ADDIS ABABA UNIVERSITY
SCHOOL OF GRADUATE STUDIES
DEPARTMENT OF CHEMISTRY

Electrochemical Determination of Trichlorophenol using Modified Electrode

BY

Ephrem Hailemariam

Advisor: Merid Tessema (PhD)

June, 2014

Addis Ababa University

School of Graduate Studies
Department of Chemistry

Electrochemical Determination of Trichlorophenol using Modified Electrode

By:

Ephrem Hailemariam

A Thesis Submitted to the Department of Chemistry, Addis Ababa University In Partial Fulfillment of Requirements for Masters of Science in Chemistry (Analytical Chemistry)

June, 2014

Addis Ababa, Ethiopia

Addis Ababa University

School of Graduate Studies

Department of Chemistry

Electrochemical Determination of Trichlorophenol using Modified Electrode

By: Ephrem Hailemariam

Approved by the examining board:	Signature	date
Merid Tessema (PhD) (Advisor)	_____	_____
Theodros Solomon (Prof.) (Examiner)	_____	_____
Solomon Mehretie (PhD) (Examine	_____	_____

Acknowledgements

First and foremost, I want to thank GOD for helping me during all the difficult times of my life and giving me the strength to do the project work**.**

My sincere gratitude goes to my advisor, Dr. Merid Tessema for his professional guidance, valuable advice, encouragement and strong support. Dr. Merid continuously supported this work from its initial stage. He gave me a good insight from the very beginning. Despite, his tight schedule he patiently read the first draft of the thesis and gave very valuable comments, corrections and suggestions. I have a high regard for his interest in this work and am grateful for all clues in my studies, which were always very valuable.

Specially I would like to express my deepest gratitude to, Ato Negussie Negash (PhD Student at Department of Chemistry in Electrochemistry lab, AAU) for valuable guidance, consistence encouragement and constructive criticism throughout my thesis work and for his unrestricted intellectual advice in shaping the study and also I would like to thank again Ato Negussie with deepest gratitude for allowing me to work with his PhD laboratory Material and Instrument as a result of which I was free to use all the required materials available in the laboratory. Next, I would like to thank Ato Molla Tefera and Alemnew Geto (PhD. Students at Department of Chemistry in Electrochemistry lab, AAU) for their valuable comments. Finally I am extremely thankful for Addis Ababa University providing me tuition free scholarship award.

I would like to express my deepest sense of gratitude and respect to my beloved family and I would like to thank all those who directly or indirectly helped me to complete this thesis, especially to all my friends and BGI Ethiopia Quality Control lab staffs.

Table of contents Page

List of Tables

page

List of Figures page

Abbreviations and symbols

BAS	Bioanalytical system
CMEs	Chemically modified electrodes
CV	Cyclic voltammetry
E_{pa}	Anodic peak potential
E_{pc}	Cathodic peak potential
E_{sw}	Square wave amplitude
GCE	Glassy carbon electrode
HPLC	High performance liquid chromatography
I	Current
I_p	Peak current
LOD	Limit of detection
mGCE	modified glassy carbon electrode
PEDOT	poly(3,4-ethylenedioxythiophene)
RSD	Relative standard deviation
SWCNT	Single wall carbon nanotube
SWV	Square wave voltammetry
TCP	Trichlorophenol
W_p	Pulse width
USEPA	US Environmental Protection Agency
μM	Micro molar

Electrochemical Determination of Trichlorophenol using Modified Electrode

Abstract

The electrochemical behavior of 2,4,6-Trichlorophenol was investigated using single-wall carbon nanotubes (SWCNT) and poly(3,4-ethylenedioxythiophene) (PEDOT) composite modified glassy carbon electrode (SWCNT/PEDOT/GCE. 2,4,6-Trichlorophenol was studied by using cyclic voltammetry at different scan rates over the potential range of -0.2 to 1.2 V. The results indicated that the reactions of the 2,4,6-Trichlorophenol at modified glassy carbon electrode is irreversible and charge-transfer controlled. Optimization of different variables such as pH of working solution, and modifier composition were carried out. Square wave voltammetry parameters such as frequency, amplitude and step potential were also adjusted to improve the method efficiency during the experiment. The modified electrode exhibited very good analytical performance due to this the analysis of sample showed best operational stability. Under this experiment, analytical parameters were calculated from the calibration curve of measured square wave responses as a function of concentrations of 2,4,6-Trichlorophenol. The reproducibility for the ten repeated measurements of 80 μM of 2,4,6-Trichlorophenol gave a relative standard deviation of 3.65%, indicating excellent reproducibility of the method. Linear calibration plots were obtained in the range 6 - 80 $\mu molL^{-1}$ with ($R^2 = 0.999$) and the detection limit with (S/N=3) was as low as $7.5x10^{-8}$ $molL^{-1}$.

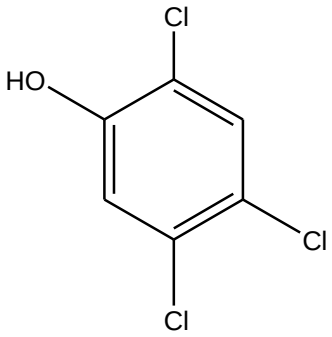
KEY WORDS

2,4,6- Trichlorophenol, PEDOT, SWCNT, Glassy carbon electrode and square wave voltammetry.

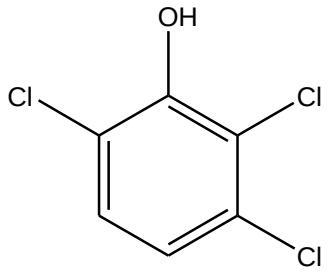
1. Introduction

Chlorophenols are a group of chemicals that are produced by substitution of chlorines to phenol. Phenol is an aromatic compound derived from benzene [1]. The presence of chlorinated compounds in aquatic environments has caused several environmental pollution problems. Chlorinated phenols being widely used constitute a major class of organic pollutants that contaminate the ecosystem and accumulate in the food chain [2]. Various industrial effluents contain up to 18 mg/l of Chlorophenols, while the municipal wastewater contains from 1 to 21 mg/ml of these compounds [2]. There are five basic types of Chlorophenols and 19 different isomer Chlorophenols. Several chlorinated phenols such as 2-chlorophenol, 2,4-dichlorophenol, 2,4,6-trichlorophenol, and pentachlorophenol have been classified as the priority pollutants [2]. Most Chlorophenols are solid at room temperature. They have a strong, medicinal taste and smell. Small amounts can be tasted in water. Some Chlorophenols are used as pesticides and antiseptics and in bleaching wood pulp with chlorine to make paper. Small amounts are produced when water is disinfected with chlorine. Moreover, the 2,4,6-TCP is considered a priority pollutant by both the US Environmental Protection Agency (USEPA) and the European Union (EU). Thus, the residue of 2,4,6-TCP amounts in our living environment is very important. Many official methods were published for determination of 2,4,6-TCP including EPA methods 604 [1] and 8270 [11]. Many researchers also have reported chromatographic methods and spectrophotometry for the determination of Chlorophenols, including high performance liquid chromatography (HPLC) [11, 12], gas chromatography-mass spectroscopy (GC-MS) [13,14] and gas chromatography-electron capture detector (GC-ECD)[7]. This work present a good sensitivity, selectivity and reproducibility based on SWCNT/PEDOT glassy carbon electrode using square wave voltammetry to 2,4,6-TCP determination.

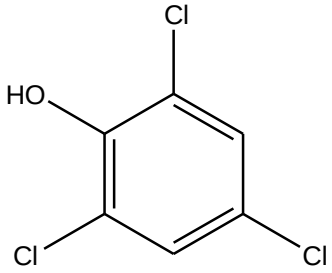
Trichlorophenol is a class of Chlorophenols that contains three covalently bonded chlorine atoms. Trichlorophenol are produced by electrophilic halogenations of phenol with chlorine. [5]. Different isomers of Trichlorophenol exist according to which ring positions on the phenol chlorine atoms are contained. For example 2,4,6-Trichlorophenol, has two chlorine atoms in the *ortho* positions and one chlorine atom in the *Para* position. There are different isomers of Trichlorophenol those are 2,3,4-Trichlorophenol, 2, 3, 5-Trichlorophenol, 2,3,6-Trichlorophenol 2,4,5-Trichlorophenol, 2,4,6-Trichlorophenol 3,4,5-Trichlorophenol [7].



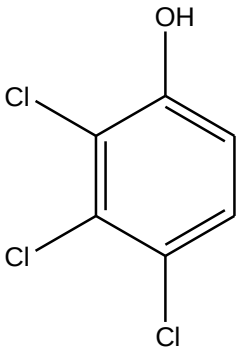
2,4,5 Trichlorophenol



2,3,6 Trichlorophenol



2,4,6 Trichlorophenol

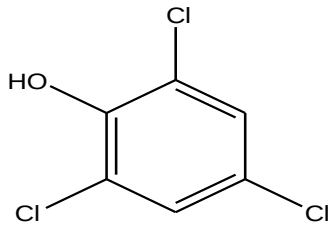


2,3,4 Trichlorophenol

Figure 1. Different isomer of Trichlorophenol

1.1. Physical and Chemical Properties of 2,4,6-Trichlorophenol

2,4,6-Trichlorophenol is a chlorinated phenol compound that exists at room temperature as colorless to yellow crystals with a strong phenol odor. A chemical formula for 2,4,6-Trichlorophenol is $C_6H_3Cl_3O$, and it has a molecular weight of (197.45 g/mol)



2,4,6 Trichlorophenol

It is practically insoluble in water but. It is soluble in acetone, benzene, carbon tetrachloride, diacetone, alcohol, ether, denatured alcohol formula, methanol, pine oil, toluene, turpentine and water [9]. It is also soluble in hot acetic acid. It is stable under normal temperatures and pressures [13].

Table 2. Physical properties of 2,4,6-Trichlorophenol

Properties	Information
Molecular weight	197.5
Specific gravity	1.4901
Melting point	69°C
Boiling point	246°C
Log K_w	3.69
Water solubility	0.800 g/L at 25°C
Vapor pressure	0.008 mm Hg at 25°C
Dissociation constant (p*K*a)	6.23 at 25°C

2,4,6-Trichlorophenol is organochloride of phenol that contains three covalently bonded chlorine atoms. 2,4,6-Trichlorophenols are produced by electrophilic halogenation of phenol with chlorine. Other name of 2,4,6-Trichlorophenol is 1-Hydroxy-2,4,6-trichlorobenzene. 2,4,6-Trichlorophenol is incompatible with acid chlorides, acid anhydrides and oxidizing agents. 2,4,6-Trichlorophenol can be converted to the sodium salt by reaction with sodium carbonate. It forms ethers, esters and salts by reaction with metals and amines. It undergoes substitution reactions such as nitration, alkylation, acetylation and halogenations. It can be hydrolyzed by reaction with bases at elevated temperatures and pressures. It reacts with alkalis at high temperatures. It is volatile with steam but unable to from alkaline solution [12].

1.2. Health effects of 2,4,6-Trichlorophenol

2,4,6-Trichlorophenol is the most toxic chlorophenol compounds, on breathing, it can irritate the nose, throat and lung causing coughing ,wheezing and shortness of breathing, high exposures may cause restlessness, weakness, rapid breathing and shaking [12]. The routes to potential human exposure to 2,4,6-Trichlorophenol are inhalation, ingestion, and dermal contact. The general population may be exposed to 2,4,6-Trichlorophenol through ingestion of contaminated food or water or inhalation of contaminated air [11].

2,4,6-Trichlorophenol is considered a priority pollutant by both the US Environmental Protection Agency (USEPA) and the European Union (EU). According to the National Cancer Institute (NCI 1979), substantial exposure of the general population was questionable; however, residues may be present throughout the environment.

2,4,6-Trichlorophenol can also form compounds when industrial wastewater containing phenol or certain aromatic acids are treated with hypochlorite or during the disinfection of drinking water. 2,4,6-Trichlorophenol has been detected in ambient air and in samples of river water, landfill leachate, chemical plant effluent water, sewage treatment plant effluent, and tap water. Generally directly or in directly 2,4,6-Trichlorophenol has significant impact on the human health.

1.3. Economic importance of 2,4,6-Trichlorophenol

2,4,6-Trichlorophenol is used as a broad range pesticide against insects, fungi, vegetation and bacteria and as a fungicide in paper and pulp mills, as an herbicide, germicide for preservation of wood, leather, glue, and textiles ingredients of germicidal soaps and as an intermediate in the manufacture of other pesticides [11,12].

2. Literature Review

2.1. Chemically modified electrodes (CMEs)

A Chemically modified electrode (CME) is an electrode made of conducting or semiconducting material that is coated with a selected monomolecular, multimolecular, ionic or polymer film. There are different electrode modifiers, including: polymers, permselective membranes, organic ligands, lipids, surfactants, organic and inorganic micro particles, organometallic or inorganic catalysts, ion exchangers, biological materials, clays, zeolites silicates and so on [20, 23]. Thus chemically modified electrodes prepared by surface modification to produce an electrode suited for a particular function and show different properties to those of the unmodified electrodes.

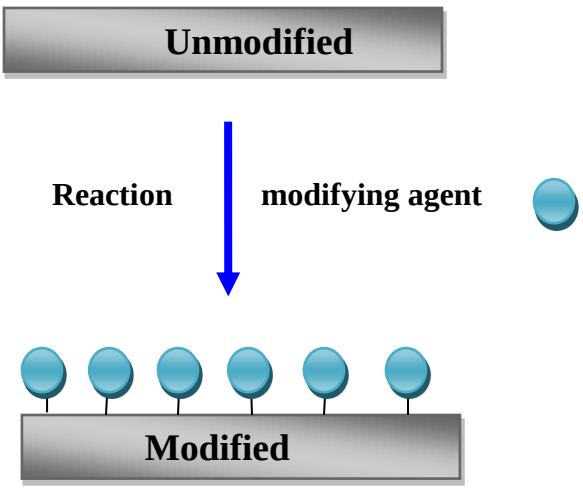


Figure 2. Immobilization of a modifying agent on the electrode surface

Purposeful covering on electrode surface with adsorbed layers or film can change electron transfer rate on an electrode surface. Electrocatalysis at modified electrode is usually an electron transfer reaction between the electrodes and the nearby solution mediated by an immobilized modifier, which proceeds at lower over potential, otherwise occur at bare electrode [23].

Electrodes are usually chemically modified by one of the following four approaches.

(1) Chemisorptions adsorption: Here the forces involved are the valence forces of the same as those operating in the formation of chemical compounds**.** The chemical film is strongly ideally and irreversibly adsorbed (chemisorbed) onto the electrode surface. This approach usually yields monolayer (or less) coverage. This type of modification are the substrate-coupled self-assembled monolayers (SAMs) in which uncorrelated molecules spontaneously chemisorbs at specific sites on the surface of the electrode to form a super lattice [23].

(2) Covalent bonding: Linking agents, such as, organosilanes or cyanuric chloride are used to covalently attach from one to several monomolecular layers of the chemical modifier to the electrode surface [23].

(3) Composite: The chemical modifier is simply mixed with an electrode matrix material, as in the case of an electron-transfer mediator (electro-catalyst) combined with the carbon particles (plus binder) of a carbon paste electrode. Alternatively, intercalation matrices such as certain Langmuir-Blodgett films, zeolite, clays and molecular sieves can be used to contain the modifier. The composite of PEDOT and SWCNT are example [26].

(4) Polymer film coating: Electron conductive and nonconductive polymer films are held on the electrode surface by combination of Chemisorptions and low solubility in the contacting solution or by physical anchoring in a porous electrode. The polymer film can be organic, organo metallic or inorganic [27].

Polymer film-coated electrodes may be further subdivided by the process used to apply the film, namely.

Dip-coating - this procedure consists of immersing the electrode material in a solution of the polymer for a period sufficient for spontaneous film formation to occur by adsorption. The film quantity in this procedure may be augmented by withdrawing the electrode from the solution and allowing the film of polymer solution to dry on the electrode [27].

Solvent evaporation - a droplet of a solution of the polymer is applied to the electrode surface and the solvent is allowed to evaporate. A major advantage of this approach is that the polymer coverage is immediately known from the original polymer solution concentration and droplet volume [26].

Spin coating - also called spin casting; a droplet of a dilute solution of the polymer is applied to the surface of a rotating electrode. Excess solution is spun off the surface and the remaining thin polymer film is allowed to dry. Multiple layers are applied in the same way until the desired thickness is obtained [30].

Electrochemical polymerization- a solution of a monomer is oxidized or reduced to an activated form that polymerizes to form a polymer film directly on the electrode surface. This procedure results in few pinholes since polymerization would be accentuated at exposed (pinhole) sites at the electrode surface. Unless the polymer film itself is redox active, electrode passivation occurs and further film growth is prevented [26].

