FABRICATION OF PORTABLE CHEMICAL SENSOR FOR HIGHLY SELECTIVE THIOCYANATE DETECTION BASED ON Co(II) PORPHYRIN COMPLEX

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FABRICATION OF PORTABLE CHEMICAL SENSOR FOR HIGHLY SELECTIVE THIOCYANATE DETECTION BASED ON CO(II) PORPHYRIN COMPLEX

ABSTRACT

Fabrication of portable chemical sensor for highly selective thiocyanate (SCN) detection based on Co(II) porphyrin complex with self-plasticizing membrane involves 5,10,15,20-tetrakis-(4-methoxyphenyl)-21H,23H-porphyrin Co(II) or CoTMeOPP as the ionophore, n-butyl acrylate (n-BA) and 2-Hydroxyethyl methacrylate (HEMA) self-plasticizing polymers as the membranes, tetradodecylammonium chloride (TDDA) as additive and glassy carbon electrode (GCE) as the transducer. This ionophore was proven to diverge from the classic Hofmeister theory, have highly selectivity towards thiocyanate anion and stable to work with the self-plasticizing membrane. The new optimized potentiometric self-plasticizing of CoTMeOPP highly selective complexation with SCN^- has been studied by using potentiometer and the stoichiometric of the complexation reactions of $Co^{2+/3+}$ and SCN^- is 2:1 [$Co^{2+/3+}$: SCN^-]. The optimum membrane composition was obtained by using silver/silver chloride (Ag/AgCl) as the first layer, 95.14 wt.% monomer (HEMA) and 4.86 wt.% photo initiator (DMPP) as the second layer and 98.86 wt.% monomer (n-BA), 0.08 wt.% photo initiator (HDDA), 0.34 wt.% ionophore (CoTMeOPP), 0.16 wt.% lipophilic salt (TDDA) and 0.56% photo initiator (DMPP). The fabricated SCN-ISE was tested with standard solution of SCN^- and exhibits a good Nernstian behavior with the slope of -60.76 mV/decade and detection limit of 9.55×10^{-8} mol/L over a wide concentration range of 1.0×10^{-7} mol/L to 1.0×10^{-1} mol/L. The fabricated sensor displayed wide working pH range from 2 to 11, short response time (68 seconds), low resistance and excellent selectivity relative to a wide variety of other anions, good repeatability and reproducibility. The fabricated electrode can be used over a period of seven days provided kept in refrigerator without itself showing any significant drift in working concentration range, slope and response time. The fabricated SCN-ISE sensor was successfully applied for the determination of thiocyanate in real samples with satisfactory recovery ranges i.e more than full percentage.

Keywords: Self-plasticizing polymeric membrane; Thiocyanate; Glassy carbon electrode; Sensor.

FABRIKASI PENDERIA KIMIA MUDAH ALIH BAGI MENGESAN KETERPILIHAN TINGGI TIOSIANAT BERDASARKAN KOMPLEKS Co(II) PORFIRIN

ABSTRAK

Pemfabrikan penderia kimia mudah alih bagi mengesan keterpilihan tinggi tiosianat (SCN) berdasarkan kompleks Co(II) porfirin bersama polimer mempolimerasi sendiri melibatkan 5,10,15,20-tetrakis- (4-metoksi fenil) -21H, 23H-porfirin Co (II) atau nama ringkas nya CoTMeOPP sebagai ionofor, n-butil akrilat atau dengan singkatan n-BA dan juga 2-hidroksietil metakrilat atau dikenali sebagai HEMA polimer mempolimerasi sendiri sebagai membran, tetradodesilammonium klorida sebagai bahan tambahan dan elektrod karbon kaca (GCE) sebagai transduser. Ionofor ini telah terbukti mencapah dari teori klasik Hofmeister, mempunyai keterpilihan yang tinggi terhadap anion tiosianat dan stabil untuk digunakan bersama polimer mempolimerasi sendiri. Reaksi tindak balas tersebut telah dikaji dengan menggunakan potensiometer dan hasil dari kajian, didapati stoikiometri daripada tindak balas pembentukan kompleks antara $Co^{2+/3+}$ dan SCN^- adalah 2: 1 [$Co^{2+/3+}$: SCN^-]. Komposisi membran optimum diperoleh dengan menggunakan argentum/argentum klorida (Ag/AgCl) sebagai lapisan pertama, 95.14 wt.% monomer (HEMA) dan 4.86 wt.% pemula foto (DMPP) sebagai lapisan kedua dan 98.86 wt.% (n-BA), 0.08 wt.% pemula foto (HDDA), 0.34 wt.% ionofor (CoTMeOPP), 0.16 wt.% garam lipofilik (TDDA) dan 0.56 wt.% pemula foto (DMPP). SCN-ISE yang difabrikasi telah diuji dengan menggunakan larutan piawai SCN^- anion dan menunjukkan tindak balas Nernstian yang baik dengan kecerunan -60.76 mV/dekad, had kepekatan pengesanan yang rendah iaitu serendah 9.55×10^{-8} mol/L dengan julat kepekatan bekerja yang luas iaitu antara 1.0×10^{-7} mol/L hingga 1.0×10^{-1} mol/L. Penderia yang difabrikasi mempamerkan kebolehan untuk bekerja dengan julat pH yang luas iaitu dari 2 hingga 11, masa tindak balas yang pantas (68 saat), rintangan yang rendah dan kepilihan anion yang sangat baik berbanding dengan pelbagai anion lain, keterulangan dan kebolehulangan yang baik. Elektrod yang dicadangkan juga boleh digunakan dalam tempoh tujuh hari jika disimpan di dalam peti sejuk tanpa menunjukkan sebarang bacaan hanyut yang besar dan berfungsi dalam julat kepekatan kerja, cerun dan masa tindak balas yang boleh diterima sepanjang tempoh 7 hari tersebut. Penderia yang telah difabrikasi telah berjaya digunakan untuk penentuan tiosianat dengan menggunakan sampel sebenar dengan julat pemulihan yang memuaskan iaitu melebihi peratusan penuh.

Kata kunci: Polimer mempolimerasi sendiri; Tiosianat; elekrod karbon kaca; penderia.

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LIST OF SYMBOLS AND ABBREVIATIONS

%	Percentage
α	Alpha
β	Beta
γ	Gamma
δ	Delta
π	Pi
(FeTPP)$_2$O	Oxo bridge iron porphyrin
µL	Micro litre
2-NPOE	2-nitrophenyloctylether
AAB15C5	4-acryloylamidobenzo-15-crown-5
AAB18C6	4-acrylotlamidobenzo-18-crown-6
AAS	Atomic absorption spectroscopy
Ag	Argentum (Silver)
AgCl	Silver chloride
AgNO$_3$	Silver nitrate
AISAL	Aluminium (III) 3,5-ditert-butyl-salophene
AITAP	Chloro-2,7,12,17-tetra-tert-butyl-5,10,15,20-tetraazaporphine aluminium (III)
Al($^{2+/3+}$)	Aluminium ($^{2+/3+}$)
Al(II/III)	Aluminium (II/III)
Al$_2$O$_3$	Aluminium oxide
Au	Gold
AU-1	N,N-Dicyclohexyl-N′-phenyl-N′-3-(2-propenoyl)oxyphenyl-3-oxapentanediamide

BTCH	1,2-bis-(N'-benzoylthioureido cyclohexane)
Ca^{2+}	Calcium cation
Ca-ISE	Calcium-Ion Selective Electrode
CGE	Coated graphite electrode
Cl^{-}	Chloride anion
ClO_4^{-}	Perchlorate anion
CN	Cyanide
CN^{-}	Cyanide anion
$Co(^{2+/3+})$	Cobalt $(^{2+/3+})$
Co(I)	Cobalt (I)
Co(II/III)	Cobalt (II/III)
$Co(O\text{-}NH_2)TPP$	Cobalt (II) tetrakis (o-aminophenyl)porphyrin
CoOBTAP	2,3,7,812,13,17,18-octakis(benzylthio)-5,10,15,20-tetraazaporphyrin Cobalt (III)
CoTMeOPP	5,10,15,20-tetrakis-(4-methoxyphenyl)-21H,23H-porphyrin Cobalt (II)
CP	Carbon paster
CPE	Carbon printed electrode
$Cu(^{2+/3+})$	Copper $(^{2+/3+})$
Cu(II/III)	Copper (II/III)
CV	Cyclic Voltammetry
DBBP	Dibutyl butyl phosphate
DBP	N-butylphthalate
DBS	Dibutyl sebacate
DDS	Dodecyl Sebacate
DMA	Decylmethacrylate

DMPP	Dimethylphenylpiperazinum
DOP	Dioctylphthalate
DPV	Differential Pulse Voltammetry
e	electron
e.m.f	Electro magnetic field
E_{cell}	Cell potential (standard condition)
E°_{cell}	Cell potential (non-standard condition)
ETH-70750	4,5-Dribromofluoresceinoctadecylster
F	Faraday’s constant
F^-	Fluoride anion
Fc-MH	Ferrocene poly (methyl-methacylate-(Co-2-hydroxylehtyl methacrylate)
$Fe(^{2+/3+})$	Ferum $(^{2+/3+})$
Fe(II/III)	Ferum (II/III)
FeOBTAP	2,3,7,812,13,17,18-octakis(benzylthio)-5,10,15,20-tetraazaporphyrin Ferum (III)
FESEM	Field Emission Scanning Electron Microscope
FIA	Flow Injection Analysis
F-ISE	Fluoride-Ion Selective Electrode
FT-IR	Fourier Transform Infra-red Spectroscopy
$Ga(^{2+/3+})$	Gallium $(^{2+/3+})$
Ga(II/III)	Gallium (II/III)
GC	Glassy Carbon
GCE	Glassy Carbon Electrode
H^+	Hydroxide cation
HCl	Hydrochloric acid

HDDA	1,6-haxanedioldiacrylate
HEMA	(Hydroxyethyl)methacrylate
Hg	Mercury
Hg-ISE	Mercury-Ion Selective Electrode
HPO_4^{2-}	Phosphate anion
HTAB	Hexadecyl trimethyl ammonium bromide
I^-	Iodide anion
i.e	That is
I-ISE	Iodide-Ion Selective Electrode
$In(^{2+/3+})$	Indium $(^{2+/3+})$
In(II/III)	Indium (II/III)
IO_4^-	Periodate anion
ISE	Ion Selective Electrode
ISM	Ion selective membrane
K	Kelvin
K^+	Potassium cation
KCl	Potassium chloride
K-ISE	Potassium-Ion Selective Electrode
KNO_3	Potssium nitrate
KSCN	Potassium thiocyanate
L	Litre
LiOAc	Lithium acetate
MH	poly (methyl-methacylate-(Co-2-hydroxylehtyl methacrylate)
min	Minute(s)
MMA	Methyl methacrylate
$Mn(^{2+/3+})$	Manganese $(^{2+/3+})$

Mn(II/III)	Manganese (II/III)
Mn(II/III)	Manganese (II/III)
Mn(III)-salophen	2,2'-[(4,5dimethyl-1,2-phenylene) bis[(E)-nitrilomethylidyne]] bis [4,6-bis (1,1-dimethylethyl) Manganese(III)
Mn(TPPP)Cl	5,10,15,20-tetrakis(2,4,6-triphenyl)porphyrinato-manganese (III) chloride
Mo(V)	Molybdenum (V)
mol	Mole
mol/L	Mole per litre
MoO_4^{2-}	Molybdate ion
MPM	Matched potential method
mV	mili volt
n	Number of moles of electron
N	Nitrogen
N_2	Nitrogen gas
N_3^-	Azide anion
Na^{2+}	Sodium cation
Na-ISE	Sodium-Ion Selective Electrode
NaOH	Sodium hydroxide
NaTPB	Sodium tetraphenylborate
n-BA	N-butyl acrylate
NH_3	Ammonia
NH_4^+	Ammonium cation
Ni($^{2+/3+}$)	Nickel ($^{2+/3+}$)
Ni(II/III)	Nickel (II/III)
nm	Nano meter

NO_2^-	Nitrite anion
NO^{2-}	Nitrate anion
OBTAP	2,3,7,812,13,17,18-octakis(benzylthio)-5,10,15,20-tetraazaporphyrin
OH^-	Hydroxide anion
o-NPOE	2-Nitrophenyl octyl ether / o-nitrophenyloctylether
PANi	Poly-aniline
PCTFE	Polychlorotrifluoroethylene
$Pd(^{2+/3+})$	Paladium $(^{2+/3+})$
Pd(II/III)	Paladium (II/III)
PEDOT	poly(3,4-ethylenedioxythiophene)
PME	Polymeric membrane electrode
POPs	Persistent organic pollutants
PSS	polystyrene sulfonate
Pt	Platinum
*p*TSC	*Para*-tolualdehydesemicarbazone
PU	Polyurethane
PVC	Poly(vinyl) chloride
Q	Reaction quotient
R	Gas constant
Ru	Ruthenium
s	seconds
S^{2-}	Sulphide
SCN	Thiocyanate
SCN-ISE	Thiocyanate-Ion Selective Electrode
SeO_3^{2-}	Selenite ANION

SiO_2	Silicon dioxide
Sn	Tin
SO_4^{2-}	Sulphate anion
SP	Screen printed
SPE	Screen Printed Carbon Paste Electrode
STFPB	Sodium tetrakis (trifluoromethyl)phenylborate
T	Temperature
TBP	Tri-n-butyl phosphate
TCP	Tricresylphosphate
TDDA	Tetra-dodecyle Ammonium Chloride
T_g	Glass temperature
THF	Tetrahydrofuran
$Tl(^{2+/3+})$	Telurium $(^{2+/3+})$
Tl(II/III)	Telurium (II/III)
TOTCl	Tri-n-octyltin chloride
UO_2SAL	uranyl 4,5-naphthyl-salophene
UV	Ultraviolet
UV-vis	Ultraviolet-Visible spectroscopy
V	Voltage
VOC	Volatile Organic Compounds
wt.%	Weight percentage
XRD	X-Ray Diffraction
$Zn(^{2+/3+})$	Zinc $(^{2+/3+})$
Zn(II/III)	Zinc (II/III)
ZrTAP	dichloro 2,7,12,17-tetra-tert-butyl-5,10,15,20-tetraazaporphine zirconium (IV)

LIST OF APPENDICES

CHAPTER 1: INTRODUCTION

1.1 General Introduction

The development of sensor for the detection of ions i.e anion and cations have grown rapidly. In this research, the fabrication of anion sensor based on Cobalt(II) or Co^{2+} ionophore by using glassy carbon electrode (GCE) as transducer with the aid of self-plasticizing polymeric membrane was conducted. There are many reported sensors that are highly selective towards ions that have been successfully fabricated and manufactured. Amongst the many ions i.e cations and anions presence in the environment, thiocyanate or SCN anion has been chosen to be the target anion. The SCN anion was chosen as the target anion owing to its various applications in the industries including its involvement in printing, dyeing, acrylic fiber production, herbicide, insecticide as well as solvents, catalysts, additives and pigments (Namasivayam & Prathap, 2006; Zhang et al., 2007). This SCN anion has been proven to prevail in the environment mainly due to untreated chemical wastes from the industries. The breakage of this anion to the environment causes many drawbacks especially to human. Elevated amount of SCN anion in the body can cause severe health problems such as hypothyroxinemia, chronic liver, kidneys and muscle cells destruction, delay in neurological system and may cause delay growth in infants. SCN anion also was found in dairy products, human milk and significant amount can be found in smokers' saliva. Thus, the detection of SCN anion in the environment becomes very crucial in improving the health problems for future generations as well as improving the quality of the environment.

Since the concentration of SCN anion presence neither environment nor human was never found to be high, SCN anion still presence in the environment and human in traces amounts. There are various detection methods reported for the detection of SCN anion in trace amounts. The methods including spectrometry method (Hovinen et al., 1999), amperometry (Cookeas & Efstathiou, 1994; Wang et al., 2004), head-space gas chromatography (Lee et al., 2003), Aldrige method (Pettigrew & Fell, 1972) , anion exchange resins (Kononova et al., 2007), ion chromatography (Michigami et al., 1992), potentiometry (Ni & Wu, 1999; Zamani et al., 2006) , polarography (Brown et al., 1995; Cai & Zhao, 1988), high performance gas and liquid chromatography (Brown et al., 1995), capillary zone electrophoresis (Tanaka et al., 2004), ferric nitrate reagent method (Bowler, 1944), spectrofluorometry (Afkhami et al., 2007), UV-vis spectrometry (Hovinen et al., 1999), Raman spectroscopy (Pienpinijtham et al., 2011), electrochemistry (Ozoemena & Nyokong, 2005), photokinetic voltammetry (Li et al., 2000), atomic absorption spectrometry (AAS) (Chattaraj & Das, 1992) and fluorescence (Zhang et al., 2007). Despite the low detection limit and wide woking range of the reported works, these methods involve costly instruments, require sophisticated operating skills, essential to be conducted at the laboratory, time consuming, tedious, and do not suitable for in place testing.

Thus, one of the methods that is commonly used in developing sensor that resolve all the problems with the prior methods that is electrochemical sensor was studied. There are three types of electrochemical sensors which are potentiometric, amperometric and conductometric sensors. In this work, GCE was chosen as the transducer to translate the activities between the ionophore and SCN anion in standard and real samples. GCE was chosen as the transducer to enhance the performance of the sensor as it is very stable and easy to operate. The SCN detection by using this method is cheap, does not need any sophisticated skills to operate, in place detection of target analyte can be done, environmental friendly, very low amount of chemical waste, facile preparation, portable and precise.

Active materials called ionophore have been widely studied as ‘ligand’ or the sensor for the detection of target materials i.e anions, cations and metals. Thus, in this work, an ionophore was utilized as the main and active material in fabricating the SCN-ISE for the detection of SCN anion in the environment. The ionophore chosen in this research is 5,10,15,20-tetrakis-(4-methoxyphenyl)-21H,23H-porphyrin-Co(II) or CoTMeOPP. In order for the ionophore to be fabricated onto the GCE and work at its optimum, polymeric membrane has been introduced as the adherence and supporting material in detecting the target analyte. Previous reported polymeric membrane used in the fabrication of ionophore onto the transducer commonly involve polyvinyl chloride (PVC) polymer which involves plasticizers. The utilization of plasticizer commonly leads to leaching problems and the leaching of plasticizer to the environment involves long term effects as it is highly toxic, harmful to the environment and will eventually shortened the life time of the sensor (Heng & Hall, 2000b). Thus, in this work, fabrication of the ionophore onto the GCE was aided by using self-plasticizing membranes of hydroxyethyl methacrylate (HEMA) and n-butyl acrylate (n-BA) polymers to prevent the drawbacks of utilizing plasticizer in the fabricating of ionophore onto the GCE. The self-plasticizing polymeric membranes of HEMA and n-BA also enhances the performance of the SCN sensor as it is conductive and hydrophobic that helped in the reduction of interference from hydroxide anion (OH^-).

Optimization of the composition of the active materials in the membrane was conducted to obtain the optimum performance and maximize the sensitivity, selectivity and the quality of the fabricated sensor. Other characterizations of the optimized membrane have been conducted to study the morphology, selectivity, sensitivity and the performance of the fabricated SCN-ISE. SCN-ISE has been successfully fabricated and optimized to be highly selective towards SCN anion and have been showed to have high performance when tested with real samples.

1.2 Problem Statement

Thiocyane (SCN) anion has been proven to prevail in the environment as well as in dairy products, human body fluid and serum due to its variety of applications in the industries. The elevated amount of SCN in the body are dangerous for human. Hence, a sensor that is highly selective in detecting SCN in trace amount needs to be manufactured to test the presence of SCN in the environment in trace amount. There are many researches on ISEs that have been reported and most of the reported ISEs are for cations. There are not many ISEs that reported the detection towards anions due to the active properties of the anion molecules. Thus, this research fabricated ISE that offers highly selective detection towards thiocyanate (SCN) anion. ISE have been widely used as a type of sensor in detecting many ions and also have been reported to have interference with other commonly found ions in the environment. Therefore, in this research, an electrode that is higly selective towards SCN anion as well as having relatively low interference towards other anions is fabricated to curb the prior problems of the reported SCN-ISE.

1.3 Objectives of Research

The main focus of this research is to examine the potential of Co(II) porphyrin complex towards the detection of thiocyanate (SCN) anion. The objectives are as follows:

i. To optimize the composition of 5,10,15,20-tetrakis-(4-methoxyphenyl)-21H, 23H-porphyrin Co(II) (CoTMeOPP) ionophore for the detection of thiocyanate anion by using potentiometric method.

ii. To fabricate the portable chemical sensor based on 5,10,15,20-tetrakis-(4-methoxyphenyl)-21H, 23H-porphyrin Co(II) (CoTMeOPP) ionophore using of self-plasticizing membrane.

iii. To characterize the polymeric membrane sensor by several analytical methods such as UV-vis Spectroscopy (UV-vis), Fluorescence Spectroscopy, Fourier-Transform Infrared Spectroscopy (FTIR), Field Emission Scanning Electron Microscopy (FESEM) and Energy Dispersive X-Ray (EDX).

iv. To evaluate the response and selectivity of the fabricated chemical sensor towards thiocyanate anion.

1.4 Scope of Research

This study involves the fabrication and characterisation of CoTMeOPP as ionophore using self-plasticizing polymeric membranes on glassy carbon electrode (GCE) as ISE. The response of particular ionophore with the detection of SCN anion in environmental media such as sediments from ex-mining area, river water, human serum i.e saliva as well as in marine aquatic environment. The characterization of the fabricated ionophore self-plasticizing membrane and its complexes includes the use of Ultraviolet-Visible Spectroscopy (UV-vis), Fluorescence spectroscopy, Fourier-Transform Infrared Spectroscopy (FT-IR), Field Emission Scanning Electron Microscope (FESEM) and X-Ray Diffraction (XRD).

The potentiometric study between CoTMeOPP and the target anion i.e SCN has been conducted. The purpose of this study is to investigate the detection, selectivity and sensitivity of the complexation between the ionophore and SCN anion. The composition of the ionophore self-polymeric membrane were explored to obtain the most suitable ratios for optimum detection of SCN anion in the solution system. The optimized ratio has been discovered and its stability, selectivity, life-time, working range, response time and effect of pH were conducted in separate experiments.

In order to understand the coordination of CoTMeOPP with SCN anion, their corresponding metal complex were prepared and characterized using FTIR, Fluorescence and UV-vis instruments. Finally, the application of the fabricated SCN-ISE was tested for real sample analysis with different types of samples.

1.5 Thesis Outline

This thesis divided into five chapters consist of:

Chapter 1 mentions the main purpose of this dissertation which is to fabricate CoTMeOPP self-plasticizing membrane for the detection of thiocyanate (SCN) anion for developing the novel potentiometric ISEs with enhanced selectivity and sensitivity.

Chapter 2 presents the background of CoTMeOPP and their metal complexes, followed by applications in various studies. The complexation study, potentiometric ISE function and application are also listed.

Chapter 3 describes the methodologies used in the design of the experiments, separated into four main sections. Optimization of the SCN-ISE, fabrications of the self-plasticizing membrane, characterization of the fabricated SCN-ISE and application of the SCN-ISE.

Chapter 4 discusses the successful fabrication and characterization of CoTMeOPP self-plasticizing membrane to detect SCN anion. In this chapter also includes the discussion about significant results such as stoichiometry and stability constant (K_f) obtained from potentiometric study and spectrometric identification of the ionophore and the anion of interest.

Chapter 5 summarizes the main conclusions of this thesis and presents an outlook for future work.

CHAPTER 2: LITERATURE REVIEW

2.1 Ion Selective Electrode (ISE)

Numerous research works have been conducted on the development of sensor with thiocyanate as the target anion based on many different methods. However, the research works on its applications as sensor for detection of trace amount of thiocyanate has yet been well studied, especially the incorporation of ionophore, glassy carbon electrode (GCE) and self-plasticized polymeric membrane to manufacture a highly selective and sensitive ions-selective electrode (ISE). ISE is a type of chemical sensor that indicates the activity of ions in terms of voltage by using the logarithm of its activity according to Nernst equation that will be explained in chapter 3 later on. The ISE sensor in this work is based on potentiometric ISE that plays an important role in sensing application with respect to human, environmental media and food samples. The ISE provides advantages to users i.e it provides rapid responses, high selectivity and sensitivity over a wide dynamic range (Abbaspour et al., 2002). These qualities made ISE a common transducer used in sensors and was chosen to be the transducer in this fabricated thiocyanate sensor.

In the fabrication of ISE electrode, the choice of ionophore plays a vital role to ensure the selectivity and sensitivity of a particular anion among other ions in the solution (Rezayi et al., 2014). For this reason, the novel fabrication of ionophores incoorperated with self-plasticizing polymeric membrane which is highly selective and sensitive towards SCN anion brings great attention in the ISE studies. Regarding the ability of metalloporphyrin that will be mentioned in later section, the applications of metalloporphyrin as ionophore in ISE area can be explored. Thus, this reason encourages us to develop an ISE for the detection of SCN anion based on CoTMeOPP.

Ion selective electrodes (ISEs) are also known as indicator electrode that are composed of simple membrane based on potentiometric measurements which are measured by potentiometric devices. ISEs respond directly to the analyte of interest and selectively bind and measure the activity of an ion of interest. The activities of the ion of interest in an analyte is translated by potentiometric devices. The examples of ISEs are pH electrode, Calcium (Ca^{2+}) electrode, chloride (Cl^-) electrode and many more. Potentiometric devices are devices that are able to accurately measure and translate the activity of ions in the solution. Potentiometric measurements are governed by selective ions extractions and to determine the desired components in an analyte, the potential values are monitored under a zero-current flow system (Faridbod et al., 2008). Other than that, ISEs give simultaneous insight of both ion-activities and electrical parameters of the cell, ISEs also act as a type of transducer that translate the ions activity in a solution into a measurable and readable values (Ammann, 2013). Transducers act as detector that detect the difference in signals produced by the complexation of the ionophore and the target anion that in this case is SCN anion and amplifies the signal to the potentiometer. The potentiometer acts as the translator that gives reading to the detection and selectivity of the ionophoe towards SCN anion. Selectivity of this transducers for one ion over another can be determined by the nature and composition of the membrane material used to fabricate the electrode (Meyerhoff & Opdycke, 1986).

In ISEs, assuming the local thermodynamic equilibrium at the sample membrane interface resulted in a direct dependence of the interfacial potential on the sample activity (Bakker et al., 1997). All potentiometric measurements are concerned with the differences of electro magnetic field (e.m.f) and ratios of concentrations. For all accurate work, it is fixed and that the measurement is determined by using 0.01 mol/L of hydrochloric acid (HCl), study of double cell by using silver-silver chloride electrode as the bridging electrode between the two cells. The double cell can be represented by using the schematic diagram of Ag | AgCl| HCl (0.01 mol/L | bridge electrolyte | sample membrane || inner filling solution |AgCl | Ag or the other way (Covington & Bates, 1973).

The advantages of ISEs compared to other analytical techniques are, it is relatively inexpensive, straightforward and simple to use without any special and sophisticated techniques required. Other than that, ISEs have extremely wide range of applications and wide concentration range. For rapid and ease application, preparation and operation, ISEs are the perfect choice of transducers to meet the demands. ISEs measure the activity of ions directly instead of concentrations which makes it preferable in the field of biological or medical applications. ISEs are able to detect both cations and anions that adds to the quality of ISE since very few techniques have the ability to do the same. In addition to that, ISEs can be used in wide temperature range, non-contaminating to both the samples as well as the environment, non-destructive and have short response time (Bakker et al., 1997).

Different transducer gives different responses towards the target ions. In the case of ISE, it needs an electrode that help to detect the activities of the ions inside an analyte. There are four types of ISEs' electrodes that are widely used in research namely glass membrane electrode (Kerridge, 1925) which is commonly used as pH electrode that detect the presence of hydrogen cation (H^+) , solid state electrode (Bobacka, 2006) commonly metal electrode such as Platinum (Pt), Gold (Au) and Glassy carbon (GC), liquid membrane electrode (Ganjali et al., 2003; Pioda et al., 1969) in which glass is used to contain the internal solution separated by selective membrane and lastly, gas sensing electrode that use the same electrode as pH sensor with the additional housing for detection of gas. However, this work will be focusing on solid state electrode for the detection of SCN anion. The next section will explain the electrode of choice in this research i.e glassy carbon electrode (GCE).

There are many reported works that used ISEs as sensor in detection of ions especially in detection of cations compared to anions. This work is focusing on anions as the target ion. There are numerous reported works on ISEs that involves ionophore incorporated with poly(vinyl) chloride (PVC) as the polymeric membrane for the detection of various anions. In early 1998, Steinle, Schaller, and Meyerhoff studied three different metalloporphyrin namely indium(III) (In^{3+}), thallium(III) (Tl^{3+}) and gallium(III) (Ga^{3+}) as the ionophores in the ISEs for the detection of fluoride (F^-) for Ga^{3+} ionophore and chloride (Cl^-) for both In^{3+} and Tl^{3+} ionophores. All three ISEs showed diversion from the classical Hofmeister series with highly selective and sensitive towards the target anions. PVC membrane was utilized for the manufacturing of all the ionophores in the ISEs. Despite the short response time of approxiamately 40-50 seconds for all the ISEs and long lifetime up to 1 month, the fabricated ISEs for the detection of F^- and Cl^- have many interferences towards another commonly found anions. Ga^{3+} based ISE have significant interferences from salicyclate, SCN^- and nitrite (NO_2^-) anions, meanwhile for In^{3+} and Tl^{3+} based ISEs, Cl^- anion, cyanide (CN^-), salicayclate and iodide (I^-) anions have proven to cause high interferences (Steinle, Schaller, & Meyerhoff, 1998). In 2001, Shamsipur et al., have reported the ISE based on Schiff base complex of zinc(III) (Zn(III)) for highly selective and sensitive towards sulphate anion (SO_4^{2-}). Shamsipur et al., used polymeric membrane electrode (PME) and coated graphite electrode (CGE) to study the best potential response towards SO_4^{2-} anion with the aid of PVC membrane. Different electrode showed significantly different response towards SO_4^{2-} i.e positive Nernstian response with slopes of -29.7±0.9 mV/decade and 29.3±0.7 mV/decade for PME and CGE respectively. In this work, both PME and CGE electrodes demonstrated wide linear working range of $5.0x10^{-1}$ mol/L to $1.0x10^{-1}$ mol/L and $1.0x10^{-7}$ mol/L to $1.0x10^{-1}$ mol/L and low limit of detection of $2.8x10^{-5}$ mol/L and $8.5x10^{-8}$ mol/L respectively. The low response time of less than 15 seconds, long lifetime of approximately 3 months, wide pH working range of 3-7 and almost none interfering ions effecting his electrode, this work has been successfully proved to have high selectivity towards SO_4^{2-} (Shamsipur et al., 2001).

Later on, in 2003, Prasad, Gupta and Kumar used iron(III) (Fe^{3+}) and cobalt(III) (Co^{3+}) with 2,3,7,812,13,17,18-octakis(benzylthio)-5,10,15,20-tetraazaporphyrin (OBTAP) as ionophores for manufacturing of azide (N_3^-) and nitrite (NO_2^-) anions respectively by using "Pyrex" glass tube as the transducer and PVC as the polymeric membrane. Both $[Fe(OBTAP)]^+$ and $[Co(OBTAP)]^+$ ionophores showed positive Nernstian response with slopes of 29.2±0.2 mV/decade and 30.0±0.2 mV/decade with wide working linear range of 8.9x10^{-6} mol/L to 1x10^{-1} mol/L and 1x10^{-5} mol/L to 1x10^{-1} mol/L respectively. The ISEs reported worked at optimum in the pH range of 4.3-10.5 for $[Fe(OBTAP)]^+$ and 2.8-6.4 for $[Co(OBTAP)]^+$ with very short response time of 12 to 13 seconds. The ISEs however have strong interferences towards thiocyanate (SCN^-), sulphide (S^{2-}) and chloride (Cl^-) anions (Prasad, Gupta, & Kumar, 2004). In 2005, Gupta, Ludwig and Agarwal used PVC-based membrane with 5,11,17,23,29,35-hexa-tert-butyl-37,38,39,40,41,42-hexakis(carbamoylmethoxy)calix[6]arene(I) as the ionophore and effect of different plasticizers towards the performance of the fabricated ISE in the detection of phosphate anion (HPO_4^{2-}) was studied. The fabricated electrode showed positive Nernstian response with 33.0 mV/decade with linear working range of 1.77x10^{-5} mol/L to 1.0x10^{-1} mol/L. The fabricated electrode has relatively short response time of 20 seconds, long lifetime of about 1 month and have high interference with perchlorate anion (ClO_4^-). This ISE however have been mentioned to have ionophore leaching problem when it is exposed to solvent with higher organic content (Gupta, Ludwig, & Agarwal, 2005).

Not so long after, in 2009 Sathyapalan and his co-workers used hydroxyl Schiff base of 2,20-((1E,11E)-2,5,8,11-tetraazadodeca-1,11-diene-1,12-diyl)bis(4-nitrophenol) as the ionophore with PVC as the polymeric membrane for the detection of SO_4^{2-}. The fabricated electrode showed positive Nernstian response of slope -28.0 mV/decade within wide linear range of 1x10^{-7} mol/L to 1x10^{-1} mol/L and detection limit of 1x10^{-7} mol/L. Despite the fabricated electrode able to work at acceptable pH range from 3 to 9, the fabricated electrode also able to diminish other interfering anions (Sathyapalan et al., 2009). At the same year, Rezaei came out with perchlorate anion (ClO_4^-) selective electrode by using bis(dibenzoylmethanato)-Co(II) complex as neutral carrier and PVC as the polymeric membrane. In this work, Rezaei revealed that the Co(II) based ionophore is highly selective towards ClO_4^- by giving Nernstian slope of -60.3 mV/decade within wide working range of 8x10^{-7} mol/L to 1x10^{-1} mol/L and detection limit as low as 5.6x10^{-7} mol/L. However, with the fast response time of less than 5 seconds and long lifetime of approximately 2 months with wide pH working range of 2-9, the ClO_4-ISE can be said to have high interference from periodate (IO_4^-), SCN^-, I^- and NO_3^- anions (Rezaei, Meghdadi, & Bagherpour, 2009).

