ELECTROCHEMICAL SENSORS FOR SENSITIVE AND SPECIFIC DETECTION OF ORGANOPHOSPHATE, HEAVY METAL ION, AND NUTRIENT

ELECTROCHEMICAL SENSORS FOR SENSITIVE AND SPECIFIC DETECTION OF ORGANOPHOSPHATE, HEAVY METAL ION, AND NUTRIENT

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TITLE: Electrochemical sensors for sensitive and specific detection of organophosphate, heavy metal ion, and nutrient

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Lay Abstract

The growing population and rapid industrial development are affecting the water quality worldwide. The major water pollutants are organophosphates, heavy metal ions, and nutrients. These water pollutants are harmful, and their bioaccumulation poses a major health concern. In the USA alone, water quality issues are predicted to cost \$210 billion annually. Therefore, sensors to detect water pollutants are developed to monitor their environmental footprints. Electrochemical sensors are popularly used to detect water pollutants owing to their low-cost and high sensitivity.

The objective of this dissertation was to fabricate highly sensitive and specific electrochemical sensors to detect organophosphate (e.g., fenitrothion, FT), heavy metal ion (e.g., lead), and nutrient (e.g., nitrite). The sensors were fabricated with ink based on nanomaterials like carbon nanotubes and detecting agents like metal oxides. The fabricated sensors achieved very high sensitivity and specificity and can detect water pollutants in lake and tap water.

Abstract

In an electrochemical sensor, the sensing performance is mainly dependent on the mass transport of the analyte towards the working electrode-electrolyte interface and working electrode properties. Carbon nanomaterials like carbon nanotubes are widely employed to modify the working electrode properties for sensitive detection. A simulation model is formulated to investigate the effects of modifying a planar bare electrode with carbon nanotubes on electrochemical detection of fenitrothion (FT, an organophosphate). The model revealed that porous electrodes caused the change in mass transport regime and influenced FT's electrochemical response. The results aided in understanding the influence of the porous electrode on analyte detection and thus assisted in the fabrication of an ultrasensitive electrochemical sensor.

Simulation supported synthesis of a highly sensitive ink to produce highly porous and electrocatalytic electrodes. Activated carbon (AC) possesses high porosity and surface area, but they suffer from lower electrical conductivity. To enhance their conductivity, AC was co-doped with nitrogen and sulfur. Multiwalled carbon nanotubes were incorporated to further improve their porosity and electrocatalytic properties. The synthesized nitrogen-sulfur co-doped activated carbon coated multiwalled carbon nanotube (NS-AC-MWCNT) ink produced highly porous electrocatalytic electrodes. The sensor revealed a 4.9 nM limit of detection (LOD) under optimized conditions. However, it failed to overcome the enzymatic sensors' performances. The ultrasensitive performance was achieved by incorporating a detecting agent in the ink that instilled analyte capture ability. Metal oxides

like ZrO₂, MnO₂, and MgO possessed affinity towards organophosphate (fenitrothion), heavy-metal ion (lead), and nutrient (nitrite). Metal oxides were modified with 3,4dihydroxylbenzaldehyde (DHBA) – Chitosan (CHIT) to produce well dispersed and uniformly coated stable electrodes. The ZrO₂-DHBA-CHIT/NS-AC-MWCNT sensor achieved a remarkable limit of detection of 1.69 nM for FT. The sensor's performance exceeded the enzymatic-based sensors. The commonly found chemical interferents had negligible interference. The sensor produced reliable and satisfactory performance in lake and tap water. The MnO₂-DHBA-CHIT/NS-AC-MWCNT/GCE and MgO-DHBA-CHIT/NS-AC-MWCNT/GCE sensors produced an enormous improvement in the sensor performance compared to unmodified electrodes for lead and nitrite showed the importance of the methodology in providing a platform for a new class of metal oxide-based sensors.

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Declaration of Academic Achievements

This dissertation was used to fulfill the requirement of a Ph.D. degree. All the research was conducted from September 2018 to August 2022.

The thesis has resulted in three manuscripts published or submitted, and I am the first author on all of them. These papers are listed below:

- <u>Krishna Jangid</u>, Rohit Gupta, Rakesh P. Sahu, Igor Zhitomirsky, and Ishwar K. Puri.
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- <u>Krishna Jangid</u>, Rakesh P. Sahu, Richa Pandey, Ri Chen, Igor Zhitomirsky, and Ishwar K. Puri. "Multiwalled carbon nanotubes coated with nitrogen-sulfur co-doped activated carbon for detecting fenitrothion." *ACS Applied Nanomaterials* 2021, 4(5), 4781-4789.
- <u>Krishna Jangid</u>, Rakesh P. Sahu, Sadman Sakib, Igor Zhitomirsky, and Ishwar K.
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List of Abbreviations

CV	Cyclic voltammetry
SWV	Square wave voltammetry
DPV	Differential pulse voltammetry
EIS	Electrochemical impedance spectroscopy
FT	Fenitrothion
OP	Organophosphate
AChE	Acetylcholinesterase
DDT	Dichlorodiphenyltrichloroethane
WHO	World health organization
USA	United states of America
FAAS	Furnace atomic adsorption spectrometry
GF-AAS	Grapite furnace atomic absorption spectrometry
ICP	Inductively coupled plasma atomic emission spectrometry
ICP-MS	Inductively coupled plasma mass spectrometry
LC	Liquid chromatography
GC	Gas chromatography
GCE	Glassy carbon electrode
MWCNT	Multiwalled carbon nanotubes
LOD	Limit of detection
РРу	Polypyrrole

ECR	Eriochrome cyanine R
AC	Activated carbon
CHIT	Chitosan
DHBA	3,4 - dihydroxylbenzaldehyde
APS	Ammonium persulfate
ABS	Acetate buffer solution
C _{FT}	Concentration of fenitrothion
$c_{ m FT}^{red}$	Concentration of reduced fenitrothion
$c_{\rm FT}^{ox}$	Concentration of oxidized fenitrothion
c _b	Initial bulk concentration of fenitrothion
Α	Area of the electrode
R	Universal gas constant
Т	Absolute temperature
F	Faraday's constant
Ε	Imposed electrode potential
ϵ	Radius of macrodisc electrode
D	Diffusion coefficient
rds	Rate-determining step

1 Introduction

1.1 Background

The continuous growth in the global population has seen proportionate growth in urban development and industrialization.¹ This rapid growth in the global economy without proper management of natural resources has drastically impacted the water quality.² Water, an essential commodity, has been contaminated by numerous sources, from individuals to heavy industries. The major water contaminants are organophosphorus compounds, heavymetal ions, nutrients, and pathogenic bacteria.²⁻⁴ Organophosphorus compounds (OP) have been increasingly employed as pesticides in agricultural activities since the ban on toxic dichlorodiphenyltrichloroethane (DDT) in 1960.⁵ The rapid increase in the global population has driven food production via extensive consumption of OPs.⁶ However, being widely used and contributing to 70% of overall pesticide intake, OPs affect human life and the ecosystem.^{7, 8} A significant portion of the sprinkled OPs on the fields are unused, and their run-off causes water contamination in the nearby water ecosystems. Their increasing accumulation in the environment and exposure to life are growing concerns. They have been identified to cause mental disorders like depression and anxiety, affected heart rate, coma, and death. The world health organization (WHO) has reported that 750,000 people are affected yearly by pesticide intoxication, out of which 46.6% die.⁹ Moreover, in the United States of America (USA), pesticide intoxication costs approximately \$1billion vearly.¹⁰

Heavy-metal ions like lead, zinc, cadmium, mercury, etc., are extensively consumed in the production of batteries, photovoltaics, paints, agriculture, and chemical industries.¹¹ Heavy-metal ions are toxic and non-biodegradable and accumulate in the ecosystem in numerous ways. Significant sources include their discharge into water bodies from industrial waste, minerals or ores mining, and coal combustion.¹¹ Being highly toxic, even a tiny intake can be harmful enough to damage human organs.^{2,12} For example, besides its restricted application, lead discharge from PVC piping systems and battery industries¹¹ is still a concern as lead toxication can cause neurological disorders like Alzheimer's disease and reduce IQ.^{2,12} Other water pollutants include nutrients like nitrite, nitrate, and ammonia. They are widely used in the food and chemical industry.^{13,14} With the rapid utilization, especially in crop production, their surface run-off from overuse and mismanagement is gradually contaminating the water bodies. Especially nitrite, which is considered carcinogenic by WHO, poses a significant threat to human life as its continued exposure can cause cancer.^{13,14} Overall, in the USA alone, the problem of affected water quality is projected to cost \$210 billion annually.⁴ Therefore, precise determination of these water pollutants is essential to limit their footprints in the ecosystem. Consequently, there has been a rapid increase in the demand for the sensor to detect organophosphates, heavymetal ions, and nutrients.

2 Literature Review

2.1 Water pollutants detection techniques

Determination of water pollutants (organophosphates, heavy-metal ions, or nutrients) is commonly done by typical analytical techniques, including spectroscopy, mass spectrometry, or colorimetry.¹⁵⁻¹⁹ Following is a brief description of each detection technique.

2.1.1 Spectroscopic techniques

Spectroscopy-based water pollutants detection techniques include furnace atomic absorption spectrometry (FAAS), graphite furnace atomic absorption spectrometry (GF-AAS), and inductively coupled plasma atomic emission spectrometry (ICP), and inductively coupled plasma mass spectrometry (ICP-MS).^{20,21} All these techniques differ in operating procedures; however, they all are expensive owing to their costly instruments.

The FAAS technique is based on sample vaporization and dissociation into constituent atoms with a flame. Then the light of a particular frequency is passed, which is then absorbed by these atoms. The absorbed light is then detected with a detector, determining the sample's concentration. GF-AAS technique slightly differs from FAAS, as graphite tube is used instead of flame for sample vaporization. ICP technique utilizes a plasma source for sample vaporization and dissociation. This is followed by the atoms' excitation and emission of light, which are then detected to determine their concentration. ICP-MS differs from ICP in the later stage of detection, as instead of detecting the emitted light, mass spectrometry is used to detect the vaporized ions directly.²¹ Subsequently, the

sample's concentration is determined by their mass-charge ratio measurements. All the above-mentioned spectroscopic techniques have reasonable detection limits, however, they are costly and often require skilled professionals to operate in a dedicated laboratory.²² Hence, these techniques are not popularly employed for water pollutants detection.

2.1.2 Chromatographic techniques

Chromatography-based detection water pollutants techniques include liquid chromatography (LC) and gas chromatography (GC). Both methods involve separating components of the sample followed by their analytical detection. The process starts with the addition of the sample into a mobile phase which is passed through a stationary phase, and depending upon the sample's component properties and their interaction with these phases, they get separated. In GC, the mobile and stationary phases are gas and liquid, while for LC, it is solid and liquid. Hence, GC is also denoted by gas-liquid chromatography and LC as liquid-solid chromatography. GC is used to study volatile gaseous samples, while LC is used for solvent dissolvable samples. After the component separation of the sample, they are passed through a detector which determines their identity and concentration. Mass spectrometers are generally employed as the detector owing to their good detection limits and wide range of sample applicability. Besides their high sensitivity, both GC and LC are expensive and require sophisticated instrumentation and skilled professionals for complex sample preparations. Therefore, both GC and LC are not generally utilized for sensor fabrication to determine water pollutants.

2.1.3 Colorimetry and fluorescent techniques

The colorimetric or fluorescent detection technique is based on analyte-receptor binding and measuring their changes in color or fluorescence. A light detector measures the light intensity emitted by the fluorescent receptors specific to the analyte. After binding to the analyte, the light intensity of the color or fluorescent receptor changes, and thus analyte detection is achieved. Depending on the receptors used, these sensors can be classified into several groups. This includes a polymer, peptide, DNAzyme, nanoparticle, and functional materials-based chemosensors.^{23,24} However, being simple to use, this technique has lower sensitivity and only offers qualitative detection of water pollutants, thereby, they are not employed extensively for water pollutants detection.

2.1.4 Electroanalytical techniques

Electroanalytical-based detection techniques involve the interrelationship between electrical potential and the analyte's chemistry. The potential driven chemical effects occur in the electrochemical cell consisting of electrodes and analyte-containing electrolyte solution. The external potential is applied to the working electrode relative to the reference electrode through an electrical source. The chemical effects occur at the interface of the working electrode and analyte-containing electrolyte.²⁵ The electrochemical response of current or potential nature is measured relative to the counter electrode. Hence, the three-electrode immersed in the electrolyte solution makes an electrochemical cell that can be employed in a laboratory or on-site field, as shown in the Fig. 2.1. Besides its on-site portability, electroanalytical techniques are cost-effective, sensitive, and target-specific enough to fabricate low-cost and effective water pollutants sensors.^{3,26}



Figure 2. 1. Schematic of a three-electrode electrochemical cell.

The electroanalytical methods can be categorized into enzymatic or non-enzymatic based on their detection mechanisms. The enzymatic electrochemical method utilizes an enzyme that, on target interaction, inhibits its enzymatic activity. For example, inhibition of the activity of the acetylcholinesterase (AChE) enzyme on pesticide interaction is utilized for pesticide detection. The AChE enzyme helps the breakdown of acetylcholine,²⁷ however, on pesticide exposure, the enzyme sites get irreversibly bound, which inhibits the breakdown process. The amount of inhibition of enzyme activity by target exposure is relative to the target concentration. Hence, enzymatic-based electrochemical detection is highly sensitive and specific to the target species.²⁸ Enzymatic-based sensor for

organophosphates detection has reported an excellent limit of detection up to 2 nM.²⁷ However, the chances of enzyme instability on environmental exposure (pH, temperature, chemicals, etc.) limit their utilization in real-world applications.^{3,29}

Contrarily, the non-enzymatic based electrochemical method is emerging as a promising option for water pollutants detection. Since there is no requirement for a bioelement in the sensing unit, these sensors are not prone to degradation by environmental conditions like temperature or pH. The non-enzymatic electrochemical sensors are highly stable and reproducible, unlike enzymatic-based sensors. In comparison to indirect target detection by enzymatic inhibition, the non-enzymatic-based method can rely on the direct determination of the analyte by the electrochemical response generated from the analyte's chemical changes. Therefore, we have chosen a non-enzymatic electrochemical detection technique to fabricate a water pollutants sensor.

2.2 Electrochemical sensor

2.2.1 Electrochemical cell

As discussed earlier, an electrochemical cell is required to run the electrochemical detection of the analyte in a closed circuit. Depending on the number of electrodes, there can be a two, or three-electrode electrochemical cell setup. In the two-electrode setup, the potential is applied at the working electrode relative to the reference electrode, completing the circuit. Since the current passes through not only the working electrode but also the reference electrode, it leads to a large ohmic drop (an undesirable bulk resistance) in the bulk solution and changes the potential difference at the reference electrode. Consequently,

the two-electrode system lacks the controllability of the applied potential in the electrodeelectrolyte interface. A third electrode called the counter electrode (Fig. 2.1) is introduced to solve this problem. The current can pass only from working to the counter electrode, surpassing the reference electrode. Hence, a three-electrode setup is generally employed for electrochemical sensors.

The analyte undergoes electrochemical reactions at the interface of the working electrode and electrolyte in the solution. Electrochemical reactions are electron charge transfer processes that occur at the electrode-electrolyte surface. These processes are either caused or accompanied by the application of electric potential and involve the transfer of electrons between an electronic conductor (electrode) and analyte (molecule or ions) at the electrode-electrolyte interface. The working electrode must be electrochemically inert only to let the analyte of interest undergoes reactions. Therefore, the working electrode is typically made of glassy carbon (GC), platinum, or gold. Similarly, the counter electrode completing the current flow of the electrochemical cell must also be inert to chemical reactions. Hence, platinum or gold material is generally used as the counter electrode. At last, the reference electrode, whose role is to maintain a stable electrode potential in the cell, can be made with numerous electrodes. This mainly includes saturated calomel electrodes (employing mercury and mercury chloride) or silver-silver chloride in KCL solution. All three electrodes are immersed in a solution whose role is to provide conductive passage for the reactive reagents at the interface of electrode and solution. A supporting electrolyte is also added to the solution to limit the background signal response of ionic solution, mass transport, or non-faradaic processes. Moreover, they must be chemically inert throughout the operating potential range to avoid any interference with the response from the target specie. Generally, buffers or inorganic salt are used as supporting electrolytes with 0.1 M or higher concentration.

2.2.2 Electrochemical sensor's parameters

The electrochemical response from the analyte in the electrochemical cell depends on several parameters, including properties of the working electrode and mass transport of the target species.

2.2.2.1 Working electrode properties

The primary role of the working electrode is to provide the supplied potential from the potentiostat to the electrode-electrolyte interface. Hence, the electrical property of the working electrode is one of the deciding parameters on which the electrochemical response depends. At the working electrode-electrolyte interface, a typical heterogeneous electron transfer process involves an electron exchange between the electrode and an analyte. With the applied potential, the energy levels of electrons in the electrode are modulated. By driving the electrode to more negative potentials, the energy of the electrons (in the electrode) is raised. They can reach a level high enough to transfer into vacant electronic states on species (molecule or ions) in the electrolyte solution. In that case, a flow of electrons from electrode to solution occurs and is termed a reduction current. Similarly, the energy of the electrons (in the electrode) can be lowered by applying a more positive potential, and at some point, electrons on the species in the electrolyte will find more favorable energy on the electrode and will transfer there, this is termed as oxidation current.²⁵

The above heterogeneous electron transfer process relies on the overlapping of the electrode and analyte's electronic energy levels. The former is readily acted through a potentiostat letting electrochemists monitor the kinetics and thermodynamics of the process. The extent of such overlapping of the energy levels is influenced by the density of available electronic levels (thus, band structure), which by applying an appropriate potential, gets occupied with the charge carriers, thereby favoring the electron transfer process. Therefore, electrons' mobility and concentration (free electrons in metals or conduction electrons in a semiconductor); thus, the electrical conductivity of the electrode material influences the electron transfer process for the electrochemical reactions.

Secondly, the working electrode provides the necessary surface area for the analyte to undergo heterogeneous electron transfer processes. The amount of available surface area of the working electrode thus influences the electrochemical response. Therefore, a high surface area working electrode is desirable for a better electrochemical signal from the analyte electrochemical reactions. Concisely, the electrical conductance and surface area are termed the electrocatalytic properties of the working electrode. Hence, a higher electrocatalytic working electrode is sought for a higher electrochemical response.