Radiofrequency polymerization- is a polymer-filming method in which vapors of the monomer are exposed to a radiofrequency plasma discharge. The high energetic radiofrequency may result in chemical damage, thereby producing unknown functionalities and structural modifications to the polymer [27].

Cross-linking a chemical step designed to couple chemical components of a film on an electrode to impart some desired property to the film such as increased stability, decreased permeability, or altered electron transport characteristics. Cross-linked films are often formed by copolymerization of bi-functional and poly functional monomers. Cross-linking may be activated chemically, electrochemically, photolytically, radiolytically, or thermally. These forms of modification are the substrate-decoupled (SAMs) in which adsorbate molecules are arranged on the electrode surface independently of any substrate structure [22, 23].

2.2. Conducting polymers

The terms polymer and monomer were derived from the Greek roots poly (many), mono (one) and meres (part) which is a large molecule, or macromolecule, composed of many repeated subunits, known as monomers [24].

Conductive polymers may be classified into two different categories: extrinsically conductive polymers and intrinsically conductive polymers. On one hand, extrinsically conductive polymers can involve a blend of conductive and nonconductive polymers, as well as metallic particles suspended in a polymer matrix [25]. Basically, they consist of highly conductive additives incorporated into polymer compounds, meaning that they are extrinsically enhanced to be conductive. Intrinsically conductive polymers, on the other hand, consist of a network of alternating single and double carbon bonds. This alternation of bonds produces conjugated π-bonds that result in a conductive material. Many polymers are strong, elastic, plastic, tough, friction-resistant and insulating. We encounter them in our day-to-day life in a wide range of products from most consumer goods to highly specialized applications. Conducting polymers contain π-electron backbone responsible for their universal electronic properties such as electrical conductivity, low energy optical transitions, low ionization potential and high electron affinity. Conducting polymers have potential applications in various fields such as fuel cells, biosensors, electroanalysis, electrocatalysis, electrochromic displays and others [27]. In recent years, electrochemically synthesized polymer films on electrode surface are considered for potential applications in electroanalysis of biological molecules.

A recent review by Cosnier [26], deals with recent advances and scientific progress in electrochemical immobilization procedures for biological macromolecules on electrodes via electro generated polymer films. Electrochemical synthesis and some properties of nanostructured conducting polymers and nano composites derived from conducting polymers and metals, carbon and inorganic and organic materials and their potential applications in batteries, super capacitors, energy conversion systems, corrosion protection and sensors have been reviewed by Malinauskas et al [25]. Conducting polymers are a natural choice for preparing arrays of voltammetry sensors because they have a rich electrochemical behavior and their

electrochemical properties can be modulated by introducing chemical modifications in the sensitive materials [27].

Electropolymerization is a good approach to prepare polymer modified electrodes (PMEs) as adjusting electrochemical parameters can control film thickness, permeation and charge transport characteristics. PMEs have many advantages in the detection of analytes because of their selectivity, sensitivity and homogeneity in electrochemical deposition, strong adherence to electrode surface and chemical stability of the film [28]. The electronically conducting polymers (ECPs) which cover several types of polymeric materials with electronic and/or ionic conductivity, including: (i) doped conjugated polymers, (ii) redox polymers, (iii) polymer composites and (iv) polymer electrolytes. ECPs as ion selective membrane and their applications in solid-state ion selective electrodes have been reviewed [29]. The most popular members of ECPs including polyaniline, polythiophene, polypyrrole and polyphenylene are p-type semiconductors as shown in Fig. 2 [30, 31]. Stability of polymers, conductivity and doping nature has been discussed in several reviews [32]. Conducting polymers are promising materials for a large variety of chemical sensors, their preparation and applications in several aspects have been discussed in many reviews [33, 34].

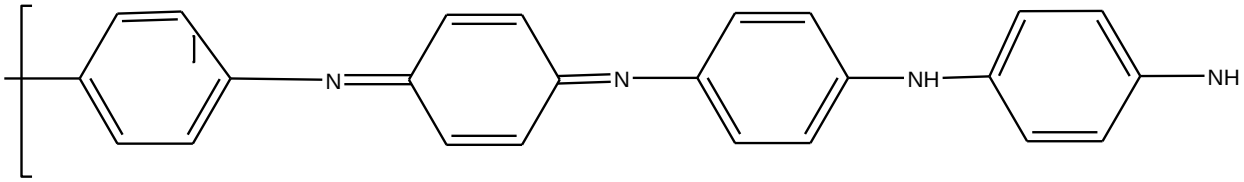
Synthesis of conducting polymers can be classified into two major categories: chemical polymerization and electrochemical polymerization. Via chemical polymerization, conjugated monomers react with an excess amount of oxidant in a suitable solvent, such as acid. The polymerization takes place spontaneously and requires constant stirring. A major advantage of chemical polymerization is the possibility of mass production at a reasonable cost this is often difficult with electrochemical methods [33].

The second method is electrochemical polymerization, which involves placing counter, working and reference electrodes into the solution containing diluted monomer and electrolyte (the dopant) in a solvent. After applying a suitable voltage, the first steps is electro oxidative formation of a radical cation from starting monomer this step is followed by dimerization process, followed by further oxidation and coupling reactions. Well-adhered film can thus be formed on the surface of working electrode. The behavior of electro polymerized film can be controlled by the polymerization conditions such as, the electrolyte, solvent, monomer

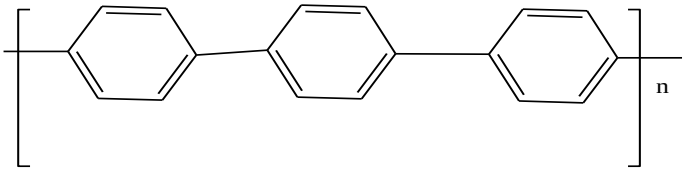
concentrations, applied potential or current and duration [30,31]. An important feature of the electropolymerization technique is the direct formation of conducting polymer films that are highly conductive, simple and suitable for use especially in electronic devices. But, not all organic monomers undergo electro-polymerization.

Certain monomers are electropolymerized due to stability of the radical ions generated in the first step and the oxidation potentials for generation of these radical cations [32].

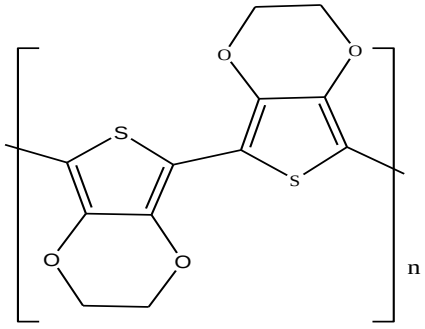
Example of polymers



Polyaniline (conductivity 1-100 S/cm)



Polyphenylene (conductivity 100 – 500 S/cm)



PEDOT

Figure 3. Some common conducting polymers [43]

2.3. Carbon nanotubes

Carbon nanotubes (CNTs) are allotropes of carbon. They have interesting properties that make them potentially useful in many applications in nanotechnology, electronics, optics and other fields of materials science, as well as potential uses in architectural fields. They exhibit extraordinary strength and unique electrical properties, and are efficient conductors of heat. CNT is a tubular form of carbon with diameter as small as 1nm and length of few nm to microns [35]. They have attracted much attention because of their high electrical conductivity, mechanical strength and chemical stability [36]. The unique properties of CNTs make them extremely attractive for electrochemical sensors and biosensors [37]. Recent investigations demonstrated that CNTs show strong electro catalytic activity and minimization of surface fouling if employed to improve the electrochemical response of some important bioactive molecules. Recently, methods based on electrochemical deposition [38] and electrochemical polymerizations [37] have been employed for loading CNTs onto electrodes. CNT-based electrodes have been demonstrated to reduce the over-potential significantly [37, 38]. The ability of carbon nanotubes to promote electron-transfer reactions suggests great promise for amperometric sensors [37]. The use of chemically modified electrodes by carbon nanotubes in electroanalysis have been reviewed in detail by Gregory et al [40]. Depending on the arrangement of their grapheme cylinders, there are two types of nanotubes:- single-walled carbon nanotubes (SWCNTs) and multi-walled carbon nanotubes (MWCNTs).

The conducting property of carbon nanotubes is dependent on the geometry of the tube. If the nanotube structure is armchair then the electrical properties are metallic. If the nanotube structure is chiral then the electrical properties can be either semiconducting with a very small band gap, otherwise the nanotube is a moderate semiconductor .In theory, metallic nanotubes can carry an electrical current density of 4×10^9 A/cm^2 which is more than 1,000 times greater than metals such as copper [38].

All carbon nanotubes are expected to be very good thermal conductors along the tube, but good insulators laterally to the tube axis. It is predicted that carbon nanotubes will be able to transmit up to 6000 watts per meter per Kelvin at room temperature; compare this to copper, a metal well-known for its good thermal conductivity, which transmits 385 watts per meter per K.

The temperature stability of carbon nanotubes is estimated to be up to 2800^{o}C in vacuum and about 750^{o}C in air [35, 38].

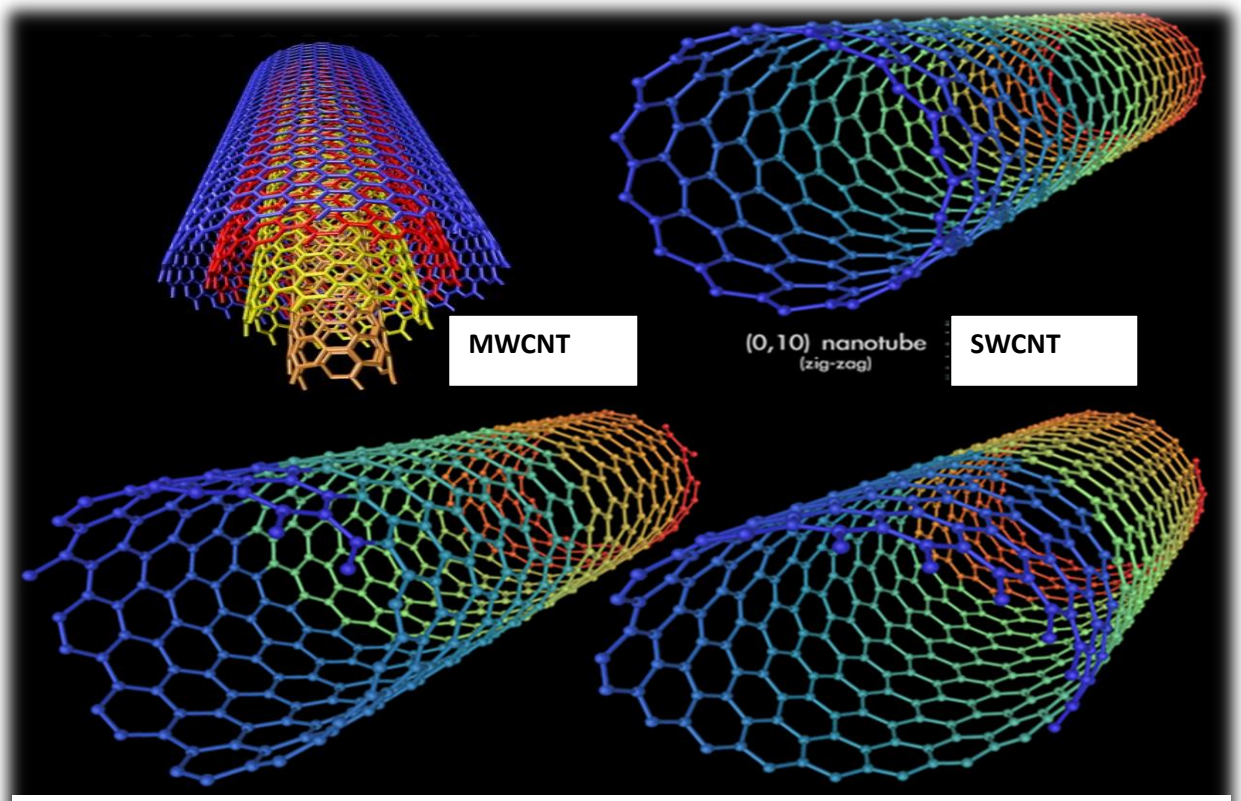


Figure 4. Structure of carbon nano-tube [33]

2.3.1. Single-walled carbon nanotubes

Single-walled carbon nanotubes are one part of carbon nanotube and they have only one single layer of grapheme cylinders; as shown in Fig.5. Most single-walled carbon nanotubes have a diameter of close to 1 nanometer, with a tube length that can be many millions times longer [37]. The structure of a SWCNT can be conceptualized by wrapping a one-atom-thick layer of graphite called graphene into a seamless cylinder. Single-walled carbon nanotubes are an important variety of carbon nanotubes; because they exhibit electric properties that are not shared by the multi-walled carbon nanotube (MWCNT) variants. In particular, their band gap can vary from zero to about 2 eV and their electrical conductivity can show metallic or semiconducting behavior, whereas MWCNTs are zero-gap metals. Single-walled carbon nanotubes are the most likely candidate for miniaturizing electronics beyond the micro electromechanical scale currently used in electronics [37].

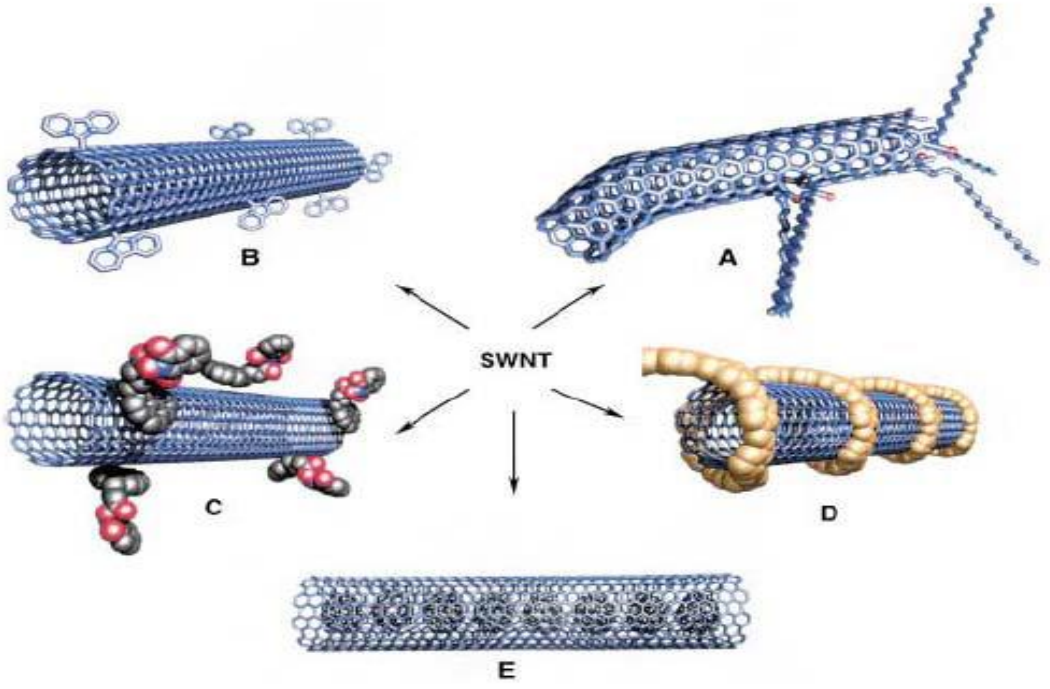


Figure 5. Structure of single-walled carbon nano-tubes [35]

2.4. Poly (3,4-ethylenedioxythiophene) (PEDOT)

Poly (3, 4-ethylenedioxythiophene) (PEDOT) is one of the most successful conducting polymers because of its low price, excellent environmental stability, high electrical conductivity, and transparency in thin oxidized films and high light transmission property. PEDOT was first synthesized in 1989 [44]. It has recently sparked much interest in the research field, especially modification of working electrodes due to its evidently superior qualities over other polymers. PEDOT is a conjugated polymer capable of both electronic and ionic charge conduction. Although PEDOT is not found in living systems, examples exist of conjugated polymers with quite similar chemistries that are common in nature [43]. Headlining these superior qualities are high stability, enhanced light transmission process ability, and simplicity of production [43]. PEDOT stands out as a material with high superiority over other polymers. The polymer backbone lacks the ability to form α-β linkages or β-β linkages, therefore the material is more regiochemically defined [44].