Recently in 2016, Aslan et al., used Schiff base of (2,2’,2’’-salicylidene-imino)-triethylamine-iron(III) ionophore with graphite rod and glass tubing as the transducer. In this work, both electrode with and without the utilization of PVC as the polymeric membrane have been fabricated and tested. The best results are based on electrode without PVC membrane. The Schiff base Fe(III) electrode has shown its positive Nernstian responses and selectivity towards three anions of I^-, SCN^- and ClO_4^- anions with Nernstian’s slopes of -55.2 mV/decade, -51.8 mV/decade and -55.9 mV/decade respectively. The poposed electrode also have wide working range of 1x10^{-5} mol/L to 1x10^{-1} mol/L for both SCN-ISE and ClO_4-ISEs and 1x10^{-4} mol/L to 1x10^{-1} mol/L for fabricated I-ISE and relatively low limit of detection of 1.23x10^{-5} mol/L for I-ISE, 2.52x10^{-6} mol/L for SCN-ISE and ClO_4-ISE with 1.18x10^{-6} mol/L. The electrodes’ lifetime was reported to be 1 month and rapid response time of 5-25 seconds (Aslan et al., 2016). The comparison made in this work between PVC-based and non-PVC based electrode showed amazing result when non-PVC based ISE gave better performance than PVC-based ISE. Another Schiff base complex was used as an ionophore by Abdel-Haleem in the same year as Aslan for the detection of I anion. Abdel-Haleem used copper (II)-sulphamethazine Schiff base complex as the neutral ionophore to study the best electrode type for the detection of I anion. Both PVC and carbon paste (CP) electrodes (PVCE and CPE) utilized PVC as the polymeric membrane meanwhile screen printed (SP) electrode (SPE) did not utilized PVC. All electrodes manifested positive Nernstian responses with each fabricated ISEs having slopes of -55.0 mV/decade, -61.2 mV/decade and -51.0 mV/decade for PVCE, CPE and SPE respectively with wide working range and low detection limit as mentioned in Table 2.1. The proposed electrode also has wide pH working range of 4-8.5 and short response time of 1-5 minutes (Abdel-Haleem & Shehab, 2016). However, the fabricated I-ISE showed high interference towards SCN anion compare to another anion which affect the performance of the I-ISE.

2.2 Ionophores for Detection of SCN Anion

Pyrroles are the main unit component in porphyrin. Pyrrole is a heterocyclic aromatic organic compound that consists of five-membered ring as shown in Figure 2.1 and existed as colorless liquid that darkens upon exposure to air. Pyrroles were first discovered and detected as a constituent of coal tar by F.F Runge in 1830s. Pyrroles have nutty odor and the chemical formula of C_4H_4NH was constructed in 1870 (Jones & Bean, 2013) and was extracted and isolated in pure form from pyrolysate by means of distillation of bone oil in 1850s. The pyrroles come with many interesting applications such as photodynamic therapy (Sternberg et al., 1998), development of novel ligands for anion, transition metals and natural substances (Depraetere et al., 1999; Turner et al., 1998). Pyrroles are not natural occurring compounds but can be found normally in vitamin B12.

ar	Anions	Ionophore	Slope (mV/ decade)	Detection limit (mol/L)	Linear range (mol/L)	Response time (s)	Lifetime (month)
8	F^-	Gallium (III)-porphyrin	-65 to -70	NR	NR	NR	1
	Cl^-	Indium (III)-porphyrin	NR	NR	NR		
		Thallium (III)-porphyrin	-57		$1x10^{-1.6}$ - $1x10^{-5}$		
1	SO_4^{2-}	Polymeric membrane	-29.7	2.8 x 10^{-5}	$1x10^{-1}$ - $5x10^{-5}$	<15	3
		Polymer Schiff base complex of Zn(II)	-29.3	8.5 x 10^{-8}	$1x10^{-1}$ - $1x10^{-7}$		
3	N_3^-	$[Fe(OBTAP)]^+$	-29.2	$1x10^{-6}$	$1x10^{-1}$ - $8.9x10^{-6}$	12	5
	NO_3^-	$[Co(OBTAP)^+$	-30.0	$1x10^{-6}$	$1x10^{-1}$ - $1.1x10^{-5}$	13	
5	HPO_4^{2-}	5,11,17,23,29,35- hexa- tert-butyl-37,38,39,40,41,42-hexakis (carbamoylmethoxy) calix [6] arene (I)	-33.0	NR	$1x10^{-1}$ - $1.77x10^{-5}$	20	1
9	SO_4^{2-}	2,20-((1E,11E)-2,5,8,11-tetraazadodeca1,11-diene-1,12-diyl)bis(4-nitrophenol)	-28.7	$1x10^{-7}$	$1x10^{-1}$ - $1x10^{-7}$	NR	NR
	ClO_4^-	bis(dibenzoylmethanato)-Co(II) complex	60.3	$5.6x10^{-7}$	$8x10^{-1}$ - $1x10^{-7}$	≤5	2
6	SCN^-	(2,2’,2’’-salicylidene-imino)-triethylamine-iron(III)	-51.8	$2.52x10^{-6}$	$1x10^{-1}$ - $1x10^{-5}$	5-25	1
	ClO_4^-		-55.9	$1.18X10^{-6}$			
	I^-		-55.1	$1.23X10^{-5}$	$1x10^{-1}$ - $1x10^{-4}$		
	I^-	copper (II)-sulphamethazine Schiff base complex	PVCE -55	$9x10^{-5}$	$1x10^{-1}$ - $1x10^{-5}$	60-180	‹1
			CPE -61.2	$1x10^{-5}$		60-300	
			*SPE -51	$1x10^{-5}$			

uation of selective electrodes, responsive towards anions.

*No PVC membrane

The extraction of pyrroles from bone oil lead to the development of industrial synthesis of pyrroles from the treatment of furan with ammonia in the presence of SiO_2 and Al_2O_3 as the catalysts in the reaction (Elming & Clauson-Kaas, 1952). The remarkable applications of pyrroles as ligands has make pyrroles as one of the most massive part of novel ligands in late 1990’s and early 2000’s.

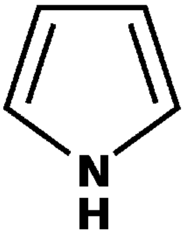


Figure 2.1: The structure of pyrrole.

Porphyrin is defined as heterocyclic macrocycle organic compound molecule that is rigid and comprises from four pyrrolic units linked by four methane bridge. In Figure 2.2 the structure of porphyrin consists of five-membered ring containing a nitrogen atom at the center that is interconnected via methane bridges ($=CH_2-$) at α carbon atom. The bulk structure of four pyrroles are arranged with square planar arrangement that made the larger ring i.e porphyrins stabilized. Other stability contributing factor of porphyrins are the existence of aromatic ring that extends over its entire structure of the porphyrin. Porphyrin normally refers to as the derivatives of porphin or in other words, porphin is the main structure of porphyrins. Porphyrins are usually prepared by two steps mechanism that uses pyrroles and aldehydes as the building blocks. These building blocks undergoes acid catalyzed condensation followed by oxidative treatment to produce porphyrins (Littler et al., 1999). Porphyrins are universal compounds that can be found in nature where they play essential role owing to the fact that their richness of their properties, they have assumed to contribute peculiar role in different disciplines ranging from medicine to material science (Milgrom, 1997).

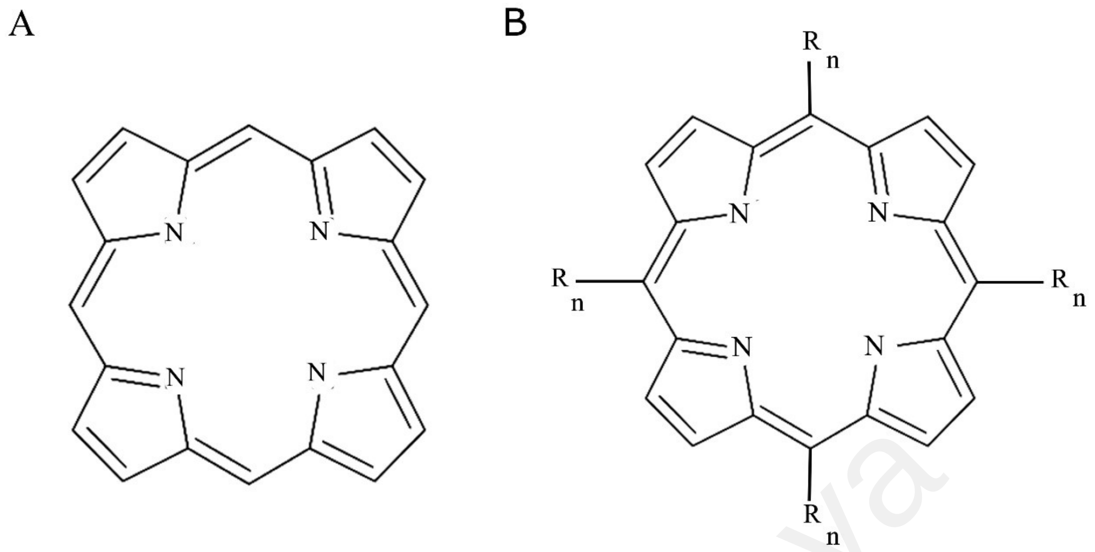


Figure 2.2: (A) The structure of porphyrin and (B) The structure of substituted porphyrin (R_n= Substituent group. n = 1,2,3..).

In recent years, porphyrins have become a common study material because of its versatility and peculiar properties that enhances the applications of researches. In 1997, porphyrins were first utilized as catalyst in catalytic asymmetric epoxidation of nonactivated olefins that has initiated preparations of many chiral porphyrins (Gross & Ini, 1997) and later on became acknowledged as catalyst in asymmetric chemical and biological synthesis up till today (Chen & Zhang, 2007; Simonneaux & Le Maux, 2002; Wilking & Hennecke, 2013). Other than catalyst, porphyrins also has been used widely as biological artificial receptors in molecular recognition (Ogoshi & Mizutani, 1998) especially in recognizing carbohydrates, proteins, enzymes and vitamins. Porphyrins in this many case studies enhanced the selectivity, binding and stability of the receptors especially when it is incorporated with different transition metals as the substituent of the porphyrin rings (Hou et al., 2011; Kremer & Lützen, 2013; Ogoshi & Mizutani, 1999). The enhanced properties of molecular recognition contributed to the detailed and further study of photosynthesis reactions.

Other peculiar application of porphyrin in research field includes the study of stereochemistry of chemical reactions. Methodology developed from porphyrins have successfully elucidated the absolute stereochemistry of many functional groups such as amines, alcohols as well as carboxylic acids (Li et al., 2012). Porphyrin worked together with surfactant has also been reported to be able to organize the stereochemistry of complex materials that improves the understanding of its reactions involved (El-Hachemi et al., 2010). With the incorporation of porphyrin in many chemical reactions, the stereochemistry of many lengthy and complicated reactions is able to be studied, proven and elaborated in more detailed manner by scientists (Yamamoto et al., 2017). Porphyrins have been used to study the electrochemistry of chemical reactions that further lead to its another unique and special quality that is as sensing material. Porphyrins has its own features that make it convenient, suitable and widely used sensing material. The ability of porphyrins to detect volatile organic compounds (VOC) that are present in the environment (Li et al., 2017) is the main contributing factor that open the sensing door of porphyrins. Other qualities include its stability towards many compounds i.e organic and inorganic compounds, reliability to be tuned or tailored by simple modifications of its molecular compounds i.e introduction of almost all metals present in the periodic table to its center and the selectivity of its versatility and enhanced properties to be selected by introducing or altering the porphyrins make porphyrin the element of choice for material sensing (D'Amico et al., 2000).

Porphyrin that is integrated as sensor is called ionophore. The ionophores are used for the detection of highly selective anion and cation (D'Amico et al., 2000; Huser et al., 1990; Ibrahim et al., 2010; Wang & Meyerhoff, 1993). In addition to the properties mentioned before, the position of nitrogen (N) atom in the porphyrin facing the center of the porphyrin facilitates the stability of the porphyrin and enhance the ability of the molecule to seize in a metal ion to form a very anchored organometallic complex porphyrin that make it highly preferable and applicable to be used as the ionophore for the detection of ions. Porphyrin that incorporates with metal ion is called metalloporphyrin (Figure 2.3(A)).

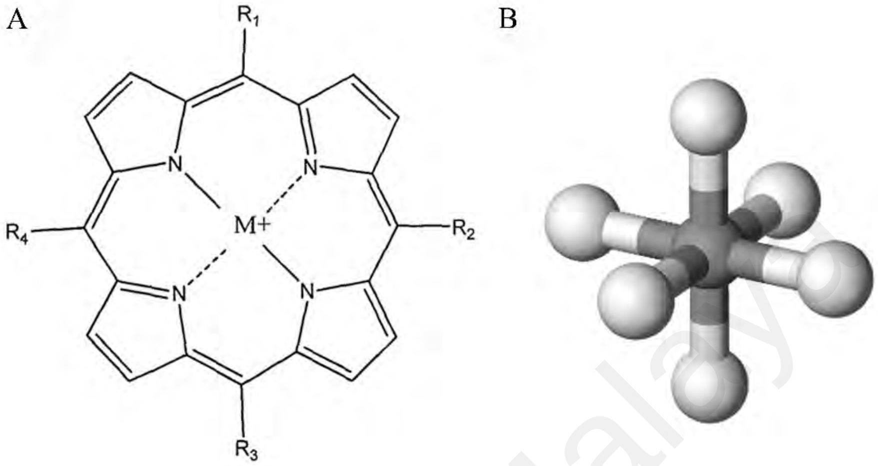


Figure 2.3: (A) The structure of metalloporphyrin (R_{1-4} = substituent group) and (B) The octahedral configuration of metalloporphyrin.

Almost all metals especially transition metals in the periodic table are able to form complex with porphyrins. There are increasing number of researches that uses the metalloporphyrins as sensors including both anions and cations. The examples of metals that has been incorporated with porphyrins are aluminium(II) and (III) (Al^{2+}, Al^{3+}) (Badr & Meyerhoff, 2005) ions, copper(II) and (III) (Cu^{2+}, Cu^{3+}) (Umar et al., 2004), zinc(II) and (III) (Zn^{2+}, Zn^{3+}) (Beer et al., 2004) and many more.

The porphyrins have coplanar configuration at the α carbon as mentioned earlier. This resulted in square planar skeleton with effective internal hydrogens bond between the pyrrolic protons. However, as the porphyrins form complexes with metals, many metals will accept six coordinating ligands resulted in an octahedral configuration (Figure 2. 3(B)). The four sites of the square planar of octahedron is occupied with nitrogen atoms and leaves the two remaining sites of the octahedron empty i.e on the top and bottom of the octahedron metalloporphyrin. The two sites are then filled by the ligands at axial position rather than equatorial position that equip the metalloporphyrin to react in specific way. There are many metals that has been incorporated with porphyrins to form metalloporphyrin and each metalloporphyrin comes with many different specific applications. The function or detection of metalloporphyrins can be tailored and exploited by incorporating different transition metals to the porphyrins center. Different metals incorporated with the porphyrins give rise to different properties of the porphyrins as the sensing agent or ionophore. The different properties will be further elaborated in the next section that further explains the metalloporphyrins and its history.

Ionophore can be defined as organic and inorganic compounds that bind to ions and can be used as an active ionic site in chemical sensory designs. In theory, ionophore behaves like enzymes that will only binds with the specific structure at the specific conditions. Prior to ionophore, liquid and polymer membrane type SCN sensor were widely studied in early 1980's and still ongoing. However, it exhibits classical Hofmeister selectivity patterns with adequate selectivity which makes it less preferable to be used as sensor that defies Hofmeister selectivity pattern. With this drawback, emerging ionophores work as a solution to this problem. There are many reported ionophores that have been used for the detection of SCN anion. In section 2.1 previously, some examples of ionophores that have been used for detection of various anions have been discussed thus, in this section, the history and the development of SCN ionophores and sensors that have been reported will be discussed.

In 1984, Wuthier et al., started an experiment to solve the problems with anion-selective liquid membrane electrodes based on classical ion exchangers such as quaternary ammonium salts, phosphonium salts, complexes of ophenanthroline and other complexing agents that exhibits the classical Hofmeister selectivity pattern. He merged Stanum or Tin (Sn) as the metal incorporated with porphyrin as the metalloporphyrin or the sensing organ. Wuthier et al., used tri-n-octyltin chloride or TOTCl in solvent polymeric membrane as the ionophore. He began to use TOTCl to create an alternative of sensing material that is able to work anti-Hofmeister selectivity pattern. This work revealed that the union of metalloporphyrin of TOTCL and solvent polymeric membrane i.e PVC as the polymeric membrane, tridodecyl(methyl)ammonium chloride or TDDA as the additive and di-n-butylphthalate (DBP) gave away the anti-Hofmeister selectivity pattern and obeys the Nernstian response with slope of -61.9 mv/decade. In this work however, the aim was to assemble an ionophore with polymeric membrane that works on anti-Hofmeister selectivity pattern. Thus, there are no detection limit that has been conducted and recorded. Based on this research, the new ideas of establishing new ionophore that are able to detect SCN anion emerges (Wuthier et al., 1984) and further developed by other researchers.

Amman et al,. in 1986 further tested new metals that bind with the porphyrin to produce the non-Hofmeister anion selectivity ionophores compared to the previous reported ionophore of lipophilic quaternary ammonium salt that obeys the Hofmeister selectivity pattern. In his work, Amman et al,. used two different metals of cobalt(III) (Co^{3+}) and manganese(III) (Mn^{3+}) to study the behavior of these two metalloporphyrins. Amman et al,. merged 5,10,15,20-tetrakis(4-carboxyphenyl)porphyrin with these two metals of Co^{3+} and Mn^{3+} respectively. PVC was utilized to anchored the ionophores onto the transducer with the help of decane-1,10-diyl-diglutarate-bis(1-butyl-pentyl) ester as the plasticizer and TDDA as the additive. The potentiometric sensor of these ionophore prevailed that the combination of the Co^{3+} and Mn^{3+} with the porphyrin mentioned above were selective towards nitrates (NO^{2-}) and SCN anions. The slopes for the utilized metalloporphyrins incorporated with Co^{3+} is -56.3 mV/decade meanwhile Mn^{3+} with the slope of -56.0 mV/decade displayed good Nernstian responses with linear working range of $1x10^{-4}$ mol/L to $1x10^{-2}$ mol/L. However, these metalloporphyrins also showed positive response of towards NO^{2-} that made the metalloporphyrins not selective only towards SCN anion but have high interference from NO^{2-}. Amman et al,. succeeded to synthesized and fabricated non-Hofmeister ionophores but not selective towards SCN anion only. Since the aim of his work was only to synthesized and fabricated new anion-selective ionophore, there were no detection limit and optimum working and pH range conducted (Ammann et al., 1986).

Not so long after that, in 1988, Hodinár et al,. used Wuthier's and Amman's ideas to merge an ionophore based on metalloporphyrin and polymeric membrane that behave as non-Hofmeister selective sensing material. Hodinár, just like Amman et al,. used Co^{3+} as the metal incorporated with porphyrin as sensing material. (α,β,γ,δ-teetraphenylporphynate) cobalt(III) or Co(III)-TPP was chosen as the sensing material with PVC as the polymeric membrane and 2-nitrophenyloctylether (2-NPOE) as plasticizer as the membrane to detect SCN anion. The mixture of ionophore and the polymeric membrane were able to detect SCN anion which behaves anti-Hofmeister selective material and gave positive Nernstian response of slope -55.0 mV/decade with detection limit of $1x10^{-5}$ mol/L to $1x10^{-1}$ mol/L concentration of SCN. This mixture was also able to give good selectivity towards SCN within the pH of 5 to 11 (Hodinár & Jyo, 1988). However, despite the desired non-Hofmeister behavior was acquired, the Nernstian slope of the sensing material towards SCN slightly deviated from theoretical value i.e -58.2 mV/decade.

In 1989, Brown et al., used Amman et al., work that collaborated Mn^{3+} as the metal incorporated with porphyrin. In Amman et al., work, Mn^{3+} porphyrin ionophore showed positive responses towards both SCN^- and NO_2^-. Thus, Brown did some modifications of Amman's work to produce a SCN-selective ISE. In his work, Brown et al., used (5,10,15,20-tetrakis(2,4,6-triphenyl)porphyrinato)-manganese(III) chloride or in short

(Mn[TPPP]Cl) together with PVC as the polymeric membrane and dibutyl sebacate (DBS) as the plasticizer. Brown used human saliva as the samples of study to differentiate between smoker and non-smoker. Brown et al., just like Hodinár et al., and Amman et al., succeed in synthesizing and fabricated the ionophore and the SCN-selective anion sensor. Brown et al., obtained a better Nernstian response than previous works of both Hodinár et al., and Amman et al., with slope of -56.0 mV/decade and worked at optimum rate within the range of $1x10^{-3}$ mol/L to $1x10^{-1}$ mol/L. Brown et al., used flow injection analysis (FIA) in the detection of SCN anion. By using Brown's method and ionophore, the working range of the sensor in sensing SCN^- is rather smaller than Hodinár et al., but the Nernstian slope is better than of Hodinár et al., with short response time of less than 60 seconds. Since Brown et al., used the FIA method, the working pH were not recorded and conducted (Brown et al., 1989). In conclusion, both Hodinár et al., and Brown et al., gave further detailed information regarding the selectivity and optimum working and pH range that Wuthier et al., and Amman et al., studies did not provide which lead to further exploration of SCN-selective ionophore.

In the following years, Daunert et al., 1991 used a different type of metal incorporated with porphyrin for the highly selective detection of SCN anion. Daunert et al., had the idea of merging cobalt(II) or Co(II) metal with amino porphyrin. The ionophore used in his work is cobalt(II) tetrakis (o-aminophenyl) porphyrin or [Co(O-NH_2)TPP]. Daunert et al., used the same transducer as Wuthier et al., i.e glassy carbon (GC) as the potentiometric sensor and fabricated the [Co(O-NH_2)TPP] GC by means of electropolymerization of [Co(O-NH_2)TPP] to poly [Co(O-NH_2)TPP]. By introducing the electropolymerization method, Daunert et al., elude the drawbacks of ionophore or plasticizer leaching that reduced the lifetime and performance of the sensor. This method grafted the ionophore to the backbone of the polymer onto the GC hence improved the lifetime of the sensor up till two months (no ionophore nor plasticizer leach).

Daunert et al., claimed that the method of polymerization is similar to of the polymerization of aniline to polyaniline. The synthesized and fabricated ionophore was able to behave as non-Hofmeister sensor which gave Nernstian slope of -43.0 mV/decade. The detection limit of the sensor is $5x10^{-7}$ mol/L and rapid response time of less than 2 seconds but however, the sensor only works at optimum when it is used at pH of 6. As the pH goes beyond 6, the detection limit is higher i.e $1x10^{-4}$ mol/L, the Nernstian graph started to levelling and diverged and hydroxide (OH^-) anion become more favorable to the ionophore instead of SCN anion (Daunert et al., 1991).

In 1994, Gao et al., had successfully synthesized an ionophore that was able to detect SCN anion. The ionophore is lipophilic metalloporphyrin derivative that used Fe(II) or Fe^{2+} as the transition metal incorporated with porphyrin. In his work, he compared the $(FeTPP)_2O$ with vitamin B12 and quarternary ammonium salt as the ionophores for the selectivity between SCN and ClO_4 anions. The tailored ionophore was experimented using three different plasticizers to find the best plasticizers that enhances the performance of the tailored ionohore. Oxo bridge iron porphyrin or $(FeTPP)_2O$ ionophore showed positive Nernstian response gradient of -58.0 mV/decade within the concentration of $1x10^{-6}$ mol/L to $1x10^{-1}$ mol/L and relatively low detection limit of $6.31x10^{-7}$ mol/L when DBP was used as the plasticizer. Gao et al., has reported that the oxo bridge iron porphyrin or $(FeTPP)_2O$ is one new neutral carrier that belongs to the anti-Hofmeister selectivity pattern family and the $(FeTPP)_2O$ ionophore was fabricated by with the aid of tetrahydrofuran (THF) as the solvent, dodecyl sebacate (DDS) and DBP as plasticizers with PVC as the polymeric membrane (Gao et al., 1994).

Metalloporphyrins began to emerge as the new type ionophore that were proven to behave as anti-Hofmeister selectivity pattern and highly selective to different types of anions. For the first time, paladium(II), (Pd(II)) metal was incorporated with ammonium complexes to acquire highly selective SCN anion ionophore. Sánchez et al., has reported the novel synthesized amine complex by utilizing the cationic cyclopalladated amine and form complex with Pd(II) metal in 1998 to prepare anion selective electrode (Sánchez at el., 1998). Based on Sánchez's work, the synthesized complex was able to perform as SCN-selective sensor just like the previously reported tailored metalloporphyrins. This work showed non-Hofmeister selectivity pattern and at the same time gave away positive Nernstian response towards anions especially SCN anion. The same common polymeric membrane of blank PVC was used in his work with two plasticizers of 2-NPOE and DBP and THF as the solvent for the cyclopalladated amine Pd(II) ionophore. Positive recognition of SCN anion based on this ionohore was able to work at low but narrow concentration of SCN i.e ranging from $1x10^{-6}$ mol/L to $1x10^{-2}$ mol/L with less than 60 seconds of response time and wide pH of 2 to 9. Beyond pH 9, the presence of OH anion become the primary ions and thus effect the selectivity of SCN anion in the sample. In addition to that, this ionophore gave deviated Nernstian slope of -29.1 mV/decade and it worked optimum when the SCN anion was conditioned in perchlorate solution. Sánchez also reported that the ionophore synthesized and fabricated was able to detect NO_2^- and I^- which might affect the detection of SCN anion when these anions are present as the interfering anions.

In 1999, Amini et el., in this work presented the utilization of two different ionophores i.e nickel (Ni^{2+}) and iron (Fe^{2+}) phthalocyanines as the ionophores for SCN selective membrane. The merging of the metals and the phthalicyanine made the new type ionophore of nickelphthalocyanines and ironphthalocyanines that were able to meet the desired properties that behaved as non-Hofmeister and also showed positive Nernstian response when it was tested with SCN anion. This SCN anion selective membrane comprised of the ionophore with PVC as the polymeric membrane as well as dioctylphthalate (DOP) as the plasticizers and TDDA with sodium tetraphenylborate

(NaTPB) as the additives. The selective membrane then conditioned in potassium thiocyanate (KSCN) for 2 hours prior to usage of the electrode. The ionophores were then fabricated onto ion-selective electrode (ISE) of graphite. The ionophores diverged from Hofmeister series and showed positive Nernstian respond of slope -58.4 mV/decade over wide working range of 1x10^{-6} mol/L to 1x10^{-1} mol/L with relatively low detection limit and wide pH range of 5x10^{-7} mol/L and pH 3 to 10 respectively. The highly SCN-selective membrane can be used for over two months and highly selective towards SCN anion compared to other anions. Amini et al., also found out the iron pthalocyanine is a more selective and sensitive towards SCN anion compared to of nickel pthalocyanine ionophore (Amini et al., 1999).

In 2001, Ganjali et al., has reported a highly selective membrane towards the detection of SCN anion by using (N,N-bis-salicyclidene-1,2,ethylenediamine) complex as neutral carriers. The deviation of the ionophore from the Hofmeister selectivity pattern was expected by Ganjali when cadmium-Schiff's base was chosen as complex for the ionophore. Ganjali further widens this research by using different plasticizer to investigate the preferable plasticizer that enhances the detection towards SCN anion. In this work, just like the previous works, PVC was chosen as the polymeric membrane together with DBP and hexadecyl trimethyl ammonium bromide (HTAB) as the plasticizers incorporated with the ionophore to form the SCN selective membrane. DBP was proven to enhanced the detection of SCN anion in the ionophore. The detection of SCN anion was performed by using potentiometric electrode as the transducer and later it was found that the fabricated electrode was selective towards SCN anion compared to other organic and inorganic anions. According to Ganjali's work, the selective membrane showed high selectivity of SCN anion within 1x10^{-6} mol/L to 1x10^{-1} mol/L and detection limit of 6x10^{-7} mol/L with the Nernstian slope of -59.1 mV/decade. The ionophore also had low response time of 15 seconds and relatively long lifetime of not more than 60 days or approximately 2 months (Ganjali et al., 2001).

After 2001, the SCN-selective membrane become a popular anion selective membrane research amongst the scientist. Different types of ionophores using different combination of metals, porphyrins, quaternary ammonium and phosphate salts, phthalocyanines and metallocenes as building blocks were explored. Throughout 2002 up till recent year, there were numerous reported works that implies further developed what Wuthier's and Hodinár's works (Chandra et al., 2008). In 2008, Chandra and his co-workers worked with the common polymeric membrane i.e PVC based ISE by using nickel(II), (Ni(II)) and Cu(II) as the metals bonded with para-tolualdehydesemicarbazone (pTSC) as the building blocks. The resultant vis, bis nitrato[4-hydroxyacetophenone semicarbazone] nickel(II) and vis,bis-nitrato[4-hydroxyacetophenone semicarbazone]copper(II) as ionophores. The ionophores were proven to show positive Nernstian responses and deviated from Hofmeister selectivity pattern towards SCN anion especially the ionophore based on Ni(II) metal. In this work, Chandra utilized PVC polymer with TDDA as the plasticizer. The SCN-selective membrane managed to exhibit positive response within wide linear range of 1x10^{-7} mo/L to 1x10^{-1} mol/L with a detection limit of 1.24×10^{-7} mol/L and a slope of -58.8 mV/decade over a wide pH range of 3 to 10 with a fast response time i.e 10 seconds and long lifetime of 90 days. The SCN-selective membrane fabricated have been used to detect SCN anion in urine and saliva especially in smokers and non-smokers and environmental samples (Chandra et al., 2008). In early 2014, Benvidi et al., merged Cu(II) metal with N-3-methyl phenyl salicylidenaminato to make a neutral ionophore of (N-3-methyl phenyl salicylidenaminato)-copper(II) for detection of SCN anion. In his work, just like many other previous works, PVC was chosen as the polymeric membrane together with DBP as the plasticizer to bind the ionophore onto the transducer. The SCN-selective electrode was able to detect SCN anion at wide linear range of 1x10^{-}

^{6} mol/L to 1x10^{-1} mol/L with detection limit of 5.0x10^{-7} mol/L. The ionophore that is selective towards SCN^{-} managed to deviate from the Hofmeister selectivity series and gave positive Nernstian response with slope of -59.3 mV/decade with the pH range of 4 to 10 with rapid detection rate of 9-21 seconds. Benvidi et al., used the fabricated SCN-selective membrane to detect the SCN anion concentration in human saliva samples to differentiate between smokers and non-smokers (Benvidi et al., 2014).

Recently in recent year of 2017, Abdel Haleem and Rizk discovered an optochemical sensor that worked on ionophore for SCN-selective sensor. Abdel Haleem used 2,2'-[(4,5dimethyl-1,2-phenylene) bis[(E)-nitrilomethylidyne]] bis [4,6-bis (1,1-dimethylethyl) Manganese(III) chloride ionophore or known as Mn(III)-Salophen ionophore with PVC as the membrane. In this work, Mn(III)-Salophen ionophore worked with chromoionophore as transducer. 4,5-Dribromofluoresceinoctadecylster (ETH-70750) was used as the chromoionophore embedded with the Mn(III)-Salophen and sodium tetrakis (trifluoromethyl)phenylborate or STFPB as the additive of the polymeric membrane together with o-NPOE and tricresylphosphate (TCP) as the plasticizers. Abdel Haleem and Rizk used human saliva that they further divide the samples to two distinct groups of smokers and non-smokers as the real samples. The fabricated SCN-selective sensor managed to produce highly selective membrane for SCN anion that worked within the range of 1x10^{-5} mol/L to 1x10^{-1} mol/L with detection limit of 1.9x10^{-6} mol/L. This novel optochemical sensor for SCN-ISE however able to work at optimum only with pH of 4.5 and beyond that, OH^{-} become the interfering anion and become more favorable to the selective membrane than SCN anion. The optochemical sensor also have poor reversibility and reproducibility. Since in his work, UV-vis was used as the method of SCN anion determination, no Nernstian slope was reported and recorded (Abdel-Haleem & Rizk, 2017).

The works presented by many researchers in order to develop new innovative and efficient sensors for the detection of SCN anion is still ongoing and continue to develop more and more. The reported works that have developed decades ago up till present years have improved in terms of its selectivity, durability, working range and more environmentally friendly. These variety reported ionophores have been proven be highly selective towards SCN anion has been used as sensors in urine, body serum, saliva and also in environmental samples. The emerges of the many types of ionophores incorporated with different types of metals clearly showed that the ionophore can be tailored to be highly selective towards an anion of interest. Later, we will look at the ionophore precisely tetrakis metalloporphyrins that has been reported to detect SCN anion. As mentioned before, metalloporphyrins are not new in many researchers as they have been widely utilized in many fields ranging of medicinal to material science. The utilization of metalloporphyrins was chosen to be the active component for the selectivity of SCN anion due to the many reasons. Table 2.2 shows the comparative evaluation of selective electrodes responsive to SCN anion based on PVC as the polymeric membrane in fabrication of the sensor.

tive evaluation of selective electrode, responsive with SCN$^-$ anion.