2.2.2.2 Mass transport in bulk solution

The electrochemical response from the analyte undergoing an electron transfer process depends not only on the working electrode's electrocatalytic properties but also on its movement from the bulk solution to the electrode-electrolyte interface.²⁵ The movement of the analyte in the solution is termed mass transport. Mass transport can occur either

naturally from random molecular motion or forcefully from a concentration gradient, etc. Generally, mass transport in an electrochemical cell is classified into diffusion, convection, and migration.^{25,30}

Diffusion is the movement of a substance (atoms, molecules, ions, or energy) from a high concentration region to a low concentration region under a concentration gradient. The substance moves from a high concentration region to a low concentration region under a concentration gradient to achieve diffusive equilibrium. Thermodynamic equilibrium is achieved when a thermodynamic potential is minimized under particular conditions. In diffusion, thermodynamic potential named Gibbs free energy (G) is minimized at constant temperature and pressure to achieve diffusive equilibrium. Since Gibb's free energy (G) possessed by 1 mole of a substance is known as chemical potential (μ), diffusive equilibrium is also said to achieve when the chemical potential of the substances is the same. The analyte under concentration gradient moves from the bulk solution to the planar electrode surface following Fick's first law^{25,30} as described below.

$$j = -D\frac{\partial C}{\partial x}$$

Where *j* is flux (mol m⁻² s⁻¹), *D* is diffusion coefficient, *C* is the concentration of the analyte, and *x* is the perpendicular distance from the electrode. The negative sign describes the species' movement from higher to lower concentration. The rate of change in concentration with time follows the Fick's second law:²⁵

$$\frac{\partial C}{\partial t} = D \frac{\partial^2 C}{\partial x^2}$$

Diffusion from the bulk solution to the electrode surface is dependent on the electrode geometry. Generally, planar, hemispherical, and cylindrical diffusion occurs on planar, hemispherical, and cylindrical electrodes, respectively.³¹Another mode of mass transport includes convection. The movement of species under internal or external forces is termed convection.³⁰ Internally forced movement is termed natural convection, which can occur either by a chemical reaction-induced change in density gradient or by thermal variations. Since natural convection is unwanted, a thermostated cell is usually employed to minimize the effect of thermal variations. Forced convection, on the other hand, is caused by external forces like the magnetic stirring of the solution or ultrasonication. Forced convection helps better the distribution and mass transport of species towards the At last, migration is the type of mass transport where species movement is electrode. under an electrical field. The charged species in the solution get influenced by the electric field generated by the potential difference at the electrode-electrolyte interface. This unwanted charged species movement can interfere with the mass transport of target species; hence, the electrochemical cell is designed to minimize migration effects. This includes the addition of a highly concentrated supporting electrolyte which nullifies the developed potential difference at the electrode interface. Generally, 0.1 M concentration or above is used for the supporting electrolyte.³⁰

2.3 Electrochemical sensor's sensitivity

2.3.1 Influence of working electrode properties

As mentioned in section 2.2.1, the electrochemical response of an analyte is influenced by the electrical property and surface area of the working electrode. The sensitivity of an electrochemical sensor is generally explained as the sensor's minimum detectable concentration above the signal's noise limits. Hence, the higher the electrochemical signal from target detection, the better the sensor's sensitivity. Therefore, the electrochemical sensor's sensitivity is greatly influenced by the signal amplification capability of the working electrode.

The working electrode signal amplification capability depends on its electrical conductivity and surface area. As discussed earlier, the electrical conductivity of the working electrode influences the analyte's electron transfer processes at the electrode-electrolyte interface. Moreover, the higher the total surface area of the working electrode, the higher the available surface area for the analyte to undergo electron transfer processes. Hence, the working electrode with higher electrical conductivity and higher surface area is desired to fabricate highly sensitive electrochemical sensors.

The bare working electrode, like glassy carbon (GCE) or gold electrode, has lower electrical conductance and surface area. Hence, the bare electrode is modified to enhance its electrocatalytic properties for an amplified signal. To increase the surface area of the bare electrode, materials possessing high surface area have been used for water pollutants detection. This generally includes porous materials like activated carbon (AC) mostly used for heavy metal ions detection.³² However, such porous material lacks electrical conductivity, limiting its electrode modification usability. Another modification strategy involves the utilization of carbon nanomaterials, metal nanoparticles, or conductive polymers.³³⁻³⁵ Widely employed carbon nanomaterials include carbon nanotubes or graphene; metal nanoparticles include gold, silver, or platinum; and conductive polymers include polyaniline or polypyrrole (PPy).

Most modified electrodes have demonstrated enhanced signals for non-enzymatic water pollutant detection but did not overcome the highly sensitive enzymatic sensor's performance. One of the reasons includes the utilization of layer-by-layer nanomaterial modification, which has shown increment in their agglomeration. The aggregate formation reduces the total surface area and passivates the electrode surface, hence, reducing the overall sensor performance. Another reason includes the poor film formation of the electrode modification due to poor bonding to the electrode surface. This also affects the stability of the fabricated sensor and raises questions about its reliability.

2.3.2 Influence of analyte's mass transport

As discussed, the sensitivity of an electrochemical sensor can be improved by enhancing the electrochemical response of the analyte. For this, carbon nanomaterials modified electrodes are widely employed. Carbon nanotubes (CNT) modified electrodes are extensively used for water pollutants detection.^{36,37} Their enhanced electrochemical response has been attributed to their higher surface area and electrical conductivity, hence higher electrocatalytic properties.38 The literature understands this faster electrode kinetics by the produced cyclic voltammogram's peak-to-peak potential separation (Δ Ep).³⁸ This

assessment is accurate only when the analyte's mass transport from the bulk solution to the working electrode is the same for unmodified and CNT-modified electrodes. Nevertheless, the analyte's mass transport is not the same for both electrodes. Hence, their claim for faster electrode kinetics in the carbon nanotube-modified electrode is not true.

As discussed earlier, parameters influencing an electrochemical response towards an analyte generally include electrical conductivity and specific surface area of the modified working electrode, mass transport of the analyte, and pH of the solution. Now for the same nanotube-modified electrode and properties of the solution, only mass transport of the analyte influences an electrochemical response, which includes diffusion, migration, and convection of the target species. Keeping migration and convection negligible and constant for comparing the planar (naked) and porous (modified) electrode, only the diffusional regime influences the analyte's cyclic voltammetric response. The diffusional regime of the analyte is inspired by the surface geometry of the electrode.³¹ Typical diffusion modes include semi-infinite planar, hemispherical, and cylindrical diffusion that occurs at planar, hemispherical, and cylindrical electrodes. The semi-infinite infinite diffusion of the analyte occurs from the bulk solution to the planar surface of the electrode, as shown in Fig. 2.2.³¹ There is another diffusion mode that can occur at carbon nanotubes modified electrodes. This mass transport is known as thin-layer diffusion, which occurs through the different nanotubes' layers encompassing confined portions of the analyte solution.38



Figure 2. 2. Schematic of (A) semi-infinite diffusion at a planar electrode, (B) thin-layer diffusion at a porous electrode.

In short, the lack of understanding behind the enhanced electrochemical response for the water pollutant at the porous electrodes makes it challenging to understand the full potential and exploits carbon nanomaterial's maximum advantages in the fabrication of highly sensitive non-enzymatic electrochemical sensors.

2.4 Electrochemical sensor's specificity

The specificity of an electrochemical sensor is crucial for its selective determination of a water pollutant in a mixture of interfering chemicals. In the enzymatic-based electrochemical detection, the specificity is offered by the attachment of the analyte to the binding sites of the enzyme. For example, organophosphate can irreversibly bind to the AChE enzyme sites and inhibits its enzymatic activity.²⁷ This provides very high specificity in the enzyme-based electrochemical sensors. On the other hand, the selective response
from the non-enzymatic electrochemical sensors can be achieved using materials possessing affinity to the target specie. This affinity-based mechanism is incorporated into the sensor by depositing material onto the working electrode, whose primary role is to increase the concentration of target specie near the electrode-electrolyte interface. We can call such materials detecting agents in non-enzymatic electrochemical sensing. Numerous detecting agents have been explored for different water pollutants detection. Generally, ferric oxide (Fe₂O₃) has been utilized for selective electrochemical detection of Zn²⁺, Cd²⁺ and Pb²⁺,³⁹ Zirconium dioxide (ZrO₂) for organophosphates,⁸ and molybdenum disulfide (MoS₂) for Hg²⁺ selective detection.⁴⁰

The detecting agent-modified electrodes have shown a good selective response to water pollutants, but they still failed to achieve the high sensitivity and selectivity of enzymatic electrochemical sensors. Achieving the enzymatic electrochemical sensor's performance in non-enzymatic sensors is essential. Even a tiny amount of water pollutants have indicated a drastic negative influence on human life, such as a small amount of lead intoxication leading to neurological disorders.^{2,12} One of the prominent reasons for this lack in performance is attributed to the nanoparticle agglomeration of the detecting agent. The nanoparticles are inclined to form agglomerates in their natural state, which reduces their overall surface area.⁴¹ The aggregate formation also reduces the overall electrode's electrical conductivity by passivating the electrode surface. The nanoparticle aggregation on deposition on the working electrode has reduced electrocatalytic performance, significantly affecting the sensor's performance. Another reason for the limited performance is their poor film formation upon electrode modification. The nanoparticles

agglomeration leads to large aggregates, which on electrode deposition, form clumps and affect their film formation. Poor film formations reduce electrochemical performances and raise questions about the sensor's reliability and repeatability. Therefore, non-enzymatic sensors are yet to achieve reliable, repeatable, highly sensitive, and highly selective electrochemical performance as enzymatic-based sensors.

2.5 Electrochemical techniques

In this section, different electrochemical techniques are described for analyte determination and electrode characterization. For detection purposes, voltammetric techniques like cyclic voltammetry (CV) and square wave voltammetry (SWV) are usually used. CV can be used as a multifunctional tool to determine analyte, study the electrochemical reactions and electrode kinetics, and electrochemical surface area evaluation. SWV, on the other hand, is mainly used as an analyte detection tool due to its high sensitivity compared to CV. At last, electrochemical impedance spectroscopy (EIS) is used to characterize the charge transfer ability of the modified electrodes.

2.5.1 Cyclic voltammetry

As mentioned earlier, CV is used for analyte determination, the study of electrode kinetics and electrochemical reactions occurring at the electrode-electrolyte interface, and electrode's electroactive surface area evaluation. In the CV, the potential applied at the working electrode varies linearly with time. The resulting current is measured, generating *I vs. E* cyclic voltammogram (as seen in Fig. 2.3). The potential is varied between initial potential, E_i , and switching potential, E_s . The CV profile starts from E_i till E_s in the forward CV scan and then switches back from E_s to E_i in the backward scan. Depending upon the scan route, oxidation or reduction reactions, hence redox reactions occur.⁴² The rate at which potential is swept is known as scan rate (v).



Figure 2. 3. Schematic of (I) time-dependent potential in cyclic voltammetry, (II) a typical cyclic voltammogram.

The CV profile can determine the type of analyte's electrochemical reactions. The different electrochemical reactions include reversible, quasi reversible, and irreversible. Their determination is mainly exercised by either measuring the peak-to-peak potential separation (Δ Ep) of the cathodic and anodic peaks generated in the complete CV run or by the relationship between peak current (I_p) and *v*. The analyte undergoing reversible reaction follows the Ip *vs. v* relationship as per the Randles-Sevcik equation:²⁵

$$I_n = 2.69 * 10^5 n^{3/2} ACD^{1/2} v^{1/2}$$

where A is electrode area, n is the number of electrons participating in the electrochemical reaction, C is concentration, and D is the diffusion coefficient of the analyte. Moreover, Δ Ep of such cyclic voltammogram is 57/n mV at 298 K.²⁵ For the irreversible reaction, Ip

vs. v follows another version of the Randles-Sevcik equation applicable to the only irreversible reaction:²⁵

$$I_p = 2.99 * 10^5 n (n' + \alpha_{n'})^{1/2} A C D^{1/2} v^{1/2}$$

where α and n' denote transfer coefficient and number of electrons transferred prior to the rate-determining step. Moreover, the irreversible reactions are sluggish, and higher overpotential is required for the reaction to occur. Hence, ΔEp of irreversible reactions is larger than 57/n mV. At last, the quasi-reversible reactions have characteristics between the two reactions.

The CV is also used as a characterizing tool to evaluate the electroactive surface area of the working electrode. The CV is varied for different scan rates for a known redox specie (generally, $Fe(CN)_6^{3-}/Fe(CN)_6^{4-}$), then the obtained data is used to measure *Ip* of each voltammogram. Then, *Ip vs.* $v^{1/2}$ graph is plotted, and using the Randles-Sevcik equation,³⁷ the electroactive surface area is evaluated. This technique helps determine the available surface area of different modified working electrodes.

The CV measures the current generation from faradaic and non-faradaic processes. The faradaic processes involve the charge transfer at the electrode-electrolyte interface compared to non-faradaic processes. The electric double layer formation near the electrode causes these non-faradaic processes.²⁵ The charge species from the electrode and electrolyte form the double layer and act as an interfacial capacitor, and this leads to a charging current in addition to the faradaic current. At a low concentration of the target species, the faradaic current can be lower than the charging current. Hence, the charging current in CV limits its sensitivity in analyte determination. Hence, the CV, apart from its valuable utilization, has the disadvantage of low sensitivity in the electrochemical detection of the analyte.

2.5.2 Square wave voltammetry

The square wave voltammetry is a type of pulse voltammetry technique.²⁵ Pulse voltammetry techniques are primarily used for analyte detection with high sensitivity. In this, a combination of staircase potential function and square wave pulses are utilized. They are superimposed such that the square wave pulse coincides staircase step in the forward direction, as shown in Fig. 2.4. The current is supplied both at the forward and reverse pulses termination. The current differences are then plotted against the staircase potential. This process eliminated the charging current that occurs in CV measurements. Hence, the SWV technique measures only the faradaic current, thereby providing high sensitivity in the analyte detection. The scan rate of the SWV is usually evaluated with the following equation:

$$v = P_{step} * f$$

where v is scan rate (mV/s), P_{step} is staircase size or step potential (mV), and f is square wave frequency (Hz). The square wave frequency, f is 1/t, where t is the time duration of one wave cycle. Moreover, the pulse height is known as amplitude (in mV). The step potential, frequency, and amplitude combined are called SWV parameters. The SWV profile of the analyte response depends on SWV parameters. They all influence the analyte's redox peak shape and hence, the current response. Thereby, all these parameters are optimized for the optimal sensor performance.



Figure 2. 4. Schematic of a potential cycle in square wave voltammetry.

2.5.3 Electrochemical impedance spectroscopy

Electrochemical impedance spectroscopy (EIS) is a crucial characterizing tool in electrochemistry. EIS analyzes the charge transfer ability of different components of the electrochemical system. The technique analyzes charge transfer processes by providing the charge transfer resistances of each sensor's component, including the bulk solution, working electrode, and double-layer near the electrode.⁴³

For a typical single electrochemical reaction described in Schematic 4, the current is generated from faradaic and non-faradaic processes. The charge transfer between the working electrode and analyte at the electrode-electrolyte interface produces a faradaic current. This occurs with the analyte surpassing the electrode activation barrier resistance or charge transfer resistance (R_{ct}) and the solution resistance (R_s). The double-layer formation leads to a charging current and acts as an interfacial capacitor (C_{dl}) . Consequently, the capacitor charging produces a non-faradaic current. The charge transfer process of the reactant species is followed by their mass transport near the electrode interface, which leads to Warburg impedance (Z_w). These numerous components are used to build electrical circuits describing the charge transfer processes of the entire electrochemical system. The Randles equivalent circuit is widely employed to build the simplest electrical circuits. Using this, the Nyquist plot is generated with the imaginary (Z_i) vs. real (Z_r) part of the complex impedance.²⁵ With Randles equivalent circuit and Nyquist plots, the charge transfer resistance (R_{ct}) of the different modified working electrodes is determined.²⁵

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3 Problem statement and research objectives

As discussed in the literature review section, the numerous fabricated sensors have failed to achieve or overcome the ultrahigh sensitivity and specificity of enzymatic-based electrochemical sensors for water pollutants determination. Moreover, non-enzymatic electrochemical sensors are growing in demand for water pollutants detection. However, their sensing performances are not excellent enough to replace the enzymatic-based sensors. Therefore, this work aims to fabricate non-enzymatic electrochemical sensors for ultra-high sensitivity and specificity for water pollutants, including organophosphate, heavy-metal ion, and nutrient. The research goal is achieved by achieving the following three primary research objectives.

3.1 Modeling porous electrode kinetics for better sensor performance (Chapter 4)

As mentioned in the previous section, in addition to the working electrode properties, the analyte's mass transport to the electrode-electrolyte interface also influences its electrochemical response. Carbon nanomaterials like carbon nanotubes are porous in nature, and hence, in addition to semi-infinite diffusion of the analyte, a thin-layer diffusion can also occur. This additional analyte's mass transport can influence its electrochemical response. This forms the basis of the first objective, which is to investigate the occurrence of thin-layer diffusion and its effects on FT detection on porous electrodes. Consequently, formulation of a simulation model of the FT electrode kinetics at the unmodified and porous modified electrodes is also proposed. A simulation model to study electrode kinetics of a redox specie that have multi-step and multi-electron reactions like FT have not yet been developed. Subsequently, this inspires another objective to formulate a new

simulation model for multi-step and multi-electron electrochemical reactions. Results from this study can assist in the understanding of the influence of the porous electrode on analyte detection and thus assist in the fabrication of an ultrasensitive electrochemical sensor.

3.2 Synthesis of a sensitive nanomaterial ink (Chapter 5)

From the simulation results of the influence of porous electrodes on fenitrothion detection, the next objective is to synthesize a highly-sensitive ink to produce highly porous and electrocatalytic electrodes. Activated carbon (AC) possesses a high surface area, but they have lower electrical conductivity. Heteroatom doping of AC can enhance its conductivity. This formulates the synthesis strategy to co-dope AC with heteroatoms for enhanced conductivity. Moreover, carbon nanomaterials like carbon nanotubes with high surface area and conductivity can assist in further their enhancement. Consequently, the electrode modifying ink using co-doped activated carbon and carbon nanotubes is proposed, where carbon nanotubes provide the supporting surface for co-doped AC coating. The objective of this work includes synthesizing an ink delivering porous electrode with high electrocatalytic properties. Hence, a novel composite nitrogen-sulfur co-doped activated carbon coated multiwalled carbon nanotube (NS-AC-MWCNT) is proposed using polypyrrole (PPy) carbonization. For an excellent coating of polypyrrole on carbon nanotubes and dispersion of carbon nanotubes, utilization of eriochrome cyanine R (ECR), a multifunctional dopant, is also proposed. The synthesized ink is used to fabricate highly electrocatalytic electrodes for fenitrothion (FT, an organophosphate) detection.

3.3 Synthesis of the specificity assay (Chapter 6)

From the early objective of synthesizing a sensitive ink, the fabricated NS-AC-MWCNT/GCE sensor failed to overcome the sensing performances of the enzymatic-based electrochemical sensors. Sensor sensitivity can be enhanced by increasing the concentration of the target species near the working electrode. Such materials are termed detecting agents and have an affinity toward the target species. However, their tendency to agglomerate limits their surface area and passivate the electrode surface, thereby affecting the electrode's electrical conductivity. Moreover, agglomeration can cause poor film formation on the electrode surface. Besides, from the knowledge and understanding of the electrode kinetics of porous electrodes on FT detection in Chapter 5, porous electrodes influence the analyte's electrochemical responses. Hence, the incorporation of the detecting agent component on the electrode surface should also enhance the porosity of the electrode. This inspires the next objective to synthesize agglomeration-free detecting agents, which produce stable electrode film of high surface area and promotes the analyte's charge transfer processes. Hence, novel composites based on metal oxides (as detecting agents, ZrO₂, MnO₂, or MgO) immobilized on chitosan (CHIT) modified catechol molecule (3,4dihydroxylbenzaldehyde, DHBA) is proposed. Detecting agents can enhance the analyte's concentration near the electrode interface. DHBA can disperse the detecting agents with catecholate type bonding and promote charge transfer processes. CHIT can promote the film-formation and enhance the porosity of the electrode. The proposed X-DHBA-CHIT (X is the detecting agent) material can be deposited on the NS-AC-MWCNT/GCE electrode to synergistically work and achieve the sensing performances of enzymatic-based electrochemical sensors for water pollutants detection.