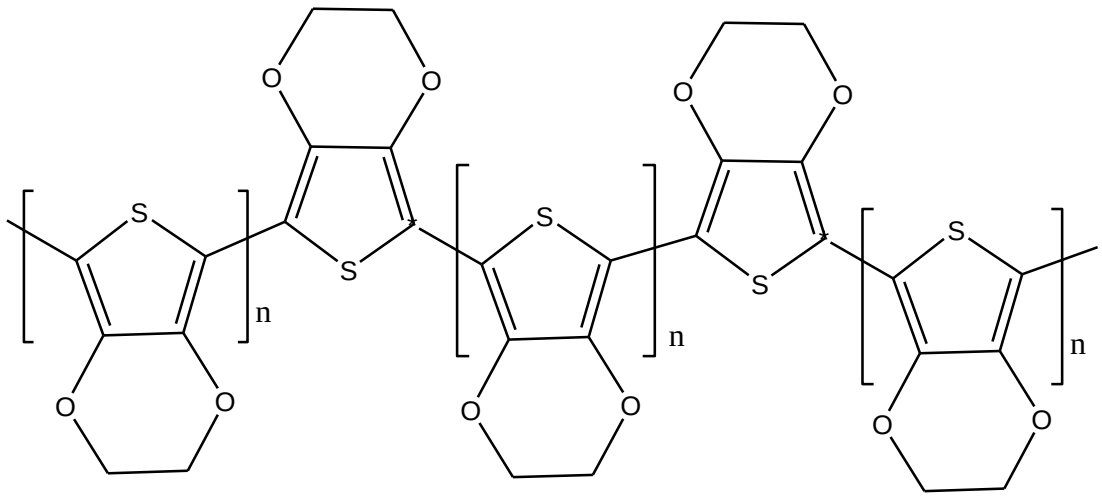


Figure 6. Poly (3, 4-ethylenedioxythiophene) Polymer

In the conventional solid electrolytic capacitors, conductive polymer was synthesized in the capacitor element. The polymerization reaction of conductive polymer is shown in Fig.7. Monomer ethylene dioxithiophene (EDOT) is synthesized into polymer (PEDOT) through oxidation polymerization.

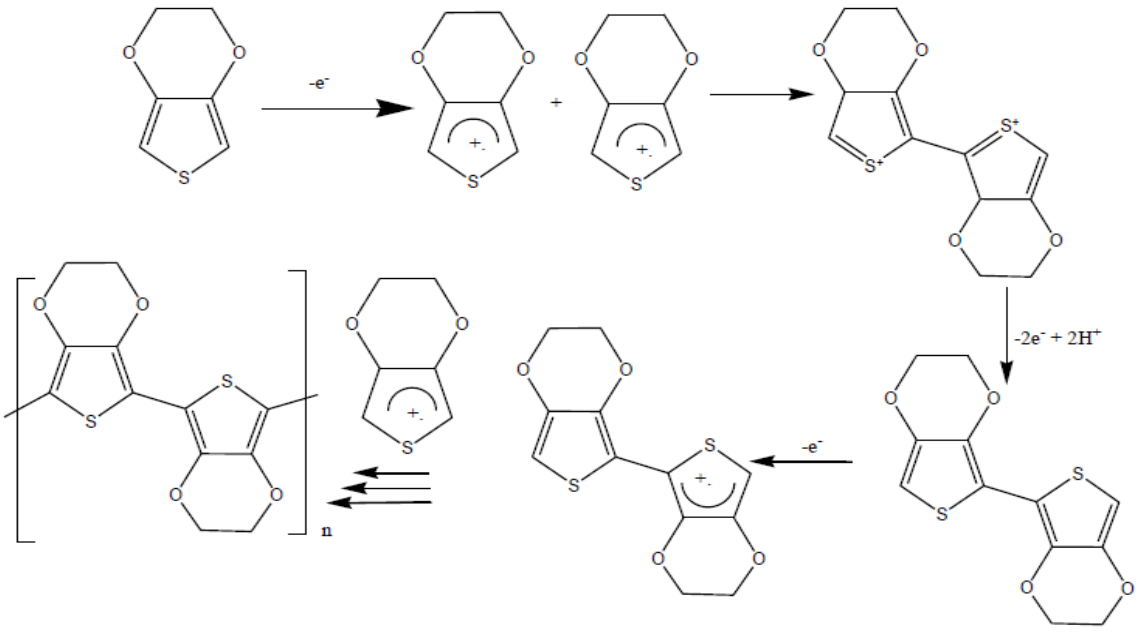


Figure 7. Poly (3, 4-ethylenedioxythiophene) Polymerization process

2.5. Voltammetric Methods

Voltammetry is an electroanalytical technique and versatile technique for research purposes based on the measurement of current flowing through an electrode dipped in solution containing electro active compounds while a potential is imposed upon it [20, 21]. Voltammetry is typically performed using a three electrode system connected to a potentiostat, which accurately controls the applied potential. The redox reaction takes places at the working electrode, because the working electrode is where the reaction of interest is taking place. This electrode could be made of several materials. Usually, it has a very little surface in order to assume quickly and accurately the potential imposed by the electrical circuit. The second electrode is a reference electrode, which maintains a constant potential throughout the experiments, and the third electrode is the counter electrode, which complete the electrical circuit. The counter electrode also known as the auxiliary electrode, is often much larger than working electrode to minimize current density at the electrode surface [20]. Generally the potential is applied between working electrode and the reference electrode whereas the current is measured between the working electrode and counter electrode. Historically, the field of voltammetry developed from polarography, which is a particular type of voltammetry that was discovered by the Czechoslovakian chemist Jaroslav Heyrovsky in early 1920s [50].

The common characteristic of all voltammetric techniques is that they involve the application of a potential (E) to an electrode and the monitoring of the resulting current (i) flowing through the electrochemical cell [50]. In many cases the applied potential is varied or the current is monitored over a period of time (t). Thus, all voltammetric techniques can be described as some function of E, i and t. They are considered active techniques (as opposed to passive techniques such as potentiometry) because the applied potential forces a change in the concentration of an electro active species at the electrode surface by electrochemically reducing or oxidizing it [50].

The analytical advantage of various voltammetric techniques include excellent sensitivity with very large useful linear concentration range for both inorganic and organic species (10^{-12} to 10^{-1} M), a large number of useful solvents and electrolytes, a wide range of temperature they allow, rapid analysis times (seconds) simultaneous determination of analytes, the ability to determine kinetics and mechanistic parameters, a well-developed theory and thus the ability to reasonably

estimate the values of unknown parameters, and the case with which different potential waveforms can be generated and small current measured[48].

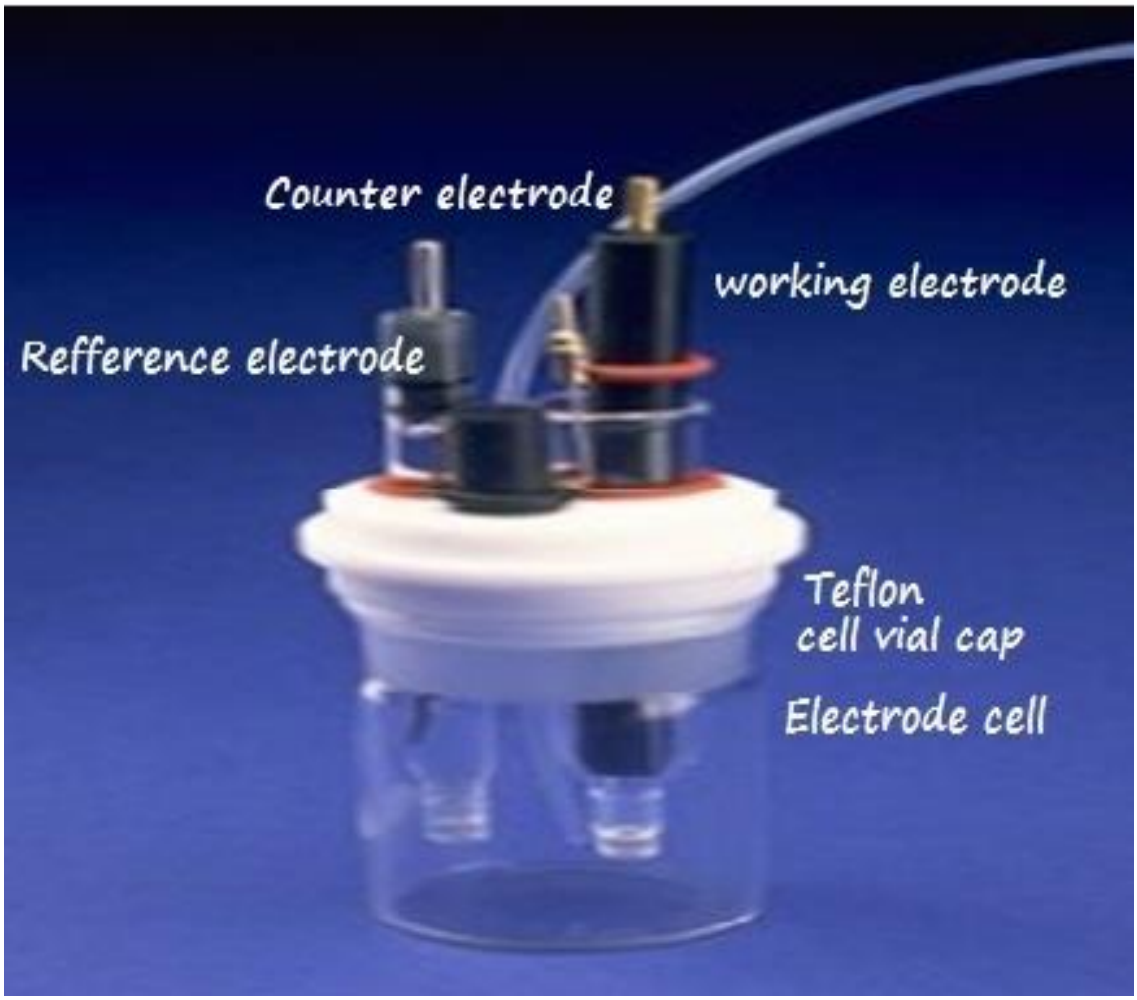


Figure 8. Three Electrodes system

2.5.1. Cyclic Voltammetry

Cyclic voltammetry is the most effective and versatile electro analytical technique for the study of electro active species. Its versatility is combined with ease of the measurement and has resulted in extensive use of cyclic voltammetry in the field of electrochemistry, inorganic chemistry, organic chemistry and biochemistry. The effectiveness of cyclic voltammetry results from its capability for rapid observing redox behavior over a wide potential range [48]. It is the most widely used technique for acquiring qualitative information about electrochemical reactions but rarely used for quantitative determinations, and it is widely used for the study of redox processes, for understanding reaction intermediates, and for obtaining stability of reaction products [48].

The power of cyclic voltammetry results from its ability to rapidly provide considerable information on the thermodynamics processes and the kinetics of heterogeneous electron transfer reactions and also on coupled chemical reactions or adsorption processes. It is often the first experimental technique to be performed. In particular, it offers a rapid location of redox potentials of the electro active species, and convenient evaluation of the effect of media on the redox process. Depending on the information sought, single or multiple cycles can be used. During the potential sweep, the potentiostat measures the current resulting from the applied potential. The resulting current-potential plot is termed cyclic voltammogram [46]. Cyclic voltammetry consists of cycling the potential of an electrode, which is immersed in unstirred solution and measuring the resulting current. The potential of the working electrode is controlled verses a reference electrode such as saturated calomel electrode (SCE) ($Hg/Hg_2Cl_2/Cl$) or Ag/AgCl/Cl.

The controlling potential that is applied across these two electrodes can be considered an excitation signal. The excitation signal for cyclic voltammetry is linear potential scan with a triangular wave form as shown in figure.9. This triangular potential excitation signal sweeps the potential of the electrode between two potentials sometimes called the switching potential [21, 22].

In Cyclic voltammetry, the direction of the potential is reversed at the end of the first scan. Thus, the waveform is usually of the form of an isosceles triangle with the advantage that the product of the electron transfer reaction that occurred in the forward scan can be probed again in the reverse scan. It is Powerful tool for the determination of formal redox potentials, detection of chemical reactions that precede or follow the electrochemical reaction and evaluation of electron transfer kinetics. Different voltammetric techniques are distinguished primarily by the potential function that is applied to the working electrode and by the material used as the working electrode [50].

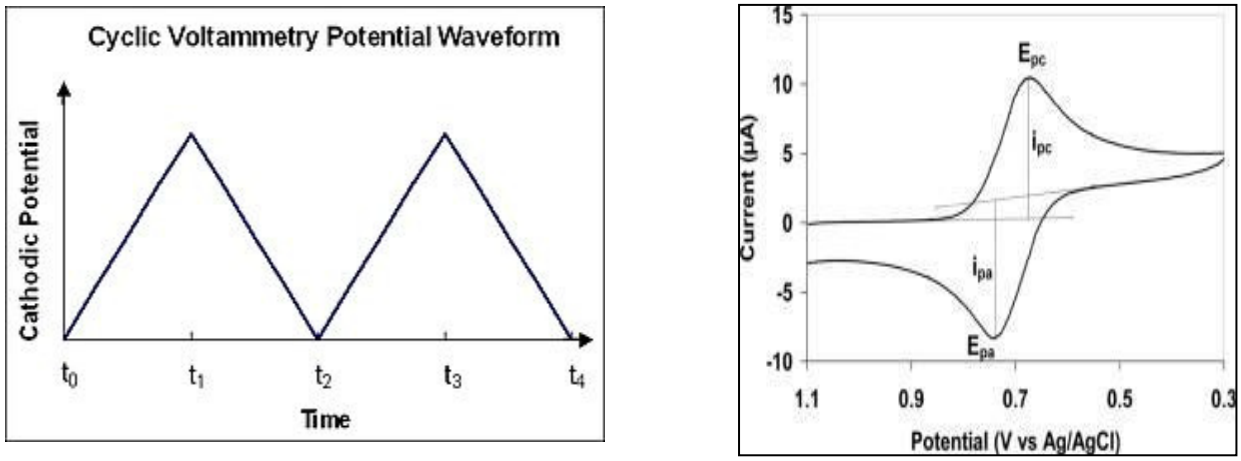


Figure 9. (a) Excitation wave form of cyclic voltammetry (b) response obtained for reversible cyclic voltammetry

A cyclic voltammogram is obtained by measuring the current at the working electrode during the potential scan. The current measured can be considered as the response signal to the potential excitation signal. The voltammogram is a display of current (vertical axis) verses potential (horizontal axis). Because the potential varies linearly with time, the horizontal axis can also be thought as time axis [45, 50]. Cyclic voltammetry has become an important and widely used electro analytical technique in many areas of chemistry. It is rarely used for quantitative determinations, but it is widely used for the study of redox process, for understanding reaction intermediate, and obtaining stability of reaction products [43, 48].

The important parameter of a cyclic voltammogram are the magnitude of the anodic peak current (I_{pa}), Cathodic peak current (I_{pc}), anodic peak potential (E_{pa}) and Cathodic peak potential (E_{pc}). The formal reduction potential E^o, for reversible couple is centered between E_{pa} and E_{pc} [45, 47].

$$E^o = \frac{Epa + Epc}{2} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (1)$$

The number of electrons transferred in the electrode reaction (n) for reversible couple can determined from the separation between peak potentials (for reverse couple) is given by

$$\Delta E = E_{pa} - E_{\mathbf{pc}} = 0.059 / n \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots \quad (2)$$

Where n is the number of electrons transferred and E_{pa} and E_{pc} are the anodic and Cathodic peak potentials, respectively, in Volts. Thus for a reversible redox reaction at 250_c with n electrons E_p should be 0.0592/nV or about 60 mV for one electron. Randles-Sevcik equation is an equation that correlates the peak current (I_p) with concentration (C), $I_p = kC$, where k is a constant that includes different cell parameters such as transfer Coefficient, number of electrons involved in the reaction, electrode area, diffusion coefficient and scan rate [53].

2.5.2. General use of voltammetric method

- Quantitative determination of organic and inorganic compounds in aqueous and non-aqueous solutions
- Measurement of kinetic rates and constants
- Determination adsorption processes on surfaces
- Determination electron transfer and reaction mechanisms
- Determination of thermodynamic properties of solvated species
- Fundamental studies of oxidation and reduction processes in various media
- Determination of complexion and coordination values
- Quantitative determination of pharmaceutical compounds
- Determination of metal ion concentrations in water to subparts-per-billion levels
- Determination of redox potentials
- Detection of eluted analytes in high-performance liquid chromatography (HPLC) and flow injection analysis
- Determination of number of electrons in redox reactions
- Kinetic studies of reactions [20]

2.6. Pulse Voltammetric Techniques

Pulse voltammetric techniques are the difference in the rate of the decay of the charging and the faradic currents following a potential step. The charging current decays exponentially, whereas the faradic current (for a diffusion-controlled current) decays as a function of the square root of time that is, the rate of decay of the charging current is considerably faster than the decay of the faradic current[45]. The important parameters for pulse techniques are Pulse amplitude, Pulse width and Sample period's numbers of different pulse techniques are available on the epsilon, which differ in their potential pulse wave forms, the number of sampling points, and whether a solid electrode (voltammetry) is used. The discrimination against the charging current that is inherent in these techniques leads to lower detection limits The discrimination against the charging current that is inherent in these techniques leads to lower detection limits and higher Sensitivity, which makes these techniques suitable for quantitative analysis [46].