Ionophore	Slope (mV/decade)	Detection limit (mol/L)	Linear range (mol/L)	Resp onse time (s)	Life time (day)	
Tri-n-octyltin chloride (TOTCl)	-61.9	-	-	-	-	Wuthier
5,10 15,20-tetrakis(4-carboxyphe)porphyrin cobalt(III)	-56.3	-	1x10^{-4} – 1x10^{-2}	-	-	Ammann
5, 5,20-tetrakis(4-carboxypheny orphyrin manganese(III)	-56		1x10^{-4} – 1x10^{-1}			
(α,β,γ,δ-tee henylporphynate) cobalt (II Co(III)-TPP]	-55	-	1x10^{-5} – 1x10^{-1}	-	>14	Hodinár
(5,1(,20-tetrakis(2,4,6-triphenyl)poı rinato)-manganese(III) chloric ır (Mn[TPPP]Cl)	-56	-	1x10^{-3} – 1x10^{-1}	<60	-	Brown
C t(II) tetrakis (o-aminophenyl) phyrin, [Co(O-NH$_2$)TPP]	-43	5 x 10^{-7}	1x10^{-8} – 1x10^{-2}	<25	>60	Daunert
Oxo bridg on porphyrin, [FeTPP)$_2$]	-43	6.31 x 10^{-7}	1x10^{-6} – 1x10^{-1}	-	-	Gao e
	-53	3.98 x 10^{-7}	1x10^{-6} – 1x10^{-1}			
	-58	6.31 x 10^{-7}	1x10^{-6} – 1x10^{-1}			

Ionophore	Slope (mV/decade)	Detection limit (mol/L)	Linear range (mol/L)	Resp onse time (s)	Life time (day)	

Pd(II) amine complex	-29.1	-	$1x10^{-6} – 1x10^{-2}$	<60	-	Sánchez et al.,
Nickel (Ni^{2+}) phthalocyanines Iron (Fe^{2+}) phthalocyanines	-58.4	$5 x 10^{-7}$	$1x10^{-6} – 1x10^{-1}$	20	>60	Amini et al., 1
(N,N-bis-salicyclidene-1,2,ethylenediamine) complex	-59.1	$6 x 10^{-7}$	$1x10^{-6} – 1x10^{-1}$	15	<60	Ganjali et al., 2
Vis, bis nitrato[4-ydroxyacetophenone semicarbazone] nickel(II) Vis, bis nitrato[4-ydroxyacetophenone semicarbazone] copper(II)	-58.8	$1.25 x 10^{-7}$	$1x10^{-7} – 1x10^{-2}$	10	90	Chandra et al.,
(N-3-methyl phenyl salicylidenaminato)-copper(II)	-59.3	$5 x 10^{-7}$	$1x10^{-6} – 1x10^{-1}$	9-21	60	Benvidi et al., 2
2'-[(4,5dimethyl-1,2-phenylene) is[(E)-nitrilomethylidyne]] bis [4,6-bis (1,1-dimethylethyl) [Mn(III)-Salophen chloride]	-	$1.9 x 10^{-6}$	$1x10^{-5} – 1x10^{-1}$	120	7	Abdel-Haleem & R

Continued from table 2.2

Pd(II) amine complex	-29.1	-	$1x10^{-6} – 1x10^{-2}$	<60	-	Sánchez et al.,
Nickel (Ni^{2+}) phthalocyanines Iron (Fe^{2+}) phthalocyanines	-58.4	$5 x 10^{-7}$	$1x10^{-6} – 1x10^{-1}$	20	>60	Amini et al., 1

2.3 Porphyrins and Metalloporphyrins Ionophore

Metalloporphyrins are used in different applications due to its versatility. They can work with wide range of functional groups and substituents introduced to the peripheral position of the porphyrin rings and also able to work with many solvents. The introduction of different functional groups to the porphyrin rings in the metalloporphyrins gave different properties and different selectivity towards different anions of interest. Thus, it can be said that metalloporphyrins can be tailored to meet the desired properties of a sensor. The presence of many porphyrin rings inside the metalloporphyrin enable the metalloporphyrin to detect the volatile organic compounds (VOC) presents in the environment. This quality makes the metalloporphyrin a good "sensing material" (D'Amico et al., 2000). Metalloporphyrins are extensively used by the researchers as the ionophores since they meet most of fitting ionophores qualities (Shirmardi-Dezaki et al., 2013b) especially in developing sensors for detection of various ions either anions and cations. The combination of metals into the porphyrin rings will determine the selectivity of metalloporphyrin either towards anions or cations. Metallporphyrins for the detection of anions have been widely explored and has grabbed the interest of many researchers all around the world. Previously, a number of papers have been reported ionophores integrated ISEs i.e metalloporphyrins with Mn(III), Molybdenum(V) (Mo(V) or Mo^{5+}), Ruthenium(II), (Ru(II)), Ag(I), Mercury(II) or Hg(II), Gold(III) or Au(III), Co(III) and Co(II) as the metal coordinated inside the porphyrin rings for the detection of SCN anion. These reported metalloporphyrins has their own drawbacks as to function as SCN anion sensors. Figure 2.3(A) showed the structure of metalloporphyrin with M at the centre of the porphyrin rings. The M represents the metal integrated in the ionophore of interests with respect to this research.

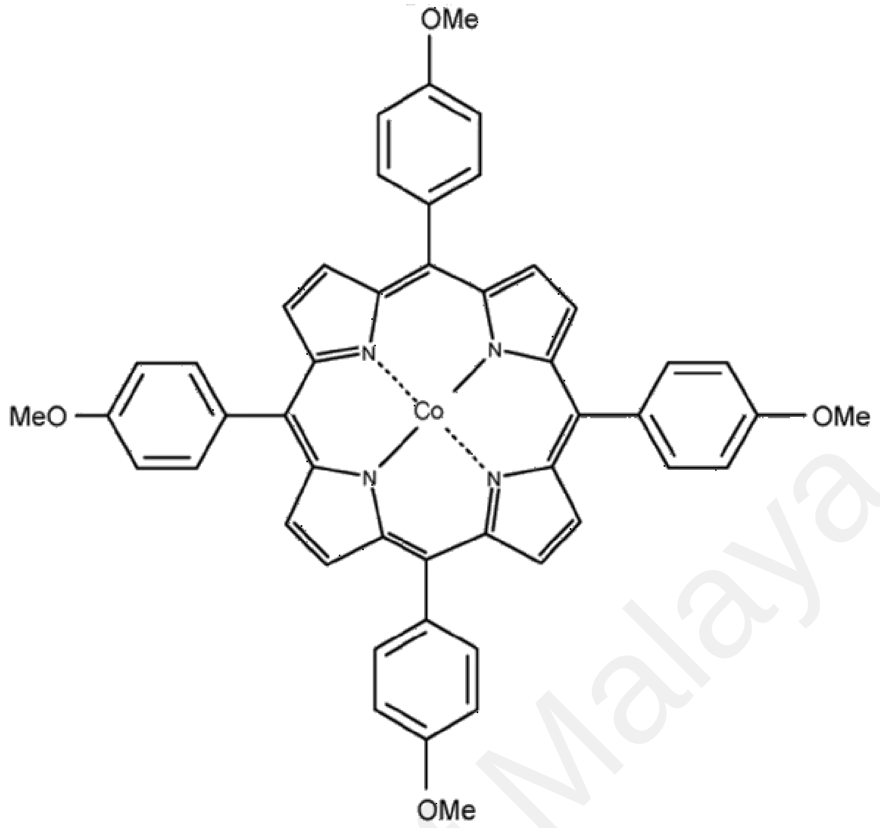


Figure 2.4: Structure of 5,10,15,20-tetrakis-(4-methoxyphenyl)-porphyrin-Co(II) (CoTMeOPP)

5,10,15,20-tetrakis- (4-methylphenyl)-21H,23H-porphyrin-cobalt(II) or CoTMeOPP ionophore (Figure 2.4) was chosen in this study as it can be used with many common functional groups and solvents without being affected by the catalytic activities of the polar groups (Gotardo et al., 2005). It is a versatile ligand that forms well-defined complexes with many types of different metals (Lu & Zhang, 2011) and offers many other qualities (Castello et al., 2008). Due to the high stability and compatibility of complexes formed between different metals with the ionophore, it explained the wide utilization and experimentation using different types of metals in the porphyrin rings.

In early 1980's i.e 1984, Wuthier et al., used Sn(II) as the metal to study the properties of metalloporphyrin that was deviated from Hofmeister selectivity pattern and this work was the pioneer of the selectivity membrane. His work revealed selectivity towards SCN anion (Wuthier et al., 1984). In late 1980's to 1990's the ionophore incorporated with Mn(III) has been widely used as ionophore for the detection of SCN anion (Ammann et al., 1986; Chaniotakis et al., 1988; Huser et al., 1990). However, by merging Mn(III) in the porphyrin rings, the ionophore became more chloride (Cl^-) selective, NO_2^- selective, iodide (I_3^-) and salicylate selective as compared to target SCN anion (Chang & Meyerhoff, 1986; Kondo et al., 1989). Nitrates (N_3^-) was also found to have serious impact as interfering anion for Mn(III) porphyrin (Huser et al., 1990) The potentiometric performance also highly influenced by the membrane solvent with the merge of Mn(III) and Mo(V) inside the porphyrin rings. Mo(V) metal has been proven to be able to give positive response towards SCN anion (Chakraborty & Das, 1988).

Despite the positive response of Mo(V) tetraporphyrins towards SCN anion, the Mo(V) tetraporphyrins also have been reported to be a OH^- carrier (Abe & Kokufuta, 1990) and with different coordinating sites, Mo(V) tetraporphyrins have enhanced response towards salicylate (Chaniotakis et al., 1988). This behaviours of Mo(V) tetraporphyrins and Mn(III) pophyrins make them not highly selective towards SCN anion and deviate from the objectives of this research i.e to fabricate the highly sensitive sensor towards SCN anion.

Another metal that has been previously used before with the porphyrin rings as ionophore for the detection of SCN anion is Ru(II) (Huser et al., 1990). However, Ru(III) porphyrin ionophore has proven to have higher interference from ClO_4 anion despite it has discriminate many other anions that has been interfering with SCN anion when tested with other metals such as Cl^- and NO_2^-. Ag(I), Hg(II) and Au(III) also form complexes with porphyrins to act as an ionophore for the detection of SCN anion (Wang & Meyerhoff, 1993). These metals are known as its high cost as well as its hazards if these metals leached to the surroundings. Other than that, Ag(I) and Au(III) porphyrins shows high selectivity towards both ClO_4^- and SCN^-. However, thiosulfate and thiourea poison this type of ionophore electrode (Bühlmann et al., 1998).

Back in 1980s, cobyrinate ionophores showed comparable selectivity towards SCN anion. Cobalt(III) porphyrin ionophore are widely investigated its selectivity towards SCN anion. With the exchange of the coordinates of Co(III) porphyrin ionophore to axial ligands, this ionophore are able to respond selectively towards SCN and NO_2 anions (Schulthess et al., 1984). This ionophore also respond significantly with I anion and in certain cases NO_2 anion interfere strongly towards Co(III) porphyrin (Hodinár & Jyo, 1988). Co(II) was also been reported to be utilized with the porphyrins to form metalloporphyrins and Co(II) porphyrin is able to discriminate the interference of I anion (Daunert et al., 1991) but no further investigations were reported with Co(II) porphyrin. Based on Table 2.2, in late 1980's and early 1990's Co(III) and Co(II) have been used as the active material for the detection of SCN anion. This is due to the interaction formed between the metal center of the metalloporphyrin and SCN anion. SCN anion has linear geometric arrangement meanwhile the CoTMeOPP have Co(II) metal at the center with (Ammann et al., 1986; Daunert et al., 1991; Fleischer et al., 1968; Hodinár & Jyo, 1988; Vlascici et al., 2006)

In early 2002, Gupta et al., were the pioneer scientists that worked with CoTMeOPP as an ionophore for detection of anions (Gupta et al., 2002). In his work, Co(I) were used to be incorporated with the porphyrin. The synthesized Co(I) porphyrins were used to detect molybdate anion (MoO_4^{2-}). Gupta has successfully synthesized and fabricated liquid membrane ISE based on Co(I) porphyrins that is MoO_4^{2-} selective with PVC as the polymeric membrane, sodium tetraphenyl borate (NaTPB) as anion excluder and (DBP), dioctyl phthalate (DOP), dibutyl butyl phosphate (DBBP), tri-n-butyl phosphate (TBP) and 1-chloronaphthalene (CN) as plasticizing mediators. This field of study was not just focusing on selectivity of Co(I) porphyrin selectivity towards MoO_4^{2-}, but this work also emphasized on different types of plasticizers with the potentiometric response of Co(I) porphyrin towards the target anions. DBP presented the best response as plasticizer that works best in the concentration range of 5.0×10^{-5} mol/L to 1.0×10^{-1} mol/L with a near Nernstian slope of 32.0±1.0 mV/decade of activity with pH over the range of 5.4 to 10.5. In addition to the qualities, this Co(I) porphyrin ISE possesses facile Nernstian response of 18 seconds with a lifetime of four months.

Since then, CoTMeOPP has been widely utilized as it provides extremely versatile synthetic base for various applications in different disciplines ranging from medicine to material science. Other than CoTMeOPP been used as ionophore, CoTMeOPP has also been utilized as dechlorination of tetrachloroethylene (Dror & Schlautman, 2003), incorporated in sensors (Kimura et al., 2006; Zhang et al., 2007) and also a material in cathode (Zhu et al., 2009). In 2006, Co(II) was eventually incorporated with the porphyrin ring after being left out in early 1990s. Vlascici et al., reported a new composition for Co(II)-porphyrin based membranes ISEs that detects SCN anion. As Daunert et al., proved that Co(II) porphyrins are able to detect SCN anion and successful eliminated the interfering of I anion, Vlascici et al., in this work fabricated CoTMeOPP with PVC as the polymeric membrane and o-nitrophenyloctylether (o-NPOE) as plasticizer to produce SCN-selective ISEs (Vlascici et al., 2006). The facile fabrication of the ISE using CoTMeOPP as ionophore are shown to exhibit high selectivity for SCN anion with a near Nernstian slope of -65.8 mV/decade which close to the theoretical value in the working range of 1×10^{-5} mol/L to 1×10^{-1} mol/L and detection limit of 6×10^{-6} mol/L with good stability in time.

The latest utilization of CoTMeOPP as ionophore was operated by Ibrahim et al., in the year of 2010. In this work, Ibrahim et al., used the same ionophore as both Gupta et al., and Vlascici et al., but detecting different type of anions. Selenite anion (SeO_3^{2-}) was chosen as the target anion in this study. Despite the charge of SeO_3^{2-} is 2-, in the pH range of 5.59 to 7.38, the SeO_3^{2-} anion was categorized as monovalent anion. Just like previous works, Ibrahim et al., also worked with PVC as the polymeric membrane and he was also focusing on different types of plasticizers that increases the selectivity and response of the SeO_3^{2-} selective membranes. TCP, DBP, DOP, o-NPOE and DBBP were the selection of plasticizers utilized in this work. It was then revealed that the collaboration of CoTMeOPP, PVC and TCP as the plasticizer exhibits the Nernstian slope -57.0 mV/decade at pH of 6.47 and linear working range of $1x10^{-5}$ mol/L to $1x10^{-2}$ mol/L with detection limit of $3.4x10^{-5}$ mol/L. Beyond the pH of 6.47, the selenite-selective electrode showed detection limit lowered to $4.7x10^{-5}$ mol/L i.e pH 11. This work clearly showed that the SeO_3^{2-} selective membrane depends on pH to work at optimum rate (Ibrahim et al., 2010).

After Ibrahim et al., the utilization of CoTMeOPP as ionophore was not reported in any papers. In conclusion, in these previous works of Gupta et al., Vlascici et al., and Ibrahim et al., all of them used the same ionohophore i.e CoTMeOPP, polymeric membrane i.e PVC and also the same electrode i.e liquid-membrane electrode. These works reported were able to synthesized the ionophore, optimized and fabricate the ionophore onto the electrode to produced anion selective electrode. Table 2.3 shows the comparative evaluation of selective electrode with CoTMeOPP as the ionophore.

Table 2.3 : Comparative evaluation of selective electrodes based on CoTMeOPP ionophore.

Year	**2002**	**2006**	**2010**
Target anion	MoO_4^{2-}	SCN^-	SeO_3^{2-}
Polymeric membrane		PVC	
Plasticizer	DBP	o-NPOE	TCP, DBP, DOP, o-NPOE, DBBP
Detection limit (mol/L)	5.0×10^{-5}	1.0×10^{-5}	$3.4x10^{-5}$
Refs	Gupta et al.	Vlascici et al.	Ibrahim et al.

Thus, in this work, further exploration of CoTMeOPP ionophore is conducted in fabrication of SCN-selective electrode by using self-plasticizing membranes to replace commonly used PVC membrane. The selective and sensitivity of the CoTMeOPP ionophore towards SCN anion compared to other commonly found anions will be further investigated and the evaluation and comparison of this work will be further reported in Chapter 4 later on.

2.4 Analyte of Interest

Thiocyanate or rhodanide with anion of SCN is one of the Persistent Organic Pollutants or POPs that is commonly found in trace amounts in water bodies and food industry especially crops in Malaysia and it is one of the many catalysts and additives used in many fields (Bhunia et al., 2000). SCN is one of the most important cyanide complexes (Garson & Simpson, 2004, Zhang et al., 2012) and widely produced by microorganisms by means of cyanogenesis (Mak et al., 2005) either to detoxify or utilize it as a source of nitrogen for growth (Drochioiu et al., 2008). Cyanide being very toxic to the environment is broken down by various means and produce SCN as the end product. Being metabolism product of cyanide does not abolish the toxicity of SCN. The derivatives of SCN commonly exist in the form of sodium, potassium or ammonium salts that is colorless and normally tasteless. The wide applications of SCN in the industries made it preferable as it is cheap, easy to acquire and manufactured. SCN has made its way as one of the most applied ‘assistance’ and ‘seasoning’ in the industries (Jeong & Chung, 2006; Namasivayam & Prathap, 2006; Staib & Lant, 2007).

The wide applications of SCN anion includes printing, dyeing, acrylic fiber production, herbicide, insecticide. In many other industries, SCN is used as solvent, catalysts, additives and pigments (Namasivayam & Prathap, 2006; Zhang et al., 2007). The wide utilization of SCN in the industries has its drawbacks when it managed to seeped out to the environments especially in marine aquatic environment via untreated waste from the industries, leaching from the soil especially from agricultural region and waste water discharged from coal conversion (Jeong & Chung, 2006), untreated and leaching from mine and ex-mining sites (Hassan et al., 2015).

The outbreak of the SCN to the environment have reached the level where it pollutes the aquatic lives as well as the consumer i.e human and the pollution rate is increasing over time. There are many ionophores that have been utilized as sensors in various applications especially in detecting harzardous materials. As mentioned before, metalloporphyrin have been used widely as ionophore for the detection of both anions and cations. As different metal contributes to different sensitivity towards different anions and cations, the selection of metals to be incoorperated with the porphyrin needs to be made thoroughly. Co(II) has been reported to show good response towards SCN detections (Vlascici et al., 2006). Thus, this research will be focusing on Co(II) porphyrin or specifically 5,10,15,20-tetrakis-(4-methoxyphenyl)-21H,23H-porphyrin-Co(II) (CoTMeOPP) as the ionophore for the detection of SCN anion.

In the past decades, there has been increasing interest in the trace determination of thiocyanate. Its derivatives commonly exist in colorless salt of potassium and sodium (Dash et al., 2009) and proven to prevail in the crops (Tonacchera et al., 2004), human milk (Kirk et al., 2007), dairy products (Arvand et al., 2007), atmospheric air and water bodies (Recalde-Ruiz et al., 2000). In some countries, SCN is added to milk as preservative (Fernández et al., 2005; Ponce, 2013). Traces amount of SCN infiltrates human body by means of food (Brown et al., 1995; Lanno & Dixon, 1996; Sanchez et al., 2007; Vaz, 2011), medicine and mainly by cigarette (Connolly et al., 2002). SCN is excreted out of the body though saliva, urine and serum via the kidneys (Singh et al., 2008; Zhang et al., 2012b).

SCN comes with variety of applications in the industry such as photofinishing, herbicide and insecticide production (Bhunia et al., 2000), dyeing, acrylic fiber production, manufacturing of thiourea, coke industry (Staib & Lant, 2007), metal separation and electroplating (Jeong & Chung, 2006), soil sterilization and corrosion inhibition (Namasivayam & Prathap, 2006). However, it comes with many drawbacks too. The exudation of SCN into marine aquatic environment significantly impacted the food chain. Bioaccumulation of SCN anion that is taking place in human body can be summarized in Figure 2.5. Human that is placed on top of the food chain resulted in bioaccumulation of SCN anion in the body.

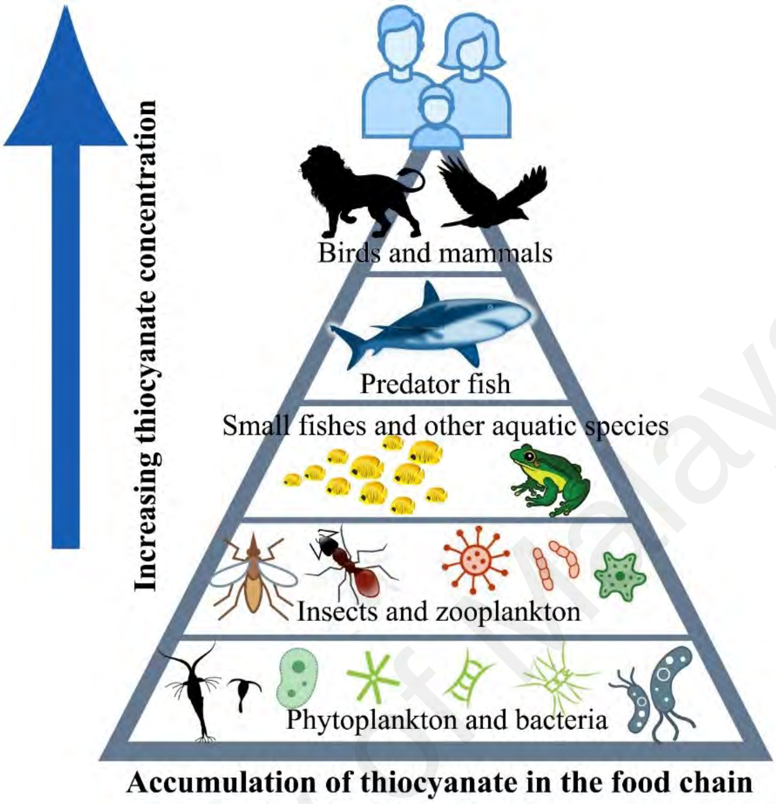


Figure 2.5: Bioaccumulation of thiocyanate in the food chain (Yu & Zhang, 2013).

As the food pyramid goes up, the amount of SCN anion increases and accumulated in human body as human stands at the top of the food chain. Elevated amount of SCN anion in the body block the iodine uptake by the thyroid gland (Zhang et al., 2007) leading to iodine deficiency in the body, hypothyroxinemia, body cell destruction as the SCN anion is very toxic to the body, chronic change in liver, kidneys and muscle cells. As SCN has been proven to prevail in dairy products as well as human milk, the infants may suffer from delay or impairment in neurological system (Ponce, 2013). This SCN that has polluted the marine environment has been proven to prevail beyond elevated amount (Deocadiz, 1999; Turner, 2010).

Thiocyanate as many other contaminants has their own proved natural decomposing mechanism by means of anaerobic and aerobic environment with the aid of pure and mixed cultures of indigenous soil bacteria (Chakraborty & Veeramani, 2006; Ebbs, 2004; Touch et al., 2004). This mechanism however has it flaw, the SCN is not readily removed through the biological treatment (Dash et al., 2009) and when it is mixed with other contaminants such as phenols, it will inhibit other’s removal in the rotating bioreactor (Chakraborty & Veeramani, 2006; Staib & Lant, 2007) and results in accumulation of both SCN and phenol in water bodies. This hazardous contaminant sourced from various chemical processes such as untreated agricultural herbicide (Kwon et al., 2002) and insecticide (Staib & Lant, 2007), untreated chemical waste from mills (Namasivayam & Prathap, 2006) and factories (Bhunia et al., 2000) and the major SCN contributor in the body is from cigarette smoke (Scherer, 2006; Toraño & van Kan, 2003). Figure 2.6 shows the summarization of sources of SCN pollution in the environment.

Several analytical methods for quantitative determination of SCN have been previously reported which includes spectrophotometry (Hovinen et al., 1999), amperometry (Cookeas & Efstathiou, 1994; Wang et al., 2004), head-space gas chromatography (Lee et al., 2003), Aldrige method (Pettigrew & Fell, 1972), anion exchange resins (Kononova et al., 2007), ion chromatography (Michigami et al., 1992), potentiometry (Ni & Wu, 1999; Zamani et al., 2006), polarography (Brown et al., 1995; Cai & Zhao, 1988), high performance gas and liquid chromatography (Brown et al., 1995), capillary zone electrophoresis (Tanaka et al., 2004), ferric nitrate reagent method (Bowler, 1944), spectrofluorometry (Afkhami et al., 2007), UV-vis spectrometry (Hovinen et al., 1999), Raman spectroscopy (Pienpinijtham et al., 2011), electrochemistry (Ozoemena & Nyokong, 2005), photokinetic voltammetry (Li et al., 2000), atomic absorption spectrometry (AAS) (Chattaraj & Das, 1992) and fluorescence (Zhang et al., 2007). Table 2.4 shows the summarization of the reported methods in determining trace amount of SCN anion.

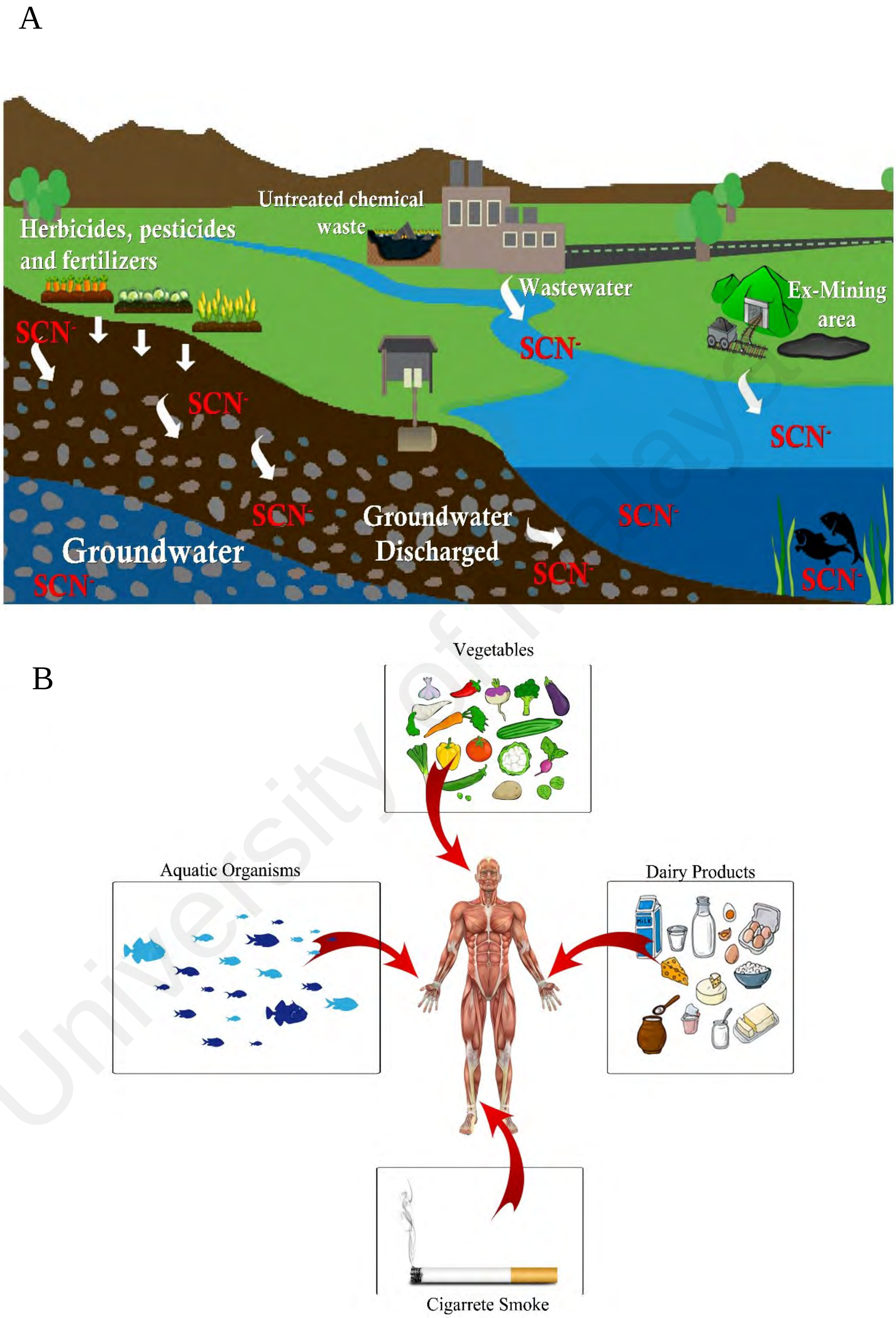


Figure 2.6: (A) The sources of SCN outbreaks in the environment and (B) Sources of SCN entering human body.

Table 2.4 : The summarization of reported methods in determining trace amount of SCN anion.

Methods	Spectro-metry	Fluorimetry	Electro-chemistry	Raman Scattering	Chromato-graphy
Detection limit (mol/L)	6.89×10^{-5}	9.00×10^{-7}	1.00×10^{-9}	1.00×10^{-6}	8.60×10^{-11}
Refs	(Themelis & Tzanavaras, 2002)	(Song et al., 2015)	(Shehab & Mansour, 2014)	(Wu et al., 2014)	(Ammazzini et al., 2015)

In analytical chemistry, rapid and simple method of detection of trace amounts of ionic species is a compelling interest. The detection methods of thiocyanate reported need separation processes before measurement, time consuming and involve complicated, expensive sample preparation. Moreover, the detection limit is high and this adds to the cut-breaks of the detection tools. To curb the limitations, ion selective electrodes (ISEs) emerged as a type of tools to deter the cut-breaks of the prior detection methods (Bakker & Qin, 2006).

2.5 Sensor Fabrication

2.5.1 Membrane Composition

The composition of the polymeric membrane is crucial to the selectivity, sensitivity and performance of the sensor towards detection of the target anion. The difference in composition of the compounds in the polymeric matrix has been proven to show significant influence towards the selectivity and sensitivity of the fabricated sensor (Ibrahim et al., 2010). Thus, the construction of the electrode membrane of ISE solely rely on the polymer matrix which contains mixture of active components such as ionophore ionic additives and photo-initiator.

2.5.1.1 Polymer matrix

The fabrication of ionophore to the GCE is aided by polymeric matrix or also called polymeric membrane. Polymeric membrane containing electroactive species that are coated directly on the surface of the electrode is the most common way of fabricating an ionophore onto the electrode. Since the electrode helps in detecting the ion activities in sample solution based on the charge emitted by the target anion bonded with the ionophore, by definition, the selection of the polymeric membrane needs to be made based on certain properties that aid and ease the ion to electron detection by both the ionophore and GCE. The main and crucial property of choosing the polymeric membrane is that it is able to conduct electricity. Conducting polymer are commonly used as polymeric membrane for the fabrication of ionophore to the electrode. Other than that, the polymeric membrane needs to be soft polymer to avoid the rupture of the highly selective polymerized membrane onto the GCE due to certain conditions such as pH or temperature. In addition to the properties of polymeric membrane, the polymer must not affect the selectivity of the ionophore and must not react with the ionophore that will reduce the performance of the sensor. The polymeric membrane also cannot be easily dissolved by common solvent whether organic nor inorganic solvent to ensure the reproducibility of the sensor, increasing the lifetime of the sensor and to prevent leakage of the ionophore to the sample solution. There are many types of polymer that meets the desired properties of polymer to be used as the polymeric membrane in the fabrication of ionophore onto the GCE such as PVC, silicon rubber (Kimura et al., 1992), polyurethane (PU) (Malinowska et al., 2001) and polyaniline (PANi) (Evtugyn et al., 2007).

(a) Self-polymerized polymeric membrane

Based on section 2.2, almost most of the researchers that have utilized different ionophores for the detections of SCN anion and also another anion worked closely with PVC as the polymeric membrane which aided and enhanced the performances of the sensors. PVC was used hand in hand with plasticizers and highly volatile solvent commonly tetrahydrofuran (THF) that plasticized the PVC onto the desired electrode. These mixtures of PVC, plasticizers, additives, ionophores and THF are drop-casted onto the GCE surface and let to self-vaporized and plasticized. This is the most common method for fabrication of ionophore onto the electrode of interest. In recent years, there are new types of polymeric membrane that merged as self-plasticizing polymers to replace PVC, namely (PU) and silicon rubber and PANi as polymeric membrane. The utilization of self-plasticizing polymeric membranes is to avoid the utilization of plasticizers in fabrication of ionophore onto the electrode. The mixture of the polymeric membrane with ionophore, additives, highly volatile solvent and a significant amount of plasticizer often lead to leaching and exudation of ionophore or plasticizers to the sample solution. The leaching of the membrane especially the plasticizer decreased the performance of the sensor by shortening the lifetime of the sensor and lowering the selectivity of the sensor (Heng & Hall, 2000b). The leaching of plasticizers leads to adhesion problem of the polymeric membrane onto the GCE and it has been reported that the commercially used plasticizers to be toxic to the environment and this will have many drawbacks in the future especially pollution of highly toxic chemicals to the environment (Heng & Hall, 2000c). The development of green sensor emerged rapidly to curb the problems arose by the plasticizer in the polymeric membrane.

In this research, the approach of choosing self-plasticized polymers is one of the main focus to overcome the leaching problems to enhance the performance of the SCN-ISE. Hence, the monomer must have low glass temperature T_g in order to be able to self-photocurable. The utilization of self-plasticizing polymer as the polymeric membrane simultaneously keeping and maintaining other desired qualities lead to the discovery of non-plasticized based silicon rubber as membrane in early 1970’s to curb the problems related to plasticizer that reduced the performance of the sensor (Pick et al., 1973). There are many types of self-plasticizing polymers that have been used by researchers ever since the discovery of silicon rubber membrane and acrylic polymer being one of the mostly utilized self-plasticizing polymers replacing plasticized-based electrode membranes. Back in 1994 Reinhoudt et al., utilized (hydroxyethyl)methacrylate (HEMA) hydrogel layer as the membrane matrix without the use of plasticizer for manufacturing K^+ selective electrode with K-selective ionophore of hemispherand. In his work, the fabricated K-ISE managed to give positive Nernstian respone of 59.0 mV/decade with wide working range of $1x10^{-1}$ mol/L to $1x10^{-1}$ mol/L and long lifetime of 10 months (Reinhoudt et al., 1994).