4 Influence of Conductive Porous Electrodes on the Apparent Electrode Kinetics of Fenitrothion

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4.1 Abstract

Planar bare electrodes fabricated with surface-modified multiwalled carbon nanotubes (MWCNT) are used to detect organophosphate (OP) compounds, which are used as herbicides and fungicides but are harmful to human health. Deposition of carbon nanotubes on the surfaces of bare electrodes enhances electrocatalysis, increasing the electrode to analyte current response and narrowing peak-to-peak potential separation during cyclic voltammetry (CV). We hypothesize that, as the thickness of the deposited MWCNT layer decreases, FT mass transport between the layers of porous multiwalled carbon nanotubes changes from semi-infinite to thin-layer diffusion. This influences the electrode electrode multiwalled response to the analyte. Using simulations and experiments, we show that when porous MWCNT are deposited on a conductive glassy carbon electrode the mass transport of fenitrothion (FT, an OP) changes from semi-infinite to thin-layer diffusion during CV. This alters the electrochemical response of the electrode and reduces peak-to-

peak potential separation. To simulate CV response to FT, both the semi-infinite and thinlayer diffusion models are employed for the planar bare and modified porous surface electrodes. The transition from thin layer to semi-infinite diffusion is clear when the nanotube layer thickness on the bare electrode increases. The model is applicable to other toxic chemicals, such as 4-nitrophenol, parathion, or methyl parathion that have similar electrode kinetics.

4.2 Introduction

Organophosphorus (OP) compounds are widely used to improve agricultural production, contributing to 70% of pesticide consumption in agriculture.⁵⁻⁶ The OP residue that persists in the environment has a serious public health impact, since it can lead to mental disorder, coma and death.⁷ Due to human poisoning and associated illnesses from OP, an annual economic cost of \$1 billion is estimated in the U.S. alone.⁸ The typical analytical techniques to monitor OP include mass spectrometry⁹ and liquid or gas chromatography,¹⁰ but these require complex sample processing, skilled professionals, and are costly.¹¹ Due to their lower cost, rapid response and high sensitivity, electroanalytical methods have emerged as promising methods for OP monitoring.

To lower the detection limit and improve sensor sensitivity, the electrochemical response of an electrode to an analyte must be enhanced. When nanomaterials, such as carbon nanotubes (CNT), are deposited on a bare electrode, the electrode specific surface area and electrical conductivity also increase,^{12,13-14} which improves sensitivity by augmenting electrocatalysis,¹⁵ enhancing OP detection.^{12,16-17} These faster electrode

kinetics have been interpreted through the peak-to-peak potential separation of cyclic voltammograms, a comparison is valid when the mass transport of the analyte towards the electrode is the same for both the bare and modified electrode.¹⁵ However, the mass transport regime is not the same in both cases, making the argument in favor of faster electrode kinetics influenced by a CNT-modified electrode questionable.

The additional mass transport of analytes on the surfaces of CNT-modified electrodes occurs through thin-layer diffusion in contrast to semi-infinite diffusion for bare electrodes.^{15,18-19} Semi-infinite diffusion of the analyte from the bulk solution occurs at the planar surface of the electrode, while thin-layer diffusion occurs between the layers of nanotubes containing trapped portions of the solution, as depicted in Fig. 4.1. Both the semi-infinite and thin-layer diffusion models for ferrocyanide demonstrate that the thin-layer model produces a smaller peak-to-peak potential separation in cyclic voltammogram for a porous CNT/glassy carbon electrode (GCE) as compared to an unmodified GCE.¹⁵ Further evidence for thin-layer effects at the surface of a porous MWCNT/GCE electrode has been provided based on semi-infinite and thin-layer diffusion mixed mass transport for nicotine.¹⁸ Therefore, mass transport effects must be considered while explaining the electrochemical response of a porous modified electrode towards an analyte.

To simulate a cyclic voltammetry (CV) response, we select fenitrothion (FT) as the OP that undergoes multi-step and multi-electron electrochemical reactions. FT has a redoxactive $-NO_2$ group that undergoes multi-step multi-electron electrochemical reactions, as shown in Fig. 4.2 (B). Application of a negative cathodic potential of -0.6 V vs. Ag/AgCl during cyclic voltammetry reduces the $-NO_2$ group of FT irreversibly to -NHOH producing

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FT^{red 20} as shown in Fig. 4.7 (A). During subsequent scans, the -NHOH and -NO groups undergo quasi-reversible oxidation and reduction, forming FT^{ox} and FT^{red}, respectively. We are unaware of the existence of a theoretical model to simulate such multi-step and multi-electron electrochemical reactions like, FT.

Our objective is to simulate FT's electrochemical response at bare and porous modified electrodes to investigate the occurrence of thin-layer effects. We hypothesize that the change in the FT mass transport regime between the layers of porous multiwalled carbon nanotubes from semi-infinite to thin-layer diffusion influences the electrode electrochemical response to the analyte. This thin-layer effect lowers peak-to-peak potential separation during CV with a MWCNT/GCE compared to a bare GCE. We simulate the FT electrode kinetics at the surfaces of bare and porous modified electrodes to characterize the effects that arise due to thin-layer diffusion through the porous MWCNT deposit. The theoretical model should apply to other toxic compounds with similar electrode kinetics, like 4-nitrophenol, parathion, or methyl parathion, that are typically used to synthesize pesticides, pharmaceuticals, and dyes.^{12,21-22}



Figure 4. 1. Schematics of the two different mass transport regimes present at the surfaces of (A) a glassy carbon electrode (GCE) and (B) a multiwalled carbon nanotube/glassy carbon electrode (MWCNT/GCE). (C) TEM image of the MWCNT.

4.3 Mathematical formulation

The CV responses to FT at the surfaces of the unmodified planar GCE and porous MWCNT/GCE are simulated using semi-infinite diffusion and thin-layer cell models.¹⁵ In contrast to the semi-infinite planar diffusion model, the thin-layer cell model contains both planar conductive and insulating layers.¹⁵ The solution containing the electroactive species is confined within these thin-layer cells, representing pockets of solution contained between MWCNT layers. The FT electrochemical reactions occur when electron transfer between FT and the electrode is thermodynamically and kinetically favorable, which generates redox peaks in the CV profiles.

We assume an idealized scenario, where only FT is present in the solution exposed to the electrodes. The starting, vertex and ending potentials used in the simulation are -0.2, -0.8 and 0.5 V, respectively. Table 1 contains a list of the dimensionless variables used to simulate the electrochemical process.

Table 4. 1: List of all the dimensionless variable used for simulating the electrochemical reaction kinetics of the FT, where c_{FT} , c_{FT}^{red} , and c_{FT}^{ox} denote the concentrations of FT, reduced FT, and oxidized FT, respectively. c_b initial bulk concentration of FT, ϵ radius of the macroscale disc electrode, *A* area of the electrode, *x* perpendicular coordinate from the electrode, *R* universal gas constant, *T* absolute temperature, *F* Faraday's constant, *E* imposed electrode potential, E_1^0 apparent formal potential for step A (see Fig. 4.2 (B)), $E_{rds}^{0'}$

and $E_{non-rds}^{0'}$ formal potentials for the second and first electron transfers of step B, $\Delta E^{0'} = E_{rds}^{0'} - E_{non-rds}^{0'}$, *D* diffusion coefficient of the species (D = 6.9× 10⁻⁶ cm² s^{-1 23}), *t* time, k_1^0 and k_2^0 apparent standard heterogeneous rate constants for step A and B (see Fig. 4.2 (B)), respectively, *v* scan rate, and *I* current.

Dimensionless quantity	Expressions
Concentration	$C_{\rm FT} = \frac{c_{\rm FT}}{c_b}$, $C_{\rm FT}^{red} = \frac{c_{\rm FT}^{red}}{c_b}$, and $C_{\rm FT}^{ox} =$
	$\frac{c_{\rm FT}^{ox}}{c_b}$
Distance	$X = \frac{x}{\epsilon}$
Potential	$\theta_1 = \frac{F(E-E_1^0)}{RT}, \theta_2 = \frac{F(E-E_{rds}^{0\prime})}{RT}$ and
	$\theta_{\Delta E^{0'}} = \frac{F \Delta E^{0'}}{RT}$
Rate constant for step A of reaction (see Fig. 4 2 (B))	$K_1^0 = \frac{k_1^0 \epsilon}{D}$
Rate constant for step B of reaction (see Fig.	$K_{2}^{0} = \frac{k_{2}^{0}\epsilon}{k_{2}^{0}\epsilon}$
4.2 (B))	
Time	$\tau = \frac{Dt}{\epsilon^2}$
Scan rate	$\sigma = \frac{F\epsilon^2 v}{PTP}$
Layer thickness	$L = \frac{\beta \sqrt{Dt_{max}}}{\epsilon}$
Current	$j = \frac{i\epsilon}{AFDc_{h}}$

The diffusion of electroactive species is represented through Fick's second law, and c_{FT} , c_{FT}^{red} , and c_{FT}^{ox} , are determined at the bare electrode and thin-layer cell by applying Eqs. (4.1)-(4.3). The diffusion coefficients for FT, FT^{red}, and FT^{ox} are assumed identical for sake of simplicity.

$$\frac{\partial C_{FT}}{\partial \tau} = \frac{\partial^2 C_{FT}}{\partial x^2},\tag{4.1}$$

$$\frac{\partial c_{FT}^{red}}{\partial \tau} = \frac{\partial^2 c_{FT}^{red}}{\partial x^2}, \text{ and}$$
(4.2)

$$\frac{\partial C_{FT}^{ox}}{\partial \tau} = \frac{\partial^2 C_{FT}^{ox}}{\partial X^2}.$$
(4.3)

Two boundary conditions (at X = 0 and X = L for $\tau > 0$) and an initial condition (at $0 \le 1$ X < L for $\tau = 0$) are required to solve these equations. At the electrode surface (X = 0), the multi-electron multi-step kinetics are governed by the IUPAC approved Butler-Volmer kinetic equation²⁴⁻²⁵ (Eqs. (4.4)-(4.6)) which assumes that the second step of the irreversible reduction of FT to FT^{red} (i.e., step A) is the rate determining step (Fig. 4.3 A). This assumption is consistent with the behaviors of the experimentally measured cyclic voltammograms of bare and carbon nanotube modified electrodes. For the two-electron transfer reaction (step B), the second electron transfer is assumed to be the rate determining step (rds), while the first electron transfer remains in equilibrium (Fig. 4.3 B). Hence, kinetic equations guided by Bard & Faulkner²⁶ are derived, where $E_{rds}^{0'}$ and $E_{non-rds}^{0'}$ denote the formal potentials for the second and first electron transfers, respectively. In the bulk (X = L), the boundary condition differs for two scenarios, (a) pre-specified concentration of FT for semi-infinite diffusion and (b) no-flux boundary condition of the species (see Fig. 4.2 (A)) for thin layer diffusion. The $L \rightarrow \infty$ boundary condition is imposed at a reasonably large distance compared to the diffusion length scale $\sqrt{Dt_{max}}$ by introducing a multiplier $\beta = 2.^{24}$ Further,

$$\frac{\partial C_{FT}}{\partial X}\Big|_{X=0} = K_1^0 \exp[-(1+\alpha_1)\theta_1] C_{FT}, \qquad (4.4)$$

$$\frac{\partial C_{FT}^{red}}{\partial X}\Big|_{X=0} = -K_1^0 \exp[-(1+\alpha_1)\theta_1] C_{FT} + K_2^0 \exp[(1-\alpha_2)\theta_2] C_{FT}^{red} - K_2^0 \exp(-\theta_{\Delta E^{0'}}) \exp[-(1+\alpha_2)\theta_2] C_{FT}^{ox}, \text{ and}$$
(4.5)

$$\frac{\partial C_{FT}^{ox}}{\partial X}\Big|_{X=0} = -K_2^0 \exp\left[(1-\alpha_2)\theta_2\right] C_{FT}^{red} + K_2^0 \exp(-\theta_{\Delta E^{0'}}) \exp\left[-(1+\alpha_2)\theta_2\right] C_{FT}^{ox}.$$
(4.6)

Equations (4.1)-(4.3) coupled with Butler-Volmer electrode kinetic equations, Eqs. (4.4)-(4.6), are discretized using the finite volume method with equally spaced grid points. Time discretization is based on an implicit time scheme where current concentrations values at neighboring node are used for time marching. The resulting set of linear algebraic equations is cast in a classical tridiagonal matrix form and solved using the tridiagonal matrix algorithm (TDMA), which provides the one-dimensional spatiotemporal concentration for different species. All simulations are conducted in the MATLAB R2015a environment and run on a personal computer (Intel Core i7 processor @ 2.11 GHz, 16 GB RAM) for \approx 2 seconds to generate each voltammogram. The MATLAB code is available at https://github.com/Krishnajangid1996/Fenitrothion-electrode-kinetics. Then, the overall electrochemical current response is calculated as follows,

$$j = -4\frac{\partial C_{FT}}{\partial X} + 2\frac{\partial C_{FT}^{ox}}{\partial X}$$
(4.7)



Figure 4. 2. (A) Schematic of the boundary conditions for FT transport for the onedimensional semi-infinite diffusion and thin-layer cell model where the species undergoes chemical reactions and produces electroactive species FT^{red} and FT^{ox} . (B) Schematic of the FT electrochemical reactions where the redox-active -NO₂ group produces electroactive FT^{red} and FT^{ox} through steps A and B.



Figure 4. 3. Schematics of the mechanism of (A) step A, and (B) step B of the FT electrochemical reactions producing electroactive FT^{red} and FT^{ox} .

4.4 Experimental

4.4.1 Chemicals and Instruments

Analytical grade FT and sodium acetate buffer solution (ABS) reagents were obtained from Sigma-Aldrich (USA). MWCNTs (length 1-2 μ m, outside diameter 13 nm, and inside diameter 4 nm) were purchased from Bayer Inc. (Germany). Deionized water was used for all solution preparation. 0.1M NaOH and 0.1M HCl were used to adjust the pH of the buffer solution. FT stock solutions were prepared in ethanol and stored at 4°C. Electrochemical measurements were executed with a PARSTAT 2273 potentiostat, Princeton Applied Research, USA. A three-electrode arrangement consisting of a GCE (bare working electrode), platinum wire (counter electrode) and Ag/AgCl (reference electrode) immersed in buffer solution was used for the cyclic voltammetric experiments. Transmission electron microscopy (TEM) characterization was performed with FEI Titan 80-300 LB instrument.

4.4.2 Preparation of MWCNT/GCE

MWCNTs were suspended in ethanol to form a 1 mg/mL suspension. Several volumes of the MWCNT suspension were drop casted on the GCE and dried in air to form MWCNT-modified GCEs. The electrochemical response of the electrodes was obtained by scanning them using cyclic voltammetry from -0.2 to -0.8 V, then -0.8 to 0.5V, followed by 0.5 to -0.2V. A 1 mM FT concentration and a 0.2 M ABS (pH 5) were used for all experiments. Scan rates of CV were varied from 50 to 175 mV/s.

4.5 Results and discussion

4.5.1 Theoretical results from the mechanistic model

The FT cyclic voltammetric simulations are performed and revealed the influence of thinlayer effects on the voltammetric response. Apparent standard rate constants for step A and B (Fig. 4.2 B) of FT at the bare planar and porous modified electrode are $k_{1,b}^0$, $k_{2,b}^0$ and $k_{1,m}^0$, $k_{2,m}^0$ respectively. We have assumed that rate constants of thin-layer diffusion model ($k_{1,m}^0 = 6.5*10^{-2}$ cm/s, $k_{2,m}^0 = 8.2*10^{-3}$ cm/s) are greater than the semi-infinite diffusion model ($k_{1,b}^0 = 4.2*10^{-2}$ cm/s, $k_{2,b}^0 = 5.1*10^{-3}$ cm/s). We first compare the peak-to-peak potential separation ΔE_{pp} for both semi-infinite diffusion and thin-layer diffusion models. Three different sets of rate constant values are considered, i.e., using (1) $k_{1,b}^0$, $k_{2,b}^0$ for semiinfinite diffusion and $k_{1,m}^0$, $k_{2,m}^0$ for thin-layer diffusion, (2) $k_{1,b}^0$ and $k_{2,b}^0$ for both models, and (3) $k_{1,m}^0$ and $k_{2,m}^0$ for both models. Hence, Case 1 uses different rate constants for semiinfinite and thin-layer diffusion, while Cases 2 and 3 use the same values for both models. For Case 1, ΔE_{pp} is lower for the thin-layer diffusion model than the semi-infinite diffusion model (Fig. 4.4 A). Use of the same rate constants for Cases 2 and 3 provides similar results (Figs. 4.4 B, C). Next, the experimental CVs of GCE (Fig. 4.6 A) and MWCNT/GCE (Fig. 4.6 E) are compared with the simulated CVs, where the kinetic parameters, including apparent standard heterogenous rate constants, transfer coefficients and formal potentials $E_{rds}^{0'}$ and $E_{non-rds}^{0'}$, are varied to obtain similar peak current ratios (I_A/I_{C2} and I_{C1}/I_A), as shown in Figs. 4.5 A, B. Again, peak-to-peak potential separation ΔE_{pp} is lower for the thin-layer diffusion model than the semi-infinite diffusion model (Fig. 4.5 C). Considering the timescale of the experiments, when FT is considerably depleted in the thin-layer cell, less current is drawn,^{15,18} which explains why ΔE_{pp} is lower for a thin layer.



Figure 4. 4. Simulated cyclic voltammograms for the semi-infinite and thin-layer diffusion models for (A) Case 1 that uses $k_{1,b}^0$, $k_{2,b}^0$ for the semi-infinite diffusion model and $k_{1,m}^0$, $k_{2,m}^0$ for the thin-layer diffusion model, (B) Case 2 using $k_{1,b}^0$ and $k_{2,b}^0$ for both models, and (C) Case 3 using $k_{1,m}^0$ and $k_{2,m}^0$ for both models, (D) Plot of peak-to-peak potential separation ΔE_{pp} vs. Log of the dimensionless layer thickness, *L*. The simulation parameters for all curves are $D = 6.9 \times 10^{-6}$ cm² s⁻¹, v = 0.1 V s⁻¹, $E_{rds}^{0\prime} = 5$ mV, $E_{non-rds}^{0\prime} = 1$ mV, and $C_b = 1$ mM.



Figure 4.5. Superimposed plots of simulated and experimental CV of (A) GCE and (B) MWCNT/GCE. (C) Simulated cyclic voltammograms for the semi-infinite and thin-layer diffusion models for the experimentally matched CVs. The simulation parameters for all curves are $D = 6.9 \times 10^{-6}$ cm² s⁻¹, v = 0.1 V s⁻¹, $E_{rds}^{0'} = 7$ mV, $E_{non-rds}^{0'} = 2$ mV, and $C_b = 1$ mM.

The FT response towards varying electrode surface characteristics is simulated for thin layers, where the dimensionless layer thickness *L* is varied and ΔE_{pp} for the simulated CVs plotted *vs*. Log(L) (Fig. 4.4 D). All other parameters are identical to those used for Case 1. As shown in Fig. 4.4 D, with the increase in layer thickness, the peak-to-peak potential separation ΔE_{pp} increases since the diffusion layer is now larger.¹⁵ For sufficiently large layer thicknesses, ΔE_{pp} approaches the value simulated by the semi-infinite model. Since, the apparent standard heterogenous rate constants are held constant, changes in electrode geometry alter FT mass transport, changing ΔE_{pp} .