2.6.1. Square Wave Voltammetry

Square wave voltammetry is the most sophisticated technique in the family of pulse voltammetric techniques. Square wave voltammetry is a large-amplitude differential technique in which the wave form composed of a symmetric square wave, superimposed on a base staircase potential applied to the working electrode. The current is sampled twice during each square wave cycle, once at the end of the forward pulse and once at the end of the reverse pulse. Since the square wave modulation amplitude is very large, the reverse pulses cause the reverse reaction of the product [49]. Below are the potential versus time wave form of square wave system.

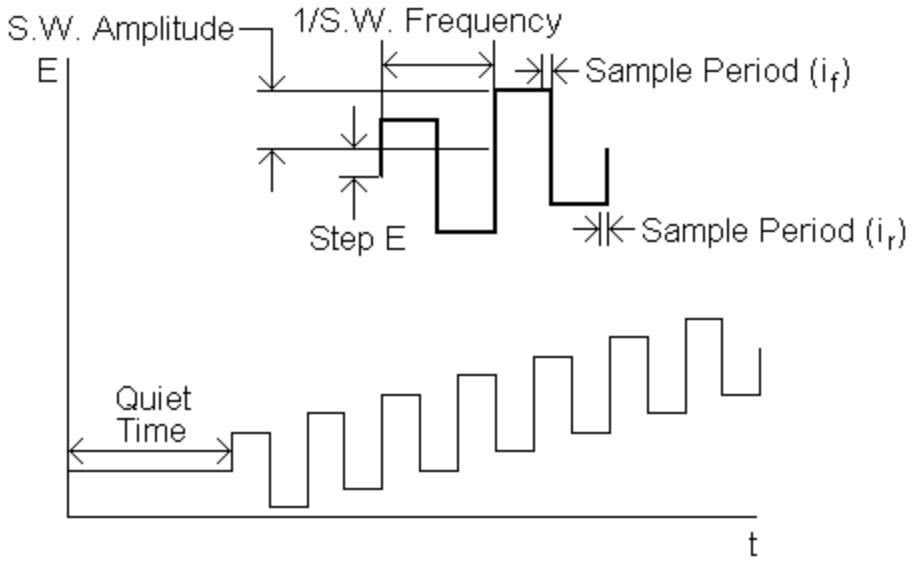


Figure 10. Applied potential wave form for square wave voltammetry.

In general, square wave voltammetry possesses excellent sensitivity. The sensitivity is higher than from differential polarographic techniques coupled to the effective discrimination against the charging background current, very low detection limit can be attained. The major advantageous of square wave voltammetry is its speed. The effective scan rate is given by fΔEs. The analysis time is very short, complete voltammogram can be recorded within a few seconds, as compared to 2-3 min in differential pulse voltammetry. An additional advantage of square wave voltammetry is that its fast scans capability to determine changes in voltammetric responses with time nondestructively [41, 42].

There are two advantages to measuring the difference current. First, it increases the discrimination against the charging current, since any residual charging current is subtracted out. Second, the shape of the current response is a symmetric peak (Fig.11). At potentials well negative of the redox potential, the current is diffusion-controlled, and the potential pulse has no effect; hence, the forward and reverse currents are equal, and the difference current is again zero. The largest difference between the forward and reverse currents (and hence the largest current response) is at the redox potential [49].

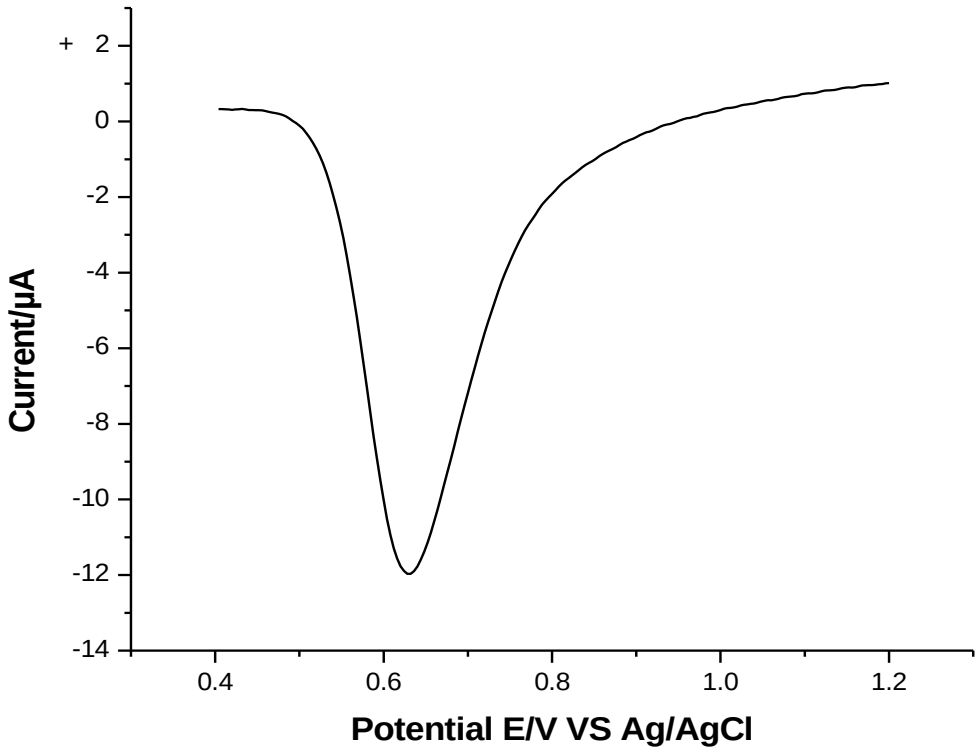


Figure 11. Square wave voltammogram (2,4,6-Trichlorophenol)

2.7. Electrodes

Electrochemical measurements initially involved three electrodes: the working electrode, Reference and counter electrode, the electrochemical reactions in the form of oxidation or reduction occurs at working electrode, and the second electrode ,counter electrode which was large enough not to be polarized during the measurement. The potential of the working electrode was thus calculated from the difference between the applied potential and the potential of the counter electrode [22]. However, distorted voltammogram were obtained when the resistance of the electrolyte was high. A third electrode was thus introduced whose potential remained constant throughout the entire measurement. This was known as the reference electrode. Generally the potential is applied between working electrode and reference electrode whereas the current is measured between working electrode and counter electrode [23].

2.7.1. Working electrodes

The condition of the surface of the working electrode can have a significant effect on the current response in voltammetry experiments. The working electrode is often used in conjunction with an auxiliary electrode and a reference electrode in a three electrode system. Depending on whether the reaction on the electrode is a reduction or an oxidation, the working electrode can be referred to as either Cathodic or anodic [22]. Working electrode is renewable which is easily pretreated and polished. The most common method for surface cleaning of glassy carbon electrode is mechanical polishing. Some example of polishing materials are diamond, alumina, silicon carbide There are different kind of working electrode those are Platinum electrode (Pt), Gold electrode(Au), Glassy carbon electrode (GCE), Silver electrode (Ag) and Carbon paste electrode (CPE) [20].

Glassy carbon electrodes (GCEs) has been widely used in the preparation of modified electrodes, Such electrodes offer the advantages of versatile bulk modification, fast response, renewability and a low background current high temperature resistance, hardness, low density, low electrical resistance, low friction, low thermal resistance, extreme resistance to chemical attack and impermeability to gases and liquids. Glassy carbon is widely used as an electrode material in electrochemistry [20].

The ease of fabrication and the control of modifiers loading (organic and inorganic molecules, enzymes, cofactors or mediators) have been valuable for electrochemical studies and analytical applications of modified glassy carbon electrodes.

Figure 12. Glassy carbon electrode

2.7.2. Reference electrodes

Reference electrodes are electrodes which have a stable and well known electrode potential and used to give a value of potential to which other potentials can be referred in terms of a potential difference, (as potentials can only be registered as differences with respect to a chosen reference value). They should provide a reversible half-reaction with Nernestain behavior, be constant over time and easy to assemble and maintain [21].

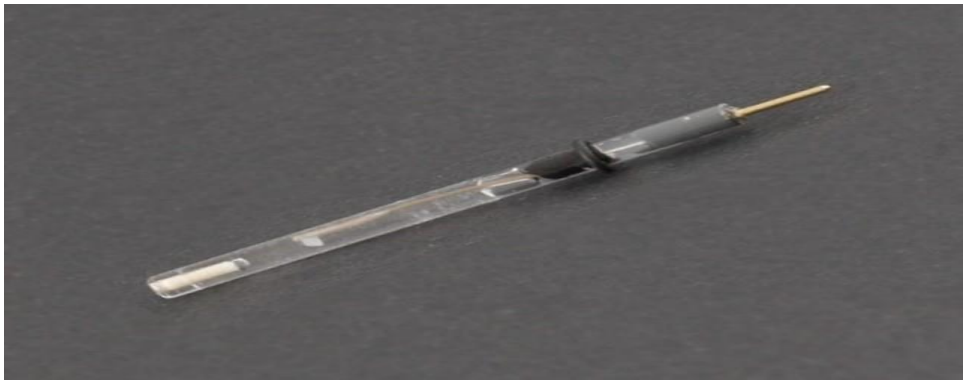


Figure 13. Reference electrode

2.7.3. Counter electrodes (Auxiliary electrodes)

The counter electrode must be inert in the conditions to which it is exposed in order to avoid Contamination of the solution [20]. Good examples of counter electrodes are platinum and graphite. The fundamental requirement is that to pass sufficient current into the solution without needing an excessive cell voltage or creating a non-uniform current distribution on the working electrode [23].

Figure 14. Counter electrode

3. Objectives of the Study

3.1. General Objective

To prepare modified GCE using SWCNT and PEDOT composite for the determination of 2,4,6-Trichlorophenol

3.2. Specific Objectives

- To modify a glassy carbon electrode using SWCNT and PEDOT composite for the determination of 2,4,6-Trichlorophenol using square wave voltammetric technique.
- To investigate the performance of SWCNT/PEDOT/GCE electrode for the determination of 2,4,6-Trichlorophenol compared to bare glassy carbon electrode.
- To optimize factors that have major effect on the sensitivity of the method such as square wave parameters (amplitude, frequency, step potential) as well as composition of the modifier and pH of working solution.
- To construct calibration curves for 2,4,6-Trichlorophenol at SWCNT/PEDOT/GCE using SWV technique and determine performance characteristics.

4. Experimental

4.1. Reagents

2,4,6-Trichlorophenol was purchased from Fluka (Busch, Switzerland), SWCNT was obtained from Saarchem Holparo Analytical Ltd, 3,4-ethylenedioxythiophene (EDOT) monomer, was purchased from Aldrich, di-potassium phosphate and di-hydrogen phosphate were purchased from (Fulka, Germany) and (Changshu Yangyuan chemicals, China), NaOH was purchased from (Labmerk chemicals, India) and HCl was purchased from (Riedel-De Haen, Germany). Stock solution 2,4,6-Trichlorophenol ($1x10^{-3}$ M) was prepared in 0.1 M phosphate buffer of PH 6. All experiments were carried out in phosphate buffer (0.1 M, pH 6). The stock solution of 2,4,6-Trichlorophenol prepared was stored in the refrigerator to avoid exposure to air and light. All solutions and subsequent dilutions were prepared using double de-ionized water. All chemicals were analytical grade and were used throughout this experiment without purification.

4.2. Instruments and Apparatus

The Voltammetric experiments were performed using CHI840C Electrochemical analyzer and BAS CV-50 W potentiostat/galvanostat analyzer which were turn interfaced to a personal computer with conventional three electrodes cell was used to measurements, with a PEDOT/SWCNT/GCE used as the working electrode, Ag/AgCl (3M KCl) as the reference electrode and a platinum wire serving as counter electrode and the pH of solution was measured using ion meter.

4.3. Preparation of SWCNT/PEDOT/GCE Modified Electrode

The preparation of new composite materials that have distinct properties which were not observed in the individual components was the objective of this work. The study led to the development of SWCNT/PEDOT modified GCE for the determination of 2,4,6-Trichlorophenol. The modified electrode was used as square wave detector in batch system with improved qualities such as simplicity of electrode preparation, wider linear range, low detection limit, and good stability. To the best of our understanding the modification and application of the SWCNT/PEDOT/GCE modified electrode. First of all GCE was cleaned by polishing it by alumina powder with 0.05 micron. Back ground CV was taken using 0.1 M phosphate buffer solution as supporting electrolyte, the bare GCE was polished with a 0.05μm alumina slurry and washed with double distil water and washed by using acetone to dried it then the electropolymerization of 0.1 M (EDOT) on the surface of GCE was carried out using cyclic voltammetry between -0.9 and 1.5 V for ten cycles. Ten cycles was chosen as the optimum number of cycles to decrease the memory effect due to adsorption of 2,4,6-Trichlorophenol on the PEDOT/GC surface electropolymerization of EDOT on surface of working electrode or glassy carbon electrode. The PEDOT film was allowed to grow on the Glassy carbon electrode surface for ten successive scans as can be seen from the increasing anodic and Cathodic peak current densities. The polymer film was washed with acetonitrile to stabilize radical cations and oligomers on the surface of GC and also to remove unreacted monomer. After drying 10 μL SWCNT (1mg SWCNT: 1mL DMF) were coated onto the PEDOT/ GCE and dried for an overnight to get the SWCNT/ PEDOT /GCE. In order to determine the optimum amount of polymer coating 5, 10, 15, 20, and 30 cycles of CV scans of PEDOT polymerization were carried out before coating with SWCNT dispersion. Finally 10 cycles of CV scans of PEDOT polymerization was selected as show in (Fig 15) , due to high peak current responses for 2,4,6-Trichlorophenol .

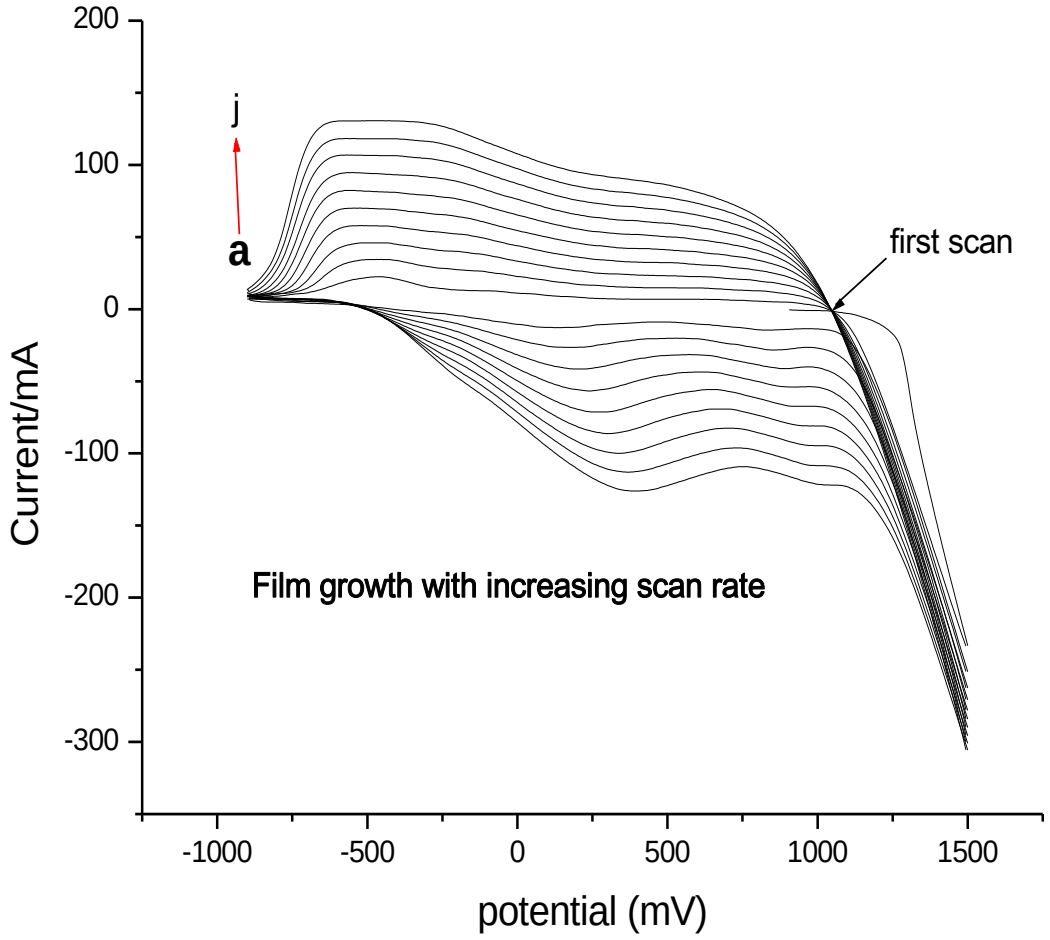


Figure 15. Repeating cyclic voltammograms of PEDOT film growth at a glassy carbon electrode in a solution of 0.1M Tetrabutylammonium per chlorate in acetonitrile containing 0.01M EDOT, at scan rate of 0.1 Vs^{-1} and potential range − 0.9 to 1.5V

5. Results and Discussion

5.1. Investigation of the Electrochemical Behavior of 2,4,6-Trichlorophenol at the Modified Glass Carbon Electrode by using cyclic voltammetry

The responses for 2,4,6-Trichlorophenol at bare glassy carbon electrode, at PEDOT modified electrode and at SWCNT/PEDOT/GCE using cyclic voltammetry are shown in Fig.16. The bare GCE gave no response current for 2,4,6-Trichlorophenol with no oxidative or reduction peaks. But the PEDOT modified electrode gave higher current responses, compared to the bare GCE, for 2,4,6-Trichlorophenol oxidation and reduction due to the electrocatalytic effect of the polymer. The oxidation and reduction peak currents for 2,4,6-Trichlorophenol increased markedly with successive runs at the PEDOT/GCE.