By using Reinhoudt's idea, in 2000, Heng and Hall came up with K-selective electrode by using acrylic copolymers of n-butyl acrylate (n-BA) and methyl methacrylate (MMA) and two different K-selective ionophores of 4-acryloylamidobenzo-15-crown-5 (AAB15C5) and 4-acrylotlamidobenzo-18-crown-6 (AAB18C6). Their fabricated K-ISEs showed good Nernstian response with slope of 59.16 mV/decade with relatively low response time of less than 10 seconds and lifetime of less than 7 days. Both ionophores of AAB15C5 and AABC18C6 have wide working range of $1x10^{-5}$ mol/L to $1x0^{-1}$ mol/L and detection limits of $4.8x10^{-5}$ mol/L and $5.8x10^{-5}$ mol/L respectively (Heng & Hall, 2000a). Later on, Heng and Hall used the same K-selective ionophores and polymeric membranes (Table 2.5). But this time, in their work, inner layer of HEMA was introduced and utilized with additional another commonly used Ca^{2+} and Na^+-selective ionophores to study the performances of the self-plasticizing membranes ISEs compared to PVC based ISE. In their work, all fabricated cation-selective electrodes showed positive Nernstian responses of 59.6 mV/decade for Na-ISE, 33.6 mV/decade for Ca-ISE and 61.9 mV/decade for the K-ISE with relatively wide working range from $1x10^{-1}$ mol/L to $1x10^{-5}$ mol/L for all fabricated ISEs (Heng & Hall, 2000b).

elective electrodes based on self-plasticizing polymeric membranes.

Year	Polymeric membranes	Ionophore	Target ions	Slope (mV/decade)	Linear range (mol/L)	Detection limit (mol/L)	
1994	HEMA	Hemispherand	K+	59	$1x10^{-4}$ to $1x10^{-1}$	NR	Reinho
2000	n-BA MMA	AAB15C5 AAB18C6	K+	59.16	$1x10^{-5}$ to $1x10^{-1}$	$4.8x10^{-5}$ $5.8x10^{-5}$	Heng
	HEMA n-BA MMA	AAB15C5	K+	61.9	$1x10^{-5}$ to $1x10^{-1}$	$5.7x10^{-5}$	Heng
		;is[(12-crown-4)methyl]- lodecylmethylmalonate] (B12C4)	Na^+	59.6	$1x10^{-5}$ to $1x10^{-1}$	$5.6x10^{-5}$	
		N,N-dicyclohexyl-N,N- :tadecyl-3-oxapentanamide (ETH5234)	Ca^{2+}	33.6	$1x10^{-5}$ to $1x10^{-1}$	$5.5x10^{-5}$	
2002	MMA DMA	Na(X)	Na^+	55.1	$1x10^{-5}$ to $1x10^{-1}$	NR	Qin
		BME-44	K^+	58.8	NR	NR	
		;,6-dibenzyl-14-crown-4	Li^+	60.4			
		ETH 1117	Mg^{2+}	30.1			
		ETH129	Ca^{2+}	28.8			
2003	MMA DMA	)icyclohexyl-N′-phenyl-N′-3- -propenoyl)oxyphenyl-3- :apentanediamide (AU-1)	Ca^{2+}	32.1	$1x10^{-6}$ to $1x10^{-2}$	NR	Qin
2012	n-BA	BTCH	Ag^{2+}	28.1	$1x10^{-5}$ to $1x10^{-2}$	$2.5x10^{-6}$	Juma
	PEDOT PSS	AITAP	F^-	-58.5	$1x10^{-5}$ to $1x10^{-1}$	NR	(Ma
		ZrTAP		-56	$1x10^{-5}$ to $1x10^{-1}$		
		AlSAL		-55	$1x10^{-3}$ to $1x10^{-1}$		
		UO_2SAL		-40	$1x10^{-4}$ to $1x10^{-1}$		
2016	Poly(3-octylthiphene)	1,2,3-triazole (iL)	I^-	-56.1	$1x10^{-7}$ to $1x10^{-1}$	$6.3x10^{-8}$	(Me

Later on, in 2002, Qin and co-workers used MMA with decyl methacrylate (DMA) as the polymeric membranes in fabricating Na-selective electrode with Na-selective ionophore of 4tert-butylcalix[4]arenetetraethylester (Na(X)). This work also studied different types of cation selective ionophores of K^+, Li^+, Mg^{2+} and Ca^{2+} cations with MMA-DMA self-polymerized membranes to replace the commonly used PVC-based ISEs. The main Na-ISE gave Nernstian slope of 55.1 mV/decade with wide linear range of $1x10^{-1}$ mol/L to $1x10^{-5}$ mol/L. Other cation-ISEs that was studied by Qin showed positive Nernstian slopes as seen in Table 2.5 (Qin et al., 2002). In 2003, Qin and his co-workers again studied the same self-polymerized polymeric membranes of MMA-DMA by using new Ca-selective ionophore of N,N-Dicyclohexyl-N′-phenyl-N′-3-(2-propenoyl)oxyphenyl-3-oxapentanediamide (AU-1). The fabricated Ca-ISE showed good Nernstian response of 32.1 mV/decade within Ca^{2+} concentration range of $1x10^{-2}$ mol/L to $1x10^{-6}$ mol/L. The fabricated electrode also has very fast response time of less than a minute but relatively short lifetime of 5-8 hours (Qin et al., 2003). In 2012, Jumal used n-BA self-plasticizing polymer as the polymeric membrane for mercury (Hg^{2+}) selective electrode by using Hg-selective ionophore of 1,2-bis-(N'-benzoylthioureido cyclohexane) or BTCH. His electrode showed positive Nernstian response with slope of 28.1 mV/decade and wide linear working Hg^{2+} concentration range of $1x10^{-1}$ mol/L to $1x10^{-5}$ mol/L with low detection limit of $2.5x10^{-6}$ mol/L. His electrode also has short response time of 50-100 seconds and lifetime of 1 week (Jumal et al., 2012).

Later on, Matusevich et al., used new kind of self-polymerized polymers of poly(3,4-ethylenedioxythiophene) (PEDOT) and polystyrenesulphapte (PSS) polymers as the conductive polymer replacing PVC in detection of fluoride (F^-) anion. In his work, he utilized many different ionophores to collaborate with PEDOT:PSS polymeric membranes to obtain the best ion selective membrane (ISM) composition for fabrication of F-ISE. Four types of ionophores that are widely used as F-selective ionophores namely chloro 2,7,12,17-tetra-tert-butyl-5,10,15,20-tetraazaporphine Aluminium(III) (AITAP), dichloro 2,7,12,17-tetra-tert-butyl-5,10,15,20-tetraazaporphine Zirconium(IV) (ZrTAP), Aluminium(III) 3,5-ditert-butyl-salophene (AlSAL) and Uranyl 4,5-naphthyl-salophene (UO_2SAL) were used in this study. Based on his study, all fabricated F-ISE gave positive Nernstian responses and wide working range with acceptable slopes except for AlSAL (Table 2.5) (Matusevich et al., 2012). There are many other reports regarding self-plasticized ISE in the detection of cations reported after the year of 2002 by using ionic liquids as the replacement of ionophores. However, as mentioned before, there are few reported works on anion-ISE and they are even fewer reported works on self-plasticized polymeric membranes reported for the fabrication of anion-ISEs. The latest work on self-plasticized membrane was reported in the year of 2016, an anion-ISE based on self-plasticized polymer by Mendecki et al. His work involves poly(3-octylthiphene) polymer and 1,2,3-triazole ionic liquid (iL) for the detection of iodide (I^-) anion. The utilization of iL in his work replacing other ligands and ionophores showed positive Nernstian response towards I^- with slope of -56.0 mV/decade within wide working range of $1x10^{-3}$ mol/L to $1x10^{-7}$ mol/L and relatively low detection limit of $6.3x10^{-8}$ mol/L. The lifetime of his fabricated I-ISE was approximately 2 weeks (Mendecki et al., 2016).

There are no recent reports on anion-ISE reported by using self-plasticizing polymeric membranes since Mendecki's. Thus, this work emphasizes the usage of self-plasticizing polymeric membrane to replace the widely used PVC in the fabrication of ISEs. Based on Mendecki's work, it can be said that it is possible to obtain a better ISEs performances without the utilization of plasticizer in the fabrication of ISEs. Table 2.5 shows the comparative evaluation of selective electrodes based on self-plasticized polymeric membranes in the fabrication of the sensors. As can be seen, there are researches in developing self-plasticizing based ISEs but mainly are focusing on cation-ISE. Thus, in this work, we are developing new anion self-plasticized ISE. In this research, there are two polymeric membranes chosen to enhance the performances of the SCN-selective sensor namely n-BA and also HEMA polymers. These two self-plasticizing polymers were chosen as both fit the properties of self-plasticizing required to fabricate the ionophore onto the GCE and the utilization of self-plasticizing polymer based on CoTMeOPP ionophore was never published by researchers for the detection of SCN anion.

2.5.1.2 Ionic additive

Ionic additives are ion exchangers that are added to the self-plasticizing membrane as performance enhancer of an ISE. This ionic additive aid in the selectivity of the fabricated sensor and have been proven to significantly increases the performance of a sensor by inducing themselves a selective response with or without the presence of ionophore (Wegmann et al., 1984). The persuasive properties of the ionic additives that are able to give the appropriate Nernstian response that deviates from the classical Hofmeister series responding to the target analyte contributes to the sensitivity of the ISEs. The neutral carrier of the CoTMeOPP ionophore needs lipophilic sites of opposite charge of the target anion in the polymeric membrane that helps to increase the selectivity and sensitivity of the highly selective membrane (Ammann et al., 1985). This is where ionic additive serves as a performance enhancer in an ISE. Additionally, the addition of additives increases the response behavior and ionic strength in the membrane that helps the neutral carrier membrane to function properly i.e sensitive towards the target analyte (Zhang et al., 2006). Ionic additive of ammonium salt namely tetradodecylammonium chloride (TDDA) is the additive chosen to aid the neutral carrier of CoTMeOPP in selecting the target anion of SCN. The TDDA is widely used as additive for both polymerization of plasticized and non-plasticized membranes (Heng & Hall, 2000b; Schaller et al., 1994).

2.5.1.3 Ionophore

Ionophore is the organic and inorganic compounds that bind to ions. The ionophore acts as the active component in the polymeric membrane. Ionophore is the carrier either neutral or ionic that helps in the selectivity of the target anion in the sensor. The basic concept of ionophore is the same as metal-ligand and enzymes-substrate theory. The fabricated ionophore should ideally form strong binding, highly selective towards the target anion and form reversible complexes only with the target anion. This will prevent the problem of ion-exchange between the ionophore and the interfering anions. The ionophore should be lipophilic to reduce the possibility of ionophore leaching sample solution as low as possible. These properties of ionophore add to the performance of the ionophore in terms of selectivity, sensitivity, reproducibility as well as the life time (Bühlmann & Chen, 2012). CoTMeOPP serves as neutral ionophore that acts as the active component in the selective membrane and helps in the detection of target anion. This ionophore was chosen to be fabricated and incorporated in the sensor as the active material.

2.5.1.4 Photo-initiator

The photo-initiator is needed in order to initiate the photocurable polymeric membrane. The self-plasticizing polymeric membrane of n-BA has low T_g to self-polymerized but still needs an initiator to initiate the process of polymerization of the polymer onto the electrode of GCE. The simple photocurable process of n-BA and HEMA normally use dimethylphenylpiperazinum (DMPP) as well as 1,6-hxanediol diacrylate (HDDA) as the photo-initiator (Kisiel et al., 2008).

2.5.2 Fabrication of Electrode Membrane

The potentiometric ISE can be further classified into three main group of glass membrane, crystal or solid membrane and polymeric or liquid membrane (Faridbod et al., 2007). The liquid membrane ISEs as mentioned before has been widely used as electrode but however, the emerges of solid-state electrode has capture the attention of researchers as a way to prevent leaching of plasticizers and the sample solution to the sample solution and environment. Solid state electrode (Figure 2.7) have been proven to provide low electrical resistance in addition to the absence of the internal solution that can cause leaching of analyte of interest to the surroundings (Bobacka, 2006), ease method of preparation and handling.

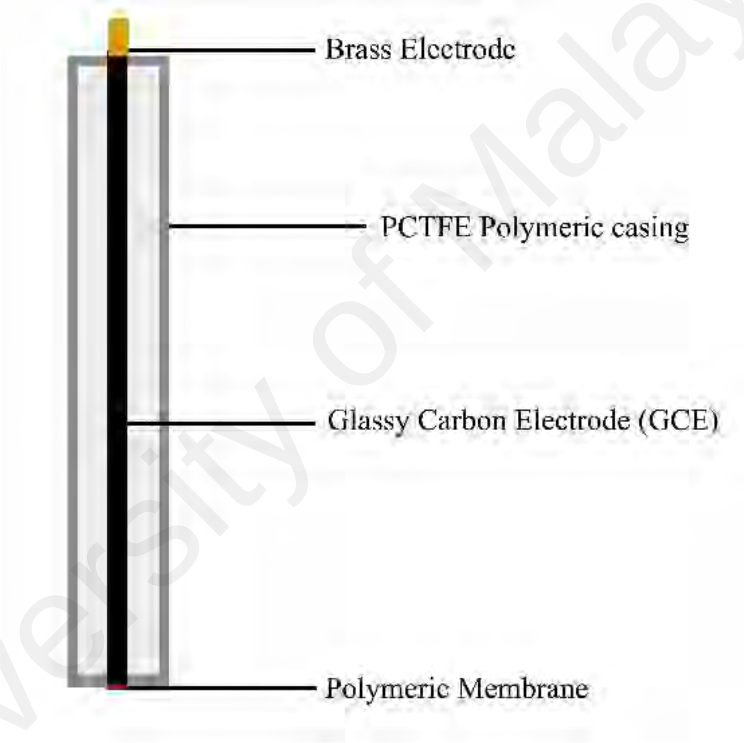


Figure 2.7: Schematic diagram of solid state electrode i.e GCE.

As shown in Figure 2.7, the polymeric membrane consisting of the active compounds of ionophore, additive and photo-initiator are fabricated onto the carbon disk on the surface of the electrode without the need of internal solution and plasticizer. This added up to the qualities of the fabricated sensor. The solid-state electrode that has been studied and utilized in this work is GCE and the facile preparation of the polymeric membrane offers a fast, non-complicated, simple and easy to work with without the need of any sophisticated training. The fabrication of the active compounds onto the GCE was done by means of photocuring by using ultra-violet (UV) radiation for 500 seconds. The method of photocuring has been introduced since the slf-plasticizing membrane was introduced as the new type of membrane to replace polymer that needs plasticizer in early 1990's (Antonisse et al., 1996).

The polymer of polysiloxane used by Antonisse and his co-workers has been proved to be able to polymerized without the utilization of plasticizer and able to perform the same way and even better than PVC membrane. Ever since, growing interest in self-plasticizing polymeric membrane to be incorporated in the fabrication of sensors has developed and new findings on the new type of polymers has emerged as new alternatives to increase the variability and performance of the sensor. The fabrication of polymeric membrane by photocuring method has developed to be a new facile, simple and fast method. Despite the presence of plasticizer in the polymeric matrix, the self-plasticizer still needs initiator and cross-linker to be able to polymerize when exposed to UV radiation (Heng & Hall, 2000a). DMPP and HDDA are the initiators and the cross-linker that has been proven to aid in initiating and cross-link the polymers in order to have successful polymerization of the active components onto the GCE (Kisiel et al., 2008). There are few numbers of research that reported the photocurable method in fabricating the ionophore onto the electrode.

Bean and his co-workers in 2004 have reported to fabricate ferrocene ionophore onto the solid-state electrode via one- step photocuring method by using methyl methacrylate (MMA) polymer for the detection of glucose (Bean et al., 2005). In his work, HEMA and MMA polymers are mixed together with DMPP as the photo-initiator in a vial to get homogenous mixture of polymer and ferrocene was added to the mixture and fabricated onto SPE before exposure to UV radiation. Bean has managed to fabricated poly (methyl-methacylate-(Co-2-hydroxylehtyl methacrylate) or MH with ferrocene ionophore embedded onto the polymeric membrane producing Fc-MH membrane onto the SPE electrode. The performance of the electrode was studied and by using photo-polymerization method, it is proven to delay the process of leaching of the polymeric membrane with high reproducibility and repeatability.

In 2006, Alva and his co-workers also worked with SPE just like Bean. However, Alva’s work is more delicate as he worked with four layers of membranes to optimized the sensor. The Ag and AgCl has already been printed onto the SPE and he collaborated the HEMA polymer with DMPP as photo-initiator as first layer. The first layer then exposed to UV radiation for three minutes with constant N_2 gas flowing throughout the photocuring process. The second layer of n-BA was aided by HDDA, DMPP and two active ionophores of valinomycin and sodium-tetrakis[bis-3,5(trifluoromethyl)phenyl] borate. This mixture was then photocured in UV radiation with continuous N_2 gas for three minutes. The fabricated electrode managed to deviate from the classical Hofmeister selectivity series and able to detect the ion of interest of potassium (K^+) ion with Nernstian response of 58.5 mV/decade. The schematic diagram of his work is as shown in Figure 2.8 (Alva et al., 2006). The fabricated sensor was able to work with wide range of K^+ concentration of $1x10^{-1}$ mol/L to $1x10^{-5}$ mol/L with detection limit of 2.5×10^{-6} mol/L with good reproducibility.

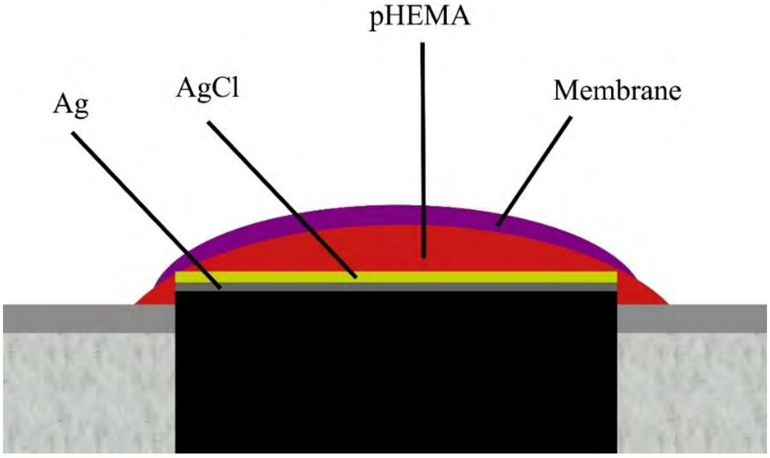


Figure 2.8: The schematic diagram of Alva et al., self-platicizing membrane.

Based on these previous photocuring techniques reported, there still no published work on self-plasticizing membrane for the detection of thiocyanate. Thus, this needs further exploration and research in searching for green sensor without the utilization of plasticizer membrane for fabrication of the sensor.

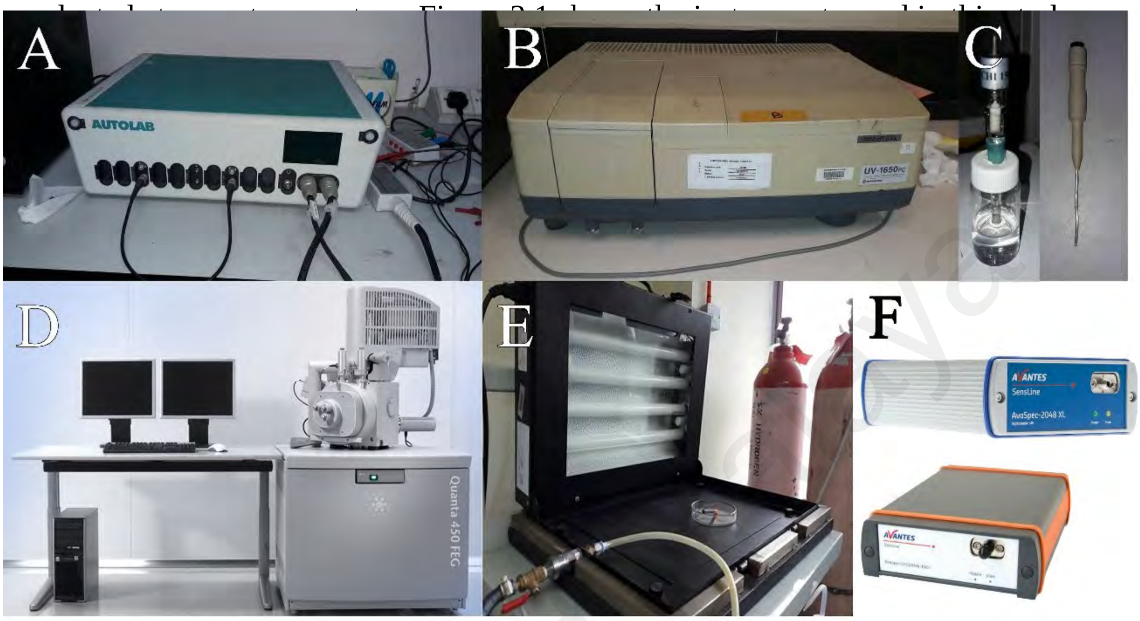
CHAPTER 3: METHODOLOGY

3.1 Chemicals and Instruments

All glasswares were dried in an oven overnight prior to use. Membrane materials such as 5,10,15,20-tetrakis(4-methoxyphenyl)-21H,23H-porphyrin-Co(II) (CoTMeOPP) as an ionophore, 1,6-hexanediol di-acrylate (HDDA) as a cross linker, 2,2-dimethoxy-2-phenylacetophenone (DMPP) as initiator, and tetradodecylammoniumchloride (TDDA) as an additive were purchased from Sigma-Aldrich, USA. Hydroxyl-ethyl methacrylate (HEMA) and n-butyl acrylate (n-BA) were purchased from Merck, Germany and used as the monomer of initial and membrane layers of electrode, respectively. Silver nitrate ($AgNO_3$) from Merck, USA and potassium chloride (KCl) from Lobal Chemie, India were purchased. The acetone and absolute ethanol solvents were used throughout the research work from R&M Chemicals, U.K and J Kollin Chemicals, respectively. The potassium thiocyanate (KSCN) and all other chemical salts from Sigma-Aldrich, USA and Fluka, USA were of analytical grade and used without further purification. The optimized fabricated highly selective membranes were characterized by Fourier-Transform Infrared (FT-IR) spectroscopy, fluorescence spectroscopy, UV-vis spectroscopy, Field Emission Scanning Electron Microscope (FESEM) and Dispersive X-Ray Analysis (EDX).

The fabrication of the membrane was aided with Autolab Potentiostat (PGSTAT-302N) from Metrohm, Platinum (Pt) rod, Ag/AgCl electrode and UV LED exposure box (220/240Hz). The electromotive force (E/mV) of fabricated sensors were measured relative to Ag-AgCl double junction reference electrode (Thermo Orion 900200) using a digital pH/ISE meter (Orion Star A214) with Orion reference electrode fill solution as the internal solution. All measurements were carried out at room temperature. The morphology study of the fabricated sensors was carried out by using Sputter Coater (Polaron CPD 7501 Critical Point Dryer Biorad Sputter Coater), FESEM and EDX

(Quanta FEG 450, EDX-OXFORD) instruments and the binding properties of the fabricated sensor were studied by using fluorescence spectroscopy (AvaSpec- 2048 XL) and UV-vis spectroscopy (UV-1650 PC). All measurements and characterizations were conducted at room temperature. Figure 3.1 shows the instruments used in this study.



A- Autolab Potentiostat (PGSTAT-302N), Metrohm
B- UV-Vis spectroscopy (UV-1650 PC)
C- Electrodes of Ag/AgCl and Pt
D- FESEM and EDX (Quanta FEG 450, EDX-OXFORD)
E- UV-LED exposure box (220/240Hz)
F- Fluorescence (AvaSpec-2048 XL)

Figure 3.1: Instruments used in this study.

3.2 Fabrication of Ion Selective Electrodes (ISE) Based on CoTMeOPP Ionophore

The sensor membranes were fabricated and photocured according to the modification method reported previously (Alva et al., 2006). The procedure of preparing ISE was as follows. First, the GCE are cleaned from any impurities by using 0.1 μm alumina on a nap less cloth. The GCE was ensured to be free and cleaned from the alumina powder and foreign materials prior to use. Then, the deposition of Ag(s)/AgCl(s) as internal reference electrode on the surface of GCE was conducted by using cyclic voltammetry method by using Autolab potentiostat (PGSTAT-302N), Metrohm (Figure 3.1 (A)). The aqueous solution of 2×10^{-3} mol/L of silver nitrate ($AgNO_3$) containing 1.0 mol/L potassium nitrate (KNO_3) at pH 5.93 was prepared and placed inside the solution vial as the coating solution i.e the source of Ag. Cyclic voltammetry was used by applying a single cycle program for finding out the potential intervals of reducing the Ag(I) to Ag(0) and oxidation of silver took placed onto GCE, respectively. The Ag was then deposited onto the GCE (Figure 3.2 and 3.3) with cyclic voltammetry in the range of -0.5 V to -0.08 V for 40 cycles with a scan rate of 0.1 mV/s.

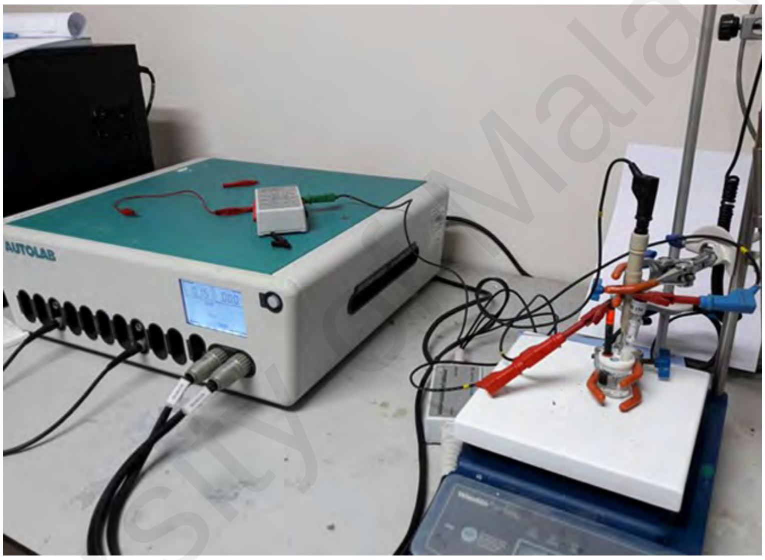


Figure 3.2: The instrument set-up for the electrodeposition of Ag onto the GCE as first layer.

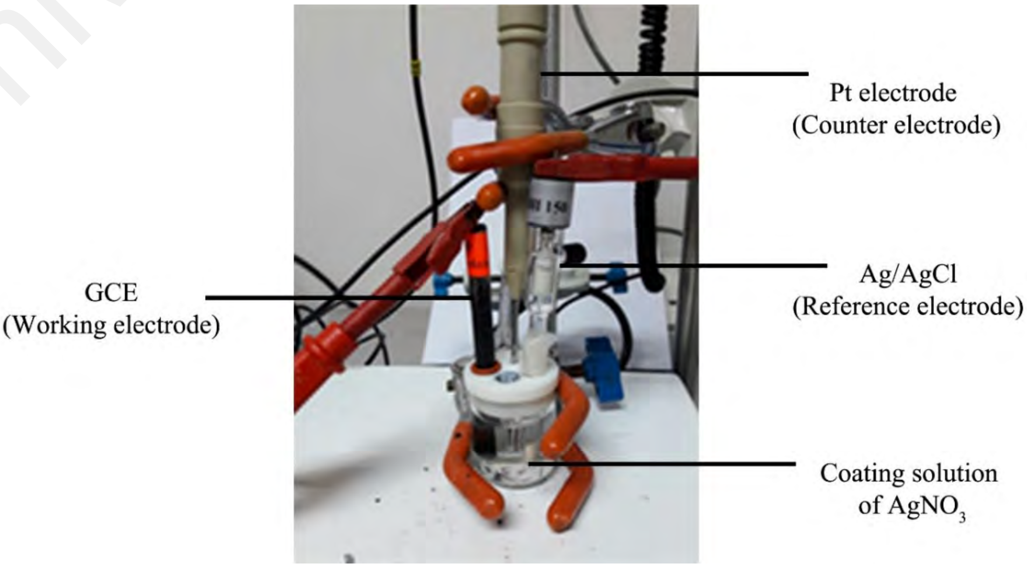


Figure 3.3: The cell setup of Ag coating onto GCE.

Modified glassy carbon electrode with first Ag deposited layer and secondly was dipped for 15 minutes into a solution of 0.01 mol/L KCl at pH 6.85 then dried at room temperature. After that, mixture of inner layer was prepared. 95.14 wt.% of HEMA and 4.86 wt.% of DMPP were mixed thoroughly in a vial and sonicated to get a homogenous mixture. 0.5 µL of the colorless mixture of HEMA and DMPP were drop-casted by using micropipette onto the Ag(s)/AgCl(s) deposited GCE as second layer. The polymeric membrane of HEMA was then polymerized by photocuring onto the modified Ag(s)/AgCl(s) deposited GCE by using UV box with a constant flow of nitrogen gas (N_2) for 500 seconds (Figure 3.4). The inner membrane of poly-(HEMA) was polymerized by photocuring method.

Figure 3.4: The photopolymerisation of self-plasticizing membrane by using UV radiation.

The inner membrane was then dipped or conditioned into solution of $1x10^{-2}$ mol/L of potassium thiocyanate (KSCN) solution for 15 minutes and dried at room temperature. Then, the last layer of the membrane was prepared by mixing 0.34 wt.% CoTMeOPP, 0.56 wt.% DMPP, 0.08 wt.% HDDA, 0.16 wt.% TDDA and 98.86 wt.% n-BA. The same procedure of sonicating to ensure homogenous mixing of the active components of the outer layer. 0.50 µL of the purple-colored mixture was then dropped into the hydrated poly-(HEMA) modified Ag(s)/AgCl(s) deposited GCE and photocured for 500 seconds by using UV box with continuous flow of N_2 gas. The sensor was fabricated with CoTMeOPP as the ionophore using self-plasticizing membrane. The schematic diagram of the fabricated are as shown in Figure 3.5.

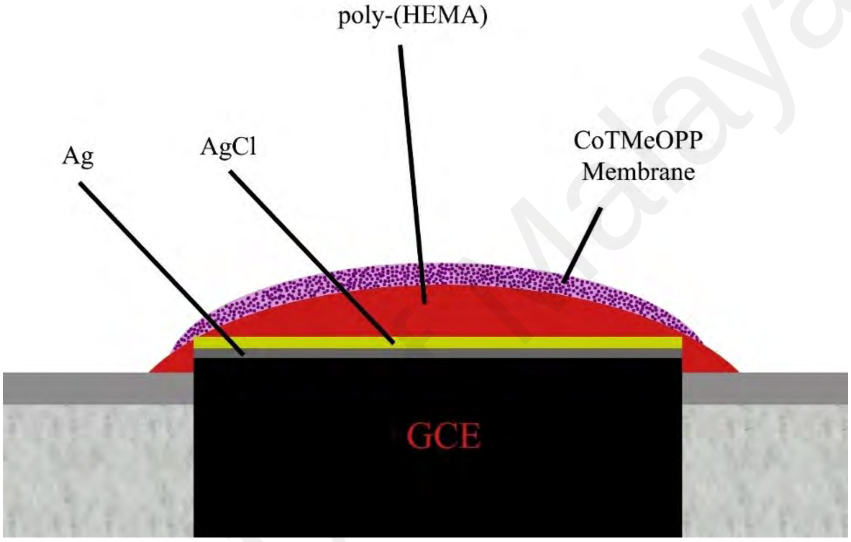


Figure 3.5: The schematic diagram of the fabricated membrane onto GCE.

3.3 Sensor Evaluation

The ISEs potentiometric measuring setup consists of a galvanic cell. A galvanic cell refers to two electrodes that are connected to the two terminals of a voltmeter. The two electrodes are an indicator electrode and a reference electrode. Both electrodes are immersed into the sample solution and connected to the two terminals of a voltmeter to be translated into readings (Lindner & Pendley, 2013). The setup of ISEs are as shown in Figure 3.6. The simple and facile setup and processes of ISEs make it a preferable method of measuring potential that involves ion selective membrane for a target ion. The common reference electrode used by the ISEs is Ag/AgCl and there are a number of different ISEs that have been widely used by researchers as transducers in the potentiometric set up of

ISEs. The ISEs are commonly used as detectors for different types of ions such as hydrogen ion (H^+), OH^-, NH_2^-, NH_4^+, Cl^-, Br^- and many other ions.

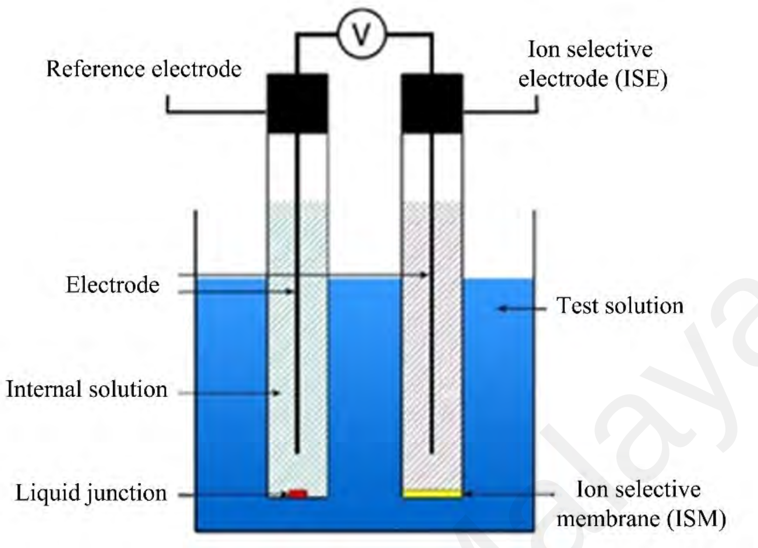


Figure 3.6: The setup of Ion Selective Electrode (ISE).