4.5.2 Results obtained from CV experiments

Cyclic voltammetry is routinely used with GCE and MWCNT/GCE to characterize mass transport changes. The dependence of peak current on scan rate shows whether the process involves semi-infinite (*I vs.* $v^{1/2}$) or thin-layer (*I vs.* v) diffusion.²⁷ Both cases are evaluated

for all three voltammogram peaks (C_1 , A, and C_2) with the two electrodes. For the bare GCE (see Figs. 4.6 B, C and D), the peak current is linearly dependent on the square root of scan rate, confirming semi-infinite diffusion. For the MWCNT/GCE (Figs. 4.6 F, G and H), the peak current is instead linearly dependent on the scan rate itself, confirming thin-layer diffusion across the deposited nanotube interface on the GCE. Since FT transport across relatively small thin-layers is adsorption limited,²⁷ the measurements are made immediately after MWCNT/GCE exposure to the solution, which minimizes FT adsorption.



Figure 4. 6. Cyclic voltammetry response of 1 mM FT in 0.2 M ABS (pH 5) at various scan rates ranging from 50 mV/s to 175 mV/s at (A) GCE and (E) MWCNT/GCE. Variation of peak current with respect to the square root of scan rate, I vs. $v^{1/2}$, for three different peaks (B) C₁, (C) A, and (D) C₂ for 1 mM FT detection with a GCE. Plot of peak current *vs.* scan rate, I vs. v, for the (F) C₁, (G) A, and (H) C₂ peaks when 1 mM FT is detected by an MWCNT/GCE. The background solution contains 0.2 M ABS (pH 5) and the scan rates are varied from 50 mV/s to 175 mV/s.

Nanotube deposition on a bare electrode surface influences the peak-to-peak potential separation. The responses of bare planar and porous modified electrodes are experimentally validated, as shown in Fig. 4.7A, where a GCE modified with 1.5 µg MWCNT has $\Delta E_{pp} = 0.040$ V compared to 0.066 V for the GCE. The influence of varying MWCNT mass, a surrogate for *L*, is investigated for different amounts of nanotube deposition on GCEs (Fig. 4.7B). As the MWCNT mass on the GCE increases from 1.5 µg, both the oxidation (A) and reduction (C₂) peaks shift to higher over-potentials, increasing peak-to-peak separation. For 4.5 µg of MWCNT and up, the peak-to-peak separation reaches that of the GCE ($\Delta E_{pp} = 0.066$ V). That ΔE_{pp} saturates after a sufficiently large layer thickness is consistent with the simulation in Section 4.1. Hence, we induce that while thin-layer effects influence the FT electrochemical response of an MWCNT/GCE, the electrocatalytic property of the MWCNT remains unaltered.



Figure 4. 7. (A) Cyclic voltammograms showing peak-to-peak potential separations. The ΔE_{pp} responses of the GCE and MWCNT/GCE are compared for 1 mM FT. (B) Variation of ΔE_{pp} with respect to the MWCNT mass deposited on a GCE for 1 mM FT. The inset represents cyclic voltammograms for increasing MWCNT mass on a GCE for 1 mM FT. The scan rate is 50 mV/s.

4.6 Conclusion

MWCNT are deposited on GCE to modify planar electrodes to detect Fenitrothion (FT), an organophosphate (OP). We hypothesize that, as the thickness of the deposited MWCNT layer decreases, FT mass transport between the layers of porous multiwalled carbon nanotubes changes from semi-infinite to thin-layer diffusion. This influences the electrode electrochemical response to the analyte. Using simulations and experiments, we demonstrate thin-layer effects during FT detection by an MWCNT/GCE. These effects alter FT mass transport and influence the electrode electrochemical response to this chemical species. The change in FT diffusion as the deposition layer thickness decreases reduces the peak-to-peak potential separation during cyclic voltammetry. Simulations of the CV response of an electrode to FT are conducted with both the semi-infinite and thinlayer diffusion models for the planar (GCE) and modified porous (MWCNT/GCE) electrodes. We show that, along with the contribution of altered electrocatalysis, changes in FT diffusion should also be considered to understand the electrochemical response of nanotube-modified electrodes. The model is applicable for other toxic chemicals like 4nitrophenol, parathion and methyl parathion.

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4.8 Appendix/supplementary information

Simulation code for Fenitrothion at planar electrode

clc

clear all

close all

tic

```
%% user inputs (non dim)
```

sigma = 5643;	% dimensionless scan rate
dTheta = 0.194;	% dimensionless potential
r1 = 617;	% dimensionless rate constant for Step A
r2 = 75;	% dimensionless rate constant for Step B
epsilon = 1E-01;	% Electrode radius (cm)
D = 6.9E-06;	% Diffusivity (cm ² sec ⁻¹)
R = 8.314;	% Universal gas constant (J mol ^{^-1} K ^{^-1})
F = 96485.332;	% Faraday's constant (C mol^-1)
T = 298;	% Standard temperature (K)

$\mathbf{v} = (\mathrm{sigma} * \mathbf{R} * \mathbf{T} * \mathbf{D})$	/(epsilon^2 * F); %scan rate (V s^-1)		
dE = (dTheta*R*T)/	F; %Potential (V)		
k01 = (r1*D)/(epsilon)	on^2); %Apparent rate constant for Step A		
k02 = (r2*D)/(epsilon)	on); %Apparent rate constant for Step B		
%% dimensional solver			
Cba = 1E-06;	%Bulk concentration of FT (mol cm^-3)		
alfa1 = 0.3;	%Cathodic transfer coeff for Step A		
alfaP1 = 1-alfa1;			
alfa2 = 0.9;	%Cathodic transfer coeff for Step B		
alfaP2 = 1-alfa2;			
Area = 0.0706;	%Electode area (cm^2)		
EL = -0.8;	%Voltametry starting voltage		
ER = 0.5;	%Voltametry ending voltage		
EM = -0.2;			
Ef01 = -0.63;	%Apparent formal electrode potential for Step A		
EOrds = 0.005;	%Formal potential for second electron trasnfer of step B		
E0nonrds = 0.001;	%Formal potential for first electron trasnfer of step B		

tmax = 2*(abs(EL-ER)/v); %Maximum duration of simulation (s)

dt = dE/v;	%Duration of each time step (s)
N = 500;	%Number of grid points
L = 2*sqrt(D*tmax)	; %Span of simulation domain (cm)
dx = L/N;	%Space steps
t = dt:dt:tmax;	%Time vector
COA = Cba*ones(1,	N); %Initial concentraion field
aPO = dx/dt;	%Mass diffusion coeff for time neighbour
aE = D/dx;	% Mass diffusion for east face
aW = D/dx;	%Mass diffusion for west face
E(1) = EM;	%Initialization of potential vector
CA = C0A';	%Initialization of concentration vector
CB = (zeros(1,N))';	% Initialization of concentration vector
CC = (zeros(1,N))';	% Initialization of concentration vector
%% Voltage vector	evaluation for transient boundary condition

for m=1:length(t)

if m < (0.25*length(t))

E(m+1) = E(m) - dE;

elseif m < (0.75*length(t))

E(m+1) = E(m) + dE;

else

E(m) = E(m-1) - dE;

end

end

%% Main time loop

for m=1:length(t)

% coefficient matrix

% for specie A

MatA(1,1) = aE + aP0 + k01*(exp(((-(1+alfa1)*F)/(R*T))*(E(m)-Ef01)));

MatA(1,2) = -aE;

for i = 2:N-1

MatA(i,i) = aE + aW + aP0;

MatA(i,i-1) = -aW;

MatA(i,i+1) = -aE;

end

MatA(N,N-1) = -aW;

MatA(N,N) = aW + aP0 + ((2*D)/dx);

% for specie B

MatB(1,1) = aE + aP0 + k02*exp((1-alfa2)*((F)/(R*T))*(E(m)-E0rds));

MatB(1,2) = -aE;

for i = 2:N-1

MatB(i,i) = aE + aW + aP0;

MatB(i,i-1) = -aW;

MatB(i,i+1) = -aE;

end

MatB(N,N-1) = -aW;

MatB(N,N) = aW + aP0 + ((2*D)/dx);

% for specie C

MatC(1,1) = aE + aP0 + k02*exp(((-F)/(R*T))*(E0rds-E0nonrds))*exp((-F)/(R*T))*(E0rds-E0nonrds))*(E0rds-E0nonrds))*exp((-F)/(R*T))*(E0rds-E0nonrds))*(E

(1+alfa2))*(((F)/(R*T))*(E(m)-E0rds)));

MatC(1,2) = -aE;

for i = 2:N-1

MatC(i,i) = aE + aW + aP0;

MatC(i,i-1) = -aW;

MatC(i,i+1) = -aE;

end

MatC(N,N-1) = -aW;

MatC(N,N) = aW + aP0 + ((2*D)/dx);

%% source terms (spatial and temporal sources)

% for specie A

SourceA(1) = aP0*CA(1);

for i = 2:N-1

SourceA(i) = aP0*CA(i);

end

SourceA(N) = aP0*CA(N) + ((2*D*Cba)/dx);

% for specie B

SourceB(1) = aP0*CB(1) + CA(1)*k01*exp(-(((1+alfa1)*F)/(R*T))*(E(m)-Ef01)) + CC(1)*k02*exp(((-F)/(R*T))*(E0rds-E0nonrds))*exp((-(1+alfa2))*(((F)/(R*T))*(E(m)-E0rds)));

for i = 2:N-1

SourceB(i) = aP0*CB(i);

end

SourceB(N) = aP0*CB(N);

% for specie C

SourceC(1) = aP0*CC(1) + CB(1)*k02*exp((1-alfa2)*((F)/(R*T))*(E(m)-E0rds));

for i = 2:N-1

SourceC(i) = aP0*CC(i);

end

SourceC(N) = aP0*CC(N);

% use matrix inversion to find the concentration of species at current time

invMA = inv(MatA);

invMB = inv(MatB);

invMC = inv(MatC);

CA = invMA*(SourceA');

CB = invMB*(SourceB');

CC = invMC*(SourceC');

% plot concentration profiles in realtime

figure(1)

%subplot(3,1,1)

plot(CA,'r');

hold on

plot(CB,'b');

plot(CC,'k');

hold off

grid on

 $legend('FT (sp A)', '{FT_{red}} (sp B)', '{FT_{ox}} (sp C)')$

% Calculation of current repsonse at the electrode surface

I(m) = (F*D*(1/dx)*(4*(-CA(2)+CA(1))+(2*(-CC(2)+CC(1))))*Area); % dimensional current

pause(0.01)

end

%%

toc

Istar = (I*epsilon)/(F*Area*D*Cba); %dimensionless current

figure(2)

plot(E,Istar)

Istar = Istar';

 $\mathbf{E} = \mathbf{E'};$

Simulation code for Fenitrothion at porous electrode

clc

clear all

close all

tic

%% user inputs (non dim)

sigma = 5643;	% dimensionless scan rate
dTheta = 0.194;	% dimensionless potential

- r1 = 950; % dimensionless rate constant for Step A
- r2 = 120; % dimensionless rate constant for Step B

epsilon = $1E-01$;	% Electrode radius (cm)
D = 6.9E-06;	% Diffusivity (cm ² sec ⁻¹)
R = 8.314;	% Universal gas constant (J mol^-1 K^-1)
F = 96485.332;	% Faraday's constant (C mol^-1)
T = 298;	% Standard temperature (K)
v = (sigma*R*T*D	D)/(epsilon^2 * F); %scan rate (V s^-1)
dE = (dTheta*R*T))/F; %Potential (V)
k01 = (r1*D)/(epsi	lon^2); % Apparent rate constant for Step A
k02 = (r2*D)/(epsi	lon); %Apparent rate constant for Step B
%% dimensional s	olver
Cba = 1E-06;	%Bulk concentration of FT (mol cm^-3)
alfa1 = 0.95;	%Cathodic transfer coeff for Step A
alfaP1 = 1-alfa1;	
alfa2 = 0.775;	%Cathodic transfer coeff for Step B
alfaP2 = 1-alfa2;	
Area = 1;	%Electode area (cm^2)
EL = -0.8;	%Voltametry starting voltage

- ER = 0.5; %Voltametry ending voltage
- EM = -0.2;
- Ef01 = -0.61; % Formal electrode potential for Step A
- E0rds = 0.005; % Formal potential for second electron trasnfer of step B
- E0nonrds = 0.001; % Formal potential for first electron trasnfer of step B
- tmax = 2*(abs(EL-ER)/v); %Maximum duration of simulation (s)
- dt = dE/v; %Duration of each time step (s)
- N = 500; %Number of grid points
- L = 0.2*sqrt(D*tmax); %Span of simulation domain (cm)
- dx = L/N; % Space steps
- t = dt:dt:tmax; % Time vector
- COA = Cba*ones(1,N); % Initial concentration field
- aPO = dx/dt; % Mass diffusion coeff for time neighbour
- aE = D/dx; % Mass diffusion for east face
- aW = D/dx; % Mass diffusion for west face
- E(1) = EM; % Initialization of potential vector
- CA = C0A'; % Initialization of concentration vector (species A)

CB = (zeros(1,N))'; %Initialization of concentration vector (species B)

CC = (zeros(1,N))'; %Initialization of concentration vector (species B)

%% Voltage vector evaluation for transient boundary condition

for m=1:length(t)

if m < (0.25*length(t))

E(m+1) = E(m) - dE;

elseif m < (0.75*length(t))

E(m+1) = E(m) + dE;

else

E(m) = E(m-1) - dE;

end

end

%% Main time loop

for m=1:length(t)

% coefficient matrix

% for specie A

MatA(1,1) = aE + aP0 + k01*(exp(((-(1+alfa1)*F)/(R*T))*(E(m)-Ef01)));

MatA(1,2) = -aE;

for i = 2:N-1

MatA(i,i) = aE + aW + aP0;

MatA(i,i-1) = -aW;

MatA(i,i+1) = -aE;

end

MatA(N,N-1) = -aW;

MatA(N,N) = aW + aP0;

% for specie B

MatB(1,1) = aE + aP0 + k02*exp((1-alfa2)*((F)/(R*T))*(E(m)-E0rds));

MatB(1,2) = -aE;

for i = 2:N-1

MatB(i,i) = aE + aW + aP0;

MatB(i,i-1) = -aW;

MatB(i,i+1) = -aE;

end

MatB(N,N-1) = -aW;

MatB(N,N) = aW + aP0;

% for specie C

MatC(1,1) = aE + aP0 + k02*exp(((-F)/(R*T))*(E0rds-E0nonrds))*exp((-(1+alfa2))*(((F)/(R*T))*(E(m)-E0rds)));

MatC(1,2) = -aE;

for i = 2:N-1

MatC(i,i) = aE + aW + aP0;

MatC(i,i-1) = -aW;

MatC(i,i+1) = -aE;

end

MatC(N,N-1) = -aW;

MatC(N,N) = aW + aP0;

%% source terms (spatial and temporal sources)

% for specie A

SourceA(1) = aP0*CA(1);

for i = 2:N-1

SourceA(i) = aP0*CA(i);

end

SourceA(N) = aP0*CA(N);

% for specie B

SourceB(1) = aP0*CB(1) + CA(1)*k01*exp(-(((1+alfa1)*F)/(R*T))*(E(m)-Ef01)) + CC(1)*k02*exp(((-F)/(R*T))*(E0rds-E0nonrds))*exp((-(1+alfa2))*(((F)/(R*T))*(E(m)-E0rds)));

for i = 2:N-1

SourceB(i) = aP0*CB(i);

end

SourceB(N) = aP0*CB(N);

% for specie C

SourceC(1) = aP0*CC(1) + CB(1)*k02*exp((1-alfa2)*((F)/(R*T))*(E(m)-E0rds));

for i = 2:N-1

SourceC(i) = aP0*CC(i);

end

SourceC(N) = aP0*CC(N);

% use matrix inversion to find the concentration of species at current time

invMA = inv(MatA);

invMB = inv(MatB);

invMC = inv(MatC);

CA = invMA*(SourceA');

CB = invMB*(SourceB');

CC = invMC*(SourceC');

% plot concentration profiles in realtime

figure(1)

plot(CA,'r');

hold on

plot(CB,'b');

plot(CC,'k');

hold off

grid on

 $legend('FT (sp A)', '{FT_{red}} (sp B)', '{FT_{ox}} (sp C)')$

% Calculation of current repsonse at the electrode surface

 $I(m) = (F^*D^*(1/dx)^*(4^*(-CA(2)+CA(1))+(2^*(-CC(2)+CC(1))))^*Area); \% \text{ dimensional current}$

pause(0.01)

end

%%

toc

Istar = (I*epsilon)/(F*Area*D*Cba); %dimensionless current

% Theta = $E^{*}(F/(R^{*}T));$

figure(2)

plot(E,Istar)

Istar = Istar';

E = E';

5 Multiwalled Carbon Nanotubes Coated with Nitrogen-Sulfur Co-Doped Activated Carbon for Detecting Fenitrothion

This chapter is reproduced from *Multiwalled Carbon Nanotubes Coated with Nitrogen-Sulfur Co-Doped Activated Carbon for Detecting Fenitrothion, ACS Applied Nano Materials* 2021, 4,5, 47814789, <u>Krishna Jangid</u>, Rakesh P. Sahu, Richa Pandey, Ri Chen, *Igor Zhitomirsky, and Ishwar K. Puri, <u>https://doi.org/10.1021/acsanm.1c00376</u>, Copyright © 2021, with permission from ACS. The author of this thesis is the first author and the primary contributor to this publication.*

5.1 Abstract

A nonenzymatic electrochemical sensor is drop casted for sensitive and specific fenitrothion (FT) detection, where a glassy carbon electrode (GCE) is modified by depositing an ink containing nitrogen-sulfur co-doped activated carbon coated multiwalled carbon nanotubes (NS-AC-MWCNT). We provide a method for NS-AC-MWCNT synthesis as follows. First, polypyrrole is coated on multiwalled carbon nanotubes (PPy-MWCNT). Next, the PPy coating is carbonized and chemically activated to enhance specific surface area. The activated carbon coating, co-doped with nitrogen-sulfur improves its surface electrical conductivity and electrocatalytic response as an electrode. Electrochemical impedance spectroscopy (EIS) confirms enhanced charge transfer for the NS-AC-MWCNT/GCE in comparison with MWCNT/GCE and GCE. Scanning electron

microscopy (SEM), and high-resolution transmission electron microscopy (HRTEM) with fast Fourier transform (FFT) elucidate the morphology of the material, and X-ray photoelectron spectroscopy (XPS) and Raman spectroscopy its chemical and structural characteristics. An NS-AC-MWCNT-containing ink is prepared and deposited on a GCE to fabricate a sensor to detect FT. The volume of NS-AC-MWCNT ink deposited, buffer solution pH, accumulation potential V_{pp} and time t_{pp} , and square wave voltammetry (SWV) parameters are optimized. For 0.05 - 40 μ M FT concentrations, the sensor provides a linear current response with a 4.91 nM limit of detection (LOD) with a signal to noise (S/N) ratio of 3. Chemical interferents have negligible influence on FT detection. The sensor detects FT in real lake and tap water samples.