The excellent mechanical and electrical properties of carbon nanotubes help to transport electrons over long distances and this quality gives the advantage of fabricating polymer nano composites that have good mechanical and electrical properties [37]. The responses for 80 µM 2,4,6-Trichlorophenol obtained at SWCNT/PEDOT/GCE multilayer film are shown in Fig.16. The result indicates that the SWCNT/PEDOT increased the effective surface area of the modified electrode and the response becomes reproducible and constant after the ten potential cycles with E_{pa} = 0.612 V and higher peak currents were obtained compared to the PEDOT/GCE.

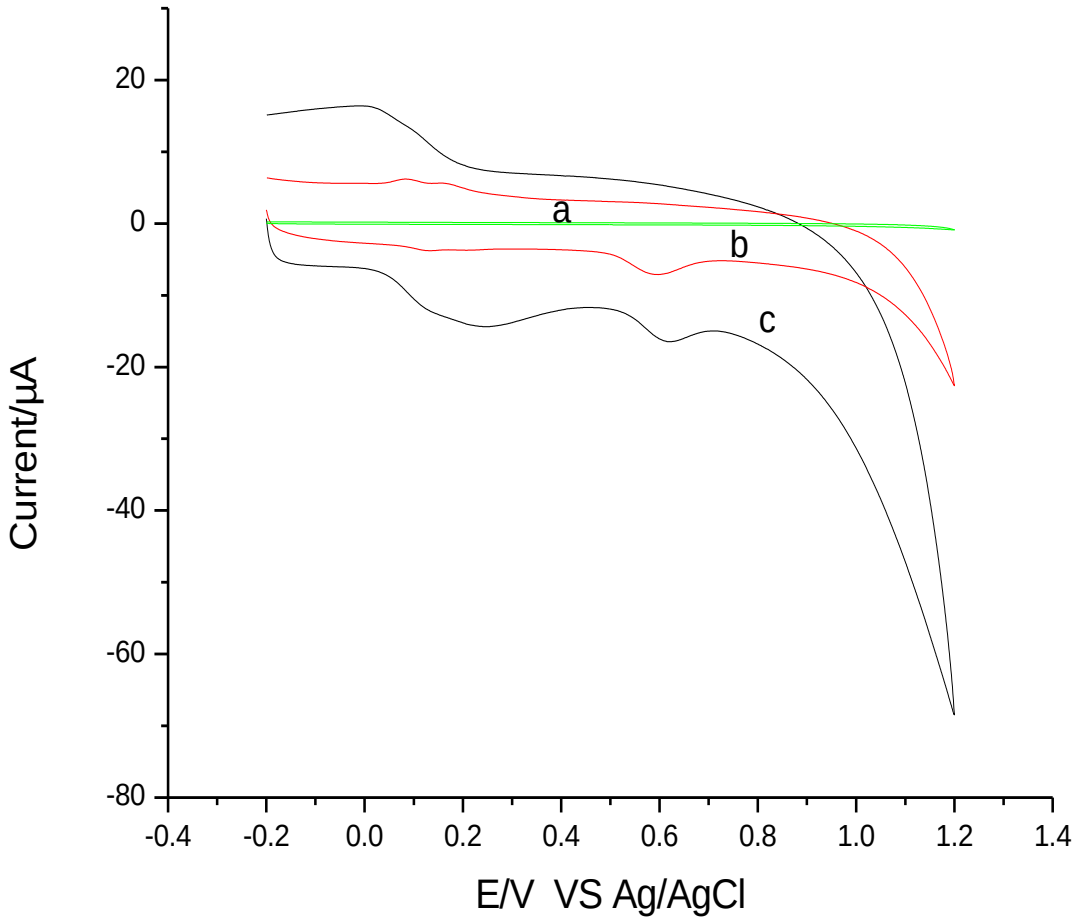


Figure 16. Cyclic voltammograms of 80 µM 2,4,6-Trichloropheno at bare GCE (a) at PEDOT Modified GCE (b) and at SWCNT/PEDOT/GCE (c), 0.1 M phosphate buffer pH 6 Recorded at scan rate of 50 mVs^{-1}

5.2. Investigation of the Electrochemical Behavior of 2,4,6-Trichlorophenol at the Modified Glass Carbon Electrode by using square wave voltammetry

The responses for 2,4,6-Trichlorophenol at bare glassy carbon electrode, at PEDOT modified electrode and at SWCNT/PEDOT/GCE using square wave voltammetry were recorded in 0.1M phosphate buffer solution. The voltammograms for 2,4,6-Trichlorophenol at the SWCNT/ PEDOT modified GCE using square wave voltammetry showed significantly increased current peaks, because square wave voltammetry possesses excellent sensitivity, and gives very low detection limit with high speed compare to other pulse voltammetric techniques. The SWCNT/PEDOT modified GCE highly improved the detection sensitivity for 2,4,6-Trichlorophenol as shown in Fig.17. The response for 2,4,6-Trichlorophenol at the unmodified GCE (curve a) is lower as compared to PEDOT modified electrode (curve b).The SWCNT/PEDOT modified GCE (curve c) gave a much higher current response and the peak potential also showed a shift to lower potential value . The magnitude of the signal (peak current) enhancement was calculated and was found 48% compared to the unmodified electrode.

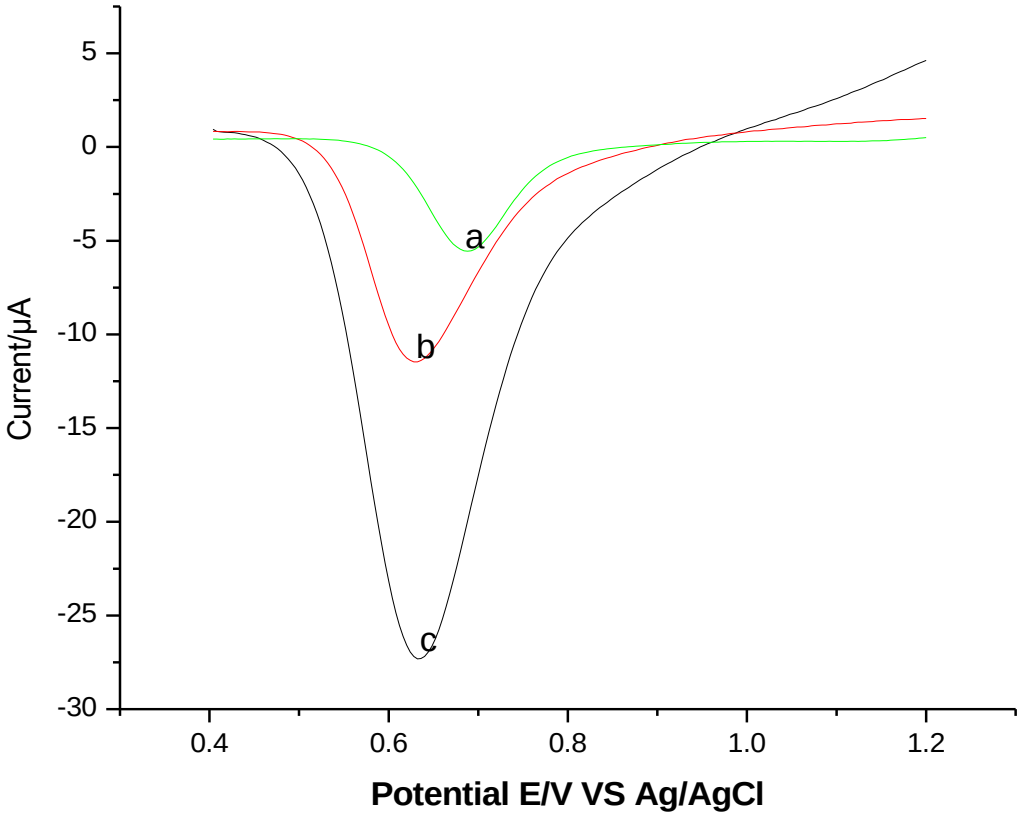


Figure 17. Square wave voltammograms of 80µM 2,4,6-Trichlorophenol, bare GCE at (curve a) PEDOT modified electrode at (curve b) and SWCNT/PEDOT/GCE at (curve c) in 0.1 M phosphate buffer

5.3. Effect of scan rate on the peak current of 2,4,6-Trichlorophenol at SWCNT/PEDOT modified GCE

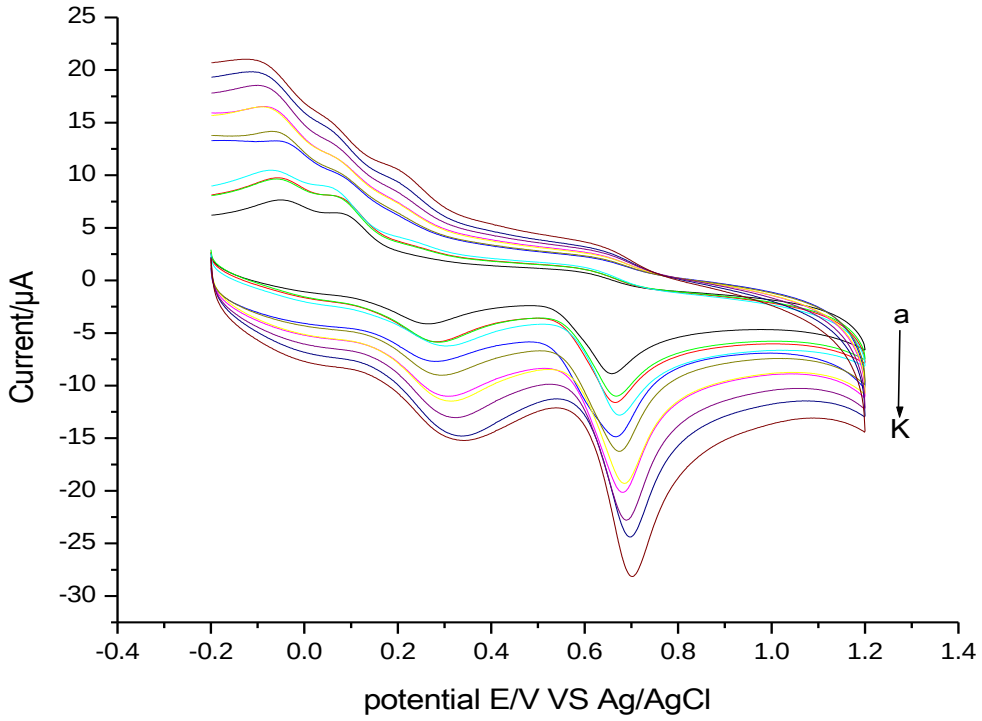


Figure 18. Cyclic voltammograms for 80 µM 2,4,6-Trichlorophenol in 0.1 M PBS solution pH 6 At SWCNT/PEDOT/GCE at scan rates from 10 up to 350 mVs^{-1}

Fig.18 shows the cyclic voltammograms for 80 µM of 2,4,6-Trichlorophenol of in 0.1M Phosphate buffer solution (pH =6.0) at SWCNT/PEDOT/GCE at different scan rates. From table 2, below it can be observed that the scan rate affects the position of oxidation peaks. Inspection of the curve shows increase in the scan rates increases the Cathodic peak current of 2,4,6-Trichlorophenol and shifts the Cathodic peak potential toward more negative potentials, whereas the anodic peak potential shifts toward more positive potentials. In addition the oxidation peak current for the oxidation of 2,4,6-Trichlorophenol exhibited a linear relation to the square root of scan rate, in the range from 10 to 350 mVs^{-1} . The relationship between the oxidation peak current and square root of the scan rate indicates that the oxidation of 2,4,6-Trichlorophenol at the composite modified electrode is a charge controlled process.

Table 2. The corresponding peak current and peak potential of different scan rate

scan rate (mvs^{-1})	$(mv/S)^{½}$	I_{pa}(A)	I_{pc}(A)	I_{pa}/I_{pc}	E_{pc}(V)	E_{pa}(V)	E_{pa}/E_{pc}
20	4.47	-3.860E-6	1.318E-6	2.928	0.047	0.635	13.51
50	7.071	-5.635E-6	2.971E-6	2.160	0.057	0.656	11.51
80	8.944	-7.048E-6	3.508E-6	1.860	0.073	0.665	9.11
100	10	-7.728E-6	4.164E-6	1.856	0.074	0.670	9.05
130	11.402	-8.638E-6	5.446E-6	1.586	0.075	0.671	8.94
150	12.247	-9.206E-6	5.837E-6	1.577	0.076	0.673	8.94
180	13.416	-9.839E-6	6.261E-6	1.571	0.078	0.681	8.73
200	14.14	-1.026E-5	6.613E-6	1.552	0.080	0.688	8.60
250	15.81	-1.133E-5	8.044E-6	1.408	0.082	0.689	8.40
300	17.32	-1.232E-5	9.171E-6	1.343	0.084	0.690	8.21
350	18.71	-1.303E-5	9.868E-6	1.320	0.093	0.700	7.52

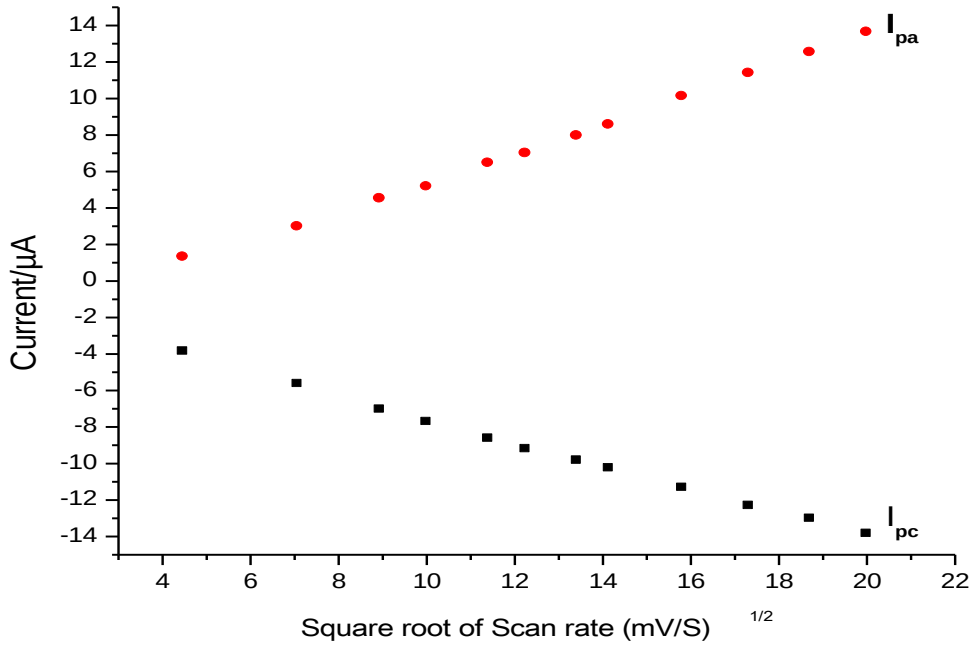


Figure 19. Effect of scan rate on the anodic and Cathodic peak current of 80 µM of 2,4,6-TCP 0.1 M phosphate buffer at a pH 6.0 at a scan rate from 10 up to 350 mV/s

5.4. pH effect

The pH value of the solution has a significant effect on the peak current and peak potential of catalytic oxidation and reduction of an analyte. The effect of pH on the electrochemical response of 2,4,6-Trichlorophenol was investigated using phosphate buffer of pH ranging from pH 5 to 9. The position and peak current for 80 μM of 2,4,6-Trichlorophenol in the investigated pH range is illustrated in Fig.20. With increasing pH 6 the peak potential shifted to more negative potentials showing the electrocatalytic effect of the modified electrode. The peak current was found to increase from pH 5 to 6 with a maximum at pH 6 and then decreased progressively as the pH increased. Therefore pH 6 was chosen for further analysis of 2,4,6-Trichlorophenol.