The translation of ions' activities is calculated via Nernst's equation as follow:

$E_{cell} = E°_{cell} – (RT/nF) \ln Q$ …………….. (Eq. 3.1)

Based on Equation 3.1;

E_{cell} = cell potential under non-standard conditions (V),

E^O_{cell} = cell potential under standard conditions,

R = Gas constant i.e 8.31 (volt-coulomb) / (mol-K),

T = Temperature (K),

n = number of moles of electrons exchanged in the electrochemical reaction (mol),

F = Faraday's constant (96500 coulombs / mol),

Q = Reaction quotient.

$$E_{cell} = E_O - \frac{0.0591\,V}{n} \log_{10} Q \text{...............} \quad \text{(Eq 3.2)}$$

Theoretically, the Nernst's equation will give the voltage that showed the activities of the ions inside the system within a concentration range normally from 1x10^{-1} mol/L to 1x10^{-8} mol/L of standard and a graph of standard was plotted. For the detection of cation, the fabricated ionophore selective membrane will give a positive slope and negative slope for the detection of anion. In the case of SCN anion, the n is equal to the charge that is -1 and thus, the standard graph will give a slope that shows positive response that diverge from the Hofmeister selectivity series and obeys the Nernstian equation of (Eq 3.2) (Figure 3.7).

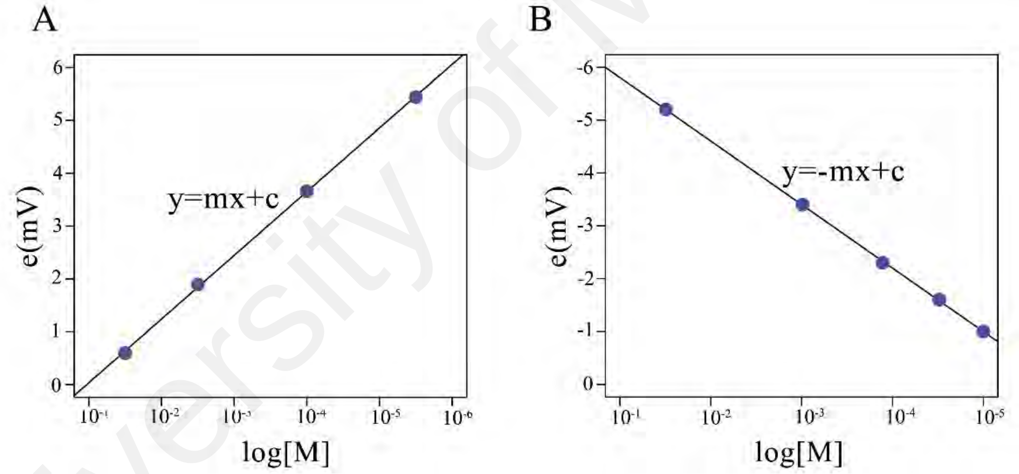


Figure 3.7: The example of standard calibration curve based on ISE for (A) Cation and (B) Anion.

By comparing the Eq 3.2 with linear formula of y = mx + c, the gradient of the standard graph of SCN anion will give negative slope of -60.0 mV/decade. However, literature value of Nernstian slope is -58.2 mV/decade (Ammann et al., 1986).

The responses of the sensors were measured in an electrochemical cell setup. The external reference electrode of double junction Ag/AgCl was used with 3.0 mol/L KCl as the inner solution of the reference electrode (Orion 900002) and 0.10 mol/L of lithium acetate (LiOAc) as the gel that bridges the electrolyte. The ISE membrane was prepared according to the methods of fabrication of ionophore onto the GCE in section 3.1 and the GCE was used as the working electrode in the system. Both electrodes were connected to an Orion pH/ISE meter as shown in Figure 3.8 and Figure 3.9.

Figure 3.8: The electrochemical setup of fabricated sensor.

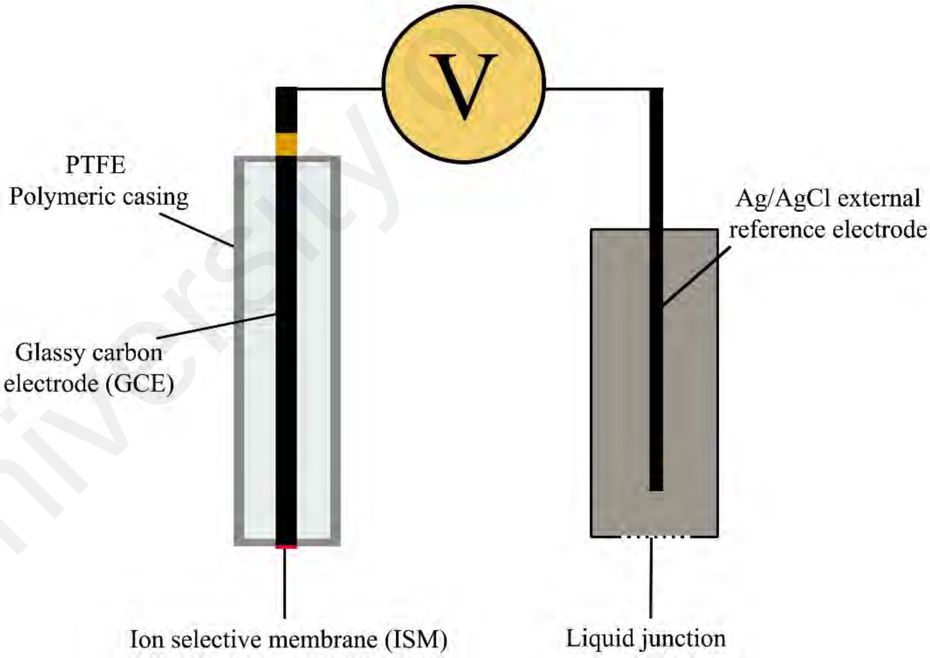


Figure 3.9: Schematic diagram of an ion-selective electrode in this research.

The SCN standard or test solutions were prepared by dissolving colorless KSCN salt with deionized water with concentrations ranging from 1 x 10^{-8} mol/L to 1 x 10^{-1} mol/L, respectively. The potential readings for the fabricated SCN-selective membrane with different compositions of active materials were taken by using the prepared standards of SCN anion solutions. The tests were conducted starting from low to high concentrations of standard solution of SCN anion and the potential readings were recorded once stable reading (mV) was obtained at approximate 1.13 minutes to 3.8 minutes. The summarization of cell potentials measured can be represented by using a schematic galvanic cell as follows:

Ag | AgCl (s) | KCl (3 mol/L) | bridge electrolyte | SCN- solution | CoTMeOPP membrane | Inner filling membrane layer | AgCl(s) | Ag

The optimization of the SCN-selective membranes was conducted to study the best composition of the active membranes to work at optimum in determining the concentration of SCN anion in the test solutions as well as in real samples. The different compositions of the active materials in fabrication of SCN-ISE are conducted by varying the amount of the active materials as shown in Table 3.1

Table 3.1: The different composition of ISE membrane based on CoTMeOPP ionophore for the potentiometric detection of SCN anion.

SCN-ISE	Inner layer			Outer layer					
	HEMA (mol%)	DMPP (mol%)	Total (mol %)	n-BA (mol%)	HDDA (mol%)	CoTMeOPP (mol%)	TDDA (mol%)	DMPP (mol%)	Total (mol%)
E1	95.14	4.86	100	99.20	0.08	0.00	0.16	0.56	100
E2	0.00	0.00	0	98.86	0.08	0.34	0.16	0.56	100
E3	95.14	4.86	100	98.86	0.08	0.34	0.16	0.56	100
E4	95.14	4.86	100	99.02	0.08	0.18	0.16	0.56	100
E5	95.14	4.86	100	99.01	0.08	0.35	0.00	0.56	100
E6	95.14	4.86	100	98.84	0.08	0.36	0.16	0.56	100
E7	95.14	4.86	100	98.75	0.08	0.45	0.16	0.56	100
E8	95.14	4.86	100	98.89	0.08	0.45	0.02	0.56	100
E9	95.14	4.86	100	98.81	0.08	0.45	0.10	0.56	100

Continued from Table 3.1

E10	95.14	4.86	100	98.77	0.08	0.45	0.14	0.56	100
E11	95.14	4.86	100	98.71	0.08	0.45	0.20	0.56	100
E12	95.14	4.86	100	98.66	0.08	0.54	0.16	0.56	100
E13	95.14	4.86	100	98.30	0.08	0.90	0.16	0.56	100
E14	95.14	4.86	100	97.95	0.08	1.26	0.16	0.56	100

3.4 Effect of pH on Electrode Performance

The effect of pH on potential responses was evaluated in two different concentrations of $1x10^{-5}$ mol/L and $1x10^{-2}$ mol/L test solutions at room temperature. The pH of SCN anion test solutions was adjusted from pH 1.0 to 12.0 by the addition of hydrochloric acid (HCl) and sodium hydroxide (NaOH) solutions respectively (Abbaspour et al., 2002). Stable readings were obtained before potential readings were taken and recorded.

3.5 Determination of Selectivity Coefficient

The potentiometric selectivity coefficient $\left(\mathrm{K}^{\mathrm{pot}}_{\mathrm{X}^{\mathrm{Y}-},\mathrm{M}^{\mathrm{n}-}}\right)$ was evaluated using the matched potential method (MPM) according to IUPAC recommendations (Arvand et al., 2007). This method measures the difference in potential of both working and reference electrodes according to the two separate solutions of X^{Y-} and M^{n-} in which X^{Y-} is the target anion of SCN and M^{n-} is the interfering anions at the same activity i.e $(a_{X^{Y-}} = a_{M^{n-}})$. The selectivity coefficient is as follow:

$$K^{MPM}_{X^{Y-},M^{n-}} = a^{\left(1-\left(\frac{y}{n}\right)\right)} e^{\left(E_{M^{n-}} - E_{X^{Y-}}\right) yF/\,(RT)V} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad \text{Eq. 3.3}$$

Where the notations above refer to:

$E_{M^{n-}} =$ potential measured values of interfering anions

$E_{X^{Y-}} =$ potential measured values of target anion

F = Faraday constant (96 485 J)

R = Universal gas constant (8.314 J/mol K)

T = Room temperature at 298 K

3.6 Response Time of Fabricated Electrode

The response time of a sensor was considered as the required time needed by the sensor to give a stable potential reading within ±1 mV reading during the immersion of the sensor in a series of respective test solutions. In this study, the response time for the sensor to reach stable state and provide stable reading was conducted. The time taken after each successive immersion of the electrode in the series of SCN stock solutions ranging from 1.0×10^{-8} mol/L to 1.0×10^{-1} mol/L was recorded and analyzed. The measurement was conducted from low concentration to a higher concentration at room temperature (Hassan et al., 2003).

3.7 Repeatability, Reproducibility and Stability of Fabricated Electrode

The parameters of repeatability, reproducibility and stability of the fabricated sensor need to be evaluated in order to validate the performance of the sensor. The repeatability of the fabricated sensor was tested by using the same optimized electrode to undergo three repetitive potential measurements of SCN standards in a day provided the tip of the electrode was washed with deionized water thoroughly after each measurement (Shirmardi-Dezaki et al., 2013a). While the reproducibility of SCN-ISE was examined using three different fresh electrodes with optimum composition and each electrode was used only once with the test solution of SCN anion within the linear range of the optimized SCN-ISE on the same day. The stability of the electrode was evaluated by repeated calibration of the electrode in KSCN standard solution within the working linear range with freshly prepared electrode and a few consecutive days using the same electrode (Hassan et al., 2003).

3.8 Analytical Application of Fabricated ISEs

3.8.1 Potentiometric Titration

The sensor was treated with $1x10^{-2}$ mol/L of $AgNO_3$ (25 ml) solution. A freshly prepared solution of $1x10^{-2}$ mol/L of SCN (20 ml) was placed in a clean beaker and titrated with the $AgNO_3$ freshly prepared earlier by using calibrated micropipette in a stepwise manner. The mixture was mixed thoroughly using magnetic stirrer before the potential reading was taken. The potential reaction solution mixtures were measured by using the same digital pH/ISE meter at each titration by using the same reference electrode of Ag/AgCl (Arvand et al., 2007). Stable and static reading of the sensor was taken.

3.8.2 Real Sample Applications

The applicability of the fabricated selective membrane fabricated on GCE electrode was tested directly in sample solutions of Klang river water, sediments from ex-mining area in Lahad, Perak, fish samples and saliva of both smokers and non-smokers (Arvand et al., 2007; Hovinen et al., 1999; Michigami et al., 1992). The determination of SCN anions in Klang river water, sediments and fish samples were conducted by using spike method (Eymard & Genot, 2003). Eight arrays of SCN concentration solutions were prepared ranging from $1x10^{-8}$ mol/L to $1x10^{-1}$ mol/L for the standard calibration curve preparations. Fish samples have been treated prior to the analyzing process due to the relatively low pH of the samples. The determination of the SCN concentrations were determined directly without any pre-treatment for other samples. Each sample was analyzed in triplicate and the analysis was repeated under identical conditions. The sampling location of river water and sediments are as shown in Appendix 1 and 2.

CHAPTER 4: RESULTS AND DISCUSSION

4.1 Optimization of CoTMeOPP Based SCN-ISE

In this part, the potential capability of the CoTMeOPP ionophore to be used as ionophore for the detection of SCN anion was investigated and developed to work at optimum. Therefore, a novel potentiometric self-plasticizing acrylate polymer based on CoTMeOPP as the active compound for the determination of SCN anions was fabricated, optimized and studied.

4.1.1 Optimization of the Composition of Fabricated Membranes

The composition of self-plasticizing membranes with the presence of ionophore can affect the performance of a sensor i.e the positive potentiometric response that deviates from the classic Hofmeister series and the detection limit of the sensor. Gupta et al., and Ibrahim et al., have reported significant influence on the performance of the fabricated sensor especially the selectivity, sensitivity and detection limit when the amount of ionophore incorporated in the polymeric membrane varies. Other active components were also proven to aid in enhancing the performance of the fabricated sensor (Gupta et al., 2002; Ibrahim et al., 2010) such as additives and polymeric membranes. As the CoTMeOPP ionophore is said to be the active component that detects the active anion of SCN in a sample solution, the amount of the ionophore to be incorporated in the self-plasticizing membrane plays a vital role. The right amount of ionophore utilized in fabrication of anion selective electrodes needs to be determined to maximize the sensitivity and the performance of the sensor. In this study, in order to find the optimized membrane composition for the SCN-ISE, different compositions of the active materials i.e CoTMeOPP ionophore and TDDA additive were incorporated onto the GCE with some modifications. These electrodes with different compositions were tested with the sample solution of SCN range from $1x10^{-8}$ mol/L to $1x10^{-1}$ mol/L (Table 4.1).

omposition (mol%)							Electrode characteristics		
Inner Lay		Outer Layer					Slope (mV/decade)	Linear Range (mol/L)	Detection limit (mol/L)
HEMA	D	n-BA	HDDA	CoTMe OPP	TDDA	DMPP			
95.14	4	98.86	0.08	0.34	0.16	0.56	31.15	1x10^{-6} – 1x10^{-1}	1.02x10^{-6}
95.14	4	99.20	0.08	0.00	0.16	0.56	-22.93	1x10^{-3} – 1x10^{-1}	9.77x10^{-4}
0.00	0	98.86	0.08	0.34	0.16	0.56	-30.83	1x10^{-7} – 1x10^{-1}	8.51X10^{-8}
95.14	4	98.86	0.08	0.34	0.16	0.56	-60.76	1x10^{-7} – 1x10^{-1}	9.55x10^{-8}
95.14	4	99.02	0.08	0.18	0.16	0.56	-37.98	1x10^{-4} – 1x10^{-1}	9.77x10^{-5}
95.14	4	99.01	0.08	0.35	0.00	0.56	-62.69	1x10^{-4} – 1x10^{-1}	8.51x10^{-5}
95.14	4	98.84	0.08	0.36	0.16	0.56	-54.82	1x10^{-5} – 1x10^{-1}	8.32x10^{-6}
95.14	4	98.75	0.08	0.45	0.16	0.56	-65.96	1x10^{-5} – 1x10^{-1}	8.91x10^{-6}
95.14	4	98.89	0.08	0.45	0.02	0.56	-36.79	1x10^{-5} – 1x10^{-1}	8.91x10^{-6}
95.14	4	98.81	0.08	0.45	0.10	0.56	-72.87	1x10^{-4} – 1x10^{-1}	7.94x10^{-5}
95.14	4	98.77	0.08	0.45	0.14	0.56	-49.18	1x10^{-4} – 1x10^{-1}	7.08x10^{-5}
95.14	4	98.71	0.08	0.45	0.20	0.56	-50.59	1x10^{-4} – 1x10^{-1}	8.13x10^{-5}
95.14	4	98.66	0.08	0.54	0.16	0.56	-67.95	1x10^{-5} – 1x10^{-1}	9.44x10^{-6}
95.14	4	98.30	0.08	0.90	0.16	0.56	-67.48	1x10^{-5} – 1x10^{-1}	9.55x10^{-6}
95.14	4	97.95	0.08	1.26	0.16	0.56	-62.07	1x10^{-5} – 1x10^{-1}	8.32x10^{-6}

CoTMeOPP ionophore for the potentiometric detection of SCN anion.

num membra omposition **Absence of Ag/AgCl layer

Composition (mol %)		Electrode characteristics		
Inner Layer	Outer Layer	Slope	Linear Range	Detection limit

The fabricated ion selective membranes (ISM) onto GCE electrode of E0 to E14 based on Table 4.1 have been tested for the determination and selectivity of SCN anion (SCN-ISE) at different concentrations of standard SCN anion solutions ranging $1x10^{-8}$ mol/L to $1x10^{-1}$ mol/L. As seen in Table 4.1, for E0, the SCN-ISE has been fabricated without the Ag/AgCl layer i.e the first layer with the optimized membrane composition. The potentiometric test was conducted and showed negative Nernstian response i.e positive gradient of 31.15 mV/decade. Fabricated SCN-ISE without Ag/AgCl layer showed wide working range of $1x10^{-6}$ mol/L to $1x10^{-1}$ mol/L despite its negative Nernstian response. Figure 4.1 shows the assembly of the ISE that is commonly used which involves Ag/AgCl as external reference electrode with the desired anion or cation for the internal solution and the ion-selective membrane (ISM). Silver (Ag) is widely used as it is a conductive noble metal that is commonly used as electrode as it is stable and highly conductive. GCE was also proven to be highly conductive and stable electrode. The conductivity and the sensitivity of the transducer was assumed to increase by depositing the Ag/AgCl layer onto the GCE. Thus, in this study, Ag/AgCl layer was incorporated in fabrication of ISM as it enhanced the performance of the SCN-ISE based on the desired response i.e deviates from Hofmeister series and showed positive Nernstian response. The summarization of the modification can be seen in Figure 4.1.

Figure 4.1 shows the schematic diagram of the working principle of the fabricated SCN-ISE based on GCE as transducer. The polymeric membrane (PM) in Figure 4.1 is HEMA (inner layer), ISM is the optimized ion-selective membrane of n-BA, CoTMeOPP and TDDA active materials, e is the electron, + is the “hole” of the oxidized PM and L is the ion-recognition site of the ionophore. The primary ions are circled and counterions are squared. Based on the schematic diagram in Figure 4.1, the signal transfer can be portrayed by the arrow. The signal transfer from the target anion in solution are transferred to ISM and hence towards PM that will eventually reach GCE (right to left).

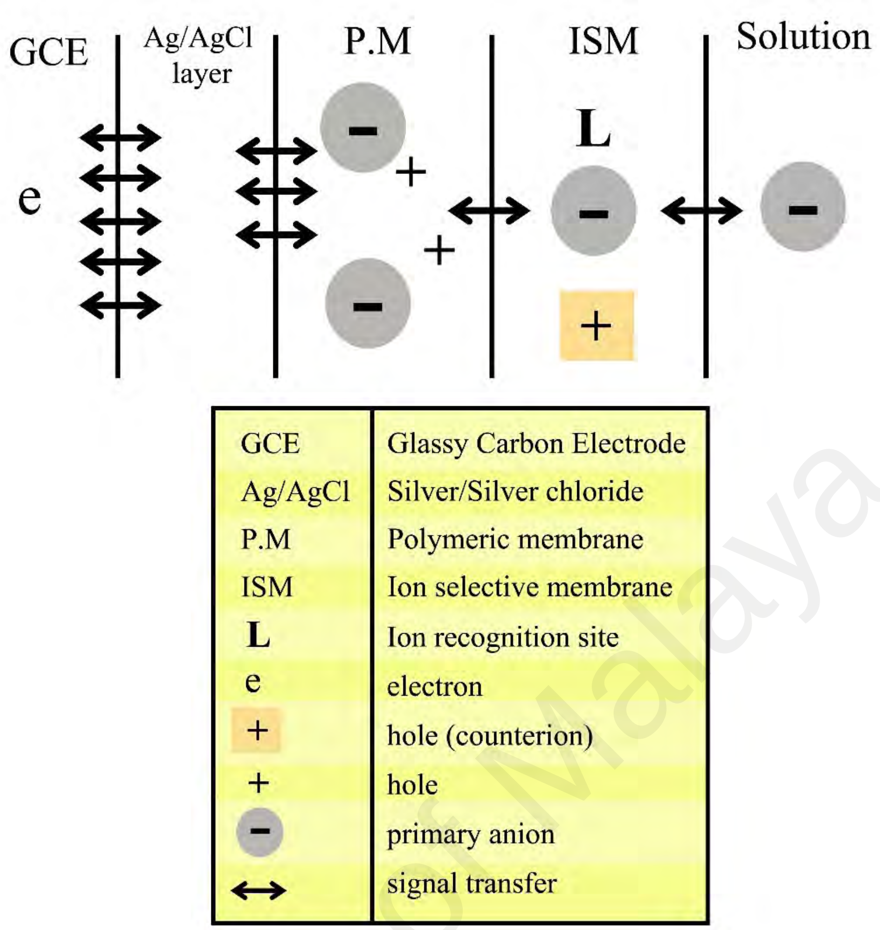


Figure 4.1: Operating principle of solid-contact SCN-ISE.

As the signals flow from the solution up till PM i.e from right to left, the signal was relatively slow (represented by few number of arrow), but as the signal is approaching Ag/AgCl layer and GCE, the signal is rapid and enhance (by referring to the number of arrows in the Figure 4.1). Ag/AgCl layer is highly conductive and plays a vital role in transferring and enhancing the signal rapidly to GCE. This can be proven by observing the obvious improvement in the Nernstian response (slope) as well as other parameters of E3 that has the same composition of inner and outer layer as E0 but E3 was modified by deposition of Ag/AgCl first layer. The SCN-ISE with Ag/AgCl first layer has successfully showed anti-Hofmeister behavior that obeys the Nernstian response with negative slope. In addition to that, there are numerous reported works that employ the Ag/AgCl layer as the fundamental layer before depositing ISM onto the electrodes to enhance the performance of the ISEs as well as obtaining the positive Nernstian response and better slopes (Heng & Hall, 2000a, 2001). Based on this result, the Ag/AgCl aid in the electron transfer or signal transfer for a better performance of the fabricated SCN-ISE and thus, the fabrication of SCN-ISM onto GCE was modified by deposition of Ag/AgCl layer as the first layer.

The ISM of E1 to E14 was fabricated with Ag/AgCl as the first layer, HEMA and DMPP as the inner layer and n-BA, HDDA, CoTMeOPP, TDDA and DMPP as the outer layer (Figure 3.5). To investigate the selectivity and the sensitivity of the ionophore towards SCN, the SCN-ISM was fabricated without ionophore in the outer layer (E1). E1 shows a positive Nernstian response i.e negative gradient of the standard graph that deviates from the Hofmeister series but however displayed a deviated potentiometric response towards the target anion of SCN with slope of -22.93 mV/decade. This result clearly showed that CoTMeOPP ionophore plays a vital role in sensing SCN anion and the absence of CoTMeOPP as the active materials resulted in relatively low response of the E1 ISM. Other than that, it can also be seen that Ag/AgCl first layer assisted and enhanced the electron transfer between SCN anion, ion-selective membrane and the transducer as it managed to give desired type of Nernstian response i.e negative standard graph despite the narrow working range and high detection limit. The negative slope of the standard Nernstian graph also manifested that the self-polymerized membranes of HEMA and n-BA and the additive used in fabricating the SCN-ISE contributes in the selectivity and the performance of the SCN-ISE. Despite the slope being negative as what expected response of ionophore towards anion of interest i.e SCN, the slope of E1-ISE deviates from theoretical value i.e -58.2 mV/decade (Ammann et al., 1986). Other than demonstrating CoTMeOPP is the essential sensing mechanism for SCN anion and the deposition of Ag/AgCl assist in acquiring the desired Nernstian slope, it also shows that the fabricated polymeric membranes itself (HEMA and n-BA) are able to response towards anions but in small percentage of sensitivity. However, there are previous studies to show the responsive behavior of the self-plasticized polymeric membranes towards ions in the fabrication of ISEs. In 2000, Heng and Hall actively studied the response of self-plasticizing acrylic polymer responses towards cation in the fabrication of cation ISEs. In his work, the utilization of acrylic monomers of n-BA and MMA without ionophore in the ISM showed slight responses towards the target cations. This somehow match the condition in E2 in which no ionophore was incorporated on to the membrane and the polymeric membranes of HEMA and n-BA able to give slight response towards the target anion of SCN (Heng & Hall, 2000a, 2001).

Addition of various wt.% of ionophore in the construction of ISM from E2 to E14 showed different results in Nernstian responses, working ranges and detection limits. The consecutive E2-E14 ISM, ionophore were incorporated together with the conductive polymeric membranes shown in Figure 3.5 and the Nernstian responses of the fabricated ISM electrodes showed relatively fitter and better slopes than E0 and E1 i.e electrode without Ag/AgCl layer and ionophore respectively. This clearly proves that the ionophore plays a vital role as the active sensing agent for the detection of SCN anion in the fabricated membrane and can be said to be SCN-selective ionophore. In E2, there was no inner layer of polymeric membrane HEMA used in the fabrication of the SCN-selective membrane to study the effect of HEMA inner layer with the performance of the SCN-ISE. The fabricated electrode without the HEMA inner layer shows Nernstian response of -30.83 mV/decade which is half of the theoretical value and can be said to give negative Nernstian response towards SCN anion. However, despite the slope deviated from the theoretical value of -58.2 mV/decade, E2 ISM can be observed to have relatively wide linear working range of $1x10^{-7}$ mol/L to $1x10^{-1}$ mol/L with relatively low detection limit of $8.51x10^{-8}$ mol/L.

Other than that, by comparing ISMs of E2 and E3, an apparent and significant improvement in the Nernstian responses and slopes of E3 (-60.76 mV/decade) compared to E2 (-30.83 mV/decade) when an amount of CoTMeOPP ionophore is incorporated onto the ISM. As can be seen, when comparing E2 and E3, in E2 ISM, no HEMA inner layer was deposited onto the ISE and in E3, HEMA inner layer was deposited together with the same amount of ionophore used in E2. Based on these results, it can be said that HEMA highly influence and contributes to the sensitivity of the SCN-ISE and the outer layer works in sync with Ag/AgCl first layer in detection of target anion to give positive Nernstian response. HEMA was utilized previously by Reinhoudt in 1994 as mentioned previously in fabrication of K-ISE. In his work, the HEMA polymer was conditioned with the target cation solution to improves the sensitivity, selectivity and the performance of the ISE (Reinhoudt et al., 1994). Other than him, Qin, Heng and Hall also conditioned the polymeric membrane in the target cation solution to enhance the performance of the ISEs (Heng & Hall, 2000c; Qin et al., 2003). This important step of conditioning the polymeric membrane inside the target ions can help the self-plasticize polymeric membrane to create a signal 'pathway' of the target ion in the test solution into the transducer for better detection. In other words, the conditioning HEMA layer or inner layer will cause an adjustment of the inner phase boundary potential and as the ion transport between test solution and the outer layer, it will alter the composition of the inner 'solution' i.e HEMA, resulting in a drift of the membrane potential and thus aids and enhance the signal of the target ion (Michalskan et al., 2004).

From the results obtained from E2 ISM, the membrane showed to have almost good response towards SCN anion but the sensitivity towards SCN anion is low. This eventually demonstrated that the inner layer of HEMA aids and contributes in the highly selective detection of SCN anion for the SCN-ISE as the conductive polymer and additional inner phase boundary as mentioned in Chapter 2 before. Based on previous works, it was found that the conductive polymers of HEMA and n-BA chosen in this work play a significant role in the performance of the ISEs by providing a medium of transfer between the stock solution and the transducer and the hydrophilic HEMA conditioned in aqueous KSCN allowed the saturation of the membrane with SCN (inner phase boundary layer) and ensured a better Nernstian slope thus increasing the sensitivity of the SCN-ISE.

As can be observed in E3, E4, E6 and E12-E14, different percentage of ionophore was incorporated with the electrode. In ISM E4, the smallest amount of CoTMeOPP ionophore of 0.18 wt.% was fabricated onto the electrode and the Nernstian response of E4 ISM compared to E1 ISM i.e without the CoTMeOPP ionohore has increased significantly from -22.93 mV/decade to -37.98 mV/decade and the working range has also broadened from $1x10^{-3}$ mol/L - $1x10^{-1}$mol/L to $1x10^{-4}$ mol/L - $1x10^{-1}$mol/L. It proves that the addition of ionophore onto the SCN-ISM enhanced the selectivity and sensitivity of the electrode towards SCN anion and the previous reported works of Vlascici et al., that showed CoTMeOPP ionophore is selective towards SCN anion can be proven (Vlascici et al., 2006). The increase in the performance of SCN-ISE as soon as the ionophore is incorporated into the ISM is a solid indicator that the CoTMeOPP ionophore is definitely a SCN-selective ionophore. However, with this narrow linear working range and relatively high detection limit, the fabricated SCN-ISE is far behind compared to another SCN-ISEs reported.

As the percentage of CoTMeOPP ionophore increases, in E3 ISM i.e 0.34 wt.%, major difference can be observed in Nernstian response, working range as well as the detection limit of the E3 ISM electrode. The E3 ISM gave the nearest Nernstian’s value as the reported theoretical value of -60.76 mV/decade with wide linear range of $1x10^{-7}$ mol/L to $1x10^{-1}$ mol/L with relatively low detection limit of $9.55x10^{-8}$ mol/L. The Nernstian response for E3 ISM can be said to fit the theoretical value and have the best fit for the detection of SCN anion. Based on previous studies PVC-based ISEs with different types of ligands in detection of SCN anion mentioned in Chapter 2 (Table 2.1), the best Nernstian response with slope nearest to the theoretical value was reported by Amini et al., Ganjali et al., Chandra et al., and Benvidi et al., with slopes of range -58.8 mV/decade to -59.1 mV/decade. These reported works based on PVC polymeric membrane however have relatively narrow linear working range and higher detection limit as compared to of the fabricated SCN-ISE in this work (Amini et al., 1999; Benvidi et al., 2014; Chandra et al., 2008; Ganjali et al., 2001). Based on Chandra's work i.e PVC-based ISE, the lowest detection limit reported was $1.25\text{x}10^{-7}$ mol/L within linear working range of $1\text{x}10^{-2}$ mol/L to $1\text{x}10^{-7}$ mol/L. Compared to fabricated self-plasticized SCN-ISE (E3), the electrode showed tremendous improvement in the linear working range and detection limit even without the utilization of the plasticizer. Based on Gupta et al., the utilization of plasticizer increases the performance of the cation ISE due to the presence of lipophilic site of the plasticizer (Gupta et al., 2006). Other than Gupta, Vlascici also reported the work with the detection of SCN anion by using CoTMeOPP ionophore with PVC-based membrane. In his work, the detection limit was reported to be $1\text{x}10^{-5}$ mol/L which is relatively higher compared to the fabricated SCN-ISE with E3 ISM (Vlascici et al., 2006). However, based on E3, enhancement in the detection limit of the ISE i.e relatively low detection limit can be observed compared to other reported PVC-based ISEs by using self-plasticized polymeric membrane.

The fabricated SCN-ISE fabricated successfully precede the previously reported SCN-ISE that involves plasticized PVC membrane in its fabrication. As mentioned previously in Section 2.5.1.1, in the year of 2012 and 2016, there are two reported works that utilized the self-plasticized membranes in the detection of anions of F and I (Matusevich et al., 2012, Mendecki et al., 2016) and there are no further reports on self-plasticized SCN-ISE have been reported. Based on their works, the linear working ranges and the detection limits of their purposed F-ISE and I-ISE also showed better working range and lower detection limit as compared to the previously reported fabricated PVC-based ISEs. The fabricated SCN-ISE in this work has also successfully showed a good and comparable sensitivity towards the target anion with the previous works reported by Matusevich and Mendecki. These three reported ISEs of F-ISE, I-ISE and SCN-ISE proves that no utilization of plasticizer is in a fabrication of ISEs to obtain a good Nernstian response with low detection limit.

On the other hand, by comparing E3 and E4 ISM, it can also be seen that a little difference in the amount of the active material of CoTMeOPP ionophore showed a significantly different response towards SCN anion. The fabricated SCN-ISE response towards SCN anion can be interpreted by analyzing the Nernstian response i.e the slope of the standard graph. Based on comparison between E3 and E4 ISMs, in terms of the selectivity, response and sensitivity, E3 ISM clearly showed better response towards SCN anion compared to E4 ISM (by analyzing the standard graph slope). Based on Gupta and Ibrahim, the amount of ionophore is essential in an ISM to get the maximum performance of the fabricated ISE. Thus, further optimization in the amount of ionophore is conducted to obtain the right amount of CoTMeOPP ionophore required in an ISM for maximum response.