5.2 Introduction

The use of pesticides to control weeds, insect infestations and diseases has increased steadily to meet rising global food demand, contributing to pesticide accumulation in the environment that is harmful to humans and other life forms.¹ Pesticide runoff contaminates surface and groundwater, increasing its toxicity towards microorganisms, which reduces soil fertility.²⁻³ Since their introduction in the 1960s, the consumption of organophosphorus compounds (OP) that replace dichlorodiphenylytrichloroethane (DDT) has consistently increased.⁴ Used in agricultural fields, gardens and commercial areas, OP are the most used pesticides, contributing 70% towards overall pesticide consumption.⁵⁻⁶ Continuous OP exposure produces severe and fatal physiological effects in humans, including mental disorders such as depression and anxiety, headache, blurred vision, nausea, vomiting, lowered heart rate, loss of coordination, fever, coma, and death.⁷ The World Health

Organization reports that over 750,000 people suffer from pesticide poisoning yearly, of whom 350,000 die.⁸ Human pesticide poisonings and related illness are estimated to have an annual economic cost of \$1 billion in the United States alone.⁹

Several analytical methods are used to detect OP, including gas or high performance liquid chromatography,¹⁰ and mass spectrometry.¹¹ These techniques are typically complicated, costly and require complex sample procedures and trained professionals to operate, limiting their operation in field conditions.¹² An electroanalytical method based on inhibition of immobilized acetylcholinesterase (AChE) enzyme¹³⁻¹⁴ is used for rapid onsite pesticide analysis. However, it cannot provide real-time pesticide analysis with confidence because of the poor chemical and physical stability of enzymes due to their denaturation, sensitivity to the chemical environment (e.g., pH), and chemical degradation during fabrication, storage, and use.¹⁵

The sensitivity of an electroanalytical sensor is influenced by its specific surface area, electrical conductivity, thereby, electrocatalytic property of the working electrode.¹⁶⁻¹⁹ The higher surface area available with nanomaterials amplifies the analyte signal response. Carbon nanomaterials (e.g., carbon nanotubes and graphene), conductive polymers (e.g., PANI, PEDOT and Polypyrrole) and metal nanoparticles (e.g., Au. Ag and Pt) have been investigated in this regard.²⁰⁻²² Porous materials, such as activated carbon (AC), with a higher available surface area, are used to detect heavy metal ions, e.g., Cd (II), Pb (II), Cu (II) and Hg (II).²³ The conductivity of porous carbon materials can be increased with heteroatom doping.²⁴⁻²⁸ The nitrogen-sulfur co-doping of AC also supplies additional valence electrons and enhances conductivity. Since it is more electronegative

than carbon, nitrogen doping polarizes surrounding carbon atoms, enhancing their electrocatalytic activity through a higher charge density. The atomic size difference between sulfur and carbon changes interlayer spacing and orbital matching in AC, creating additional active sites that enhances its electrocatalytic behavior.²⁹

Our objective is to synthesize and fabricate an AC coated MWCNTs for detecting organophosphates with low resistance and high sensitivity. We hypothesize that the activated carbon coating co-doped with nitrogen-sulfur on multiwalled carbon nanotubes, increases both the surface area and surface electrical conductivity. Concisely, MWCNTs act as a supporting structure for the NS-AC coating, where AC coated MWCNTs provide a higher electroactive surface area, enhancing the number of active sites available for OP detection. NS co-doping of AC improves the electrical conductivity of the electrode surface and promotes faster charge transfer between FT and the electrode. The enhanced electrocatalysis provides the motivation for modifying a glassy carbon electrode (GCE) to detect OP. Hence, we synthesize a novel nitrogen-sulfur co-doped activated carbon coated multiwalled carbon nanotube (NS-AC-MWCNT) composite and deposit it in the form of an ink over GCE to fabricate a sensor that serves as an electrode to detect the OP surrogate fenitrothion (FT) with SWV.

Synthesis of N-AC-MWCNT has been accomplished by carbonizing PPy-MWCNT, prepared using cetrimonium bromide (CTAB) and ammonium persulfate (APS) as dispersant and oxidant, respectively.³⁰⁻³¹ Sulfur and nitrogen co-doped carbon nanotubes were synthesized using methyl orange as a dopant and aniline as a nitrogen source.³² We have a conceptually different approach by utilizing pyrrole as the nitrogen source and Eriochrome cyanine (ECR) as the multifunctional dopant to prepare our material. ECR has advantages over CTAB and methyl orange, since the structure of the aromatic compound promotes carbon nanotube adsorption, and the charged groups promote electrostatic repulsion between the nanotubes, improving nanotube dispersion. Charged groups on aromatic compound are also beneficial for polymerization. Therefore, as a multifunctional dopant, ECR provides better nanotube dispersion and a better polypyrrole coating.³³⁻³⁴

Below, we (1) describe the synthesis of NS-AC-MWCNT by carbonizing PPy-MWCNT prepared with ECR and APS, a new method to fabricate a composite that has higher conductivity and surface area for effective electrochemical sensing; (2) report the first use of an NS-AC-MWCNT ink to detect FT with square wave voltammetry; (3) characterize the influence of the amount of ink deposited on the GCE, buffer solution pH, accumulation potential and time, and SWV parameters on the wave shape and current response of the FT sensor; (4) confirm sensor sensitivity and specificity for FT detection; (5) confirm use of the sensor to detect FT in real lake and tap water samples; and (6) compare the sensing performances of differently modified electrodes..

5.3 Materials and Methods

5.3.1 Chemicals and instruments

Fenitrothion (FT, analytical grade), pyrrole (Py, purity > 98%, stored at 4°C before use), sodium acetate buffer solution (ABS, molecular biology grade), potassium hydroxide (KOH), dichlorvos, chlorpyrifos, ammonium persulfate (APS), and eriochrome cyanine (ECR) were purchased from Sigma-Aldrich (Canada). Multiwalled carbon nanotubes (MWCNTs) with an outside diameter of 13 nm, inner diameter of 4 nm and lengths between 1-2 µm were obtained from Bayer Inc. (Germany). Deionized water (DI) and all reagents were used without further modification. The pH of the buffer solution was adjusted using a pH meter with 0.1 M NaOH and 0.1 M HCl solutions. Stock solutions of FT were prepared in ethanol and stored in a refrigerator at 4°C before use. Laboratory tap water and Lake Ontario lake water from Bayfront Park in Hamilton, Canada were used as is to detect spiked FT.

Cyclic voltammetry (CV), square wave voltammetry (SWV) and electrochemical impedance spectroscopy (EIS) were performed with a PARSTAT 2273 potentiostat (Princeton Applied Research, USA) using its conventional three electrode configuration that consists of a glassy carbon electrode as the bare working electrode, platinum wire as counter electrode and Ag/AgCl as reference electrode, all immersed in ABS. Chemical analysis was performed by XPS (Physical Electronics, model Quantera II) with a monochromatic Al-K alpha X-ray source at 1486.6 eV. Raman spectroscopy was conducted with a Renishaw inVia spectrometer at a 514 nm laser excitation by drop-casting 4 μ L of the sample on a glass substrate. The synthesized material was characterized by SEM (JEOL 6610LV), HRTEM (FEI Titan 80-300 LB), and FFT performed with imageJ software.

5.3.2 Synthesis of PPy coated MWCNT

Polypyrrole (PPy) coated MWCNTs were synthesized through chemical polymerization³⁵⁻³⁶ utilizing APS as oxidant and ECR as a dopant for Py and dispersant for MWCNTs,³³ as shown in the schematic in Figure 5. 1A. 200 mg of MWCNTs were homogenously

suspended with ultrasonication in 20 mg of ECR dissolved in 200 mL DI water. The resulting suspension was stirred at 4°C in an ice bath to which 800 mg of Py was added. 3.4 g of APS was dissolved in 15 mL of DI water, added to the MWCNT suspension, and stirred for 20 h in an ice bath at 4 °C. The synthesized material was washed with DI water and dried in an oven at 70 °C.

5.3.3 Synthesis of NS-AC-MWCNT

Nitrogen-sulfur co-doped activated carbon coated MWCNT (NS-AC-MWCNT) were synthesized through chemical activation and carbonization (Figure 5. 1A).³⁰⁻³¹ After synthesis, 1 g of PPy-coated MWCNT was dispersed in 25 mL of 4M KOH and the suspension was stirred and dried in air at 80 °C to form a powder. The powder was activated and carbonized under a nitrogen atmosphere in a tube furnace, first at 700 °C for 2 h and then cooled to room temperature. The resulting filtrate was made pH neutral by washing it with 1 M HCl and DI water after which it was dried for 12 hours in an oven at 70 °C. The synthesized NS-AC-MWCNT material was dispersed in ethanol to form a uniform 1 mg/mL suspension. Different deposition volumes of the material were drop casted on GCE and dried in air to obtain electrodes.



Figure 5.1. (A) Schematic of NS-AC-MWCNT synthesis, where prepared PPy coated MWCNT are activated with KOH and carbonized at 700 °C. (B) Schematic of the sensor fabrication. NS-AC-MWCNT modification of GCE is performed with drop casting the ink on GCE which is used to detect FT in ABS via SWV.

The fabrication of the NS-AC-MWCNT/GCE electrochemical sensor is illustrated in Figure 5. 1B. The electrodes were electrochemically activated by successively scanning them in an acetate buffer (pH 5) from 0.1 V to 1.2 V (at a scan rate of 100 mV/s) until stabilized. CV or SWV were utilized to obtain the current response of FT. CVs were scanned for 50 μ M FT in a 0.2M acetate buffer (pH 5.0) at a scan rate of 50 mV/s between -0.8 and 0.5 V *vs.* Ag/AgCl. CV was also performed to determine the electroactive surface area of different electrodes in 1 mM Fe(CN)₆³⁻/Fe(CN)₆⁴⁻ / 0.1 M KCl solution for scan rates from 25 to 200 mV/s. SWV was performed in 0.2M acetate buffer to determine the optimum amount of NS-AC-MWCNT deposited, pH of the acetate buffer, accumulation potential, accumulation time, and SWV parameters, such as amplitude, step potential, and frequency. SWV was utilized to analyze the FT response. The optimised SWV conditions were a 10 Hz frequency, 10 mV step potential and 140 mV amplitude. The limit of detection (LOD) of the sensor was calculated from standard deviation σ of the current response of the NS-AC-MWCNT/GCE in ABS at optimised conditions when no FT was present and the slope of the linear regression fit of the calibration curve *m* for samples containing FT using the formula LOD = $3\sigma/m$.³⁷ Sensor selectivity was examined by introducing tenfold excess concentrations, i.e., 200 µM, above the FT concentration of nine commonly found salt interferents and organophosphates (dichlorvos and chlorpyrifos) in drinking or usable water. The salts were of Ni²⁺ (NiSO₄.6H₂O), Zn²⁺ (ZnSO₄.7H₂O), Mn²⁺ (Mn(CH₃COO)₂.4H₂O), Co²⁺ (CoSO₄.7H₂O), Fe³⁺ (H₂Fe₂O₁₃S₃), Cl⁻ (NaCl), PO4³⁻ (Na₃PO₄), NO₃⁻ (KNO₃) and SO4²⁻ (K₂SO₄), in 20 µM of FT in ABS. Lake water was filtered with 8 µm pore size filter paper. Thereafter, 0.2 M (pH =5) solutions of lake water and laboratory tap water samples were obtained by adding 2 mL of 2 M acetate buffer to each 18 mL sample. These samples were spiked with known concentrations of FT and a linear regression fit used to calculate the FT concentration.

5.4 Results and discussions

5.4.1 Material characterization

The morphology of the synthesized NS-AC-MWCNT is observed from SEM and HRTEM images and FFT sample analysis. Figure 5. 2A presents an SEM image that shows the microstructure of the randomly interconnected nanotubes forming a spongelike architecture, illustrating the three dimensionalities of the material. Carbonization of PPy coated MWCNT produces AC coated MWCNT.³⁰⁻³¹ Figure 5. 2B shows an HRTEM image of the NS-AC-MWCNT with the region of crystalline carbon nanotube and amorphous coating of AC on MWCNT being marked by solid and dashed lines respectively. The FFT

analysis³⁸ of the marked regions in the HRTEM image are presented in Figure 5. 2C,D. Well-defined spots in Figure 2C corresponds to the (002) and (100) crystalline planes of carbon nanotube.³⁹ Blurred spots correspond to the amorphous activated carbon coating on the outer surfaces of the nanotubes (Figure 5. 2D). The obtained core-shell architecture of the AC coated MWCNT is in agreement with the previously reported work.⁴¹



Figure 5.2. Morphological characterization of the composite material: (A) SEM and (B) HRTEM images, and (C, D) FFT images of the marked regions in the HRTEM image. The

crystalline (C) and amorphous (D) locations are identified by solid and dashed lines in (B) that correspond to the carbon nanotubes and activated carbon coating.

The XPS survey spectra in Figure 5. 3A show weak peaks for K 2s and K 2p that occur due to KOH treatment of the PPy-MWCNT, as well as peaks for C 1s, O 1s, O Kll, S 1s, S 2p and N 1s. Elemental composition from the XPS data reveals carbon, nitrogen, sulfur, oxygen, chlorine, and potassium in NS-AC-MWCNT are present in 78.7, 2.3, 2.3, 10.6, 1.2 and 5.1 wt %, respectively (Table 5. S1). The higher resolution N 1s spectrum of Figure 5. 3B reveals peaks at 398.4, 400.2 and 400.9 eV corresponding to the N-6 (pyridinic), N-5 (pyrrolic) and N-Q (quaternary) nitrogen groups, respectively.^{30, 40} Outernary and pyridinic nitrogen groups have a strong electron donor tendency⁴¹ that enhances electron transfer and improves sensor sensitivity. In addition to the C-C (284.7 eV), C-OH (285.7 eV), C=O (287.4 eV) and COOH (290.8 eV) peaks of C 1s of MWCNT (Figure 5. S1A), the NS-AC-MWCNT spectrum provides another C-N (285.6 eV) peak with C-C (284.8 eV) and C=O (287.5 eV) peaks (Figure 5. S1B) that correspond to nitrogen doping.³⁰ The S 2p spectra of Figure 5. 2C contain peaks at 165.7, 168.4 and 171.4 eV for sulfoxide, sulfate, and sulfite groups⁴² that confirm sulfur doping. PPy served as a source for nitrogen-containing groups while multifunctional dopant, ECR and oxidant, APS served as the sources for sulfur-containing groups. The O 1s spectra for both MWCNT (Figure 5. S1C) and NS-AC-MWCNT (Figure 5. S1D) show oxygen-containing C=O, C-OH, and COOH groups.



Figure 5. 3. (A) XPS survey spectra for NS-AC-MWCNT and pristine MWCNT. (B, C) Higher resolution XPS spectra for N 1s and S 2p for NS-AC-MWCNT confirm nitrogen-(N-5, N-6, and N-Q) and sulfur-comprising groups (sulfoxide, sulfate, and sulfite). (D) Raman spectra for pristine MWCNT and NS-AC-MWCNT reveal D and G bands, where the intensity ratio of the G band to D band I_G/I_D indicates the degree of graphitization.

Raman spectroscopy reveals the degree of ordered structure, defects, and hybridization⁴³⁻⁴⁴ and evaluates the extent of graphitization that influences the electrical conductivity.⁴⁵⁻⁴⁶ Raman spectra for MWCNT and NS-AC-MWCNT (Figure 5. 2D) show two peaks at 1343.5 cm⁻¹ and 1574.1 cm⁻¹ for the D and G bands, respectively, where the D band is associated with disordered carbon (sp³) and the G band with graphitic carbon

 (sp^2) .⁴³ The intensity ratio IG/ID is indicative of the degree of graphitization. For the MWCNT and NS-AC-MWCNT, IG/ID = 0.95 and 1.20, respectively, confirming the enhancement of graphitization, thereby electrical conductivity, in the latter composite material.

The electrochemical response of synthesized NS-AC-MWCNT composite is investigated using CV, SWV and EIS to understand the effect of different modifications on the charge transfer at the electrode-electrolyte interface. Figure 5. 4A shows the electrochemical impedance data in the Nyquist plot. Nyquist plots for GCE, MWCNT/GCE, and NS-AC-MWCNT/GCE are fitted per Randles equivalent circuit model.⁴⁷ The charge transfer resistance is calculated from the experimental data and the analysis of the equivalent circuit. There is a significant decrease in the charge transfer resistance R_{ct} at the interface of electrolyte and electrode surface when the GCE is coated with MWCNT, where $R_{ct} = 49.4 \ \Omega$ for GCE and 12.6 Ω for MWCNT/GCE. This R_{ct} decrease occurs due to the higher electrical conductivity and surface area for the MWCNT with respect to the GCE.⁴⁸⁻⁴⁹ For the NS-AC-MWCNT/GCE, R_{ct} further decreases to 5.4 Ω . This significant decrease in resistance is attributed to the associated increases in the electrochemically active surface area, therefore active sites available for FT detection, and electrical conductivity, thereby enhancing electrocatalysis by the NS-AC-MWCNT modified GCE.

The interaction of FT and the NS-AC-MWCNT is investigated with CV and SWV. We suggest π - π interactions are involved between FT and electrode. These interactions further facilitate the charge transfer by enhancing the reduction and oxidation capabilities of the electrode for the -NO₂ group on FT. Under a formal potential, the -NO₂ group first undergoes an irreversible reduction to the -NHOH group at a negative cathode potential of -0.59 V vs. Ag/AgCl, followed by a reversible oxidation at -0.005 V vs. Ag/AgCl and reduction of the -NO group at -0.06 V vs. Ag/AgCl during the subsequent reverse scan,²² as shown in Figure 5. 4B and Schematic 5. S1 in the Supplementary Information.

The CV and SWV electrochemical profiles for NS-AC-MWCNT/GCE in the presence of FT are compared with profiles for GCE and MWCNT/GCE under similar conditions in Figure 5. 4B, C. Compared to MWCNT/GCE, coating the MWCNT with nitrogen-sulfur co-doped activated carbon enhances the Faradaic current due to the higher electrocatalytic behavior of the modified electrode as discussed above. The electroactive surface areas of the GCE and modified electrodes with MWCNT and NS-AC-MWCNTT were evaluated with CV using the Randles-Sevcik equation¹⁸:

$$I_p = 2.69 * 10^5 n^{3/2} ACD^{1/2} v^{1/2}$$

where I_p denotes anodic peak current, *n* number of electrons, *A* electroactive surface area, *C* concentration of Fe(CN)₆³⁻/Fe(CN)₆⁴⁻, *D* diffusion coefficient and *v* scan rate. CV measurements are performed in a 1 mM Fe(CN)₆³⁻/Fe(CN)₆⁴⁻/0.1 M KCl solution for scan rates of 25, 50, 100, 150 and 200 mV/s. Figure 5. 4D shows the anodic peak current (I_p) *vs* square root of scan rate ($v^{1/2}$) for the GCE, MWCNT/GCE and NS-AC-MWCNT/GCE. From the slope of I_p *vs* $v^{1/2}$, the surface areas of the GCE, MWCNT/GCE and NS-AC-MWCNT/GCE electrodes are determined as 0.019, 0.037, 0.094 cm², respectively. Hence, with respect to the GCE, the electroactive surface area for the MWCNT/GCE increases 1.9-fold and by 4.9 fold for NS-AC-MWCNT/GCE.



Figure 5. 4. Electrochemical characterization of the sensor. (A) Nyquist plot for the three electrode modifications in a 1 mM Fe(CN)₆³⁻/Fe(CN)₆⁴⁻/0.1 M KCl solution $(0.1 - 10^5$ Hz, 5 mV). The inset shows the higher frequency range of the Nyquist plot for differently modified working electrodes: R_s , R_{ct} , C_{dl} and Z_w denote electrolyte resistance, charge transfer resistance, double layer capacitance and Warburg impedance, respectively. (B) Comparison of the cyclic voltammograms for (a) GCE in the absence of fenitrothion with (b) GCE, (c) MWCNT/GCE, and (d) NS-AC-MWCNT for 50µM FT in 0.2M acetate buffer solution (pH 5.0) at a scan rate of 50 mV/s. (C) SWVs for GCE (a: FT absent, b: FT present), and for (c) MWCNT/GCE and (d) NS-AC-MWCNT/GCE with 50µM FT in 0.2M acetate buffer solution (pH 5.0) at a scan rate of 50 mV/s. (D) Plots of I_p vs $v^{1/2}$ obtained from cyclic voltammograms of GCE, MWCNT/GCE, and NS-AC-MWCNT/GCE in 1 mM Fe(CN)₆³⁻/Fe(CN)₆⁴⁻/0.1 M KCl solution at scan rates: 25, 50, 100, 150 and 200 mV/s.