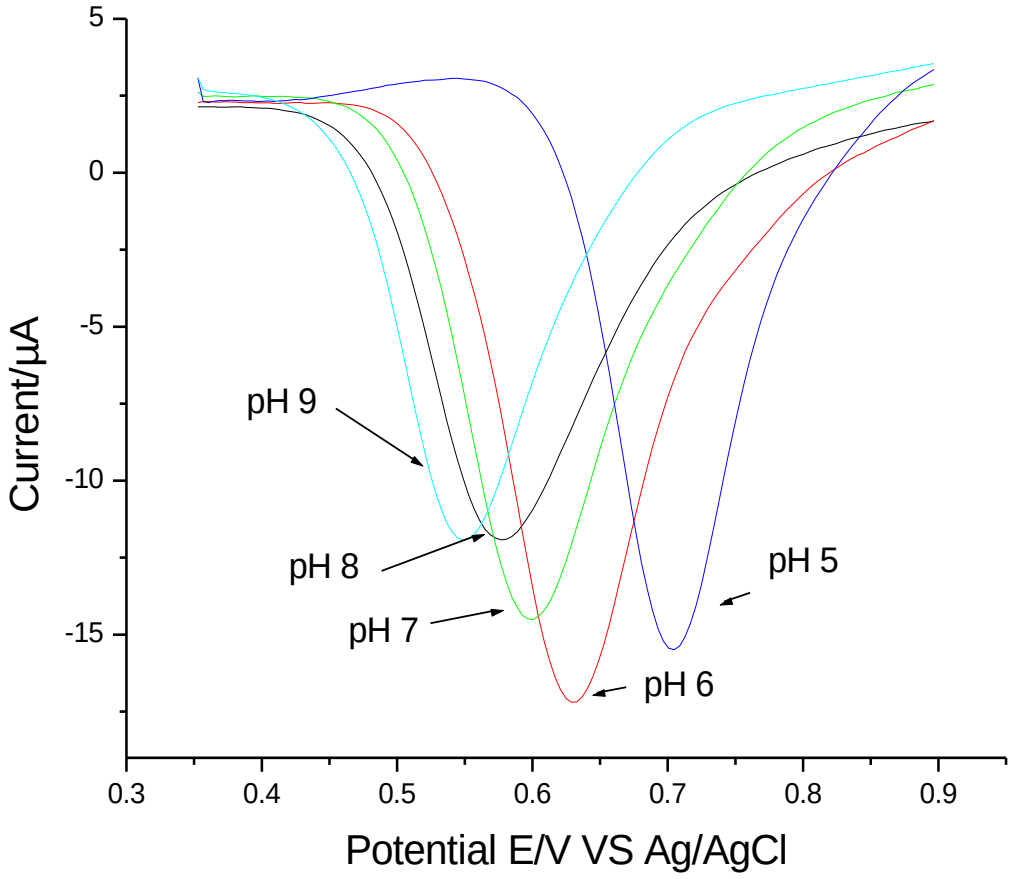


Figure 20. Square wave voltammograms showing the effect of pH at SWCNT/PEDOT/ GCE for 80μM 2,4,6-Trichlorophenol.

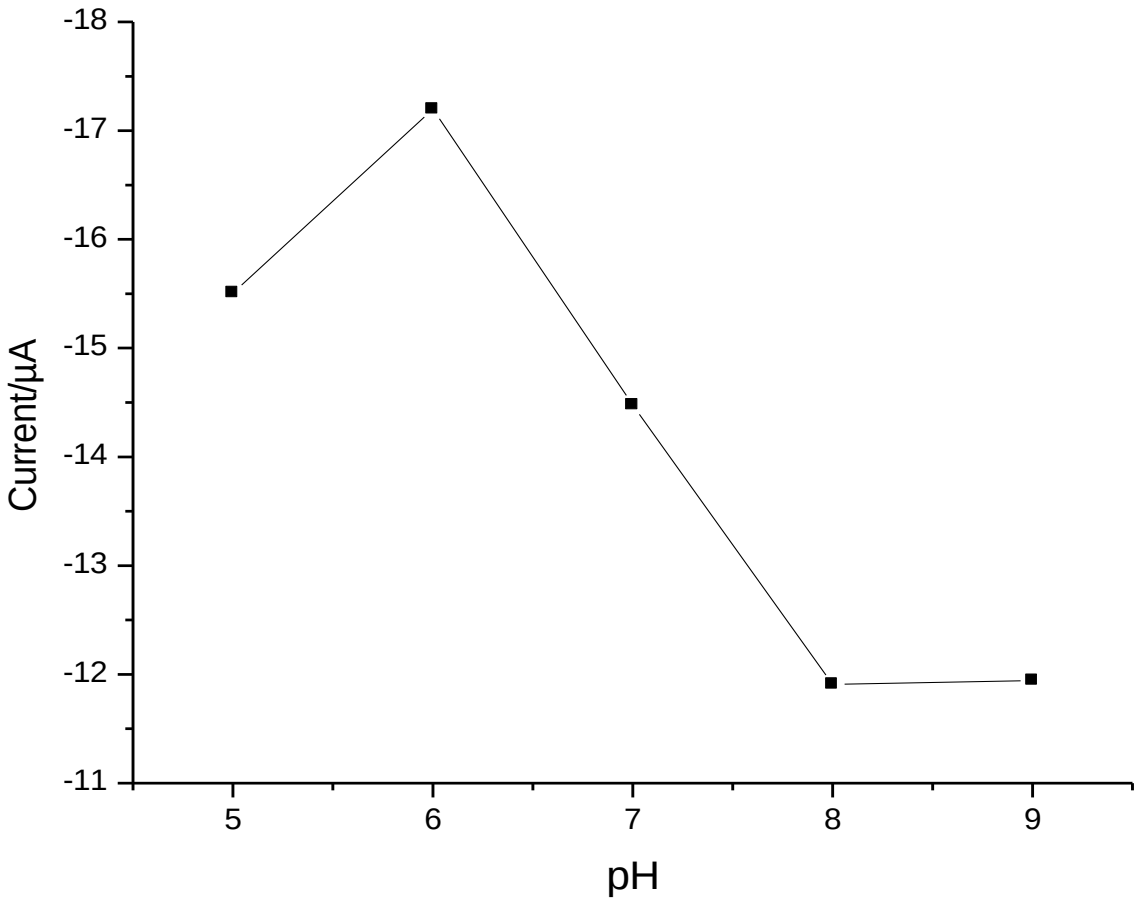


Figure 21. Current response showing the effect of pH at SWCNT/PEDOT/ modified GCE for 80 μM 2,4,6-Trichlorophenol.

The number electrons involved in electrochemical reaction of 2,4,6-Trichlorophenol was found from separation between the two peak potentials (ΔE) of 2,4,6-Trichlorophenol at SWCNT/PEDOT/GC. Since the separation between the two peak potentials was found to be 30 mV the reaction is irreversible and two electrons are involved in the reaction [21]. The peak potential also showed a shift to lower potential value with increasing the pH of the supporting electrolyte from pH 5.0 to pH 9.0 with a regression equation and correlation coefficient of E_{pa} (mV) = 86.76 – 36.7pH and R^2 = 0.972 respectively indicating the involvement of electrons in the oxidation of 2,4,6-Trichlorophenol. The slope of the equation (-36.67) mV indicates that equal number of electrons and protons are involved in the oxidation of 2,4,6-Trichlorophenol.

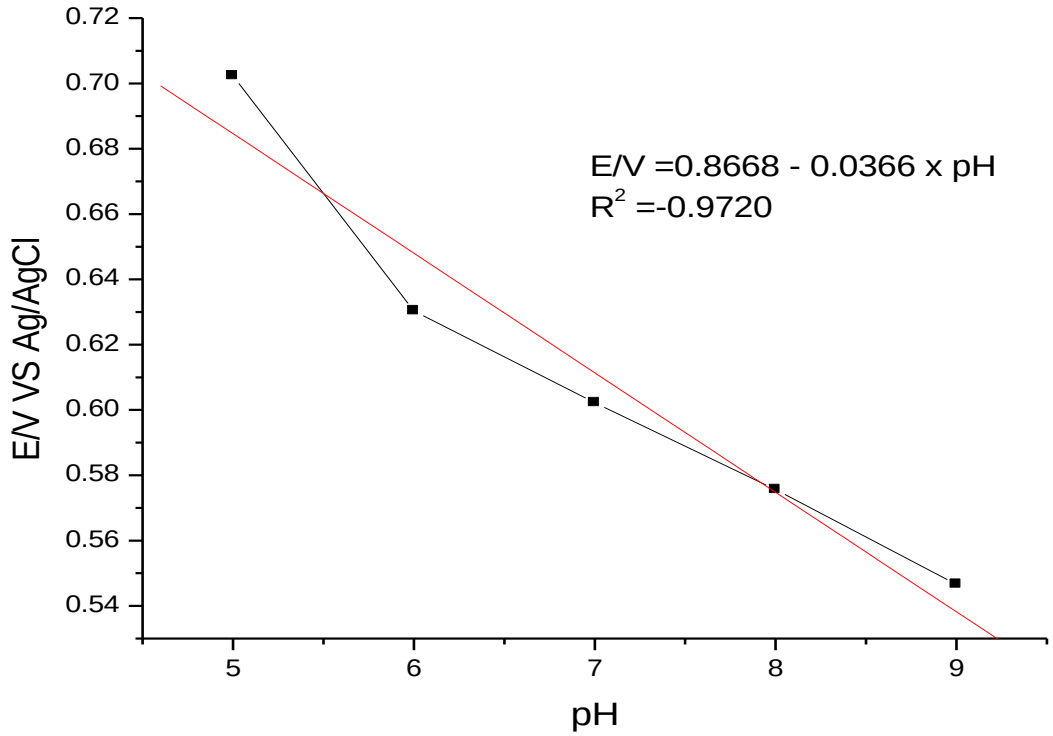


Figure 22. Potential (V) response showing the effect of pH at SWCNT/PEDOT modified GCE for 80 μM 2,4,6-Trichlorophenol.

5.5. Optimization of Square Wave Parameters for determination 2,4,6-Trichlorophenol

5.5.1. Effect of Square Wave Frequency

As it is shown in Fig 23, it was found that the 2,4,6-Trichlorophenol signal increased as the frequency increased from 10 Hz to 75 Hz. The results showed that as the frequency was increased from 10 to 60 Hz increase in the peak current was observed. However as the frequency was further increased beyond 60 Hz the peak current significantly increased.

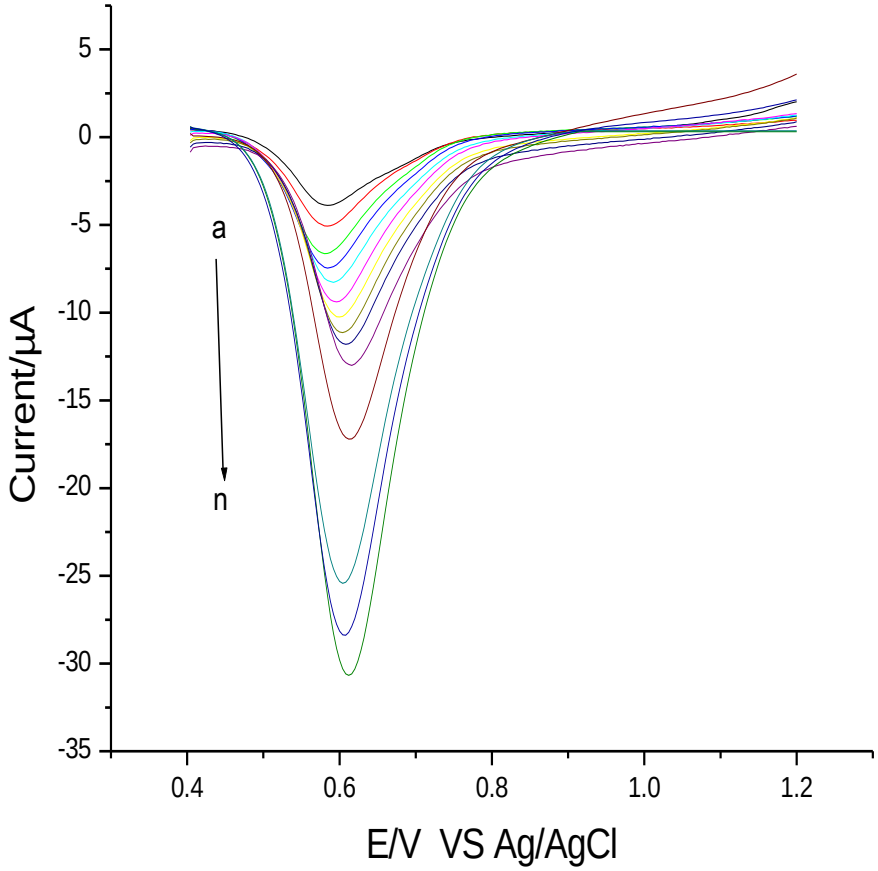


Figure 23. Square wave voltammograms showing the effect of frequency on the responses for 2,4,6-Trichlorophenol for 80 µM at SWCNT/PEDOT /GCE a) 10 Hz b) 15 Hz c) 20 Hz d) 25 Hz e) 30 Hz f) 35 Hz g) 40Hz h) 45 Hz i) 50 Hz j) 55 Hz k) 60 Hz l) 65 Hz m) 70 Hz and n) 75 Hz. Amplitude of 50 mV and step potential set at 4 mV, 0.1 M phosphate buffer pH 6

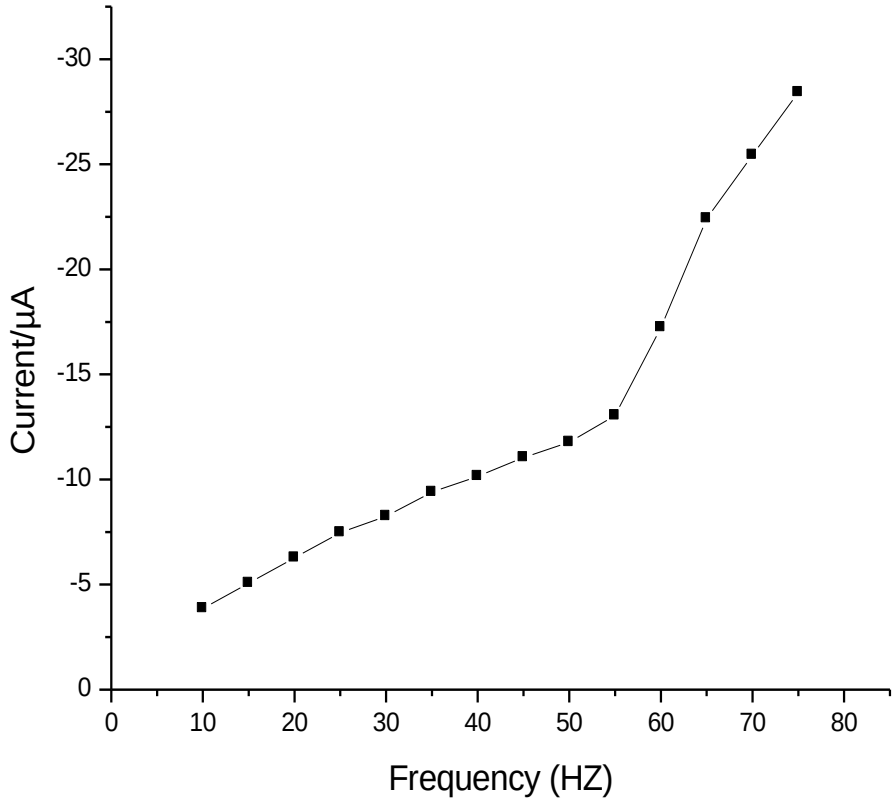


Figure 24. Current responses showing the effect of frequency for 2,4,6-Trichlorophenol 80 µM at SWCNT/PEDOT/ GCE. 0.1 M phosphate buffer pH 6.

5.5.2. Effect of Square Wave Amplitude

The effect of square wave amplitude on the current response was studied by varying the square wave amplitude from 10 to 80 mV at the frequency of 60 Hz .The result showed that as the amplitude was increased from 10 to 50 mV consequently the increase in peak current was observed. However as the amplitude was further increased from 50 mV the peak current slightly increased as it is shown in (Fig.25). Hence square wave amplitude of 50 mV was selected for further experiments for the determination of 2,4,6-Trichlorophenol.