To study whether increase in the amount of the CoTMeOPP ionophore will also increase in the sensitivity and selectivity of the SCN-ISM, a little increase of ionophore were tested in E6 i.e 0.36 wt.%. E6 ISM showed positive Nernstian response as expected but with different standard graph slope that slightly deviated from the theoretical value i.e -54.82 mV/decade with narrower linear range of 1×10^{-4} mol/L to 1×10^{-1} mol/L. Another increase in the amount of CoTMeOPP ionophore in E7 ISM i.e 0.45 wt.%

showed different response of Nernstian's slope that also slightly deviated from the theoretical value i.e -65.96 mV/decade. The linear range of 1x10^{-5} mol/L to 1x10^{-1} mol/L with low detection limit of 8.91x10^{-6} mol/L is totally lower than E6 ISM. It can be seen that, a little increase in the CoTMeOPP ionophore amount resulted in different Nernstian slope of the target analyte. However, no further trend can be observed. Further increase in the amount of CoTMeOPP ionophore was added to E12 ISM to study the relation and trend of the amount of ionophore utilized with the Nernstian's response. In E12 ISM, with 0.54 wt.% of CoTMeOPP ionophore, the Nernstian's slope is getting steeper than of E7 ISM i.e -67.95 mV/decade with the same working range of 1x10^{-5} mol/L to 1x10^{-1} mol/L and slightly higher detection limit of 9.44x10^{-6} mol/L.

Further increase in the amount of CoTMeOPP ionophore in the highly selective polymeric membrane of the SCN-ISM were conducted in E13 and E14 with 0.9 wt.% and 1.26 wt.% respectively. Both E13 and E14 ISMs exhibit different results than E3, E4, E6, E7 and E12. The Nernstian responses of both E13 and E14 did not deviate much from the theoretical value i.e -67.48 mV/decade and -62.07 mV/decade respectively and both with linear range of 1x10^{-5} mol/L to 1x10^{-1} mol/L. Despite having low detection limit of 9.55x10^{-6} mol/L for E13 ISM and 8.32x10^{-6} mol/L for E14 ISM, the Nernstian slope of both ISM that deviated from the theoretical value showed that these ISMs are not as sensitive and selective towards the target anion of SCN as compared to E3 ISM. Based on these results, it can be proven that CoTMeOPP ionophore is the active material in the detection of SCN anion and there is no readable trend of the amount of CoTMeOPP ionophore needed to utilized in the ISM for the optimized performance of the SCN-ISE. Thus, try an error was conducted to obtain the perfect amount of CoTMeOPP active material to be used in the ISM for the SCN-ISE to work at its optimum in detection of SCN anion.

It can be said that, the amount or weight percentage of CoTMeOPP ionophore utilized in a membrane does not necessarily be as much as compared to other materials as the SCN-ISE worked at its best performances only by utilizing 0.34 wt.% of the ionophore. Thus, it can be concluded that the optimum amount of ionophore need not to be the highest amount to work at optimum and` the 'right' amount of ionophore can be determined by try an error method and the 'right' amount of ionophore to be incorporated in the SCN-ISM for maximum performances is 0.34 wt.%.

As mentioned earlier, the lipophilic additive of TDDA contributes in the performance of the ISE (D Ammann et al., 1985; Heng & Hall, 2000c; Wegmann et al., 1984; Zhang et al., 2006). To study the effect of lipophilic additive towards the performance of the fabricated SCN-ISE, different amounts of TDDA additive is utilized into the SCN-ISM composition. Similar Nernstian responses can also be observed in ISM of E1 to E14 with various wt.% of TDDA cooperated in the fabrication of ISMs onto the GCE. In E5 ISM, no TDDA was utilized in the fabrication of highly selective SCN anion membrane and the Nernstian's slope of the fabricated SCN-ISE is slightly increased to -62.69 mV/decade with narrow linear range of 1x10^{-4} mol/L to 1x10^{-1} mol/L and relatively high detection limit of 8.51x10^{-5} mol/L. Other than functioning as ion exchanger in the ISE, the utilization of the additive in the fabrication of any ISEs were also proven to increase the performance of the ISEs by inducing itself a selective response towards the target ions. In this study, TDDA additive has lipophilic site with opposite charge (positively charged) of the target anion in the polymeric membrane. This condition aids in the response behavior and the ionic strength in the membrane which further increases the performance of the ISE and aids in inducing the appropriate anionic slopes and optimizes the potentiometric selectivity of the SCN-selective self-polymerizing membrane (D.Ammann et al., 1985, Bakker et al., 1994, Zhang et al., 2006). TDDA has widely utilize as additive in self-plasticized membranes as it contributes in the performance of the ISEs (Heng & Hall, 2000c, Matusevich et al., 2012, Medecki et al., 2016). Thus, it can be said that TDDA is one of the active materials in SCN-ISE for detection of the target anion.

As TDDA is proven to increase the performance of ISEs, the 'right' amount of TDDA needs to be investigated for maximum performance. In order to study the optimum amount of TDDA to be cooperated into the SCN-ISM, different wt.% of TDDA was utilized and the SCN-ISE response was studied by analyzing the response and sensitivity of the ISE (based on Nernstian's slope of the standard calibration graph). By differing the wt.% of TDDA additive in the SCN-ISM composition from low to high percentage, i.e 0.02 wt.%, 0.10 wt.%, 0.14 wt.%, 0.16 wt.% and 0.20 wt.% with the same amount of CoTMeOPP ionophore in ISMs of E7-E11, the Nernstian responses did not show any readable pattern but however, the linear working ranges were narrow with the lowest working ranges from 1x10^{-5} mol/L to 1x10^{-1} mol/L and the detection limits were higher compared with E3 ISM. The ideal amount of TDDA to be utilized in the fabrication of highly selective membrane to be able to work in the most optimum rate was also done based on try an error method. Based on these results, it can be said that the optimum amount of TDDA to be utilized in the fabrication of SCN-ISE was 0.16 wt.% to get the best performance of the SCN-ISE.

The observed potentiometric response of E1 ISM (without ionophore), E2 ISM (without HEMA inner layer), E5 ISM (without TDDA) and other SCN-ISM fabricated electrodes demonstrated the properties of CoTMeOPP ionophore, HEMA inner layer TDDA additive as the active components of electrochemical sensor towards the selectivity and sensitivity of the target anion of SCN as mentioned in the reported papers previously. The assembly of the SCN-ISE with Ag/AgCl as first layer, HEMA as the inner layer and mixtures of n-BA, CoTMeOPP and TDDA as the main components in the outer layer successfully proved that the fabricated SCN-ISE is responsive and highly sensitive towards the target anion of SCN that deviates from classical Hofmeister classical series with positive Nernstian response of slope -60.76 mV/decade (Figure 4.2).

It can be concluded that E3 SCN-ISM electrode which composed of Ag/AgCl first layer, 95.14 wt.% HEMA, 4.86 wt.% of DMPP as the inner layer and 98.86 wt.% of n-BA, 0.08 wt.% of HDDA, 0.34 wt.% of CoTMeOPP, 0.16 wt.% of TDDA and 0.56 wt.% of DMPP as outer layer worked at the most optimum rate. The membrane electrode with an optimum composition demonstrated a good Nernstian response as well as the closest to the theoretical value towards SCN anions with slope of -60.76 mV/decade, wide linear working range of 1x10^{-7} mol/L to 1x10^{-1} mol/L and relatively low detection limit of 9.55x10^{-8} mol/L. Based on the optimized fabricated SCN-ISE, it can be said that the fabricated SCN-ISE has comparable response and sensitivity (-60.76 mV/decade) towards the target anion despite the usage of self-plasticized polymeric membranes compared to the PVC-based ISEs reported by Gupta et al., Vlascici et al., and Ibrahim et al., with Nernstian responses range from -32.0 mV/decade to -65.8 mV/decade (Gupta et al., 2002; Ibrahim et al., 2010; Vlascici et al., 2006). This composition was chosen to be the optimum wt.% composition to be used in further studies in the experiment. The calibration plot of the optimized SCN ion-selective sensor are as shown in Figure 4.2.

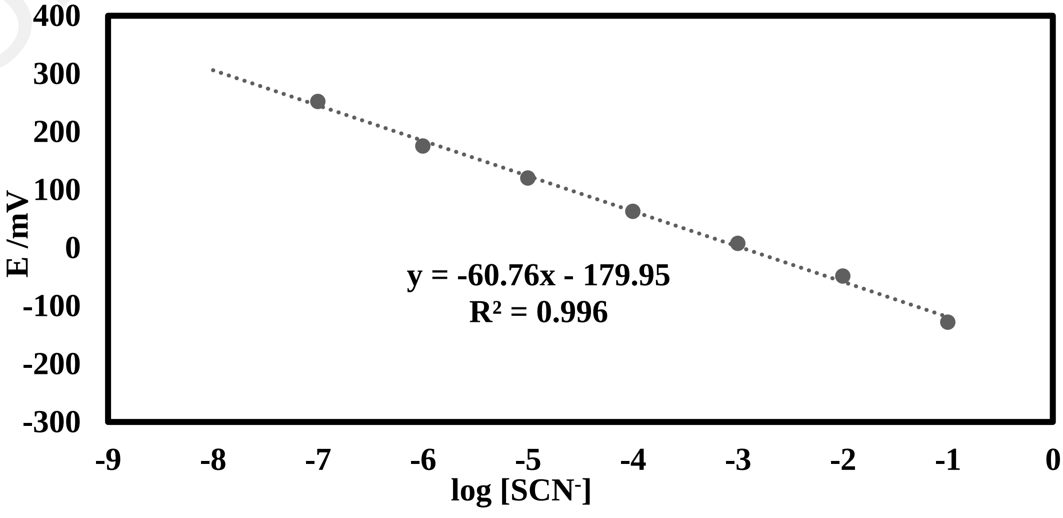


Figure 4.2: Calibration plot of the optimized SCN ion-selective sensor.

4.1.1.1 Effect of pH

The pH of the test solutions or the samples plays a very important role in determining the performance of a sensor. Since SCN has been proven to prevail in many mediums of especially in organic mediums (vegetables, milk, human serum and urine) as mentioned in Chapter 2. It can be said that the fabricated SCN-ISE will have to encounter the biological samples of variety of pH. Thus, a sensor with wide pH working range is essential in this study to conclude that the fabricated SCN-ISE is applicable to be used in determining real sample analysis without any pre-treatment before the samples were analyze. Previously, many reported works of plasticized-based ISEs in detection of SCN anion have been reported with relatively wide pH working range. Thus, the pH of the test solution on the response and sensitivity of the fabricated SCN-ISE is further studied and reported.

The pH effects of the test solutions on the potential responses of the SCN-ISEs were studied at fixed temperature and concentrations. The potential responses of the fabricated SCN-ISE in two different concentrations of SCN standard solutions of $1x10^{-2}$ mol/L and $1x10^{-5}$ mol/L were recorded over a pH of 1.0-13.0 by using the optimized SCN-ISM. The initial pH for the SCN anion dissolved in ultra-pure water was at pH 7. Hydrochloric acid (HCl) and sodium hydroxide (NaOH) with different concentrations were used to lower down as well as increase the pH of the test solutions attentively with the aid of magnetic stirring. Once the target pH of the target analyte has been constant, the stirring was turned off and the potentiometric reading was taken to get stable reading.

Figure 4.3 shows the potentiometric responses in different range of pH and it can be seen that in the test solution of $1.0x10^{-2}$ mol/L, the potential values which were obtained remained stable and constant at range of pH 2 to12. The independent, stable and constant reading manifested that the optimized SCN-ISE membrane is able to work at its optimum rate within wide range of pH with no significant drift or effect and the SCN determination can be done without treating the samples beforehand.

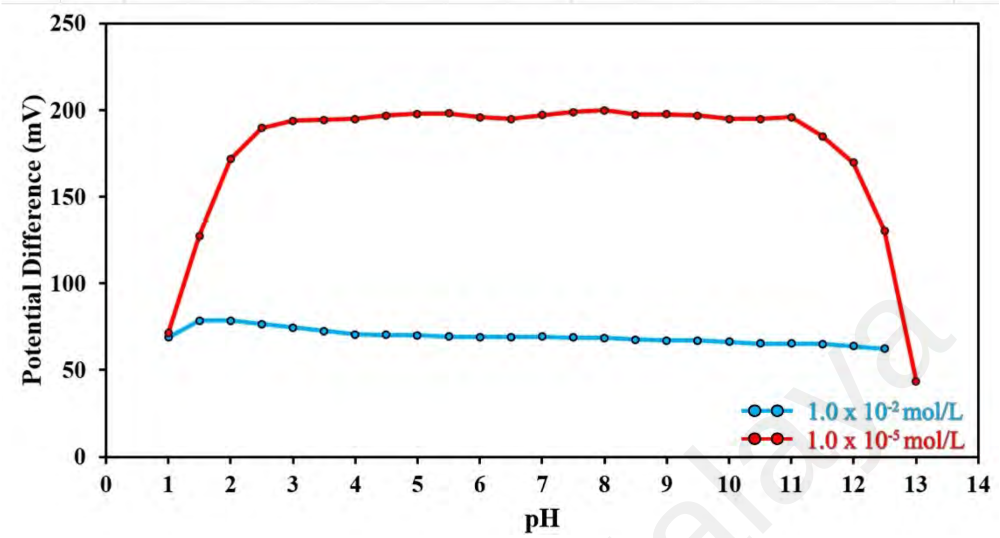


Figure 4.3: The effect of pH test solutions on the response of the fabricated SCN-ISE in different concentrations.

As can be seen in Figure 4.3, by using 1.0x10^{-5} mol/L test solution, the optimized polymeric membrane was able to give independent, constant and stable reading within the pH range of 3 to 11 which is narrower compared to the result obtained with SCN concentration of 1.0x10^{-2} mol/L. Nevertheless, it can be said at wide pH range, SCN-ISEs' performance retained without SCN-ISM being affected. Despite the wide working range of pH in test solution 1.0x10^{-5} mol/L, the optimized membrane showed tremendous change of potentiometric responses in two extreme ends i.e at relatively low pH of 1-2 and high pH of 12-13 compared to the potentiometric response obtained from the optimized membrane in test solution 1.0x10^{-2} mol/L.

This may due in very acidic and basic environment, i.e pH 1-2 and pH 11-13 in both test solutions, the concentration of H^+ increased in low pH values and OH^- ions increased in high pH values in the test solutions and the SCN-ISM showed significant drift in potential due to the partial protonation and interference of the ionophore by both H^+ and OH^- ions. This phenomenon occurred and result in loss of ionophore tendency to form ionophore complexes with the target anion of SCN (Amini et al., 1999). Other than that, the polymeric membranes of both n-BA and HEMA also reacts with strong acids and strong bases and end up to cause rupture to the membrane structure. The rupture of the SCN-ISM may lead to leaching of the SCN-ISM to the environment or the test solutions and thus cause the significant depletion in the potentiometric responses at the two pH extremes. Other than that, we can also conclude that H^+ and OH^- do not affect the SCN-ISE performance within the normal pH of the environment i.e pH 6.0-7.5.

Based on previously reported works of SCN-ISE, the pH range of the fabricated SCN-ISE has been studied and for PVC-based SCN-ISE and the pH range in which the response of SCN-ISE remained unaffected was reported by Hodinar and Jyo in 1988 with pH range of 5-11 (Hodinar & Jyo, 1988), Sánchez with 2-9 pH range (Sánchez et al.,1998) and Amini with pH range of 3-10 (Amini et al., 1999). Meanwhile, for self-plasticized polymeric membranes for F-ISE reported by Matusevich, the pH was adjusted within 5.5-6.5 and no further studies on effect of pH onto the fabricated electrode (Matusevich et al., 2012). For I-ISE, it was reported to work at its optimum without the potential being drifted at pH ≤3. Based on these results, the fabricated SCN-ISE in this work has showed tremendous improvement in the pH working range as compared to both PVC-based SCN-ISE and self-plasticized ISEs. Thus, the pH working range of this fabricated SCN-ISE is from 3-11.

4.1.2 Potentiometric Response Towards Fabricated ISE

The potential responses of the fabricated membrane electrode of SCN-ISE was studied and compared with other anions that is commonly found in the environment. The optimized sensor was used over a wide range of $1.0x10^{-8}$ mol/L to $1.0x10^{-1}$ mol/L for various types of anions. The potentiometric responses results are as shown in Figure 4.4.

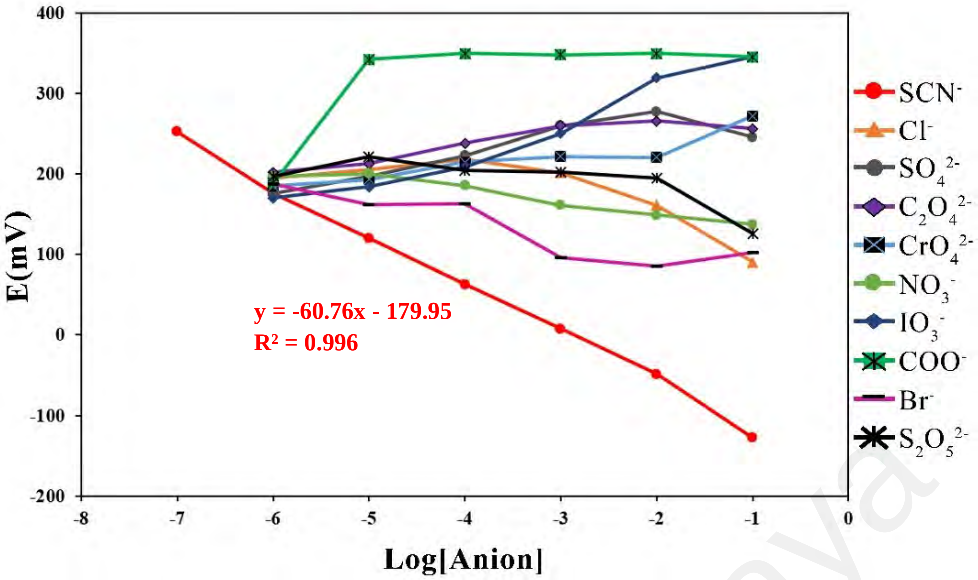


Figure 4.4: The potential responses of SC-ISE membrane electrode based on CoTMeOPP ionophore towards various anions.

Figure 4.4 clearly showed that fabricated optimized SCN-ISM exhibit a good Nernstian response with SCN anion with the Nernstian slope of -60.76mV/decade (Figure 4.2) and fits to be determined or detected by the SCN-ISM sensor based on CoTMeOPP ionophore compared to other anions. This can be justified based on the strong selective complexation behaviors of the ionophore towards SCN anion compared to another common anion present in the environment as well as the rapid exchange kinetics of the complexation between the ionophore and the SCN anion (Vlascici et al., 2006).

Other anions except SCN that have been tested for the potentiometric responses of the fabricated self-plasticized CoTMeOPP ionophore or SCN-ISM either showed negative Nernstian responses or the slopes critically deviated from the theoretical value and insignificant to be counted as affective towards the selectivity and sensitivity of the fabricated CoTMeOPP ionophore. The CoTMeOPP ionophore has been used by Gupta, Vlascici and Ibrahim for detection of different anions and based on their reported works, the fabricated CoTMeOPP ionophore based ISE showed high selectivity towards their target anions compared to other interfering anions (Gupta et al., 2002, Ibrahim et al., 2010). Vlascici et al., has reported the work based on CoTMeOPP ionophore in the detection of SCN anion and showed highly selective and sensitive respose towards the target anion and successful in eliminating I interfering anion that was reported as the main interference of CoTMeOPP ionophore previously by Gupta et al., (Gupta et al., 2002, Vlascici et al., 2006). These previous works on CoTMeOPP ionophore have showed high affinity towards the anions of interest when it is incorporated in the ISEs and this supports the result obtained for the fabricated self-plasticized SCN-ISE in this work that is highly responsive, selective and sensitive towards SCN anion.

Thus, based on Figure 4.4, it can be concluded that the fabricated SCN-ISE based on CoTMeOPP ionophore behave positively by demonstrating positive Nernstian response towards SCN^- anion i.e deviate from classic Hofmeister series compared to other anions and it is also not responsive or affected by other commonly found anions present in the environment. Further studies in the selectivity of the fabricated electrode will be elaborated in section 4.2.1.

4.2 Fabrication of SCN-Selective Electrode (SCN-ISE) Based on CoTMeOPP Ionophore

A number of parameters are required for an ISE membrane to be considered as a suitable sensor for quantitative measurement of anions. The linear working range, detection limit, selectivity, pH working range, response time, repeatability, reproducibility and lifetime are the principal and significant parameters in order to ascertain the utility of CoTMeOPP as active material or ionophore for the preparation of the fabricated SCN-ISE. In this section, selectivity coefficient, response time, repeatability, reproducibility and lifetime of the fabricated SCN-ISE is further investigated and reported.

4.2.1 Determination of Selectivity Coefficients

Selectivity coefficient explains and translates the effect of other anions towards response of the fabricated SCN-ISE i.e either the other anion affects or deviates the Nernstian response of the fabricated SCN-ISE. The selectivity coefficient of the developed SCN-ISE towards their target anions with respect to a wide range of commonly found interfering anions were determined by using matched potential method (MPM). The determination of selectivity coefficient was determined according to the explained equation in section 3.5 (Eq. 3.3). To study the selectivity coefficient of the fabricated SCN-ISE, the concentration of $1x10^{-5}$ mol/L was used for primary anion solution i.e SCN standard solution and $1x10^{-1}$ mol/L for the interfering anions solutions in constant room temperature and conditions. The selectivity coefficient results from MPM method are obtained, analyzed and summarized in Table 4.3.

Table 4.3: Selectivity coefficient values of various anions for the fabricated SCN-ISE.

Anion	$\log K_{SCN^-,M^{n-}}^{MPM}$	Anion	$\log K_{SCN^-,M^{n-}}^{MPM}$
Cl^-	-3.14	CO_3^{2-}	-2.79
Br^-	-3.71	$S_2O_3^{2-}$	-2.68
IO_3^-	-4.04	SO_4^{2-}	-4.22
COO^-	-4.69	$S_2O_5^{2-}$	-4.00
NO_3^-	-4.85	CrO_4^{2-}	-4.82

It is apparent based on Table 4.3 that the selectivity coefficients of SCN-ISE electrode towards interfering anions are lower than SCN anion and most of the selectivity coefficients are relatively low i.e less than two indicating no significant interference in the performance of the fabricated electrode for the determination of SCN anion (Rezaei et al., 2009). Based on Table 4.2, the SCN-ISE is proven to deviate from classical Hofmeister series with SCN>> $S_2O_3^{2-}$ > CO_3^{2-} > Cl^- > Br^- > $S_2O_5^{2-}$ > IO_3^- > SO_4^{2-} > COO^-

> CrO_4^{2-} > NO_3^-. Based on previously reported works on CoTMeOPP ionophore by Gupta et al., I^- and SCN^- cause high interference towards MoO_4^{2-} selective electrode and Ibrahim et al., reported in his work, the CoTMeOPP ionophore showed high interference toward SCN anion despite having selenite (SeO_3^{2-}) as the target anion (Gupta et al., 2002, Ibrahim et al., 2010). Based on Gupta and Ibrahim works, it can be clearly seen that the SCN anion is the major interference anion in the anion-ISE fabricated. It can also be said that this CoTMeOPP ionophore is selective towards SCN anion. In a work reported by Vlascici et al., the fabricated CoTMeOPP ionophore based on PVC as the polymeric membrane for SCN-ISE, he has successfully eliminated the interference from Cl anion (Vlascici et al., 2006). To add to the performance value of the fabricated self-plasticizing SCN-ISE in this study, Cl^- affect towards the fabricated SCN-ISE were also eliminated.

Based on Table 4.3, there are commonly found anions that have been tested for the selectivity coefficient values that showed no change in the potential even when a significant big volume (>30ml) that is more than the volume of primary anions (10ml) were added onto the test. This can be explained by observing the response of the fabricated SCN-ISE with other anions in Figure 4.4. As mentioned before, there is no response from the SCN-ISE with the commonly found anions i.e interfering anions which results in the highly selective of the fabricated SCN-ISE which eliminates the other effect of the interfering anions.

4.2.2 Determination of Response Time

Response time of ISE is an essential factor to be counted as one of the many performances that an ISE should have. The response time is defined as the time taken for the ISE to reach a static potential in the measurements (Ganjali et al., 2001). It is determined after the potential of one anion solution becomes constant and then the similar measurements are carried out in another solution of the same anion with 10-fold change (lower or higher) in concentration. In present studies, as shown in Figure 4.5, the practical response time was recorded over a concentration range of 1x10^{-7} mol/L to 1x10^{-1} mol/L.

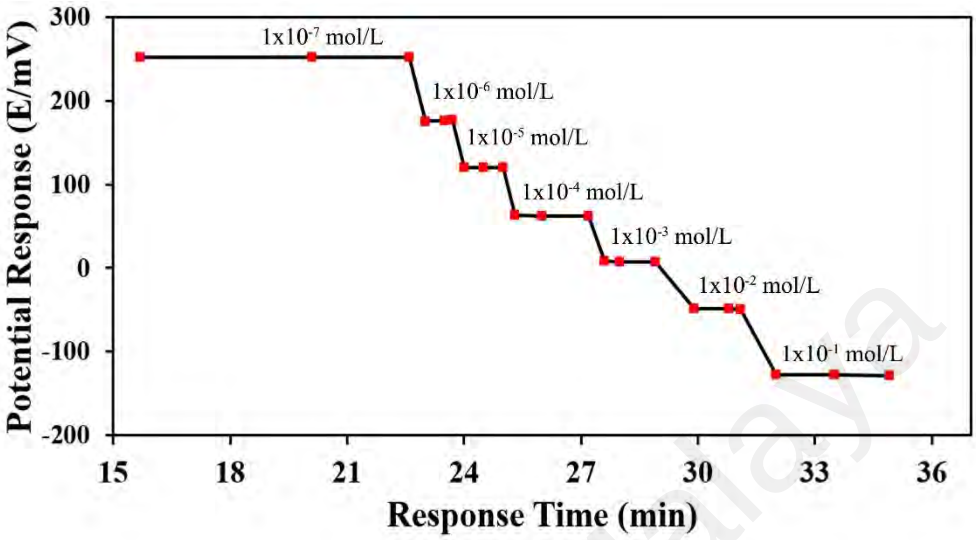


Figure 4.5: Response time curve for SCN-ISE based on CoTMeOPP ionophore.

A trend can be observed from the response time of the optimized SCN-ISE in Figure 4.5. The lower the concentration of SCN anion in the test samples, the longer the time needed for the optimized membrane to obtain a stable reading. However, as the concentration of SCN anion increases, the time taken for the SCN-ISE to become stable reduces. The longest response time of 9 minutes was observed from when 1x10^{-7} mol/L test solution was introduced to the SCN-ISE and shortest response time of 1.13 minutes was recorded by changing the anion concentration in test solution from concentration of 1x10^{-7} mol/L to 1x10^{-6} mol/L. Other response times recorded were in the range of 1.3 minutes to 3.8 minutes. It can be said that the response time for the SCN-ISE is relatively rapid for the immediate location detection of SCN anion in real sample analysis.

The PVC-based SCN-ISE has been reported to give better response time towards SCN anion. The time frame of the reported ISEs in detection of SCN-ISE is from 25 seconds to 90 seconds (Table 2.1). In self-plasticized based anion-ISEs, the fluoride-ISE (F-ISE) by Matusevich reported very fast response time of 12 seconds (Matusevich et al., 2012) and there is no reported work on the response time for Mendecki's work. It is apparent that the response time of the fabricated electrode is higher than of PVC-based SCN-ISE and self-plasticized anion-ISE but the response time of the fabricated SCN-ISE can be said to be rapid with just 1.13 minutes for the immediate location detection of SCN anion in real sample analysis.

4.2.3 Determination of Repeatability, Reproducibility and Stability

To validate the fabricated SCN-ISE, the parameters such repeatability and reproducibility of the SCN-ISE were evaluated and the results is summarized in Table 4.4. The repeatability of the SCN-ISE was tested using three measurements over the working ranges of the same optimized electrode membranes. While the reproducibility of optimized SCN-ISE was examined using three different fresh electrodes with optimum composition and each electrode was used only once.

Table 4.4: The repeatability and reproducibility of self-plasticized SCN-ISE.

ISE	Study	Nernstian Slope (mV/decade)	Average	Standard deviation	%RSD
SCN^-	Repeatability	-60.76, -60.80, -60.87	-60.81	0.056	-0.092
	Reproducibility	-60.86, -60.99, -60.97	-60.94	0.07	-0.115

*RSD = (standad deviation/average) x 100

The repeatability data showed an average slope of -60.81 ± 0.056 mV/decade with a relative standard deviation percentage (%RSD) of 0.092%. The reproducibility data showed relatively average slope of -60.94 ± 0.07 mV/decade with %RSD of 0.115%. The fabricated electrode showed %RSD values with less than one that indicates a high precision and accuracy of the fabricated procedure. The same result can be observed in the %RSD value obtained for reproducibility test of the fabricated electrode. The RSD for reproducibility showed less than 1% value which indicates that the electrode is highly reproducible. The fabricated methodology for the fabrication of the optimized SCN-ISE can be said to be able to contribute to the low %RSD value for reproducibility of the electrode. This may due to the fix amount of all the membranes to be deposited and mounted onto the GCE i.e 5.0 µL.

Long-term stabilities of the fabricated SCN-ISE were tested by performing periodic calibration with standard solutions and calculating the responses and slopes over the concentration ranges from 1x10^{-7} mol/L to 1x10^{-1} mol/L. The results are illustrated in Figures 4.6 and 4.7 and Table 4.5 below. Based on Figure 4.6, Figure 4.7 and Table 4.5, it was found that the life-time of SCN-ISE electrode was within seven days without significant drift in the concentration range, slope and response times. As can be seen in Figure 4.6 and 4.7, the slope of the optimized membrane sensor gave significant drift in the Nernstian response after 7 days from -59.64 mV/decade to -45.84 mV/decade. This may due to the self-polymerized polymeric membranes of both n-BA and HEMA has evaporated to the environment and caused change in composition of the optimized membrane thus resulted in significant drift in the potential measurements.

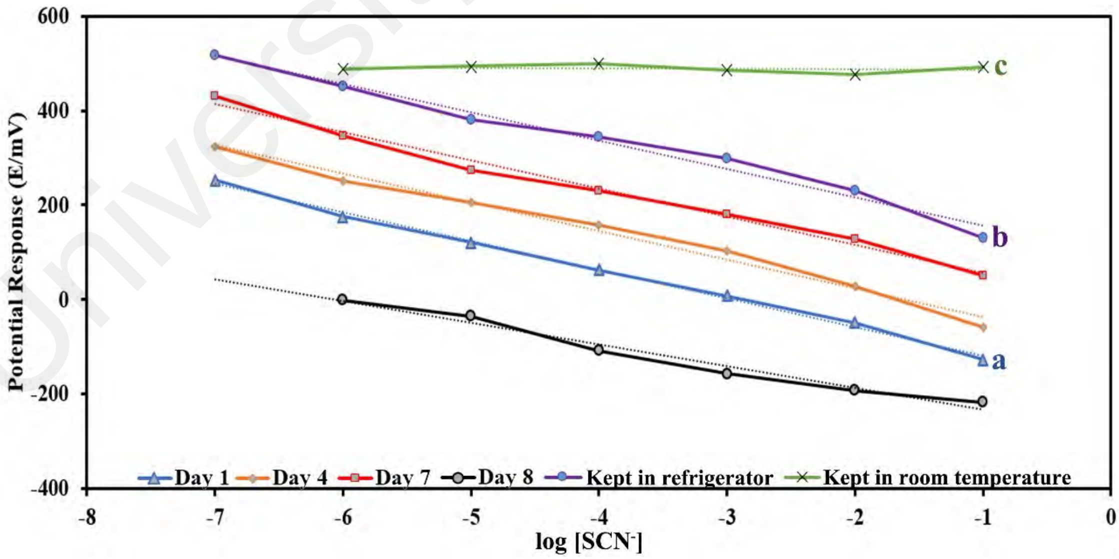


Figure 4.6: Stability response of SCN-ISE (a) freshly prepared SCN-ISE, (b) day 2 SCN-ISE (kept in refrigerator) and (c) day 2 SCN-ISE kept at room temperature.

The optimized SCN-ISE also was observed to have longer lifetime when it is stored in refrigerator. Significant change can be observed when the (a) freshly prepared optimized

SCN-ISE was (b) stored in refrigerator and used within the same concentration ranges from $1x10^{-7}$ mol/L to $1x10^{-1}$ mol/L test solution the next day i.e day 2 and when the SCN-ISE was (c) stored at room temperature. Table 4.5 shows the linear working range, detection limit and the Nernstian slopes of the fabricated SCN-ISE. All the fabricated SCN-ISEs used in determination of the ISEs' lifetime, the SCN-ISEs are kept refrigerated unless mentioned otherwise.

Table 4.5: Linear range, detection limit and slopes of the potential responses of the SCN-ISE under optimal experimental conditions.