5.4.2 Sensor optimization

The current at the redox peak for SWV conducted at 0.0002 V with varying amounts of NS-AC-MWCNT deposited on the GCE increases with increasing amounts of NS-AC-MWCNT, as shown in Figure 5. S2, which can be attributed to the increase in surface coverage of the bare electrode. The current reaches a maximum for a 15 μ L deposition volume, but for larger depositions the response remains virtually the same. Hence, the 15 μ L deposition volume is selected for all further experiments.

The effect of varying the pH of acetate buffer solution from 3-6 on the peak current and peak oxidation potential is next investigated using SWV, as shown in Figure 5. Increasing the pH from 3 to 5 increases the current at the peak oxidation potential for the redox couple, but above pH = 5 this current decreases with increasing pH, as seen in Figure 5. 5A. The lowering of current is attributed to the decrease in the proton concentration required to reduce the -NO group of FT,⁵⁰ Thus, pH = 5 is identified as the optimum acidity for our investigation. In a 50 μ M FT sample, the peak oxidation potential becomes negative as the acetate buffer pH increases due to proton participation in the oxidation of -NHOH to -NO.²² Figure 5. 4B shows a linear relationship between the peak oxidation potential and solution pH of the form Peak potential (V) = -0.0569 pH + 0.2877 with R² = 0.9937 and a 56.9 mVpH⁻¹ slope, indicating equal 1:1 contributions from protons and electrons during the electrochemical reaction of FT.^{21-22, 51}



Figure 5. 5. (A) The effect of pH on the SWV for 50 μ M fenitrothion with an NS-AC-MWCNT modified GCE in 0.2 M acetate buffer solution. (B) Plot of peak potential (V) versus pH of the 0.2M acetate buffer for 50 μ M fenitrothion.

The accumulation potential V_{pp} and accumulation time t_{pp} are relevant when an analyte reaction, i.e., of FT, is adsorption-controlled. The influence of these parameters on the Faradaic current response is investigated by varying V_{pp} from -0.5V to -0.8V vs. Ag/AgCl and t_{pp} from 40-140s with magnetic stirring of the solution. Figure 5. S3(A) shows that peak current increases as V_{pp} (applied at $t_{pp} = 30$ s) increases from -0.5V to -0.8V vs. 0.6V vs. Ag/AgCl, followed by a decrease in current when V_{pp} is further increased to -0.8V vs. Ag/AgCl. Consequently, -0.6V vs. Ag/AgCl (Figure 5. S3(B)) is selected as the optimum V_{pp} for FT detection with SWV. The effect of t_{pp} on the peak current at -0.6V vs. Ag/AgCl is shown in Figure S3(C), where the current saturates at $t_{pp} = 100$ s (Figure 5. S3(D)). Hence, we select $V_{pp} = -0.6V$ vs. Ag/AgCl with $t_{pp} = 100$ s to develop the FT sensor.

The SWV parameters are optimized for current response and peak shape as shown in Figure 5. S4. The frequency is varied between 5-25 Hz (Figure 5. S4(A)) maintaining the step potential and amplitude at 10 mV and 50 mV, respectively. The maximum current response occurs at 10 Hz (Figure 5. S4(B)) and then decreases as the frequency increases.
Hence, 10 Hz is selected as the optimal frequency to optimize the step potential that is varied between 2 - 20 mV (Figure 5. S4(C)). The maximum current occurs at 10 mV (Figure 5. S4(D)), which we set as the optimum step potential. As shown in Figure 5. S4 (E), the current increases as the amplitude increases from 20-140 mV, following which it decreases slightly with further increases in the amplitude (Figure 5. S4 (F)). On this basis, the 140 mV amplitude at 10 Hz frequency and 10 mV step potential is selected to examine the SWV response towards FT.

5.4.3 Sensor performance

The analytical sensitivity of the sensor is investigated by spiking different concentration of FT in ABS, as shown in Figure 5. 6A. The sensor response provides the calibration curve in Figure 5. 6B (Table S2). The response is linear for 0.05 - 40 μ M FT concentrations, where the peak current response I_{pc} (μ A) = 5.09C (μ M) + 26.38 with R² = 0.98. The LOD for FT is 4.91 nM for S/N = 3 based on three replicates for each concentration, n = 3. This performance is compared with previously reported results in Table 1. The analytical performance of the NS-AC-MWCNT modified GCE sensor is comparable with CeO₂/rGO /GCE and RGO/DPA/PGE and better than of several other sensors which is attributed to its larger electroactive surface area and higher electrical conductivity of NS-AC-MWCNT compared to MWCNT, AuNPs, TiO₂, and peptide nanotubes (PNT).

 Table 5. 1. Voltametric detection of fenitrothion with different electrodes.

Electrode	Technique	Linear	LOD	Reference
		range (µM)	(µM)	
CeO ₂ /rGO/GCE	DPV*	0.025 - 2.0	0.003	52
RGO/DPA/PGE	SWV	0.096 -	0.00348	53
		1.912		
ZrO ₂ /rGO/MoS ₂ -Au/GCE	SWV	0.018 - 21.6	0.00794	16
AuNPs@GNIP-GR-	DPV*	0.010 - 5.0	0.008	54
IL/GCE				
Nano-TiO ₂ /GCE	DPV*	0.025 - 10.0	0.010	55
PNT/PGE	SWV	0.114 –	0.0196	56
		1.172		
MWCNT/GCE	SWV	0.20 - 60.0	0.080	22
TiO ₂ /nafion/GCE	DPV*	0.20 - 4.0	0.0866	57
NS-AC-MWCNT/GCE	SWV	0.050 - 40.0	0.00491	This work
*DPV-Differential Pulse Vo	ltammetry			

The selectivity of the NS-AC-MWCNT modified GCE sensor is investigated by spiking organophosphates (dichlorvos and chlorpyrifos) and the salts of nine commonly found interferents in drinking or usable water, namely, Ni²⁺, Zn²⁺, Mn²⁺, Co²⁺, Fe³⁺, Cl⁻, PO₄³⁻, NO₃⁻ and SO₄³⁻ in FT containing ABS. Figure 5. 6C,D shows that a tenfold excess concentration of any salt interferent above the FT concentration has a negligible effect on the Faradaic current for the optimised FT sensing conditions. The shape of the peak was observed to be influenced by the presence of interferents. It is suggested that interactions

of ions with FT in solution likely influence the mass transport of FT and hence the kinetics of the electrode reactions. On the other hand, dichlorvos and particularly chlorpyrifos influence FT detection. The effect of the latter compound is attributed to passivation of the electrode surface due to chlorpyrifos adsorption, possibly through π - π interactions between chlorpyrifos and the NS-AC-MWCNT/GCE. Overall, the results support the selective sensing ability of the sensor for detecting FT in contaminated water.



Figure 5. 6. (A) SWVs for 0.05 to 40 μ M fenitrothion concentrations measured with the NS-AC-MWCNT modified GCE in 0.2 M acetate buffer solution at pH = 5. (B) Calibration of I_{pc} (peak current, μ A) *vs*. FT concentration (μ M) detected by the sensor. (C) Sensor SWV response to 20 μ M FT concentration at pH = 5 in ABS in the presence of different

 $200 \ \mu\text{M}$ (10× or tenfold) concentrations of interferent salts species. (D) The percentage change in the current response to the FT solution spiked with different interferent species with respect to the response towards FT alone, i.e., in the absence of interferents.

After confirming the sensitivity and selectivity of the NS-AC-MWCNT/GCE sensor, we explore its applicability for detecting FT in realistic lake and tap water samples spiked with known FT concentrations. The sensor reports FT concentrations that equal 86.4 - 90.6% of the spiked concentrations with maximum RSD of 8.2% (number of replicates, n = 3) for lake water, where this recovery is a larger 91.2-97.9% for tap water with maximum RSD of 2.7% (n = 3), as reported in Table 5. 2. The lower FT recovery for lake water are attributed to signal loss due to co-adsorption of lake water components, consisting of microorganisms, heavy metal ions and waste,²⁸ on the electrodes, leading to their subsequent passivation and fouling. Nevertheless, the sensor can detect FT in contaminated water bodies. Sensor stability was assessed by storing NS-AC-MWCNT/GCE for 6 days at 4 °C after which the sensor response towards 20 μ M FT was 86.1% of the initial current response. Overall, the sensor exhibits excellent reproducibility since the RSD is lower than 5% for all FT concentrations in the 0.05 μ M to 40 μ M range.

Table 5. 2. Recovery of various concentrations of spiked fenitrothion in lake and tap water (n = 3, where n is the number of replicates for each of the spiked FT concentration).

Sample type	Spiked fenitrothion concentration, (µM)	Found, (µM)	Recovery, (%)
T also suctors	20	18.12 ± 1.07	90.6 ± 5.3
Lake water			

	5	4.32 ± 0.41	86.4 ± 8.2
	20	19.51 ± 0.28	97.5 ± 1.4
Tap water	10	9.12 ± 0.27	91.2 ± 2.7
	5	4.89 ± 0.10	97.9 ± 2.0

5.5 Conclusion

We describe the facile fabrication of an effective, simple, and rapid electrochemical sensor to detect fenitrothion. A novel NS-AC-MWCNT composite is synthesized for use as the sensing material. The charge transfer resistance of the NS-AC-MWCNT/GCE electrode is 5.4 Ω compared to 12.6 Ω of the MWCNT/GCE electrode. The amorphous coating of activated carbon around the nanotube composite has a spongelike porous microstructure, which increases the overall surface area. Compared with GCE, MWCNT/GCE and other reported electrodes, the NS-AC-MWCNT/GCE electrode has a higher sensitivity of FT detection, as low as 4.91 nM with a signal to noise ratio of 3. Tenfold concentrations of interferents salts of Ni²⁺, Zn²⁺, Mn²⁺, Co²⁺, Fe³⁺, Cl⁻, PO4³⁻, NO3⁻, and SO4³⁻ and organophosphates (dichlorvos and chlorpyrifos) above the FT concentration do not have a significant effect on its detection. With 5 μ M FT spiked samples, the sensor satisfactorily recovers 86.4 ± 8.2 % of FT in real lake and 97.9 ± 2.0 % in tap water samples.

5.6 References

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5.7 Appendix/supplementary information



Schematic 5. S1. Electrochemical reactions of FT due to -NO₂ group.

Table 5. S1. XPS elemental composition for NS-AC-MWCNT and pristine MWCNT.

 С	Ν	S	0	Cl	K
(wt %)					

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NS-AC-MWCNT	78.7	2.3	2.3	10.6	1.2	5.1
Pristine MWCNT	98.5	-	-	1.5	-	-



Figure 5. S1. High-resolution XPS spectra for (A, C) pristine MWCNT and (B, D) NS-AC-MWCNT for (A, B) C 1s and (C, D) O 1s, confirming the C-N (285.6 eV) peak and several oxygen containing groups (C=O, C-OH, and COOH).



Figure 5. S2. Influence of the amount of NS-AC-MWCNT deposited on a GCE on the peak current (0.0002 V, SWV voltammogram) for 50 μ M fenitrothion in 0.2 M acetate buffer solution at pH = 5.



Figure 5. S3. SWVs of NS-AC-MWCNT/GCE for 50 μ M fenitrothion in 0.2 M acetate buffer solution at pH = 5 for (A) V_{pp} (-0.5, -0.6 V, -0.7 and -0.8 V *vs*. Ag/AgCl) at t_{pp} 30s and (C) t_{pp} (40, 60, 80, 100, 120, 140s) at V_{pp} -0.6 V *vs*. Ag/AgCl. (B, D) The effect of varying V_{pp} and t_{pp} on the peak current for 50 μ M fenitrothion.



Figure 5. S4. Effect of SWV frequency, step potential and amplitude with a NS-AC-MWCNT/GCE for 50 μ M fenitrothion in 0.2M acetate buffer solution at pH = 5. (A) SWVs at 5, 10, 15, 20 and 25 Hz frequencies with a 10 mV step potential and 50 mV step amplitude, where the maximum current occurs at (B) a 10 Hz frequency. (C) Effect of varying 2, 5, 10, 15 and 20 mV step potentials at 10 Hz frequency and 50 mV step amplitude on fenitrothion detection, where the maximum current occurs at (D) a step potential of 10 mV. (E) SWVs of varying 20, 40, 60, 80, 100, 120, 140, 160 and 180 mV step amplitudes at a 10 Hz frequency and 10 mV step potential, where the maximum current F occurs at a 140 mV step amplitude.

Concentration of FT (µM)	Average current (A)	Standard deviation (A)
0.05	2.53E-5	2.10E-7
0.1	2.87E-5	8.92E-7
0.5	3.05E-5	1.21E-6
1	3.53E-5	1.47E-6
		2 0

Table 5. S2. Raw data for different concentration of FT used in the calibration plot (Figure 5. 6B).

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1.25	3.95E-5	6.11E-7
2.5	5.01E-5	1.43E-6
5	5.31E-5	1.48E-6
7.5	7.15E-5	3.02E-6
10	8.17E-5	2.39E-6
20	1.30E-4	2.85E-6
40	2.24E-4	1.96E-6

6 Surface modified metal oxides deliver specific ultrasensitive electrochemical detection of organophosphate, heavy metal, and nutrient

This chapter is reproduced from the manuscript that is submitted for peer review to *Surface* modified metal oxides deliver specific ultrasensitive electrochemical detection of organophosphate, heavy metal, and nutrient, ACS Sensors (2022), <u>Krishna Jangid</u>, Rakesh P. Sahu, Sadman Sakib, Igor Zhitomirsky, and Ishwar K. Puri.

The author of this thesis is the first author and the primary contributor to this manuscript.

6.1 Abstract

Reliable on-site water pollutant detection requires highly sensitive and specific nonenzymatic electrochemical sensors. One method is to incorporate agglomeration-free detecting agents specific to the water pollutant within the ink used to print or modify an electrode. We describe a simple and effective method to produce highly electrocatalytic electrodes with well dispersed ZrO₂, MnO₂, and MgO. These inorganic metal oxides serve as detecting agents with specific affinity towards fenitrothion (FT, an organophosphate), lead (Pb²⁺, a heavy-metal ion), and nitrite (NO₂⁻, a nutrient), respectively. Our bioinspired method utilizes a catechol molecule, 3,4-dihydroxylbenzaldehyde (DHBA), and filmforming polymer chitosan (CHIT). Redox-active DHBA strongly adsorbs the metal oxide through bidentate bonding, and CHIT-linked DHBA assists in stable film formation. The metal oxide surface modification through the DHBA-CHIT chemistry enhances the electronic, film-forming, and nanoparticle dispersing properties of the electrode modifying ink. The ink is drop cast on nitrogen-sulfur co-doped activated carbon coated carbon nanotubes (NS-AC-MWCNT) modified glassy carbon electrode (GCE). This enhances the total surface area of the X-DHBA-CHIT/NS-AC-MWCNT/GCE electrode (where X denotes ZrO₂, MnO₂ or MgO) and its electrocatalytic properties. The resulting sensor with metal oxide as detecting agent and NS-AC-MWCNT as supportive electrode exhibits remarkable sensing performance. The ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE sensor has an excellent detection limit of 1.69 nM for 0.01 to 40 μ M FT, which exceeds that of enzyme-based sensing. The sensor reliably detects in lake water with a detection limit of 11.3 nM attributed to its excellent anti-fouling ability. The sensor has negligible interference from commonly-found chemicals in drinking water. Similarly, MnO₂-DHBA-CHIT/NS-AC-MWCNT/GCE and MgO-DHBA-CHIT/NS-AC-MWCNT/GCE sensors show excellent Pb²⁺ and NO₂⁻ electrochemical detection compared to unmodified electrodes. This simple, effective, non-hazardous methodology can be extended to modify the surfaces of other metal or metal oxide detecting agents.

6.2 Introduction

Population growth and urbanization have consequences for water quality¹ through the consumption and secretion of industrial chemicals into water bodies.² Organophosphorus compounds, heavy metals, and nutrients are major water pollutants of concern.²⁻⁴ Organophosphorus compounds due to pesticides used in agriculture impact human health and can lead to mental disorders, coma, and death.^{3, 5} Heavy metals like lead, cadmium and

zinc are non-biodegradable and toxic, where even tiny amounts can harm multiple organs and cause neurological diseases like Alzheimer's.^{2, 6} While inorganic nutrients such as nitrites and nitrates are widely used in the chemical and food industry, being carcinogens their bioaccumulation or uptake can lead to poisoning and even cancer with prolonged exposure.^{7,8} Water quality issues are estimated to cost \$210 billion yearly in USA alone.⁴ Hence, the accurate determination of water pollutants is necessary to control their environmental footprints.

Analytical detection for water pollutants is typically conducted through mass spectrometry,^{9,10} gas/liquid chromatography,^{10,11} spectroscopy,^{2,12} or optical colorimetry.¹³ These techniques require skilled professionals, significant labor for complex sample preparation, and expensive instruments.¹⁴ Electrochemical techniques are cost effective, and respond quickly on site with high sensitivity.^{3,15} Enzyme based electrochemical sensors are common, since they are sensitive and specific towards pollutants,¹⁶ but are prone to denaturation.^{3,17} Non-enzymatic electrochemical sensors can overcome this limitation.

The working electrode of an electrochemical sensor, which provides the electrodeelectrolyte interface necessary for redox reactions, benefits from faster electrode kinetics and a higher electrode surface area.^{17,18} Electrode modification with nanomaterials includes use of carbon nanotubes, graphene, and metal nanoparticles,¹⁹ e.g., the synthesis of highly conductive and porous nanomaterial containing nitrogen-sulfur co-doped activated carbon coated carbon nanotubes (NS-AC-MWCNT) for organophosphorus compound detection.¹⁷ Since the typical performance of an enzymatic sensor is superior to that of an electrocatalytic sensor,^{20,21} we hypothesize that a detecting agent that enhances analyte concentration near the electrode-electrolyte interface would enhance sensing performance. Therefore, incorporating analyte-capturing detecting agents is a missing key for improving the sensitivity and specificity of non-enzymatic electrochemical sensors.

Inorganic metal oxides have affinity for several water pollutants. ZrO₂, MnO₂, and MgO have strong affinity towards concerned phosphorus compounds,^{5,22} lead,²³ and nitrite,²⁴ respectively. Since metal oxides are readily functionalized²⁵ and are electrocatalytic,²⁶ electrodes modified with these materials are employed for electrochemical detection.²⁶ However, metal oxides tend to agglomerate in suspension due to van der-Waals forces, producing large aggregates in an ink suspension that result in poor film during deposition on an electrode.²⁷ This reduces the electrode surface area and electrocatalysis ability. Moreover, the modified electrode should be stable during washing and voltammetry scanning for reliable and repeatable current response. Hence, electrode modification with a metal oxide is challenging for improving sensing performance.

