. The solution can be made into a range of pH levels between pH 2-11. The ionic strength varies between 0.07-0.1.

2. Equipment

Description	Qty
1 litre glass bottle	1
200ml glass bottles	7
Magnetic stirrer	1
1 litre beaker	1
Fume hood	1
Weighing machine in fume hood	1
pH meter	1

3. Materials

Description	**Supplier**	**Qty**
Acetic acid		0.04M solution 2.3ml per 1litre water
Boric acid (H_3BO_3)	B6768	0.04M solution 2.4732g per 1litre water
Phosporic acid	04102	0.04M solution 2.74ml per 1litre water
NaOH	S5881	Xml 0.2M solution 7g in 875ml water
Distilled water		1 litre

4. Method

To be completed in fume hood:

1. Pour 900ml of distilled water into bottle
2. Dissolve the following in the distilled water:

   2.3ml of Acetic acid,2.74ml of Phosporic acid,2.4732g of Boric acid

3. Ensure the contents are mixed
4. Make up bottle to 1litre using distilled water.

To be completed on the workbench:

5. To make 0.2M NaOH solution weigh out 7g
6. Fill beaker with 875ml distilled water
7. Add the 7g of NaOH to the beaker

8. Stir until dissolved.

9. To get correct pH levels take 1litre of the buffer solution and add Xml of sodium hydroxide as shown in the table below to get suitable pH level.

pH at 18C	**X ml 0.2M Sodium hydroxide**
4.1	250
5.02	105
6.09	119.3
7	87.4
7.96	57
8.95	46.35
9.91	41.8

10. Mix with magnetic stirrer and test for pH when adding the 0.2M NaOH to get accurate pH level. Once pH level has been made pour 200ml into bottle and continue titration.

11. Store in marked 200ml glass bottles

5. Storage

Store in marked glass bottles for use

6. Safety

Ensure Boric acid is weighed in fume hood with fan turned off. Wear a P2 or better dust mask when dealing with powder.

For chemical risk see coSHH form.