	Linear range (mol/L)	Detection limit (mol/L)	Slope (mV/decade)
Fresh	$1x10^{-7} – 1x10^{-1}$	$9.55x10^{-8}$	-60.76
4 days	$1x10^{-7} – 1x10^{-1}$	$6.40x10^{-8}$	-60.63
7 days	$1x10^{-7} – 1x10^{-1}$	$9.10x10^{-8}$	-59.65
8 days	$1x10^{-6} – 1x10^{-1}$	$8.20x10^{-8}$	-46.84
Second day	$1x10^{-7} – 1x10^{-1}$	$9.40x10^{-8}$	-60.14
*Second day	$1x10^{-6} – 1x10^{-1}$	$8.8x10^{-7}$	-1.11

*Kept at room temperature

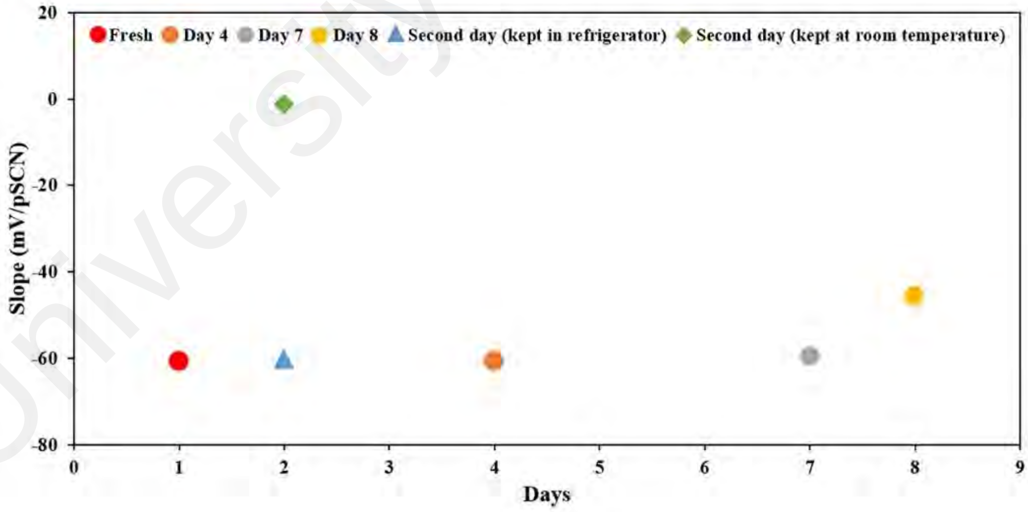


Figure 4.7: The lifetime of optimized fabricated SCN-ISE electrode.

The SCN-ISMs were prepared onto the glassy carbon disc to obtained the same result as obtained in FESEM analysis in section 4.4.3. The significant differences in the Nernstian responses especially for the SCN-ISE that was kept in room temperature was deduced to undergo evaporation of the polymeric membrane in the electrode. Further FESEM analysis was conducted and the results are as shown in Figure 4.8.

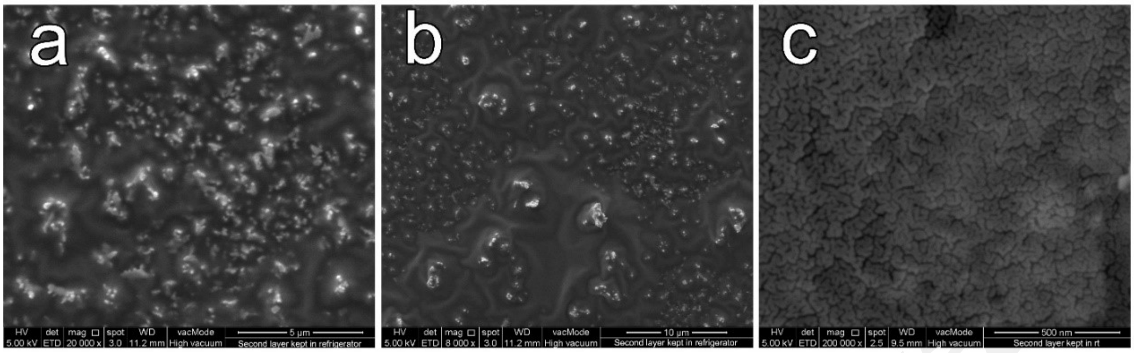


Figure 4.8: FESEM images of n-BA outer-layer that are (a) freshly prepared, second day provided (b) kept in refrigerator and (c) kept at room temperature.

The FESEM images above were obtained for freshly prepared SCN-ISM, and SCN-ISM after one day after it has been fabricated provided kept with different storage conditions i.e refrigerated and kept at room temperature. Figure 4.8 (a) and (b) shows the freshly prepared n-BA polymeric membrane as the outer-layer fabricated onto the glassy carbon disc and kept in the refrigerator respectively while Figure 4.8 (c) shows the inner-layer of n-BA kept at room temperature. The tight structure of n-BA polymeric membrane as shown in Figure 4.8 (a) as the outer-layer can be observed in section 4.4.3. The same tight-structure morphology similar to freshly prepared n-BA outer-layer can also be observed when the n-BA outer-layer that has been kept in refrigerator was observed on the second day as shown in Figure 4.8 (b). The morphology of the outer-layer of n-BA in Figure 4.8 (c) displayed cracked-like surface. This may due to the n-BA membrane has ruptured or broken due to evaporation since it is kept at room temperature. This may attribute to the major drift in potential difference shown in Figure 4.8 (a,b,c).

4.3 Characterization of Fabricated SCN-ISE

Various characterizations of the fabricated SCN-ISE were conducted for determining the complexation, interactions and morphologies of the fabricated ISE.

4.3.1 UV-vis Spectroscopy

The preliminary spectrophotometrically study of the complexation between CoTMeOPP ionophore and SCN anion were carried out. Three solutions of CoTMeOPP ionophore with concentration of $2x10^{-4}$ mol/L, $1x10^{-3}$ mol/L solution of SCN anion and their 1:1 mixture in pure DMSO solvent were prepared. The color change has been observed and recorded as shown in Figure 4.9 below. Based on Figure 4.9(b), the figure shows that the color of SCN anion salt is colorless as mentioned in Chapter 2 and the color of CoTMeOPP dissolved in DMSO in Figure 4.9(c) is red-like color that is different than the initial color of CoTMeOPP ionophore powder i.e dark purple as in Figure 4.9(a). As SCN anion was mixed with CoTMeOPP ionophore, the color changed from red color to yellowish orange color as shown in Figure 4.9(d). The initial color change can be further elaborated by using Werner's theory and supported by UV-vis spectra as shown in Figure 4.10 and 4.11.

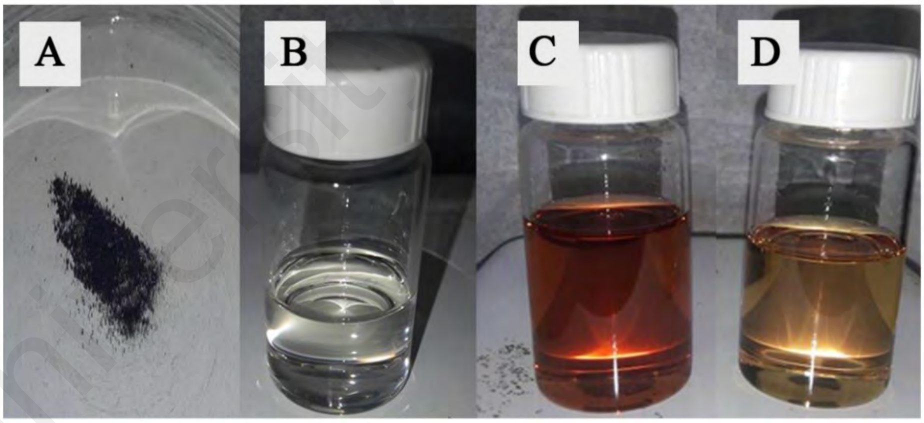


Figure 4.9: The colour of (a) CoTMeOPP ionophore powder, (b) SCN salt, (c) CoTMeOPP and (d) mixture of CoTMeOPP and SCN in DMSO.

The color of dark purple CoTMeOPP ionophore powder changed to red colour after DMSO was added to dissolve the CoTMeOPP ionophore indicates that Co^{2+} has oxidized by DMSO as well as oxygen inside the volumetric flask to become Co^{3+} (Kauffman, 1959). This further be supported by UV-vis spectra indicating the presence of Co^{3+} peaks at 395 nm-439 nm and 275 nm-297 nm in Figure 4.11. As the SCN anion added to the

CoTMeOPP ionophore solution, the solution changed from red color to yellowish orange color and based on Werner’s theory (Figure 4.10), the color represents the complexation of Co^{3+} in the solution. It can be further proved by the decrease in both peaks of 395 nm-439 nm and 275 nm-297 nm indicating the decrease in free Co^{3+}. It may due to the complexation of the Co^{3+} with SCN^- anion.

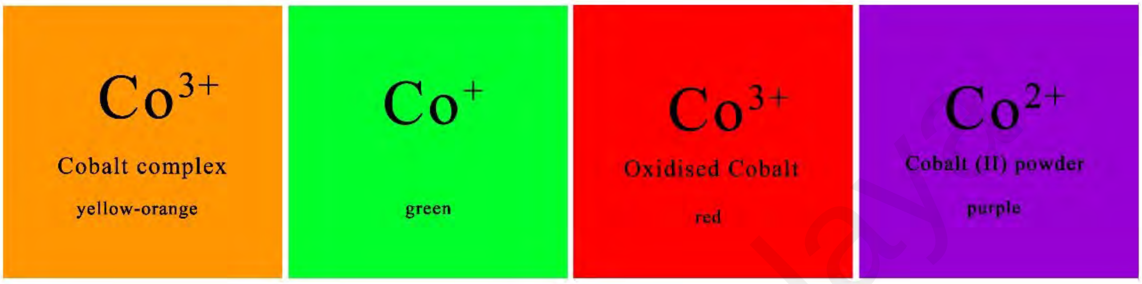


Figure 4.10: Werner's theory of Cobalt (Co) transition metal colour with different coordination number.

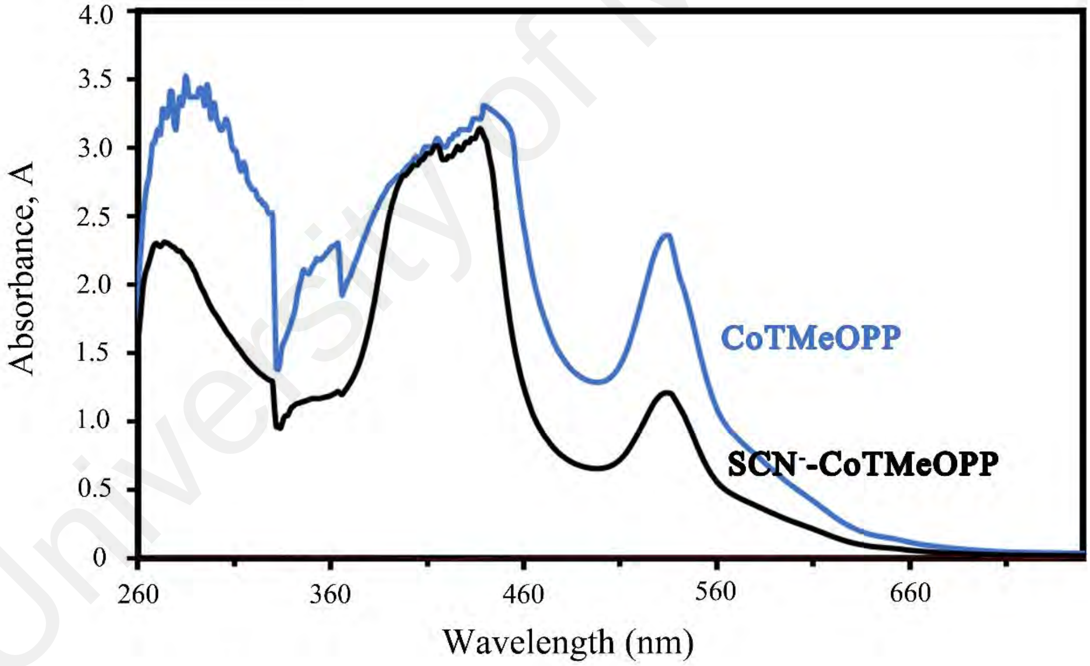


Figure 4.11: UV-Vis absorption spectra of CoTMeOPP ionophore and complex of SCN-CoTMeOPP.

The UV-vis spectra of the complexation of CoTMeOPP ionophore was recorded from 260 nm to 750 nm (Elder et al., 1978). As can be seen in Figure 4.11, the spectrum of CoTMeOPP ionophore shows three significant peaks of each 275 nm - 297 nm, 395 nm - 439 nm and 537 nm. The peak at 275 nm-297 nm indicating the charge transfer from the non-bonding orbitals of oxygen atoms in the Co(II) ligand d-orbital i.e oxidation of Co(II) has taken place and is represented by the peak with transition of $\pi \rightarrow \pi^*$ or $n \rightarrow \pi^*$ or due to the electronic transition of $^4T_1g_{(F)} \rightarrow {}^4A_2g_{(F)}$ (Ahmadi & Amani, 2012). The peaks at the visible region at 395 nm – 439 nm and 537 nm show the Soret bands i.e the peaks are at blue shift zone. This is because of the presence of Co in the metalloporphyrin that decrease the average electron density due to the delocalized π bonds. This further increases the energy available for the electron transition thus the Soret bands can be observed (Zheng et al., 2008).

The peak at 395 nm - 439 nm denoted the oxygen binding with CoTMeOPP i.e oxidation of the ionophore through axial coordination and it is assisted by axial ligand (Kalyuzhny et al., 2000) with the same transition of $\pi \rightarrow \pi^*$ or $n \rightarrow \pi^*$ or $^4T_1g_{(F)} \rightarrow {}^4A_1g_{(P)}$ and $^4T_1g_{(F)} \rightarrow {}^4A_2g_{(F)}$ (Shaker, 2010). Porphyrin ionophore has a distinguished peak at wavelength of 537 nm indicating the Q-band of free porphyrin in the solution with the transition of a_{2u} (π) → e_g* (π). The spectra SCN-CoTMeOPP in Figure 4.11 shows significant decrease in the intensities for three peaks and the decrease in these three peaks indicated the decrease in free Co^{3+} and free porphyrin (Silber & Murguia, 1985). Other than the complexation between SCN and the ionophore might be the reason behind the decrease in the intensities. The oxidation of Co^{2+} to Co^{3+} was instantaneous and crucial as the SCN anion can only be introduced by ligand exchange when CoTMeOPP ionophore contains free Co in the oxidation number of +3 (Hedtmann-Rein et al., 1987). Figure 4.11 also shows the number of absorption peaks do not change after CoTMeOPP was coordinated with SCN anion. This indicates that after SCN was coordinated with the central metal, the symmetry group of CoTMeOPP-SCN agrees with the C_{2v} symmetry group of CoTMeOPP and two anions are coordinated with two central cobalt atoms in CoTMeOPP producing $(CoTMeOPP)_2(SCN)_2$ as mentioned in section 4.3.3. The summarization of UV-vis peaks are as shown in Table 4.9.

Table 4.6: UV-vis peaks of CoTMeOPP-SCN.

Complex	Colour	Wavelength (nm)	Corresponding peak of	Transition
CoTMeOPP-SCN	Orange-yellow	275-297	Co(II)	$\pi \rightarrow \pi^*$ or $n \rightarrow \pi^*$
		395-439	Co(III)	$\pi \rightarrow \pi^*$ or $n \rightarrow \pi^*$ $^4T_{1}g_{(F)} \rightarrow {}^4A_{1}g_{(P)}$ and $^4T_{1}g_{(F)} \rightarrow {}^4A_{2}g_{(F)}$
		537	Porphyrin	$a_{2u}(\pi) \rightarrow e_g^*(\pi)$

4.3.2 Fluorescence Spectroscopy

The complexation study between CoTMeOPP ionophore and SCN anion was studied by using fluorescence spectroscopy by dissolving both solutions of 1x10^{-4} mol/L CoTMeOPP and 1x10^{-2} mol/L SCN in methanol. The metalloporphyrins itself is well-known for its luminescence property and emit apparent bands at visible region range between 390 nm - 400 nm and 550 nm – 650 nm (Harriman, 1981). Thus, the complexation between CoTMeOPP ionophore and the target anion of SCN can be studied by observing the change in the intensities of the bands. Based on Figure 4.12, the small peak at 657 nm was resulted from the transition of (π -π^*) transition of the CoTMeOPP.

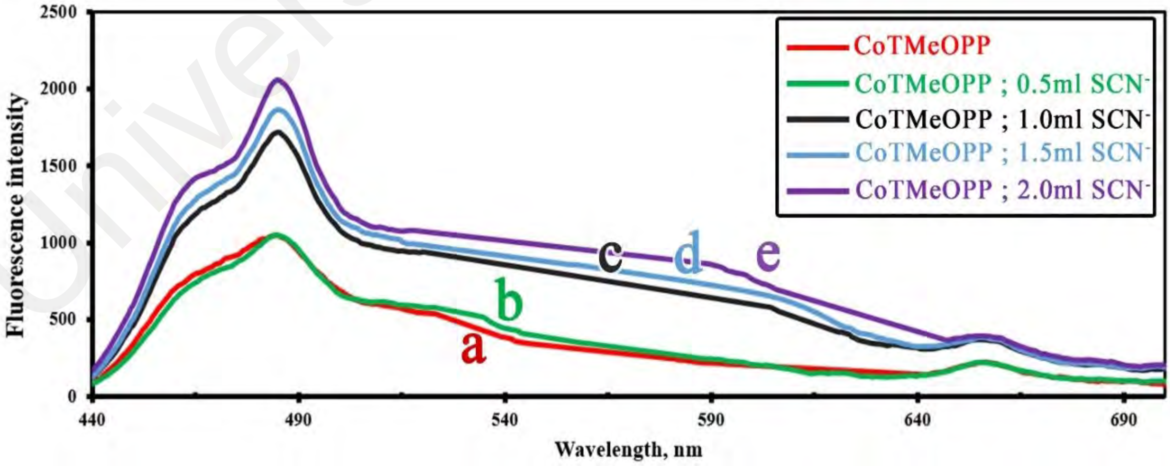


Figure 4.12: The fluorescence emission spectra of (a) CoTMeOPP with the addition of (b) 0.5ml SCN, (C) 1.0ml SCN, (d) 1.5ml SCN and (e) 2ml SCN.

Based on previously reported work by Nath & Baruah in 2014, without the metal attached to the porphyrin ring, the band was reported to show a significant high emission at the 650 nm that indicates the band for porphyrin ring. However, as Co^{2+}/Co^{3+} metal was introduced to the porphyrin ring to become metalloporphyrin, the Co^{2+}/Co^{3+} decreases the fluorescence emission of the porphyrin due to quenching. Another reason behind the small peak at 650 nm could be the central metal ion in complex i.e Co^{2+} (d7) is in +2 oxidation states and Co^{3+} (d6) is in +3 oxidation states and both are paramagnetic. Thus, the fluorescence intensity is low (Nath & Baruah, 2014). In other reported work, the SCN anion was proven to be able to increase the intensity of the porphyrin peak as it restores the fluorescence property of the porphyrin by the complexation between the central metal of the metalloporphyrin with SCN anion. The same phenomenon can be seen in this fluorescence study between CoTMeOPP ionophore and SCN anion as shown in Figure 4.12. As SCN anion is inserted into CoTMeOPP ionophore, increase in the intensities of the bands can be observed and this proved that binding or complexation between Co^{2+}/Co^{3+} metal and SCN anion occurred (Zhang et al., 2007).

The tetrakis(4-hydroxyphenyl) porphyrin derivatives i.e Co^{2+}/Co^{3+} in solid state shows a strong emission of B band indicating the second (π- π^*) excited single state at 485 nm as shown in Figure 4.12. The second (π- π^*) excited single state refers to intra-ligand transition of Co^{2+}/Co^{3+}. However, the strong emission showed at 485 nm in Figure 4.12 was drifted a bit in position than what was reported by Harriman at 1981 due to the presence of four electron donating methyl group ($-CH_3$) in the ionophore (Machura et al., 2013). Based on Figure 4.12, it can be observed clearly that the band intensity at 485 nm increases upon increase in the addition of SCN anion into the CoTMeOPP ionophore. The same situation can be seen with the band at 650 nm, the presence of Co^{2+}/Co^{3+} reduces the fluorescence property of the porphyrin and with increasing in the amount of SCN anion in the solution, the complexation of the ionophore and SCN increased hence restore the fluorescence property of phenylporphyrin. This results in increasing emission spectra. It can be deduced that the increase in fluorescence emission peak with each subsequent addition of SCN anion clearly shows interactions and complexation between the CoTMeOPP ionophore and SCN anion.

4.3.3 Fourier-Transform Infrared Spectroscopy (FTIR)

The fabricated membrane sensor was examined using FT-IR spectroscopy to observe and study the complexation between the CoTMeOPP ionophore and the target anion of SCN. The FT-IR spectra was conducted for both CoTMeOPP and the complexation between CoTMeOPP within the range of 4000 nm to 200 nm. The spectra are as shown in Figure 4.13.

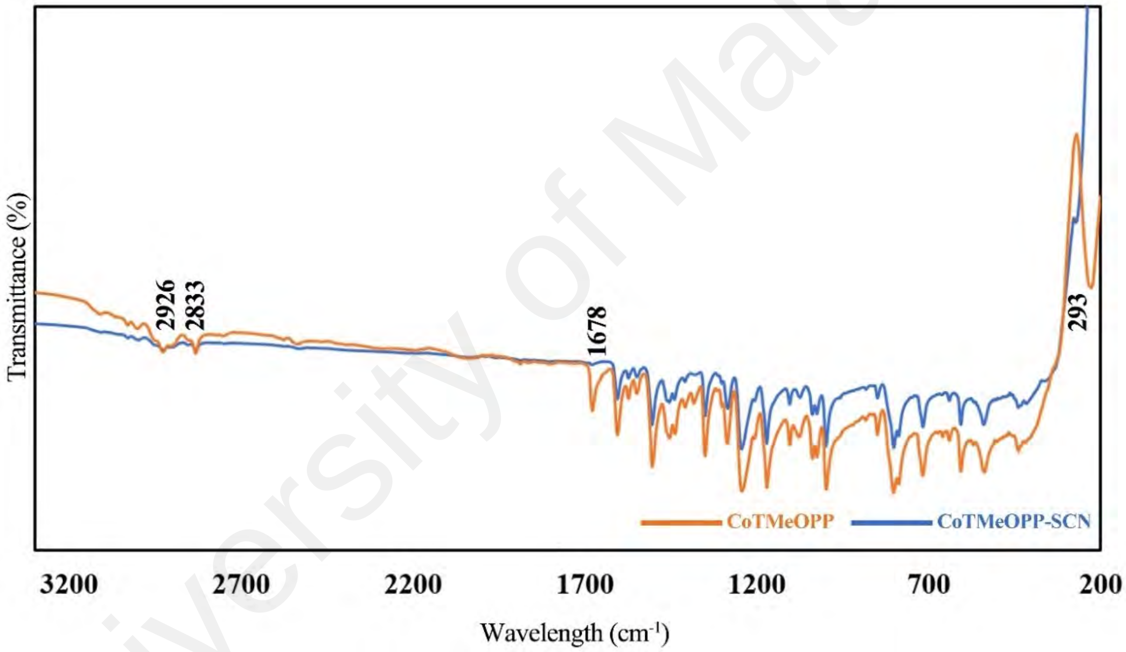


Figure 4.13: IR spectra of CoTMeOPP ionophore and CoTMeOPP-SCN complexation (4000 nm-200 nm).

Based on the Figure 4.13 and Figure 4.14, the peaks distinctive peaks of the CoTMeOPP ionophore can be seen to reduce in its absorbance once SCN anion was introduced to the solution most probably due to the complexation formed between the ionophore and the SCN target anion (Gao et al., 1994). The decreased in intensities can be observed at peaks of 2926 cm^{-1} and 2833 cm^{-1} respectively. The peaks represent C-H characteristic stretching of the ionophore. Complexation between Co and SCN anion weakens the C-H characteristic stretching of the ionophore thus decreased in the intensities at both peaks were observed.

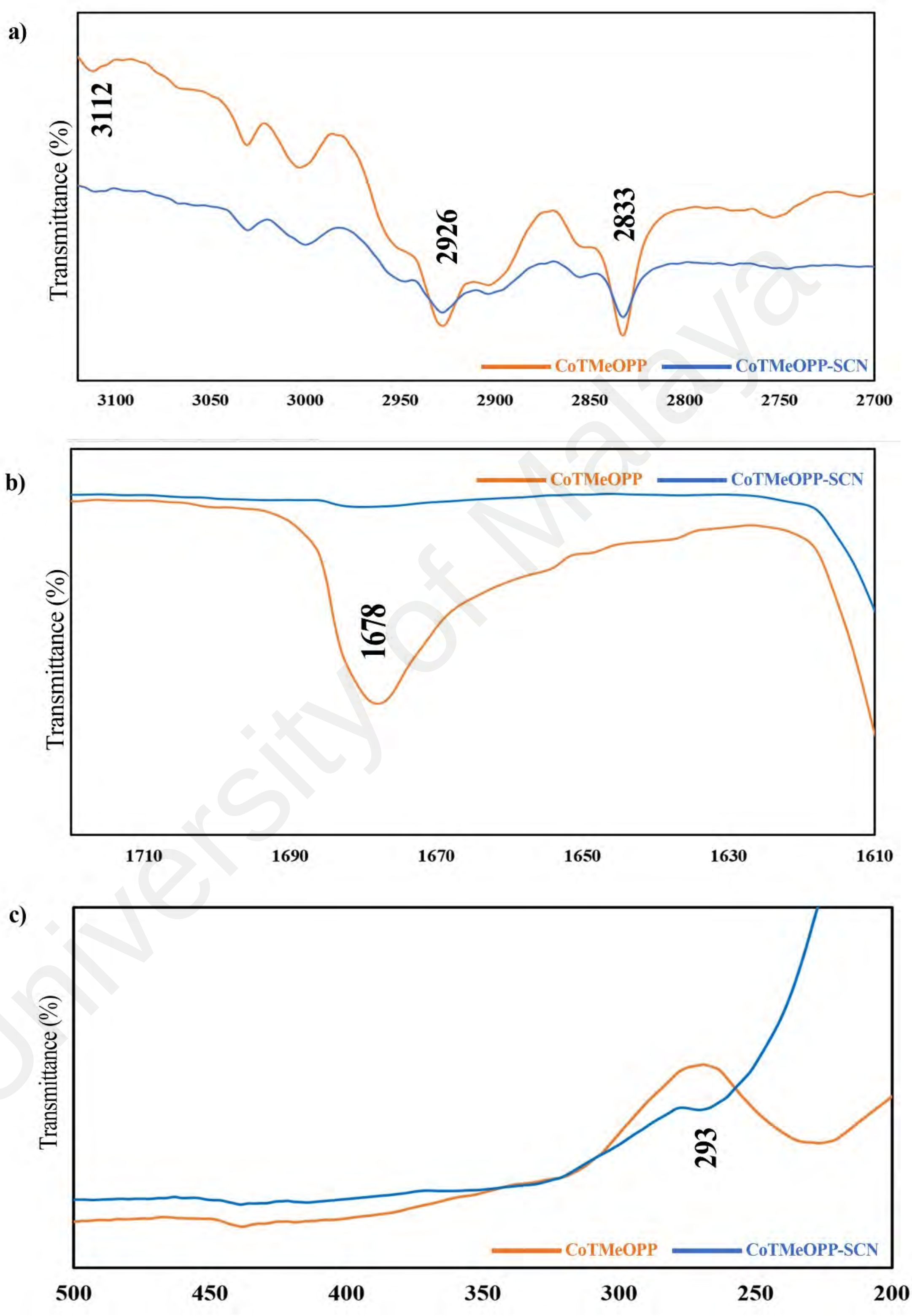


Figure 4.14: IR spectra of CoTMeOPP ionophore and CoTMeOPP-SCN complexation (a) 2926 cm^{-1} and 2833 cm^{-1}, (b) 1678 cm^{-1} and (c) 293 cm^{-1}.

Based on Figure 4.14(b) it is obvious that CoTMeOPP ionophore has a distinctive peak at 1678 cm^{-1}. The peak is pyridine characteristic stretching of the ionophore (Giacomelli et al., 2004). The peak disappears as soon as SCN anion were introduced to the CoTMeOPP ionophore. This may due to the complexation between Co metal and SCN anion weaken and diminish the pyridine characteristic stretching of the ionophore. Further confirmation of the complexation formed bwetween Co metal of the ionophore and SCN anion can be seen in Figure 4.14(c).

The peak at 293 cm^{-1} is the characteristic binding peak between SCN anion and Co metal (Forster & Goodgame, 1965). The formation of complex between Co metal and SCN anion occurred at the N atom position in this FT-IR characterisation. N atoms are arranged tetrahedrally around Co metal and formed T_d symmetry. Based on this FT-IR spectra, it can be concluded that formation of SCN-CoTMeOPP occurred.

4.3.4 Field Emission Scanning Electron Microscopy (FESEM) – Energy Dispersive X-ray (EDX)

FESEM analysis on the surface morphology of SCN-ISE selective membrane was carried out at different magnifications to study and understand the morphological changes occurred in the membrane during electrode preparation and after conditioning the fresh membrane in the SCN anionic solution. In this FESEM analysis, the glassy carbon discs were used as the base for the deposition of SCN-ISM to replace GCE to get the best realistic conditions as the SCN-ISE. In Figure 4.15, the FESEM analysis was employed onto the surface of SCN-ISM with three different conditions all with Ag(s)/AgCl(s) as the first layer and with (a) the inner-layer consisting self-polymerised HEMA and DMPP polymer, (b) outer-layer consisting optimized composition without the presence of CoTMeOPP ionophore and (c) outer-layer consisting optimized composition with

CoTMeOPP ionophore. The morphologies of the electrodes' SCN-ISMs are presented in Figure 4.15.

The surface morphologies, chemical compositions and distribution of the chemical elements of the membrane layers for the fabricated thiocyanate sensor are shown in Figure 4.15 and Figure 4.16 by FESEM images and EDX spectra. In Figure 4.15, it can be seen that the formation of the first layer of HEMA was successfully deposited and covered the solid state of Ag(s)/AgCl(s) first layer. The self-polymerized of HEMA i.e the inner-layer showed smooth structure with the presence of SCN lumps as the standard SCN anion solution was used as the inner solution of the sensor (Figure 4.15).

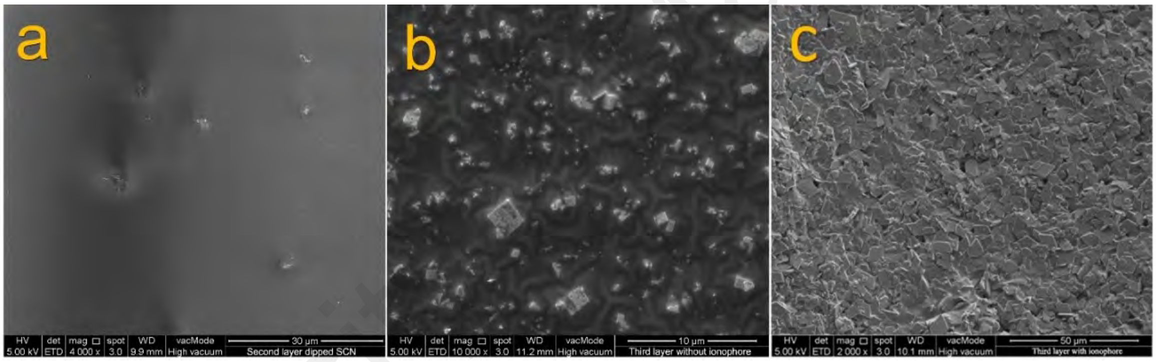


Figure 4.15: FESEM images of (a) HEMA and DMPP (inner-layer), (b) HEMA, DMPP as inner-layer and n-BA, HDDA as the outer-layer and (c) optimized membrane composition.

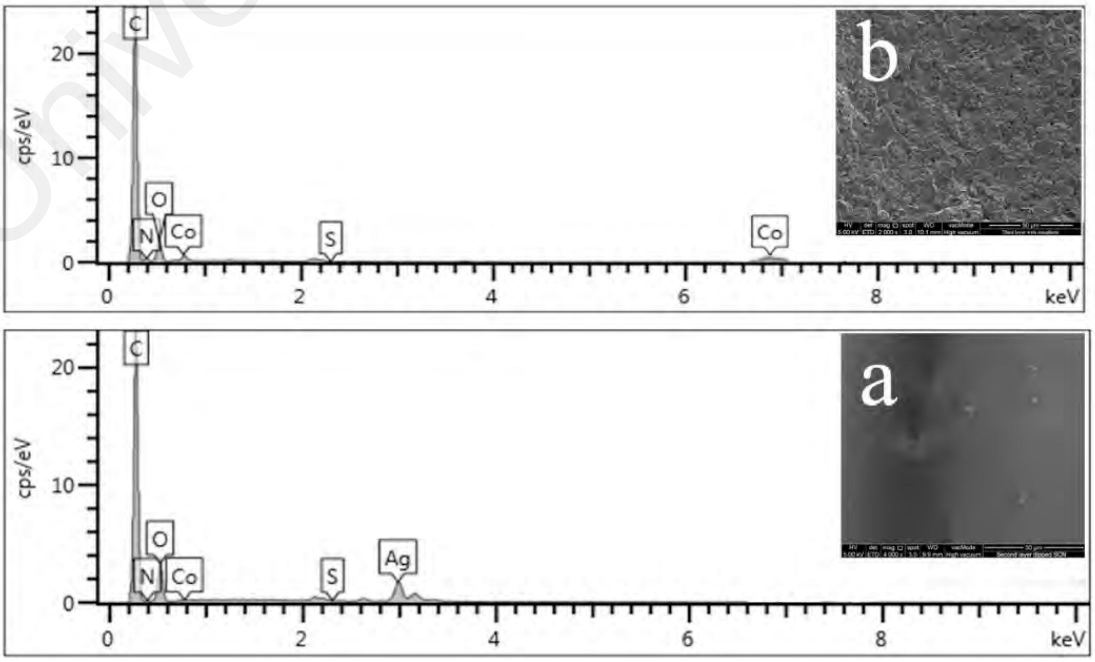


Figure 4.16: EDX spectra of (a) inner layer and (b) fresh optimized membrane.

Based on EDX spectra of the inner layer in Figure 4.16(a), the presence of SCN confirms the presence of SCN lumps in FESEM image in Figure 4.15(a). The second polymeric membrane or the outer-layer of n-BA with the additive and photo initiators were then polymerised onto the inner-layer of HEMA and DMPP. As mentioned before in Chapter 2, the n-BA monomer is a self-polymeric membrane that is conductive, soft with low T_g which makes it capable to be easily self-polymerised by means of exposure to UV-ray. Due to the conductivity of the n-BA, the samples were coated with gold layer to get the clearer and accurate FESEM images. The soft polymer of n-BA also can be seen in Figure 4.15(b), it shows the tight and porous structure of the polymeric membrane. The n-BA structure is needed to avoid rupture in the optimized SCN-selective membrane which may lead to leaching of the SCN-ISM. The addition of CoTMeOPP ionophore onto the outer-layer can be seen clearly in both Figures 4.15(c) and 4.15(b). The tight structure in Figure 4.15(b) is enhanced with the addition of CoTMeOPP ionophore in Figure 4.15(c) showing the highly-packed tight structure of the optimized polymeric membrane. In Figure 4.15(c), the optimized membrane showed fully packed, stacked, geometrical-shaped layers of CoTMeOPP ionophore onto the polymeric layer. The peak of Co can also be seen in EDX spectra in Figure 4.16(b) supporting the presence of Co in the freshly prepared optimized membrane.