Our goal was to develop a method that overcomes these challenges to fabricate highly sensitive electrodes with metal oxides to detect specific water pollutants. In our approach, the inorganic metal oxides (1) have a well-dispersed nanoparticle distribution that enhances electrode surface area and analyte capture, (2) form a stable film on the electrode for reliable faradaic current response, and (3) participate in charge transfer, thus working with conductive elements in enhancing overall electrochemical response. We address these challenges based on a bioinspired method to produce a material with filmforming, redox-active, and strong metal-oxide adhesion properties.

Catechol-containing molecules, widely studied to modify metal oxide surfaces,^{28,29} adsorb metal oxides as the catechol moiety forms a strong bidentate type bond with metal atoms.²⁹ This bonding mechanism is inspired by the firm adherence of a mussel to surfaces in aqueous conditions.³⁰ DHBA is a catechol molecule that adsorbs and disperses TiO₂ molecules with superior electronic properties31 as the redox-active DHBA forms a charge transfer complex with the adsorbed TiO₂.³² However, DHBA modified TiO₂ has poor film-forming properties.³¹ This catecholate molecule has terminating -COH groups, which can serve as linkers. Chitosan is an organic polymer with excellent film-forming properties and can be linked to DHBA using Schiff base reaction.²⁹ This is ascribed to the imine bond formation between amine and aldehyde groups of CHIT and DHBA, respectively. Hence, CHIT-linked DHBA chemistry can produce well dispersed and stable inorganic metal oxide-based electrode film.³¹

To achieve our goal, we hypothesize that, with DHBA-CHIT modification, ZrO₂, MnO₂, and MgO metal oxides with an affinity toward specific water pollutants produce uniformly coated stable electrode film that enhance the overall electrochemical performances. To assess the hypothesis, we add metal oxide powder to DHBA-CHIT solution and drop cast on a NS-AC-MWCNT modified GCE. The catechol-modified metal oxides are expected to work synergistically with the NS-AC-MWCNT nanomaterial and produce electrocatalytic electrodes with enhanced surface area and electrode kinetics.

We employ scanning electron microscopy (SEM) to investigate morphology changes and Fourier transfer infrared spectroscopy (FTIR) to characterize chemical structure during the steps of metal oxide surface modification. Electrochemical characterization, including electrochemical impedance spectroscopy (EIS), cyclic voltammetry (CV), and square wave voltammetry (SWV), are used to systematically evaluate electrode electrochemical performance. SWV is also used to optimize the faradaic current response for fenitrothion (FT, an organophosphate) detection. Finally, we investigate heavy metal (Pb²⁺) and nutrient (NO₂⁻) detection with MnO₂-DHBA-CHIT/NS-AC-MWCNT and MgO-DHBA-CHIT/NS-AC-MWCNT composite electrodes. These novel composites could be integrated into a single sensor to detect organophosphate, heavy metal ion, and nutrient in water. This simple, effective, non-hazardous metal oxide surface modification method produces a new class of specific and sensitive sensors based on metal or metal oxides.

6.3 Materials and methods

6.3.1 Chemicals and instruments

Chitosan (degree of deacetylation of 85%), 3,4-dihydroxylbenzaldehyde, acetate buffer solution (ABS), fenitrothion, lead iodide, sodium nitrite, acetic acid, zirconium oxide nanopowder, and magnesium oxide nanopowder were obtained from Sigma-Aldrich Canada. Manganese dioxide was purchased from Fisher Scientific. Multiwalled carbon nanotubes were purchased from Bayer Inc., Germany. Lake water was sourced from Bayfront Park (Hamilton, Ontario). All reagents and deionized water (DI) were utilized as-

received. The stock solution of FT was prepared in ethanol and refrigerated at 4°C until use. The stock solution of Pb^{2+} was prepared in dimethylformamide, while DI water was used for the NO_2^- stock solution.

An electrochemical potentiostat (PARSTAT 2273, Princeton Applied Research) was used for cyclic voltammetry, square wave voltammetry, and electrochemical impedance spectroscopy. A three-electrode setup, including a glassy carbon electrode (working electrode), platinum electrode (counter electrode), and Ag/AgCl electrode (reference electrode), was used to establish the electrochemical sensor. Material characterization was conducted with a JEOL 6610LV scanning electron microscope, where a silicon wafer was used for the substrate on which the synthesized material was deposited, and Fourier transform infrared spectroscopy (FTIR) was performed for powdered materials using a Bruker Vertex 70 instrument.

6.3.2 ZrO₂, MnO₂ and MgO surface modification

Surface modification of ZrO₂, MnO₂, and MgO was conducted using aqueous suspensions. For ZrO₂ nanoparticles, aqueous suspensions of bare ZrO₂, ZrO₂-CHIT, ZrO₂-DHBA, and ZrO₂-DHBA-CHIT were prepared. For MnO₂, surface modification was accomplished with aqueous suspensions of bare MnO₂ and MnO₂-DHBA-CHIT. A similar procedure was employed for MgO surface modification. A CHIT solution of 3 g/L concentration was synthesized by dissolving CHIT using 1% acetic acid in DI water. A DHBA solution of 8 g/L was synthesized by adding DHBA to DI water. For the DHBA-CHIT solution, the CHIT solution was added to the DHBA solution with a CHIT: DHBA mass ratio of 1:2. Subsequently, surface modification of ZrO_2 , MnO_2 , and MgO was performed by adding these materials to the aqueous suspensions discussed above.

6.3.3 NS-AC-MWCNT ink synthesis

NS-AC-MWCNT is prepared with KOH activation and carbonization of polypyrrole coated multiwalled carbon nanotubes (PPy-MWCNT).³³ The PPy-MWCNT is synthesized by chemical polymerization of pyrrole using ammonium persulfate as an oxidant and eriochrome cyanine that acts as a multifunctional dopant for Py and dispersing agent for carbon nanotubes.^{34,35} The detailed synthesis procedure of PPy-MWCNT and NS-AC-MWCNT ink is discussed in our previous work.¹⁷

6.3.4 Sensor fabrication

Fabrication of the electrochemical sensor is described in Schematic 6. 1. A bare GCE is polished with 0.05 μ m alumina powder and washed with DI water by ultrasonication. The NS-AC-MWCNT ink is drop cast on the cleaned GCE and subsequently air-dried. Next, the X-DHBA-CHIT (where X denotes ZrO₂, MnO₂, or MgO) ink is printed and air-dried to produce a layer-on-layer X-DHBA-CHIT/NS-AC-MWCNT/GCE electrode. The fabricated electrodes are electrochemically pretreated by performing CV from 0.1 to 1.2 V in ABS that has a pH = 5 at 0.1 V/s. Electrochemical sensing is performed with CV and SWV. The CVs are conducted in 0.2 M pH = 5 ABS from -0.8 to 0.2 V for FT, -1.0 to -0.4 V for Pb²⁺, and 0 to 1.1 V vs Ag/AgCl for NO₂⁻ at 0.05 V/s scan rate. Electroactive surface area characterization of the electrodes is also performed with CV in 0.5 mM Fe(CN)₆³⁻ /Fe(CN)₆⁴⁻/0.1 M KCl at scan rates ranging from 0.04 to 0.2 V/s. The fabricated sensor is optimized for FT detection with SWV. The limit of detection (LOD) of the ZrO_2 -DHBA-CHIT/NS-AC-MWCNT/GCE sensor for FT is calculated through the relationship $3\sigma/m$, where σ denotes the standard deviation of the SWV response for blank samples and *m* the slope of the calibration curve for FT-containing samples. Specificity of the ZrO_2 -DHBA-CHIT/NS-AC-MWCNT/GCE sensor towards FT is examined using a 20-fold excess concentration of several frequently found chemical interferents. A recovery study in ABS (0.2 M, pH 5) is performed with lake water (filtered with 8 µm pore size filter paper) and tap water.



Schematic 6. 1. (A) Schematic of metal atom surface modification with (i) DHBA, (ii) CHIT, and (iii) DHBA-CHIT. (B) Schematic of step-by-step electrode fabrication for electrochemical detection of the water pollutant.

6.4 Results and discussions

6.4.1 Material characterization

We use Fourier transform infrared spectroscopy (FTIR) to investigate the modification of bare ZrO₂, MnO₂, and MgO surfaces with DHBA modified CHIT. DHBA modification with CHIT is studied first, followed by immobilization of inorganic oxides. As seen in Figure 6. 1I, as-received CHIT powder exhibits characteristic absorptions at ~ 1550, 1062, and 1024 cm⁻¹, indicating the presence of amide II, primary -OH, and C-O-C groups.^{36,37} The 1062 cm⁻¹ absorption peak is found shifted to 1077 cm⁻¹ in ZrO₂-CHIT spectra (Figure 6. S1), revealing potential interaction between ZrO_2 and the hydroxyl group of CHIT. Synthesis of DHBA modified CHIT is confirmed by the C=N bond formed between aldehyde group of DHBA and amino group of CHIT via the Schiff base reaction.^{29,38} Corresponding absorptions of C=N bond are found in DHBA-CHIT (~ 1642 cm⁻¹), ZrO₂-DHBA-CHIT (~ 1650 cm⁻¹), MnO₂-DHBA-CHIT (~ 1635 cm⁻¹), and MgO-DHBA-CHIT (~ 1649 cm⁻¹) spectra implying Schiff base formation. The ZrO₂-DHBA-CHIT spectra showed stretching vibrations at ~ 748, and 485 cm⁻¹ for Zr-O in ZrO_2 ,^{39,40} and a sharp peak at 450 cm⁻¹ for Zr-O-Zr linkage.⁴¹ Another sharp peak at ~ 1384 cm⁻¹ indicative of the -OH group of DHBA³⁷ does not appear for ZrO₂-DHBA-CHIT but does for DHBA-CHIT and DHBA, possibly due to bidentate bonding of DHBA to surface ZrO₂ nanoparticles (as shown in Schematic 6. S1). This confirms the surface modification of ZrO_2 nanoparticles with DHBA-modified CHIT. Similarly, the ~1384 cm⁻¹ peak does not appear in the MnO₂-DHBA-CHIT and MgO-DHBA-CHIT spectra, confirming surface modification of both MnO₂ and MgO with DHBA-CHIT. Finally, the MnO₂-DHBA-CHIT spectra reveal a broad peak at 460 cm⁻¹ attributed to Mn-O vibration,⁴² and MgO-DHBA-CHIT spectra absorptions at 643 and ~ 550 cm⁻¹ ascribed to Mg-O vibrations.⁴³



Figure 6. 1. (I) Chemical structure characterization by FTIR for (A) as-received CHIT, (B) as-received DHBA, (C) DHBA modified CHIT, DHBA-CHIT modification of (D) ZrO₂, (E) MnO₂, and (F) MgO. (II) SEM images of (A) as-received ZrO₂, ZrO₂ modified with (B) CHIT, (C) DHBA, and (D) DHBA-CHIT.

ZrO₂ surface modification by CHIT, DHBA, and DHBA-CHIT is investigated with SEM. Unmodified ZrO₂ nanoparticles reveal heavy agglomeration (Figure 6. 1 (IIA)) since they do not bond as strongly to the substrate. CHIT modification of the nanoparticles promotes this agglomeration (Figure 6. 1 (IIB)). In contrast, DHBA-modified ZrO₂ nanoparticles (Figure 6. 1 (IIC)) produce more surface coverage and fewer agglomerates on a silicon substrate, behavior that is enhanced by DHBA-CHIT modification of ZrO₂ nanoparticles (Figure 6. 1 (IID)). The more even - and uniform surface coverage of the substrates is attributed to the strong DHBA-assisted immobilization of nanoparticles on the surface and the film-forming properties of CHIT.



Figure 6. 2. Fabricated electrodes characterization with (A) EIS on GCE, NS-AC-MWCNT/GCE, ZrO_2/NS -AC-MWCNT/GCE, and ZrO_2 -DHBA-CHIT/NS-AC-MWCNT/GCE electrodes in 0.5 mM Fe(CN)₆³⁻/Fe(CN)₆⁴⁻ and 0.1 M KCl in 10⁻¹ to 10⁵ Hz (5mV); (B) Peak current vs. square root of scan rate plots for electroactive surface area evaluation of GCE, NS-AC-MWCNT/GCE, ZrO_2/NS -AC-MWCNT/GCE, and ZrO_2 -DHBA-CHIT/NS-AC-MWCNT/GCE electrodes for scan rates 0.04 – 0.2 V/s for 20 μ M FT in pH 5 buffer solution; (C) CV and (D) SWV study of GCE (FT deprived), GCE, NS-AC-MWCNT/GCE, ZrO_2 -CHIT/NS-AC-MWCNT/GCE, ZrO_2 -DHBA/NS-AC-MWCNT/GCE and ZrO_2 -DHBA-CHIT/NS-AC-MWCNT/GCE in 20 μ M FT (pH 5, 0.05 V/s scan rate).

Electrochemical impedance spectroscopy (EIS) is employed to investigate the influence of surface modification of ZrO₂ nanoparticles with DHBA-CHIT by comparing the charge transfer resistance of modified ZrO₂ against unmodified ZrO₂ nanoparticles and different electrode fabricating materials (Figure 6. 2A). A Randles equivalent circuit is used to model and fit the impedance data and generate Nyquist plots.⁴⁴ The charge transfer resistance of the electrode (R_{cl}) represents the ease of electron transfer at the electrodeelectrolyte interface. As expected, Figure 6. 2A shows that modification of GCE with NS-AC-MWCNT decreases the charge transfer resistance of an electrode from R_{ct} (GCE) = 53.7 Ω to R_{ct} (NS-AC-MWCNT/GCE) = 7.7 Ω . This is attributed to the superior electrical conductivity and electroactive surface area of NS-AC-MWCNT over bare GCE.¹⁷ Deposition of unmodified ZrO_2 nanoparticles on NS-AC-MWCNT/GCE increases R_{ct} to 14 Ω , which is likely due to nanoparticle agglomeration on the electrode surface, which reduces the available electroactive surface area of the working electrode. Immobilization of the modified ZrO₂ nanoparticles with DHBA-CHIT on NS-AC-MWCNT/GCE shows a significant decrease in R_{ct} to 8.3 Ω as compared to the value for ZrO₂/NS-AC-MWCNT/GCE, indicating considerable enhancement of ZrO₂ nanoparticle immobilization through stronger bonding between nanoparticles and the electrode surface. The reduction in nanoparticle agglomeration enhances the surface area of the electrode available for electrochemistry. R_{ct} for the modified ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE is higher than for NS-AC-MWCNT/GCE, likely due to the lower electrical conductivity of CHIT.

CV is used to collect the electrochemical response of different electrodes, and the Randles-Sevcik equation⁴⁵ employed to calculate the electroactive surface area. Graphs of the anodic peak current (I_p) *vs.* square root of scan rate ($v^{1/2}$) are plotted (Figure 6. 2B), and their slopes used to determine surface area.

$$I_n = 2.69 * 10^5 n^{3/2} ACD^{1/2} v^{1/2}$$

where *n* represents the number of participating electrons, *A* is the electroactive surface area of the electrode, *C* and D are the concentration and diffusion coefficient of $Fe(CN)_6^{3-}$ /Fe(CN)₆⁴⁻, respectively. The scan rate is varied from 0.04 to 0.2 V/s. The surface areas thus determined are 0.028, 0.109, 0.055, and 0.126 cm² for GCE, NS-AS-MWCNT/GCE, ZrO₂/NS-AC-MWCNT/GCE, and ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE, respectively. NS-AS-MWCNT modification of GCE increases the surface area almost fourfold. Modification of NS-AC-MWCNT/GCE with unmodified ZrO₂ nanoparticles reduces the surface area compared with NS-AC-MWCNT/GCE, indicating formation of nanoparticles agglomerates. Conversely, deposition of ZrO₂ modified with DHBA-CHIT on NS-AC-MWCNT/GCE provides the highest electroactive surface area, a 4.5-fold increase relative to GCE. We suggest that CHIT enhances film formation of the composite and DHBA supports nanoparticle distribution by reducing agglomeration, thus enhancing the available surface area for the working electrode.

Further electrochemical characterization of the fabricated electrodes is performed with their CV and SWV responses for FT detection (Figure 6. 2 C, D). The redox-active nitro group of FT produces an irreversible reduction, quasi reversible oxidation, and reduction peaks during forward and reverse CV scans.^{17,46} Similar characteristic peaks are

observed for FT detection with all the working electrodes (Figure 6. 2 C, D). CV and SWV profiles of the electrodes are compared with their Faradaic current response. The NS-AC-MWCNT modified GCE electrode considerably improves the Faradaic current response as compared to GCE due to its superior electrocatalytic properties. Deposition of unmodified ZrO₂ nanoparticles as the FT detecting agent on the NS-AC-MWCNT/GCE shows a decrease in current in CV and SWV response. This result is supported by EIS and electrochemical surface area studies. However, deposition of CHIT modified ZrO₂ on NS-AC-MWCNT/GCE reveals an increase in current response compared to NS-AC-MWCNT/GCE. This occurs due to interaction between CHIT and ZrO₂, as observed through the FTIR study, thereby facilitating the charge transfer between FT and the electrode. An even better current response is noted with DHBA modified ZrO₂/NS-AC-MWCNT/GCE, mainly due to the addition of the redox activity of DHBA^{47,48} in the composite and improved nanoparticle dispersion. DHBA-CHIT linked ZrO₂ on NS-AC-MWCNT/GCE has the highest Faradaic current response among all electrodes due to the synergetic effect of well-dispersed ZrO₂ nanoparticles and NS-AC-MWCNT. Thus, for this case, synergetic enhancement of electrochemical surface area, the electrical conductivity of the electrode, and the strong affinity for the phosphoric group (of FT) considerably improves the current response for FT detection.

6.4.2 Sensor optimization

The optimal concentration of ZrO_2 for the DHBA modified CHIT is evaluated through the electrode SWV response for FT detection. The concentration of ZrO_2 is varied from 0.5x

to 5*x* (with x = 33.3 g/L), with concentrations of DHBA and CHIT held constant. The current response increases with increasing ZrO₂ concentration up to 2*x* (Figure 6. 3A) because of the greater availability of nanoparticles in the DHBA-CHIT matrix. Further increase in ZrO₂ concentration results in a decrement in current response since enough DHBA-CHIT ligands are no longer available to link ZrO₂ nanoparticles. This encourages nanoparticle agglomeration and reduces the electrocatalytic properties of the electrode. Thus, 2*x* is an optimal concentration for ZrO₂ in DHBA modified CHIT. Next, the optimal ZrO₂-DHBA-CHIT ink deposition on NS-AC-MWCNT/GCE is evaluated. As seen in Figure 6. 3B, the current response increases from 1 to 4 µL but does not increase thereafter with further increase in ink deposition after the electrode surface coverage has been maximized. Hence, 4 µL is chosen as the optimal deposition volume for the ZrO₂-DHBA-CHIT ink.

The pH of the acetate buffer solution is similarly varied from 3 to 6 to assess electrode performance for FT detection. The current response increases with pH increases from 3 to 5, followed by a decrease with further increases in pH (Figure 6. 3C). This is attributed to the increased proton concentration due to reduction of the FT NO group. The proton concentration decreases with increasing pH, which limits-NO reduction and thus current response. Therefore, pH=5 is selected for the acetate buffer solution as being optimal. Next, we consider the adsorption-controlled FT detection facilitated by the affinity of ZrO₂ for the phosphoric group, and effect of accumulation potential (a_p) and time (a_t) on current response. We vary a_p from -0.8 to -0.5 V (*vs.* Ag/AgCl) for $a_t = 25s$ with magnetic stirring. The peak current increases with decreasing a_p until -0.6 V (Figure 6. S2A) but a further decrease in a_p now decreases the peak current. Subsequently, we hold $a_p = -0.6$ V and vary a_t between 25-150s. The highest current response occurs at 100s and saturates with further increases in a_t (Figure 6. S2B). Consequently, a_p -0.6 V potential for 100s is selected as optimal for SWV detection of FT.