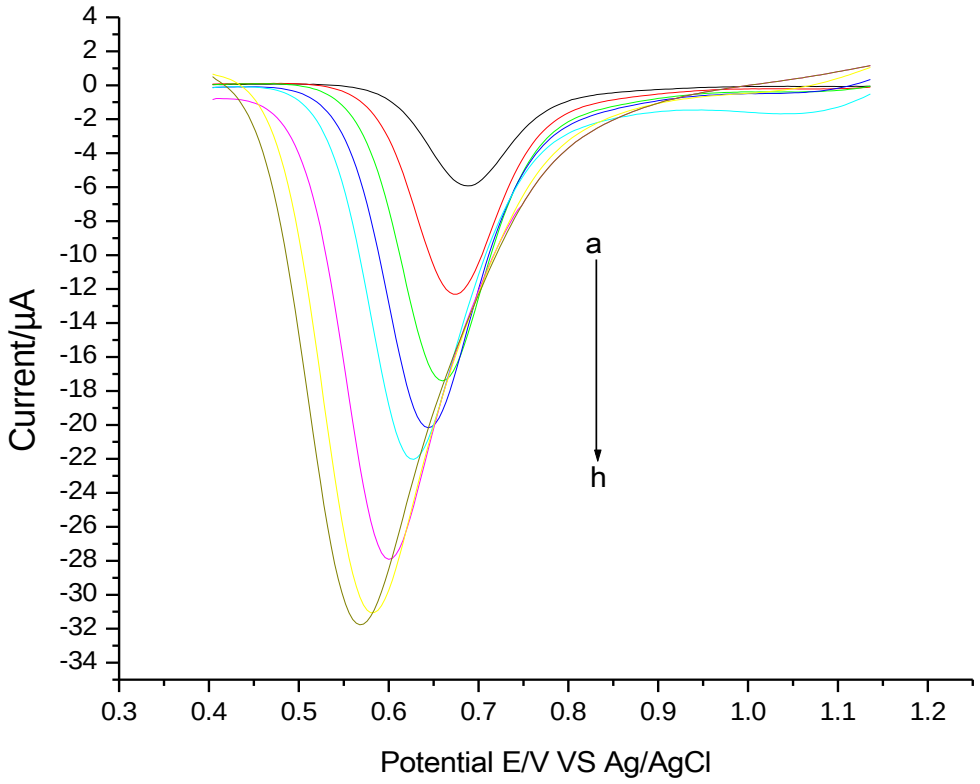


Figure 25. Square wave voltammograms showing the effect of amplitude on the responses for 2,4,6-Trichlorophenol 80 µM at SWCNT/PEDOT/GCE a) 10 mV, b) 20 mV c) 30 mV d) 40 mV e) 50 mV f) 60 mV g) 70 mV h) 80 mV. Frequency 60 Hz and step potential 4 mV, 0.1 M phosphate buffer pH 6

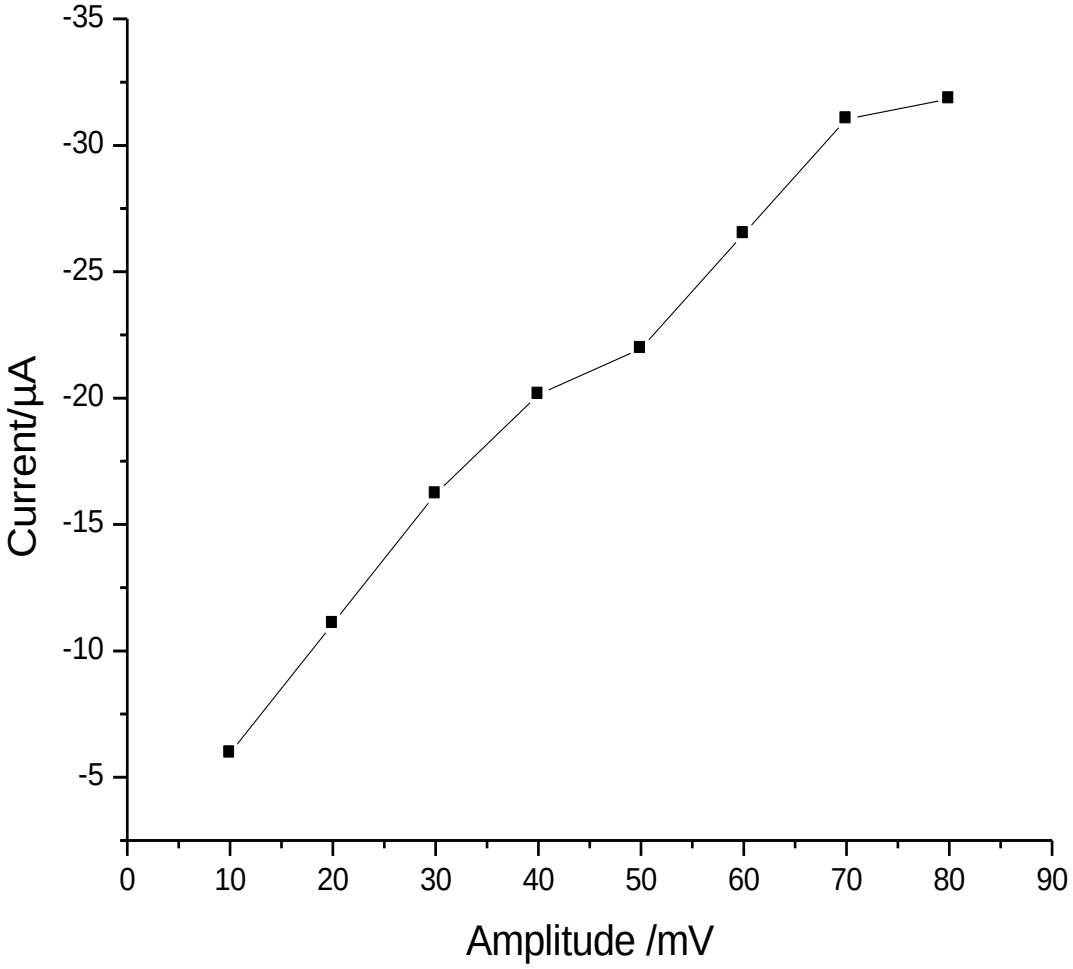


Figure 26. Current response showing the effect of square wave amplitude at SWCNT/PEDOT modified electrode for 80 µM 2,4,6-Trichlorophenol, Frequency 60 Hz, step potential 4 mV, 0.1 M phosphate buffer pH 6

5.5.3. Effect of Square Wave Step Potential

As it is indicated in (Fig.27), with increase in the step potential a linear increase in peak current was observed up to 4 mV then after the peak current declined slightly. Hence step potential of 4 mV was chosen as optimum condition for the subsequent experiments.

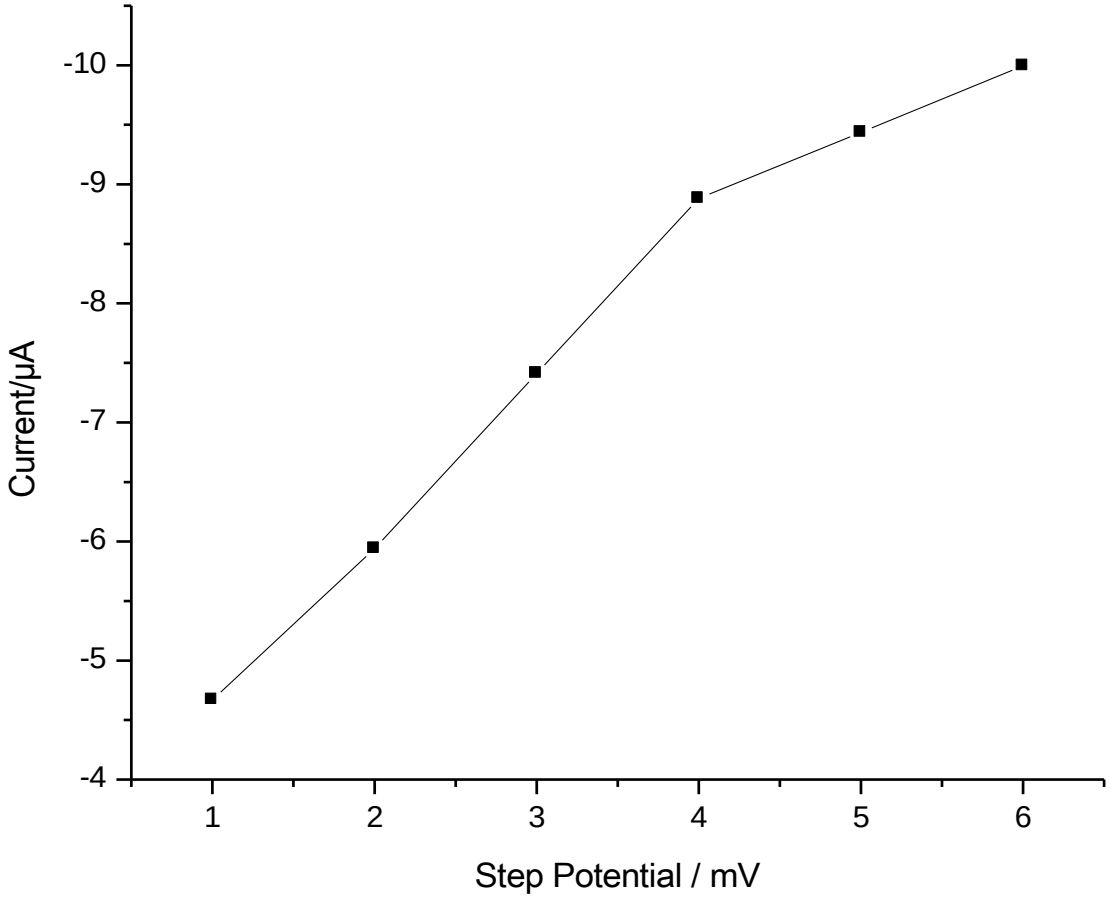


Figure 27. Peak current response showing the effect of square wave step potential at SWCNT/PEDOT/ GCE for 80 µM 2,4,6-Trichlorophenol, Frequency 60 Hz, Amplitude of 50 mV, 0.1 M phosphate buffer pH 6

5.6. Calibration Plots for 2,4,6-Trichlorophenol

Using the optimum square wave parameters described the calibration curve for of 2,4,6-Trichlorophenol was constructed. The peak height for 2,4,6-Trichlorophenol was found to increase linearly with increasing concentration of 2,4,6-Trichlorophenol from 6 to 80 $\mu molL^{-1}$ as it is shown in (Fig. 28). The calibration curve for seven average data points (n = 7) was found to be linear with R^2 = 0.999 with the regression equation of $Y = A + B*C$. Where A = 3.235 and B = 0.127. In analytical practice, calibration graphs frequently give numerical correlation coefficient values greater than 0.99 and if correlation coefficient values less than about 0.90 are relatively uncommon [53]. As the results indicated from the numerical value of correlation coefficient for this experiment, the data sets showed a good linear fit. The detection limit for 2,4,6-Trichlorophenol, considering signal-to-noise ratio of three based on below equation was found to be $7.5x10^{-8}$ mol/L.

$$LOD = \frac{3SDb}{m} \quad \ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots\ldots (3)$$

m - Slope of the regression line

SD_b - standard deviation of blank sample

.

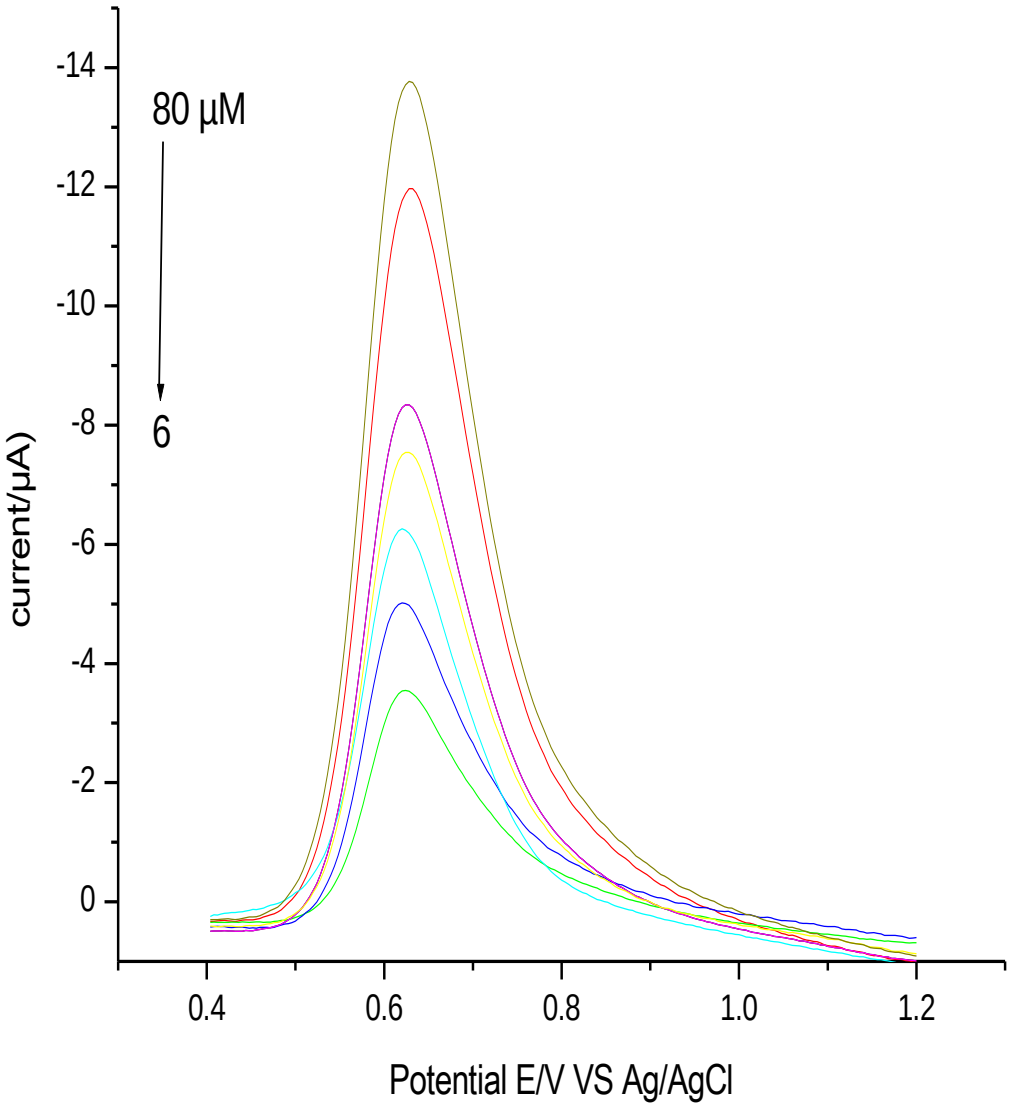


Figure 28. Square wave voltammograms for different concentration of 2,4,6- Trichlorophenol at SWCNT/PEDOT/GCE . Frequency 60 Hz, amplitude 50 mV and step potential 4 mV and 0.1 M phosphate buffer pH 6

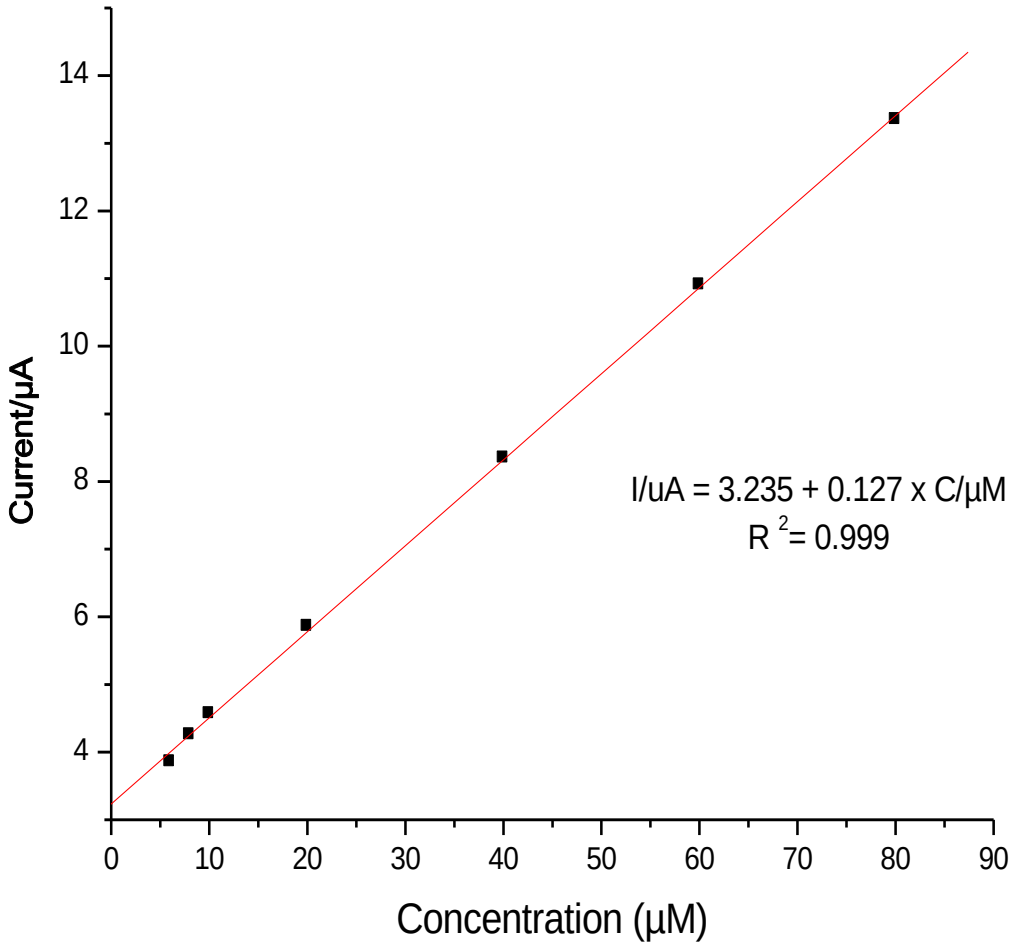


Figure 29. Calibration plot for different concentration of 2,4,6-Trichlorophenol at SWCNT /PEDOT /GCE Frequency 60 Hz, amplitude 50 mV and step potential 4 mV 0.1 M phosphate buffer pH 6.