The EDX characterization of selected elemental parts of inner layer of HEMA and DMPP and the freshly prepared outer-layer self-plasticizing SCN-ISM of the optimized membrane showed clearly the assignation of C, N, O, S, Co and Ag Kα lines. The Ag and Co are the primary evident in the presence of solid state Ag(s)/AgCl(s) and CoTMeOPP ionophore in the inner-layer and outer-layer of optimized membranes, respectively. The relative heights of the peaks for the trace elements appear in the spectra are related to their relative amounts in the membrane.

4.4 Application of Fabricated SCN-ISE

4.4.1 Analytical Application of Fabricated ISE

In order to validate the applicability of fabricated electrode of SCN-ISE, the analytical application can be conducted. Analytical application includes potentiometric titration and spike method analysis, respectively. Owing to the fact that this analytical application is simple and sensitive, these methods have been widely used and reported in literature as general methods to be applied to evaluate the applicability of new fabricated ISE sensor (Erden et al., 2006; Shamsipur et al., 1999).

4.4.1.1 Potentiometric titration

The fabricated SCN-ISE was applied as an indicator electrode in titrimetric determination of SCN^- under laboratory conditions. Ibrahim et al., has successfully apply the fabricated selenite ($SeO_{3+}{}^{2-}$) selective electrodes in potentiometric titration of silver nitrate ($AgNO_3$) to be used as indicator electrode (Ibrahim et al., 2010). In this study, it was successfully proved that fabricated SCN-ISE can be used as an indicator electrode in the potentiometric titration of 25 ml SCN solution with concentration of 1.52 x 10^{-3} mol/L and $AgNO_3$ with concentration of 0.01 mol/L. Based on Figure 4.17, it can be seen that the obtained curve is unsymmetrical and the end point of the plot corresponds to 1:1 stoichiometric of $Ag\text{-}SCN^+$ ions complex. The inert Figure 4.17 shows the plot of $\Delta E/\Delta V$ vs. mean volume of $AgNO_3$ added. Before the end point i.e 2.5 ml, the potential demonstrated logarithmic changes with the volume of $AgNO_3$ added, and remained almost constant after the end point. The obtained potential decreased with the decrease in SCN anion concentration due to their complexation with Ag^+ ion. Thus, it can be concluded that it is possible to determine the amount of SCN anions in the solution accurately using the fabricated fabricated sensor. This method is generally used to evaluate the applicability of the prepared SCN-ISE as mentioned by Erden (Erden et. al., 2006).

Based on Figure 4.17, it can be seen that the obtained curve is unsymmetrical and the end point of the plot corresponds to 1:1 stoichiometric of Ag-SCN$^+$ ions complex. The inert Figure 4.17 shows the plot of ΔE/ΔV vs. mean volume of $AgNO_3$ added. Before the end point i.e 2.5 ml, the potential demonstrated logarithmic changes with the volume of $AgNO_3$ added, and remained almost constant after the end point.

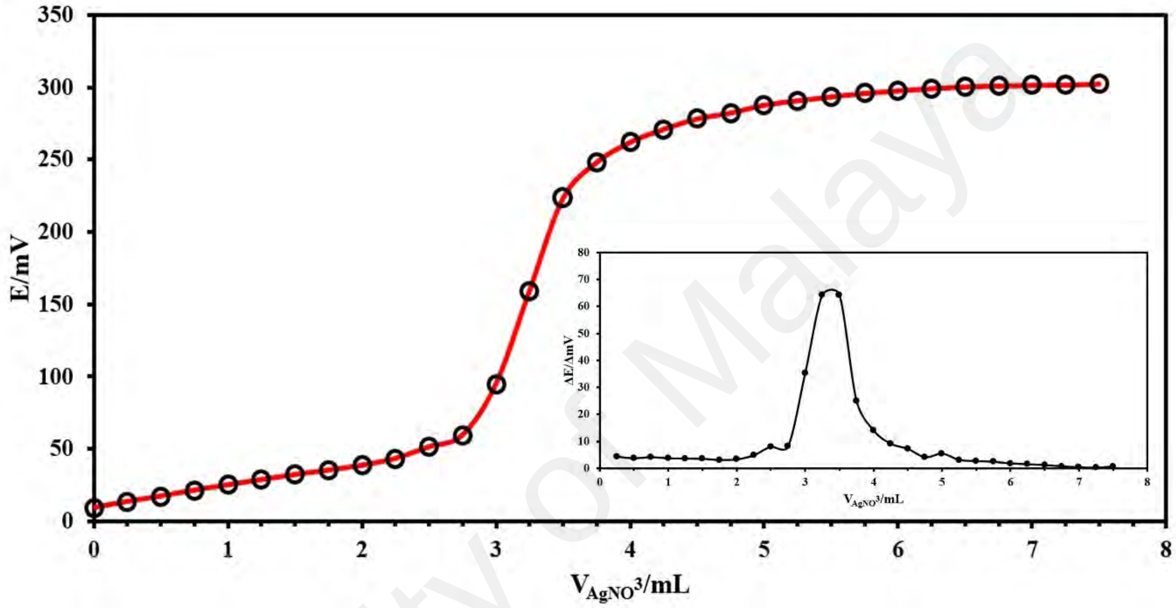


Figure 4.17: Potentiometric titration curve of 25ml $1.52x10^{-3}$ mol/L, SCN^- solution with $1x10^{-2}$ mol/L $AgNO_3$ by using fabricated SCN-ISE as indicator electrode.

The obtained potential decreased with the decrease in SCN anion concentration due to their complexation with Ag$^+$. Thus, it can be concluded that it is possible to determine the amount of SCN anions in the solution accurately using the fabricated fabricated sensor. This method is generally used to evaluate the applicability of the prepared SCN-ISE as mentioned by Erden et al., (2006).

In the study of complexation of CoTMeOPP ionophore and SCN anion, titration method using UV-vis was conducted. The titrations of Co$^{3+/2+}$ with SCN anion were recorded in the spectra as shown in Figure 4.18 below with the range from 200-800 nm about two minutes after each addition. The apparent absorbance can be seen in the absorbance range from 411 nm to 426 nm wavelength. The specific absorbance indicates the complexation of $Co^{3+/2+}$ with SCN anion. With the addition of SCN anion to the $Co^{3+/2+}$ i.e the complex mixture, the absorption band of free $Co^{3+/2+}$ decreased and shifted in comparison to the ionophore band.

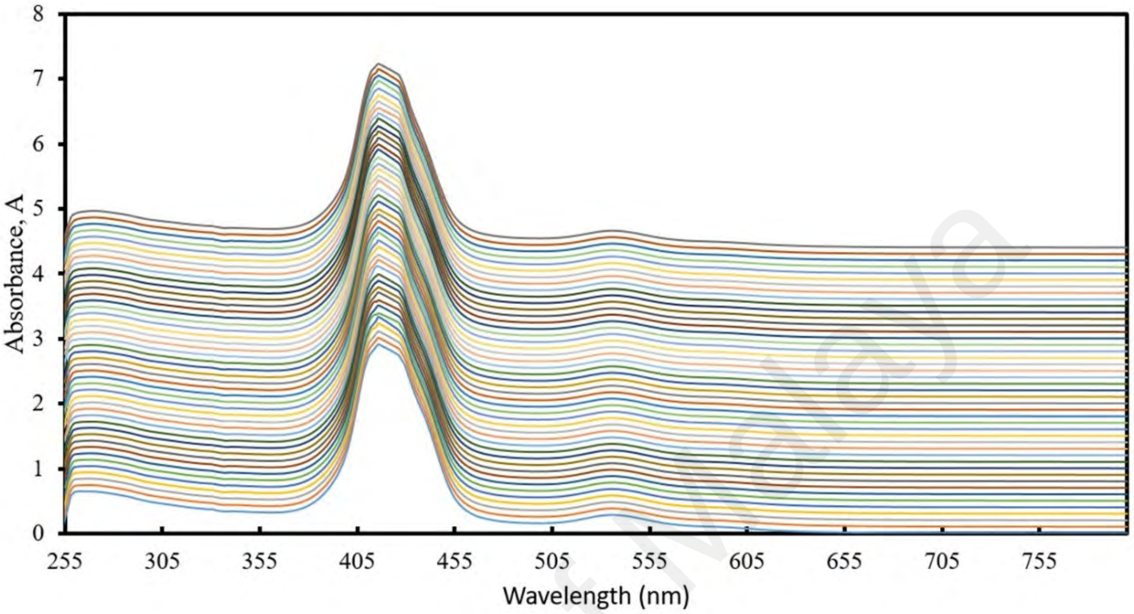


Figure 4.18: UV-vis absroption spectra of CoTMeOPP ionophore (2.3×10^{-6} mol/L) in 1:1 DI : DMSO mixture in increasing concentration of SCN^-.

This proves that complexation formed between CoTMeOPP inophore and SCN anion. The stoichiometry of formed complexes between CoTMeOPP and SCN anion was determined by titration of 2.5 ml of CoTMeOPP ionophore (2.3×10^{-6} mol/L) with SCN anion (1.0×10^{-4} mol/L) in DMSO at 25°C. Figure 4.18 represents the electronic spectra of CoTMeOPP titrated with SCN in DMSO at 25°C. The linear plot of absorbance versus the molar ratio of $[SCN^-]/[CoTMeOPP]$ shows a breaking point at 0.5. It can be seen that in the complexation process, the addition of SCN anion to the CoTMeOPP ionophore solution results in the formation of 2:1 $[CoTMeOPP : SCN^-]$. Thus, the general mechanism for the complexation process is suggested as follows:

$$2CoTMeOPP + SCN^- \leftrightarrow SCN(CoTMeOPP)_2$$

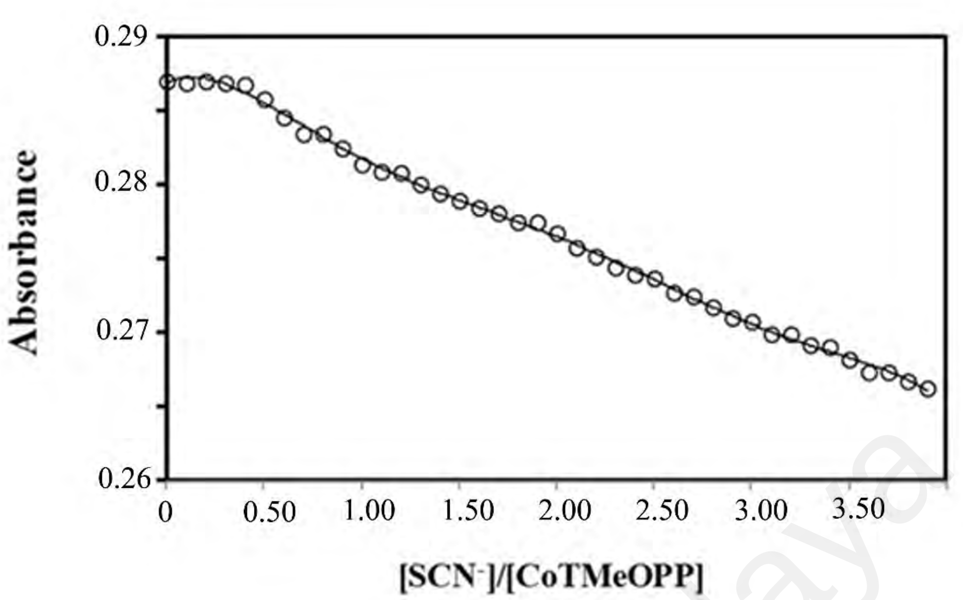


Figure 4.19: Absorbance versus the [SCN$^-$]/[CoTMeOPP] molar ratio and plots.

4.4.1.2 Analysis of sediments, river water, fish samples and human saliva

The applicability of the fabricated electrode SCN-ISE was tested through analysing direct determination of spiked SCN anion in sediments, river water and fish samples. The direct determination of SCN anion by using the fabricated optimized fabricated SCN-ISE was also conducted in the saliva samples of both smoker and non-smoker. The determination of the real amount of SCN anion in the samples was pre-determined by using Ion Chromatography (IC) and for the samples with no trace amount of SCN anion such as sediments, river water and fish samples, SCN anion were introduced by using spike method and recorded. The results in Table 4.7 support that the amount of SCN anion obtained or recovered by the fabricated SCN-ISE method agreed well with the spiked and actual amount in the recovery range of 100.1-111.6%, demonstrating the acceptable performance of the supplied electrode for the recovery of SCN anion content from all the samples with different matrices. On the other hand, the relative standard deviation (RSD) values shows readings which are lower than 1% indicating the high effectiveness of the fabricated electrode.

Table 4.7: Potentiometric determination of SCN anion in different samples by using fabricated SCN-ISE.

Sample		**SCN^{-} (mg/L)**		**^{b}Recovery, (%)**	**^{c}RSD, (%)**
		Spiked / Actual	**^{a}Found**		
Sediments		0.5	0.6	108.7	0.85
		1	1.2	109.9	0.52
		1.5	1.7	106.4	0.54
River water		5	5.4	101.5	0.79
		3	3.6	110.0	0.82
		2	2.4	108.9	0.80
Fish		4	4.5	111.3	0.57
		10	10.6	104.7	0.63
		5	5.6	109.9	0.68
Saliva	**Smokers**	6.1	6.2	110.6	0.59
		7.3	8.4	111.8	0.50
		5.9	6.8	106.9	0.34
	Non-smokers	1.9	1.9	111.6	0.75
		0.07	0.1	103.6	0.80
		0.2	0.2	100.1	0.43

^{a}Averange of three determinations, ^{b}Recovery = (found/added) x 100, ^{c}RSD = (SD/average) x 100

Based on reported works, the minimum concentration of thiocyanate that is commonly exist in sediments, river water, fishes and saliva of smokers and non-smokers are in the concentrations of 620 mol/L, 3.5 mol/L (Ecotoxicology & Chemicals, 2007), 0.01 mol/L - 40 mol/L (Cato & Brown, 2008), 159 mol/L and 72 mol/L (Tenovuo & Makinen, 1976) respectively. High reported concentration of SCN anion can be seen in sediments and saliva of smokers and non-smokers but low concentration reported for river water and fishes. As mentioned in Chapter 2, the sources of SCN in the environment and human saliva and blood serum are mainly from wash out of insecticides, pesticides, untreated waste from factories e.g paint industries, soil sterilization, dairy products and from cigarettes. Thus, concentrated amount of SCN can be observed to prevail more in primary consumers of SCN such as sediments and smokers and non-smokers rather than secondary consumer, in this case study is river water and fishes. Trace amount of SCN present in the environment occurs at 0.01 mol/L to 10 mol/L and based on the results in Table 4.7 below, it can be seen that the fabricated SCN-ISE devise has a good compatibility and it can be verified to be applicable to determine the concentration of SCN anion in solution. It can be concluded that the samples show good agreement with added and spiked concentration, hence proved the capability of fabricated SCN-ISE in sensing application.

4.4.1 Comparative Studies of SCN-ISE

All presented data manifest that the proposed SCN-ISE can be applied in direct determination of SCN anion present in solution within wide linear range, low detection limit and wide pH range. Hence, the performance of proposed electrode is compared with the published SCN-ISEs as shown in Table 4.8. All reported electrodes performances are listed in Table 4.8 and all ISEs reported are PVC-based membrane sensors for determining SCN anion. Listed reported works except for the reported SCN^- sensor by Abdel-Haleem in 2017 used different types of electrode as transducer. Abdel-Haleem used optochemical method that employed UV-vis as the transducer and did not report any linear working range of the detection of SCN anion.

Based on Table 4.8, there are many reported works on PVC-based ISEs in detection of SCN anion with relatively comparable good Nernstian responses and performances. The lowest detection limit with good Nernstian slope can be seen in Ganjali’s work in 2001 demonstrating Nernstian slope of -59.0 mV/decade detection limit of $6x10^{-7}$ mol/L. In his work, the detection of SCN can be said to be rapid with 10 seconds of response time and long lifetime of less than 60 days. His work has successfully draw and attain many good qualities in fabricating SCN-ISE.

sed on PVC membrane by using different types of ionophore.

Ionophore	Slope (mV/decade)	Detection limit (mol/L)	Linear range (mol/L)	Response time (s)	life time (day)	
ri-n-octyltin chloride (TOTCl)	-61.9		NR	-	-	Wuth
5,10,15,20-tetrakis(4-carboxyphenyl)porphyrin Cobalt(III)	-56.3	NR	$1x10^{-4}$ – $1x10^{-2}$	-	-	Amm
5,10,15,20-tetrakis(4-carboxyphenyl)porphyrin Manganese(III)	-56		$1x10^{-4}$ – $1x10^{-1}$			
(α,β,γ,δ-raphenylporphynate)Cobalt(III) or Co(III)-TPP	-55	NR	$1x10^{-5}$ – $1x10^{-1}$	-	>14	Hodin
(5,10,15,20-tetrakis(2,4,6-triphenyl)porphyrinato)-manganese(III) chloride or (Mn[TPPP]Cl)	-56	NR	$1x10^{-3}$ – $1x10^{-1}$	<60	-	DV
Cobalt(II) tetrakis (o-inophenyl)porphyrin or [Co(O-NH$_2$)TPP]	-43	5 x 10^{-7}	$1x10^{-7}$ – $1x10^{-2}$	<25	14	Daun
Oxo bridge iron porphyrin or (FeTPP)$_2$O	-43	6.31 x 10^{-7}	$1x10^{-6}$ – $1x10^{-1}$	-	-	Ga
	-53	3.98 x 10^{-7}	$1x10^{-6}$ – $1x10^{-1}$			
	-58	6.31 x 10^{-5}	$1x10^{-6}$ – $1x10^{-1}$			

Pd(II) amine complex	-29.1	NR	$1x10^{-6} – 1x10^{-2}$	<60	-	Sánchez et al., 1
Nickel (Ni^{2+}) phthalocyanines Iron (Fe^{2+}) phthalocyanines	-58.4	$5 x 10^{-7}$	$1x10^{-6} – 1x10^{-1}$	20	>60	Amini et al., 1
(N,N-bis-salicyclidene- 1,2,ethylenediamine) complex	-59.1	$6 x 10^{-7}$	$1x10^{-6} – 1x10^{-1}$	15	<60	Ganjali et al
vis, bis nitrato[4-hydroxyacetophenone semicarbazone] nickel(II) vis, bis nitrato[4-hydroxyacetophenone semicarbazone] copper(II)	-58.8	$1.25 x 10^{-7}$	$1x10^{-7} – 1x10^{-2}$	10	90	Chandra et al., 2008
(N-3-methyl phenyl salicylidenaminato)-copper(II)	-59.3	$5 x 10^{-7}$	$1x10^{-6} – 1x10^{-1}$	9-21	60	Benvidi et al
(2,2',2''-salicylidene-imino)-triethylamine-iron(III)	-51.8	$2.52x10^{-6}$	$1x10^{-5} – 1x10^{-1}$	5-25	30	Aslan
2'-[(4,5dimethyl-1,2-phenylene) bis[(E)-nitrilomethylidyne]] bis [4,6-bis (1,1-dimethylethyl) or n(III)-Salophen Manganese(III) chloride	NR	$1.9 x 10^{-6}$	$1x10^{-5} – 1x10^{-1}$	120	7	Abdel-Haleem & Riz
CoTMeOPP	**-60.76**	$\mathbf{9.55x10^{-8}}$	$\mathbf{1x10^{-7} – 1x10^{-1}}$	**68**	**7**	**This wo**

Continued from Table 4.8

However, the proposed electrode of self-plasticized SCN-ISE gave a better detection limit of 9.55 x 10^{-8} mol/L and Nernstian slope of -60.76 mV/decade than the previously reported SCN-ISEs in Table 4.8. Despite the short lifetime, the electrode has proven to be able to eliminate the interference from other anions that has been previously reported by the plasticized-based SCN-ISEs and this proposed SCN-ISE can be said to be able to give rapid detection in SCN within 68 seconds. These good qualities of proposed electrode have achieved a comparable and better qualities than of previously reported SCN-ISEs. The incorporation of the CoTMeOPP ionophore with self-plasticizing membrane i.e n-BA and HEMA was introduced in order to develop a green sensor that is environmentally friendly and does not have severe and critical effect to both environment and human health in a long run, which has been proven to possess lower detection limit. The utilization of TOMACl additive has also improved to be one of the contributing factors in the sensitivity and selectivity of the sensor that acts as the anionic exchanger in the membrane composition (Ammann et al., 1985). Despite the medium response time and life time of the SCN-ISE, we can say that the SCN-ISE has excellent performance with the linear range, detection limit as well as pH working value of the ISE.

The comparative study of SCN-ISE, it can be seen that the life time and the response time of the previously reported PVC-based SCN-ISE showed that the proposed electrode for the detection of SCN anion still suffers from short-lived lifetime as well as quite long response time. In order to increase the performance of the sensitivity and selectivity of the SCN-ISE, the incorporation of CoTMeOPP ionophore with other types of self-plasticizing polymers should be explored to avoid the evaporation and the degeneration of the self-plasticizing membrane when it is kept at room temperature. Thus, further studies about the effect of different self-plasticizing polymeric membranes on the performance of CoTMeOPP ionophore in detecting SCN anion is strongly needed.

The proposed self-plasticized SCN-selective electrode based on CoTMeOPP also have good comparable vaues compared to previous works that utilized CoTMeOPP ionophore in detection of different anions as shown in Table 4.9.

Table 4.9: Comparison of fabricated electrode based on CoTMeOPP ionophore

Year	**2002**	**2006**	**2010**	**2018**
Target anion	MoO_4^{2-}	SCN^-	SeO_3^{2-}	SCN^-
Polymeric membrane	PVC			HEMA n-BA
Plasticizer	DBP	o-NPOE	TCP, DBP, DOP, o-NPOE, DBBP	NA
Detection limit (mol/L)	5.0×10^{-5}	1.0×10^{-5}	$3.4x10^{-5}$	$9.55x10^{-8}$
Refs	Gupta et al. 2002	Vlascici et al. 2006	Ibrahim et al. 2010	**This work**

As mentioned in Chapter 2, there are only few reported works that utilized CoTMeOPP as the ionophore in the fabrication of anion selective electrode. The proposed SCN-ISE fabricated in this work has shown high selectivity towards SCN anion with relatively low detection limit compared to other fabricated anion-ISEs without the utilization of any plasticizer especially when it is compared to Vlascici et al., work that used the same target analyte. The interference caused by SCN anion in both Gupta et al., and Ibrahim et al., works, can be explained by the high interection between CoTMeOPP ionophore towards SCN anion.

The self-plasticized electrodes have been reported as shown in Table 4.10 also showed comparable performances and qualities with the proposed self-plasticized SCN-ISE in this work. The anion-selective plasticized-free electrodes reported by Matusevich et al., and Mendecki et al., by using different ionophores or ligands showed good Nernstian reponses towards the target analyte by giving acceptable Nernstian slopes (Matusevich et al., 2012, Mendecki et al., 2016). These works also showed low detection limits, rapid n of fabricated anion-ISE based on self-plasticized membrane.

meric branes	Ionophore	Target ions	Slope (mV/decade)	Linear range (mol/L)	Detection limit (mol/L)	Response time (sec)	Life time (days)	Re
DOT 12SS	AITAP ZrTAP AlSAL UO_2SAL	F^-	-58.5 -56 -55 -40	$1x10^{-5}$ to $1x10^{-1}$ $1x10^{-5}$ to $1x10^{-1}$ $1x10^{-3}$ to $1x10^{-1}$ $1x10^{-4}$ to $1x10^{-1}$	NR	11	>90	(Matu al., 2
y(3- iphene)	1,2,3-triazole (iL)	I^-	-56.1	$1x10^{-7}$ to $1x10^{-1}$	$6.3x10^{-8}$	NR	14	(Mende 20
MA BA	**CoTMeOPP**	**SCN^-**	**-60.76**	**$1x10^{-7}$ to $1x10^{-1}$**	**$9.55x10^{-8}$**	**68**	**7**	**This**

response time and long lifetime. The performance and quality of the proposed self-plsticized SCN-ISE in this work can be added to anion-ISE based on self-plasticized membanes as it on par with another reported plasticizer-free anion-ISEs. Regardless of the deficiencies in this study, all data proved that the proposed SCN-ISE can be applied in determination of SCN anions present in solution respectively.

4.4.2 Sensing Mechanism of Fabricated ISE

The CoTMeOPP ionophore is metalloporphyrin which contained porphyrin rings with Co(II) as the metal centre of the ionophore. When porphyrin rings form complexes with Co(II), the lipophilicity of the macrocycle can be easily tuned through pheripheral substituents to ensure better compatibility with the membrane materials (Prasad et al., 2004). The liphophilic quaternary ammonium salt aids in the interaction between the sensing material and the anion of interest by using the nitrogen atoms that are closely located in the ionophore compound (Gao et al., 1994). Owing to the presence of the quaternary salt and the metal, the ionophore are able to display anti-Hofmeister type of selectivity with unique anion responses. The anion ionophore, in this case the CoTMeOPP ionophore acts as neutral carrier in membrane phase i.e electrically neutral when no complexes are formed and upon the addition of SCN, the complexation between CoTMeOPP and SCN occurred and become negatively charged complex (Bakker et al., 1994) as shown in Figure 4.20.

The porphyrin rings delocalized π systems and the overlapping between ligand π orbitals and Co(II) metal orbitals of proper symmetry results in moderately high ligand field strength. Complexation between SCN anion and CoTMeOPP ligand produces back-accepting π electron density and the reduction of the oxidation state of the complexed metal-ligand occurred (Ostfeld & Tsutsui, 1974). After conditioning ISE containing CoTMeOPP ionophore with 0.01mol/L anion solution (KSCN), the ionophore in the membrane present in deprotonated form (Co^{2+}TMeOPP and Co^{3+}TMeOPP) and neutral

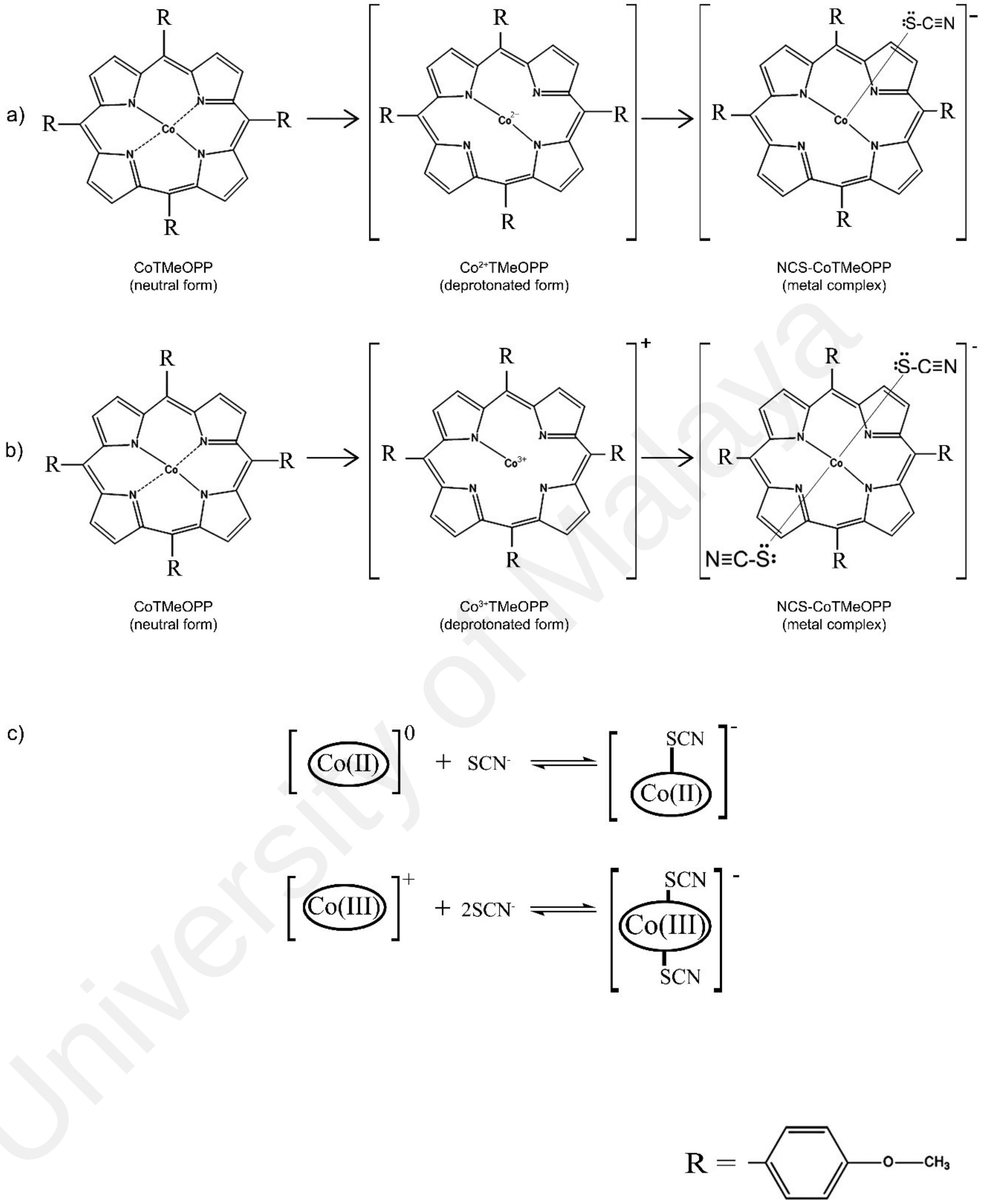


Figure 4.20: CoTMeOPP Ionophore in neutral, deprotonated and complex form (a) Co^{2+}, (b) Co^{3+} and (c) the schematic diagram of complexation between CoTMeOPP with Co(II) as the metal ionophore and CoTMeOPP with oxidised Co(II) to become Co(III) with SCN anion

form (CoTMeOPP). Therefore, the carrier mechanism of both Co^{3+} and Co^{2+} can be explained as partially disassociated ligand and the interaction of ionophore with targeted anion was confirmed using UV-vis, Fluorescence and FT-IR. All the characterisation method results suggested that the complexation occurred at $Co^{3+/2+}$ positions as illustrated in Figure 4.20.

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The SCN anion exhibits axial ligand interactions with the metal centre of the metalloporphyrin i.e $Co^{2+/3+}$ metal (Wang & Meyerhoff, 1993) and complexation formed at S-atom location of the SCN as shown in Figure 4.20 above. The formation of complex between $Co^{2+/3+}$ metal at S atom location is supported by using UV-vis spectra in Figure 4.11 (section 4.3.1). Based on the UV spectra, it can be observed in Table 4.11 that the absorption of the SCN-CoTMeOPP peaks at 300 nm is lower than of the absorption peak at 500 nm. The lower absorption peak at 500 nm compared to 300 nm and the red colour of complexation in Figure 4.10 and 4.11 indicated that the complexation occurred at S-atom position of the SCN (Buckingham, 1994).

Table 4.11: UV-vis absorption peaks of SCN-CoTMeOPP at 300 nm and 500 nm.

Spectra	Absorption (a.u)	
	300 nm	500 nm
SCN-CoTMeOPP	1.793	0.660

CHAPTER 5: CONCLUSION AND RECOMMENDATIONS

5.1 Conclusion

In this research work, the 5,10,15,20-tetrakis-(4-methoxyphenyl)-21H,23H-porphyrin Co(II) or CoTMeOPP ionophore was examined and proven to be selective and sensitive towards SCN anion. The fabrication of chemical sensor based on CoTMeOPP as the active material was successfull with the aid of self-plasticizing membranes of (hydroxyethyl)methacrylate (HEMA) and n-butyl acrylate (n-BA) and the SCN-ISE was tested for its selectivity and sensitivity by using potentiometric method. The optimum membrane composition was obtained by using silver/silver chloride (Ag/AgCl) as the first layer, 95.14 wt.% monomer (HEMA) and 4.86 wt.% photo initiator (DMPP) as the second layer and 98.86 wt.% monomer (n-BA), 0.08 wt.% photo initiator (HDDA), 0.34 wt.% ionophore (CoTMeOPP), 0.16 wt.% lipophilic salt (TDDA) and 0.56% photo initiator (DMPP). The optimized SCN-ISE exhibits a good Nernstian behaviour with the slope of -60.76 mV/ decade and detection limit of 9.55×10^{-8} mol/L over a wide concentration range of 1.0×10^{-7} mol/L to 1.0×10^{-1} mol/L. Characterisation of the fabricated SCN-ISE were conducted to study the complexation between SCN anion and CoTMeOPP ionophore by using UV-vis spectroscopy, fluorescence spectroscopy and FT-IR. Further characterization to study the morphology of the SCN-ISM were conducted by using FESEM and EDX analysis. Optimization of the fabricated SCN-ISM onto the GCE were conducted and evaluated for its responses, selectivity and selectivity towards SCN^- anion. The optimized SCN-ISE was then tested for real sample analysis in determining SCN^- anion in river water, ex-mining lake sediment, human saliva of smoker and non-smoker and fish samples. It was proven that the fabricated SCN-ISE is highly selective and sensitive towards SCN^- anion and can be used as portable sensor in determining SCN^- concentration in real sample analysis.

5.2 Recommendations

The fabrication of potentiometric SCN-ISE sensor should be done with plasticized and other self-plasticizing membranes in order to study and compare the performance between plasticized and self-plasticized SCN-ISE. Other than that, the effect of different self-plasticizing membranes with the performances of the ISEs in terms of sensitivity, selectivity, response time as well as the life time of the fabricated sensor can be further studied when more than one self-plasticized polymeri membranes are utilized.

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