5.7. Repeatability and Reproducibility of the Modified Glassy Carbon Electrode

Repeatable and reproducible are method of validation, a replicate set of data produced at a particular time point by an operator working with a particular set of equipment in a given laboratory will verify repeatability [22].

In this experiment, reproducibility was investigated by considering three modified electrodes prepared independently by taking triplicate measurements using the three electrodes. The reproducibility expressed in relative standard deviation was found to be 3.65 % for 80 μM 2,4,6-Trichlorophenol solution showing excellent reproducibility of the method. To study the repeatability of SWCNT/PEDOT/GCE, ten successive determinations of 80 μM standard 2,4,6-Trichlorophenol was made.

Table 3: Comparison of analytical performance of the calibration curves for 2,4,6-Trichlorophenol at SWCNT/PEDOT/GCE with different modified electrodes.

Detector/sensor	Linear range(μM)	LOD (μM)	R^2	Reference
Enzymeless GCE using preoxidation	0.4–750	0.04	0.997	[46]
SWCNT/GCT using flow injection	0.2 -100	0.025	0.995	[16]
SWCNT/PEDOT/GCE	6 - 80	0.075	0.999	This work

5.8. Analytical applications

The analytical application of the modified glassy carbon electrode was tested with in the linear range based on calibration graph, I/µA = 3.235 + 0.127 x C/µM. In order to assess the possible applications of the proposed method for the determination of 2,4,6-Trichlorophenol local tap water was used. The determination of 2,4,6-Trichlorophenol in the samples was carried out using square wave voltammetry in batch system at the SWCNT/PEDOT/GCE in 0.1M phosphate buffer solution as supporting electrolyte (pH 6.0). The results are given below in table 4. When known amounts of 2,4,6-Trichlorophenol were added to the tap water control samples quantitative recoveries of 99.50 % - 107.40 % were obtained. The feasible application of the SWCNT/ PEDOT/GCE for the determination of 2,4,6-Trichlorophenol is evident from their results.

Table 4: Recovery study

Sample No.	Added (μM)	Found (μM)	Recoveries (%)
1	6	5.97	99.50
2	8	8.46	105.75
3	10	10.74	107.40

5.9. Interference Study

Possible interference of phenol in the detection of 2,4,6-Trichlorophenol at SWCNT/PEDOT /GCE (0.1M phosphate buffer solution, pH 6) were investigated. The results obtained when 60 µM phenol was added to 60 µM 2,4,6-Trichlorophenol (1:1) and square wave voltammograms were recorded between 0.2 and 1.2V, amplitude 50 mV, frequency 60 Hz and step potential 4 mV are shown in (Fig. 28). The addition of phenol significantly decreased the peak current for 2,4,6-Trichlorophenol. Subsequent addition also showed further decrease in the peak currents. The addition of two fold 2-nitrophenol, 4-nitrophenol, and pentachlorophenol did not interfere with the determination of 2,4,6-Trichlorophenol. The result suggests, except for phenol that this method showed good selectivity towards the determination of 2,4,6-Trichlorophenol.

.

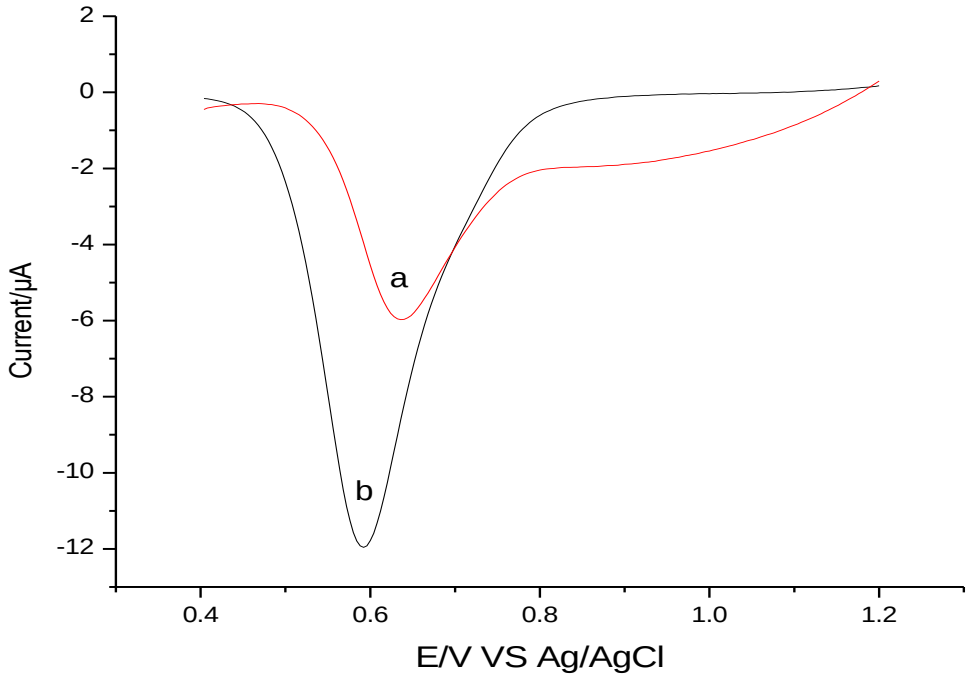


Figure 30. Square wave voltammograms for interference response using SWCNT/PEDOT modified electrode, (a) the solution of 60 µM of 2,4,6-Trichlorophenol and 60 µM Phenol (1:1 ratio) and (b) 60 µM 2,4,6-Trichlorophenol. Frequency 60 Hz, amplitude 50 mV and step potential 4 mV, 0.1 M phosphate buffer pH 6

6. Conclusion

Glassy carbon electrodes were modified with SWCNT PEDOT composites for the analysis of 2,4,6-Trichlorophenol. The modified electrodes showed excellent electrocatalytic property. Compared to the bare GCE. The SWCNT/ PEDOT/GC gave much higher current responses and shift of potential to more negative potentials. The modified electrodes were simple, fast and reproducible. The responses of the modified electrode were optimized for the square wave voltammetric determination of 2,4,6-Trichlorophenol in batch system. The results demonstrate the modified GCE exhibit a good analytical performance for the detection of 2,4,6-Trichlorophenol in the presence of other interfering substances.

7. Reference

[1]. Agency for Toxic substance and disease registration 1999.

[2]. C. Saby and J. H. T. Luong. Electroanalysis 10, 2010, 7–11.

[3]. C. Comninellis and A. Nerini. J. Appl. Electrochem. 25, 1995, 23–28

[4]. D. Pletcher and A. Alvarez-Gallegos. Electrochim. Acta 44, 1998, 853–861.

[5]. A. Alvarez-Gallegos and D. Pletcher. Electrochim. Acta 44, (1999), 2483–2492.

[6]. M. H. Elias, S. Anuar, K. Noorhana.Y. Ekramul. M. JASA. 2, 2007, 63.

[7]. Vilchez, J.L.; Blanc, R.Avidad.R.Navalon, A.J.Pharm.Biomed. Anal. 13, 2006, 1119-1135.

[8]. McEvoy, E.; Donegan. S.; Power. J. Altria, K. J. Pharm.Biomed. Anal. 2007, 44, 137.

[9]. B.J.; Mohammend. S.; waltman. Res, R. J. J. Pharm. Biomed. Anal. 27, 1983, 330.

[10]. G. Tourbillon, Garinier, F. J. Electro. Anal. Chem. 51, 1984, 161.

[11]. National Toxicology Program, Department of Health and Human Services, Report on Carcinogens, 12th edition, 2011 .

[12]. US Environmental Protection Agency, Semi volatile organics by Gas chromatography/mass Spectrometry (GC/MS), U.S. EPA Method 8270D, US Environmental Protection Agency, Which accessed from **http://www.epa.gov/sam/pdfs/EPA-8270d.pdf** .

[13]. Q.Z. Feng, L.X. Zhao, W. Yan, J.-M. J. Hazard. Matter. 2012, 167-282.

[14]. J.F. Peng, J.-F. Liu, X.-L. Hu and G.-B. Jiang, J. Chromatogr. A, 1139, 2007, 165.

[15]. J.Wang1" Enzymeless Flow Injection Analysis of 2,4,6-Trichlorophenol Based on Preoxidation by Ammonium Cerium (IV) Nitrate" $N_{0.}$ 7, 2012, 9113 – 9121.

[16]. N. Negash "Determination of Phenol and Chlorophenols at SWCNT/GCE Modified Glassy Carbon Electrode Using Flow Injection Amperometric", Article ID 926213, 2014, http://dx.doi.org/10.1155/2014/926213

[17]. M.A. Ghanem, *Electrochem. Comm., No. 9,* 2007**,** 2501-2506.

[18]. A. Shrivastava, V.B Gupta "Methods for the determination of limit of detection and limit of quantitation of the analytical Methods" Vol 2 Issue 1 ,2011, 21-25 which accessed from **http://www.cysonline.org/** on June 2,2014.

[19]. D. Vega, L. Ag¨u´ı, A. Gonz´alez-Cort´es, P. Y´a˜nez-Sede˜no, and J. M. Pingarr´on, "Electrochemical detection of phenolic estrogenic compounds at carbon nanotube-modified electrodes," *Talanta*, vol. 71, no. 3, 2007, 1031–1038

[20]. A.J. Bard, L.R. Faulkner, *Electrochemical Methods*: Fundamental and Applications, 2nd ed., John Wiley, New York, 2001, 218-220.

[21]. F.Scholz (Ed), Electro analytical Methods Guide to Experiments and applications Springer, 2005.

[22]. Douglas A.Skoog, Donald M. West, F. James Holler, Stanley R. Crouch Fundamentals of Analytical Chemistry, 8 edition, 2003.

[23]. J. Wang, *Analytical Electrochemistry*, 3rd ed. Wiley-VCH, 2006, 28-32.

[24]. Z. Spitalsky, D. Tasis, K. Papagelis, and C. Galiotis, "Carbon nanotube-polymer Composites: chemistry, processing, mechanical and electrical properties," Progress in Polymer Science, vol. 35, no. 3, 2010, 357–401.

[25]. M.A. Malinauskiene, J.; Ramanavicius, "A. Conducting polymer-based nano structured Material: electrochemical aspects. Nanotechnology", 16, 2005, 51-62.

[26]. Cosnier S. Biosensors based on electropolymerized films: new trends, Anal Bioanal Chem. 2003, 507-520.

[27]. Martin, cr. In Handbook of Conducting Polymers; Skotheim, ta; Elsenbaumer, rl; Reynolds, jr, Eds.; Dekker: New. York, 1998; 2nd ed, 409–421.

[28]. K.S.A. Chen, S.M. Electrocatalytic reduction of oxygen and hydrogen peroxide At poly (paminobenzene sulfonic acid) modified glassy carbon electrodes. Journal Of Molecular Catalysis A: Chemical, 278, 2007, 244-250.

[29]. M.A.; Garjonyt˙e, R.; Mazeikien˙e, R.; Jureviciute, I. Electrochemical response of ascorbic Acid at conducting and electrogenerated polymer modified electrodes for electroanalytical Applications: a review Talanta, 64, 2004, 121-129.

[30]. K.K.K. Diaz, A.F.;Geiss, R.H.; Gill, W.D.; Kwak, J.F.; Logan, J.A.; Rabolt, J.F.; Street, G.B. Organic metals: polypyrrole, a stable synthetic metallic polymer. J. Chem. Soc., Chem. Commun, 19, 1979, 854-855.

[31]. P.K.C. Polymers for chemical sensing, Materials Today, 8, 2005, 38-44

[32]. S. H.; Louis, E.J.; MacDiarmid, A.G.; Chiang, C.K.; Heeger, A.J." Synthesis of electrically Conducting organic polymers: halogen derivatives of polyacetylene", $(CH)_x$ J. Chem. Soc., Chem. Commun., 1977, 578-580.

[33]. I.A. "Analytical applications of conducting polymers, Electroanalysis". 3, 199 1247-254.

[34]. I.M.D.; John, R.; Riley, P.J.; Wallace, G.G. "The use of electropolymerization to produce new sensing surfaces: A review emphasizing electrode position of heteroaromatic Compounds, Electro analysis", 3, 1991, 879-889.

[35]. W. P.G.; Abrahamson, J. Carbon fiber layers on arc electrodes: Their properties and Cool-down behavior. Carbon, 16, 1997, 341-349.

[36]. B.R.H.; Zakhidov, A.; de Heer, W.A. Carbon Nanotubes the Route toward Applications Science, 297, 2002, 787-792.

[37]. W. J. Carbon-Nanotube Based Electrochemical Biosensors: A Review. Electroanalysis, 17, 2005, 7-14.

[38]. Xu, J.J.; Wang, J.L.; Chen, H.Y., electrochemically deposited nan composite of chitosan and carbon nanotubes for biosensor application, Chem. Commun., 2005, 2169-2171.

[39]. G. M.L.Chen, J.H.; Li, J.; Tao, B.; Yao, S.Z. Fabrication of polyaniline/carbon nanotube Composite modified electrode and its electrocatalytic property to the reduction of nitrite. Anal. Chim. Acta. 532, 2005, 71-77.

[40]. G.G. Wildgoose, G.G.; Banks, C.E.; Leventis, H.C.; Compton, R.G. Chemically Modified Carbon Nanotubes for Use in Electroanalysis, Microchim Acta., 152, 2006, 18-214.

[41]. R. Saito, M. Fujita, G. Dresselhaus, M.S. Dresselhaus, Appl. Phys. Letts. 60, 1992, 2204.

[42]. V. S.Vasantha and S.M. Chen, "Electrocatalysis and simultaneous detection of dopamine And ascorbic acid using poly (3,4-ethylenedioxy) thiophene film modified electrodes," Journal of Electroanalytical Chemistry, vol. 592, no. 1, 2006, 77–87.

[43]. L.B.Groenendaal, F.Jonas, D.Freitag, H.Pielartzik, and J. R. Reynolds, "Poly (3,4-ethylenedioxythiophene) and its derivative s: past, present, future," *Advanced Materials*,vol.12, 2000, 481–494 .

[44]. L. Zotti, G. Aubert, P.H. Waybright, S.M. and Reynolds, J.R, "Electrochemistry of Poly (3,4-alkylenedioxythiophene) Derivatives," Adv. Mater. Vol. 15, 2003, 855–8792 .

[45]. P. Proti, Introduction to Modern voltammetric and polarographic Analysis Techniques, Amel electrochemistry, 4th edition, New York, 2001.

[46]. S.P. Kounaves, Hand book of Instrumental techniques for analytical chemistry, John Willey and Sons, Inc., chapter 37, New York, 711.41

[47]. W.R. Lacourse, Pulsed electrochemical detection in HPLC, John Willey and Sons, Inc., New York, 199, 47-48.

[48]. http://wapedia.mobi/en/Voltammetry Electrodes; accessed on May 27, 2014**.**

[49]. L.L. Merit, J.A. Dean, F. Settle, and Willard, Instrumental methods of analysis, Litton Educational publishing, Inc., 6th edition, New York, 1981, 712-713.

[50]. P.T. Kissinger, and W.R. Heinemann, Laboratory techniques in electro analytical Chemistry, Marcel Dekker, Inc, New York, 2nd edition, 1996.

[51]. A. S. Arrobas, E. Bermejo, M. Chicharro et al., "Analytical applications of glassy carbon Electrodes modified with multi-wall carbon nanotubes dispersed in polyethylenimine as Detectors in flow systems," Analytical Chemical Acta, vol.596, no. 2, 2007, 183–194.

[52]. B. Allen J. Larry, R. Faulkner. Electrochemical Methods: Fundamentals and Applications (2nd ed.) 2000, 12-18.

[53]. Sharaf, M. A.Illman, D.L., Kowalski, B. R., Chemo metrics, Chemical Analysis Series Vol. 82: Wiley, New York, 1996.