The final step in the sensor optimization is investigation of the SWV parameters, such as frequency, step potential, and amplitude, which are varied to obtain the optimal peak current shape and highest current response for FT detection. The frequency is analyzed by varying it from 5 to 25 Hz with constant step potential and amplitude of 5 mV and 50 mV, respectively. The peak current increases to 10 Hz followed by a decrease (Figure 6. S3A) based on which a 10 Hz frequency is selected. Next, the step potential is varied from 3 to 20 mV at 10 Hz (Figure 6. S3B), where 10 mV is selected as optimal considering the peak shape and current response. Finally, the amplitude is examined by varying it between 40 to 180 mV. As shown in Figure 6. S3C, the highest current response occurs at 140 mV, which is selected as optimal at a 10 Hz frequency and 10 mV step potential for SWV detection of FT.


Figure 6. 3. SWV assisted optimization study of (A) ZrO_2 concentration in ZrO_2 -DHBA-CHIT matrix for 20 μ M FT (pH 5); (B) the amount of ZrO_2 -DHBA-CHIT ink deposition on NS-AC-MWCNT/GCE for 20 μ M FT (pH 5); and (C) pH of the buffer solution in 3 to 5.5 pH range for 20 μ M FT.

6.4.3 Sensor performance

An analytical determination of sensitivity is performed for the ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE sensor for different FT concentrations under the optimized conditions. The current response for different concentrations ranging from 50 to 0.01 μ M is used to plot the calibration curve (Figure 6. 4A), which provides the linear relation I_{pc} (μ A) = 7.91C

 (μM) + 26.38 with R² = 0.99. The limit of detection is 1.69 nM, which is calculated from the relation $3\sigma/m$, where σ denotes the standard deviation of the SWV response for blank samples and *m* the slope of the calibration curve with three replicates per FT concentration.

The sensor performance exceeds that for enzymatic-based sensors (Table. 1) for FT detection. This is because ZrO_2 nanoparticles are better dispersed through DHBA-CHIT chemistry, increasing the electrode electroactive surface area and enhancing FT capture. The improved redox activity from DHBA in the composite increases charge transfer through the modified electrode. In addition, NS-AC-MWCNT as the supporting electrode improves electrocatalysis due to its higher electrical conductance and surface area.¹⁷ Thus, the synergistic effect of ZrO_2 nanoparticles and NS-AC-MWCNT enhances the overall signal response during FT detection.



Figure 6. 4. (A) Sensor performance of ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE for FT concentrations in 0.01 to 50 μ M range (pH 5), inset represents calibration plot of peak current vs. FT concentration. (B) and (C) Interference study of ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE sensor with 20-fold concentrations of commonly found chemical interferents in consuming water in 20 μ M FT (pH 5).

The specificity of a sensor is a critical characteristic, since it must be agnostic to chemical interfering agents contained in environmental samples. We spike drinking water with interferent agents in 20-fold excess concentrations relative to FT (16 μ M). All interferents, apart from phosphate, chlorpyrifos and dichlorvos, have a negligible influence

on FT detection (Figure 6. 4B, C) by the ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE sensor due to the behavior of ZrO₂ as a detecting agent for the analyte, i.e., FT. The minor influence of phosphate occurs because of the ZrO₂ affinity towards phosphate. The relative lack of response to the chlorpyrifos and dichlorvos interferents for this sensor is better than for one fabricated with NS-AC-MWCNT/GCE¹⁷ due to the ZrO₂-DHBA-CHIT coating on NS-AC-MWCNT/GCE, which reduces π - π interactions of the sensor with chlorpyrifos. Therefore, incorporating ZrO₂ as the detecting agent in the NS-AC-MWCNT/GCE composite improves selectivity towards FT.

Electrode	Mechanism	Technique	Linear	LOD	Reference
			range	(nM)	
			(µM)		
ZrO ₂ -DHBA-	Non-	SWV	0.010 -	1.69	This work
CHIT/NS-AC-	enzymatic		50.0		
MWCNT/GCE					
1T-WS ₂ /GA/AChE-	Enzymatic	Chronoamperome	0.001 -	2.86	21
BSA/GCE		try	1.0		
Tyr/poly(2-	Enzymatic	Amperometry	0.018 -	4.7	49
hydroxybenzamide/G			3.60		
Ε					

 Table 6. 1. Comparison of modified electrodes for electrochemical detection of FT.

NS-AC-	Non-	SWV	0.050 -	4.91	17
MWCNT/GCE	enzymatic		40.0		
ZrO ₂ /rGO/MoS ₂ -	Non-	SWV	0.018 -	7.94	22
Au/GCE	enzymatic		21.6		
AuNPs@GNIP-GR-	Non-	DPV*	0.010 -	8.0	50
IL/GCE	enzymatic		5.0		
MWCNT/GCE	Non-	SWV	0.20 -	80.0	51
	enzymatic		60.0		

^{*}AChE - Acetylcholinesterase, Tyr - Tyrosinase, DPV- Differential Pulse Voltammetry

Table 6. 2. FT recovery in lake and tap water for 16, 8, and 4 μ M concentration (pH 5) with three replicates per concentration.

Water	Spiked FT	Found FT	Recovery
sample	(µM)	(µM)	(%)
Lake water	16	15.61 ± 0.84	97.5 ± 5.4
	8	7.74 ± 0.36	96.7 ± 4.6
	4	3.67 ± 0.2	91.9 ± 5.5
Tap water	16	15.79 ± 0.59	98.6 ± 3.7
	8	7.9 ± 0.38	98.8 ± 4.8
	4	3.75 ± 0.2	93.8 ± 5.5

Sensor utilization in lake and tap water, stability at room temperature and 4°C, and repeatability are examined. Tap water and filtered lake water are spiked with 16, 8, and 4 μ M FT concentrations determined with the generated calibration curve. Table 2 shows that the sensor recovers spiked FT concentrations in the range 91.9 – 97.5 % with RSD 4.6 – 5.5 % in lake water, and 93.8 – 98.8 % with RSD 3.8 – 5.5 % in tap water. The recovery performance in lake water is slightly lower due to the passivation of the electrode surface

with more waste, or contamination, in lake water. Moreover, the anti-fouling ability of the fabricated sensor tested in the lake water revealed remarkable improvement compared to the NS-AC-MWCNT/GCE sensor. The LOD measurements are performed for both the sensors with several FT concentrations from 20 to $2 \mu M$ in the lake water (Figure 6. S4). The ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE sensor revealed LOD = 11.3 nM compared to LOD = 19.8 nM from NS-AC-MWCNT/GCE. The improvement in the antifouling ability of the ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE sensor is attributed to the enhancement in the target specie capture via ZrO₂, significantly increasing specific response. Moreover, uniform coating by DHBA-CHIT assisted in blocking the interferents' access to the conductive electrode surface. Storage of the sensor at room temperature and 4° C for 4 weeks has virtually no influence on FT detection (16 μ M), where the sensor recovers 94.3% and 96.6% current responses as compared to the initial response, respectively (Figure 6. S5), i.e., the sensor is stable at both room temperature and under refrigeration for at least four weeks. The RSD for all FT concentrations was lower than 4.9 %, demonstrating excellent repeatability.

The DHBA-CHIT chemistry with ZrO₂ nanoparticles has remarkably sensitive and specific responses towards FT. Other water pollutants can also be detected using the same DHBA-CHIT method for immobilized inorganic oxides with affinity towards the analyte. We modified the MnO₂ surface with DHBA-CHIT and deposited on NS-AC-MWCNT/GCE to detect Pb²⁺ ions in an aqueous solution. CV and SWV were employed to analyze this electrode's electrochemical performances to detect 20 μ M Pb²⁺ in pH 5 ABS. The CV reveals anodic and cathodic peaks at ~ -0.53 V and ~ -0.72 V *vs*. Ag/AgCl,

respectively, in the -1 to -0.4 V potential range *vs.* Ag/AgCl (Figure 6. 5A).⁵² The SWV reveals a characteristic peak at ~ -0.62 V *vs.* Ag/AgCl (Figure 6. 5B). Both CV and SWV studies indicate Faradaic current responses in increasing order as follows for the electrodes: GCE < MnO₂/NS-AC-MWCNT/GCE < NS-AC-MWCNT/GCE < MnO₂-DHBA-CHIT/NS-AC-MWCNT/GCE. The highest current response to detect Pb²⁺ is for MnO₂-DHBA-CHIT/NS-AC-MWCNT/GCE. The detecting agent MnO₂ enhances the surface area of the electrode and significantly enhances Pb²⁺ adsorption on the electrode, providing an example of the application of DHBA-CHIT to fabricate a MnO₂ surface-modified NS-AC-MWCNT/GCE sensor that detects heavy metal ions.



Figure 6. 5. (A, B) CV and SWV detection of 20 μ M Pb²⁺ at GCE (Pb²⁺ deprived), GCE, NS-AC-MWCNT/GCE, MnO₂/NS-AC-MWCNT/GCE, and MnO₂-DHBA-CHIT/NS-AC-MWCNT/GCE in ABS (pH 5). (C, D) CV and SWV response for 20 μ M NO₂⁻ at GCE (NO₂⁻ deprived), GCE, NS-AC-MWCNT/GCE, MgO/NS-AC-MWCNT/GCE and MgO-DHBA-CHIT/NS-AC-MWCNT/CGE in ABS (pH 5). The scan rate of CV is 0.05 V/s.

Since MgO nanoparticles have affinity towards nitrite (NO₂⁻) ions, we incorporate these into the DHBA-CHIT composite to fabricate a MgO-DHBA-CHIT/NS-AC-MWCNT/GCE electrochemical sensor. CV and SWV are performed for this sensor with $20 \,\mu\text{M NO}_2$ in pH 5 ABS in the potential range from 0 to 1.1 V vs. Ag/AgCl. As shown in Figure 6. 5C, D, the CV reveals an oxidation peak at ~0.75 V (vs. Ag/AgCl), and the SWV has a characteristic oxidation peak at ~ 0.65 V (vs. Ag/AgCl) for all electrodes except GCE.⁵³ The broad oxidation peak for NO₂⁻ with GCE at a higher potential occurs due to slower reaction kinetics and limited analyte diffusion across the bare electrode. The MgO-DHBA-CHIT/NS-AC-MWCNT/GCE electrode shows the highest current response towards NO₂⁻ ions. As discussed, incorporation of the inorganic oxide into the NS-AC-MWCNT/GCE composite with the DHBA-CHIT method enhances the available surface area, analyte capture and charge transfer of the electrode. For optimal performance from MgO-DHBA-CHIT/NS-AC-MWCNT/GCE MnO₂-DHBA-CHIT/NS-ACand MWCNT/GCE, each sensor's parameter must be optimized. Besides, the preliminary results of the methodology indicate that this platform can be used to modify other metal oxides or metals for fabricating highly sensitive sensors.

6.5 Conclusion

Uniformly coated, stable and highly electrocatalytic electrodes are fabricated as ultrasensitive non-enzymatic electrochemical sensors for specific detection of water pollutants. A detecting agent with high affinity for the analyte is incorporated into the electrode modifying ink using the catechol molecule 3,4-dihydroxylbenzaldehyde (DHBA) and polymer chitosan (CHIT). A simple and effective method immobilizes metal oxides through strong adsorption by DHBA and film formation with CHIT. The detecting agents ZrO₂, MnO₂, and MgO, have specific affinities to fenitrothion (FT, an organophosphate), lead (Pb²⁺, a heavy-metal ion), and nitrite (NO₂⁻, a nutrient), respectively. Surface modification with these metal oxides enhances electronic and film-forming properties of electrodes that increase total surface area. We drop cast X-DHBA-CHIT (X: ZrO₂, MnO₂. or MgO) ink on glassy carbon electrode (GCE) modified with nitrogen-sulfur co-doped activated carbon coated carbon nanotubes (NS-AC-MWCNT). The X-DHBA-CHIT and NS-AC-MWCNT work synergistically to enhance the electrocatalytic properties of the electrode. As a result, the ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE sensor produces an excellent current response for FT detection. With a 1.69 nM detection limit, the sensor reliably detects FT in lake and tap water due to its excellent anti-fouling properties. The sensor is stable for 4 weeks at room temperature and when stored at 4° C, and experiences negligible influence from chemical interferents. The current responses of MnO₂-DHBA-CHIT/NS-AC-MWCNT/GCE and MgO-DHBA-CHIT/NS-AC-MWCNT/GCE electrodes have excellent detection capabilities for Pb^{2+} and NO_2^{-} , respectively. For the optimal performance from MnO₂ and MgO modified electrodes, each electrochemical parameter must be optimized.

6.6 References

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6.7 Appendix/supplementary information



Schematic 6. S1. Metal oxide adsorption mechanisms with catecholic DHBA via bidentate (A) chelating, (B) inner sphere bridging, and (C) outer sphere bridging bonding.



Figure 6. S2. (A) Accumulation potential and (B) accumulation time optimization study for 20 μ M FT (pH 5). Inset represents peak current vs. accumulation potential and accumulation time for (A) and (B).



Figure 6. S3. Optimization study of SWV parameters (A) frequency from 5 to 25 Hz at 5 mV step potential and 50 mV amplitude, (B) step potential from 2 to 20 mV at frequency 10 Hz and 50 mV amplitude, and (C) amplitude from 40 to 180 mV at 10 Hz frequency and 10 mV step potential for 20 μ M FT (pH 5).



Figure 6. S4. Sensor performance of ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE and NS-AC-MWCNT/GCE sensors for FT concentrations from 20 to 2 μ M in the lake water (pH 5).



Figure 6. S5. SWV responses of ZrO₂-DHBA-CHIT/NS-AC-MWCNT/GCE towards 16 μ M FT detection for four weeks storage at 4°C and room temperature (pH 5).

7 Conclusions

Numerous non-enzymatic electrochemical sensors are fabricated with ultra-high sensitivity and specificity for water pollutants detection. The working electrode modifying inks are synthesized using conceptually new methodologies. The synthesized materials modified electrodes produced excellent electrochemical performances overcoming the sensing performances of enzymatic electrochemical sensors. The key accomplishments of this dissertation are as follows:

- (1) A simulation model for studying electrode kinetics of the porous and unmodified planar electrodes is developed. The model simulated the electrode kinetics of the Fenitrothion undergoing multi-step multi-electron reactions at the electrodeelectrolyte interface. Such a simulation model can be exceptionally favorable for studying electrode mechanisms of the analyte undergoing multi-step multi-electron reactions, like 4-nitrophenol or parathion.
- (2) The simulation model revealed the existence of thin-layer diffusion at the porous electrode. Modification of bare electrode with MWCNT changed the mass transport regime of the analyte from semi-infinite to thin-layer diffusion. Thin-layer effects influenced the electrochemical response of the Fenitrothion at the porous electrode. Also, the peak-to-peak potential separation of the cyclic voltammogram was recorded lower for the porous electrode than for the unmodified planar electrode. The results of the CV experiments validated the simulation results.

- (3) Nitrogen sulfur co-doped activated carbon coated multiwalled carbon nanotubes (NS-AC-MWCNT) material is synthesized. The synthesized ink modified electrode revealed a very high electroactive surface area and faster charge transfer ability. Heteroatom co-doping enhanced the electrical conductivity of the activated carbon coating. Activated carbon-coated multiwalled carbon nanotubes produced porous modified electrodes of high surface area and electrical conductivity. Such modified working electrodes can be extremely promising for sensing applications.
- (4) The NS-AC-MWCNT is synthesized with a multifunctional dopant, Eriochrome cyanine R (ECR), utilized as a dopant and dispersing agent. ECR as a dopant for pyrrole produced uniformly coated polypyrrole. The aromatic structure of ECR promoted the adsorption of carbon nanotubes, while the presence of charge groups promoted nanotubes' electrostatic repulsion. Thus, ECR as a dispersing agent revealed enhancement in the dispersion of carbon nanotubes, which reduced their agglomeration.
- (5) The fabricated NS-AC-MWCNT/GCE electrode recorded highly sensitive electrochemical performance for Fenitrothion. The sensor revealed a 4.9 nM limit of detection under optimized conditions (amount of ink deposited, pH of the buffer solution, accumulation potential and time, and SWV parameters). This is attributed to the enhanced electrocatalytic properties of the fabricated electrode. The high electrical conductivity is ascribed to the nitrogen-sulfur co-doping and multiwalled carbon nanotubes. While the high surface area of the electrode is ascribed to the activated carbon coated multiwalled carbon nanotubes.

- (6) Final enhancement in the sensor performance is achieved by incorporating surfacemodified metal oxides (ZrO₂, MnO₂, and MgO) as detecting agents. Metal oxides were well dispersed via excellent adsorption from catecholate-type bonding of 3,4dihydroxylbenzaldehyde (DHBA). Stable films on electrode modification are achieved by incorporating chitosan linked to DHBA. Uniformly coated and stable electrodes are attributed to the DHBA-CHIT chemistry.
- (7) Modification of NS-AC-MWCNT/GCE electrode with X-DHBA-CHIT (X: ZrO₂, MnO₂, or MgO) produced enhanced electrochemical performances. Metal oxide modified with DHBA produced well-dispersed and agglomerate free electrodes. X-DHBA-CHIT modified NS-AC-MWCNT/GCE revealed enhancement in the total electroactive surface area and charge transfer ability of the modified electrodes. Enhanced electrical conductance is attributed to the redox-active DHBA's participation in the charge transfer process.
- (8) The fabricated X-DHBA-CHIT/NS-AC-MWCNT electrodes produced excellent improvement in sensor performance with respect to unmodified electrodes. The ZrO₂-DHBA-CHIT/NS-AC-MWCNT sensor achieved a remarkable limit of detection of 1.69 nM for Fenitrothion. The sensor's performance exceeded the enzymatic-based sensors. The specificity of the sensor is greatly enhanced with the incorporation of affinity-based detecting agents. The interference study revealed negligible interference from commonly found chemical interferents. The sensor produced reliable and satisfactory performance in lake and tap water detection of Fenitrothion due to its excellent anti-fouling properties.

(9) The fabricated MnO₂-DHBA-CHIT/NS-AC-MWCNT/GCE and MgO-DHBA-CHIT/NS-AC-MWCNT/GCE sensors produced enormous increment in the lead and nitrite electrochemical detection with respect to unmodified electrodes. These excellent performances from X-DHBA-CHIT/NS-AC-MWCNT/GCE electrodes are achieved from the synergistic enhancement of total surface area and charge transfer ability from X-DHBA-CHIT and NS-AC-MWCNT. The detecting agent-assisted increment in the analyte concentration near the electrode-electrolyte interface also supported the overall sensing improvements.

8 Future directions and improvements

The work of this dissertation led to the fabrication of electrochemical sensors for organophosphate, heavy metal ion, and nutrient detection. Numerous problems in the development of non-enzymatic-based sensors with ultrahigh sensitivity and specificity have been addressed. All the X-DHBA-CHIT/NS-AC-MWCNT/GCE electrodes can be combined into a single electrochemical sensor for PAN-water analysis of water pollutants. The proposed surface modification methodology can be extended to other metal oxides and metals to extend the detectable water pollutants' spectra. Moreover, combining these detecting agents modified electrodes can enable multi-analyte electrochemical determination. For sensor miniaturization, replacement of the GCE electrode system should be done. Screen-printed electrodes can be suitable alternatives. A water sampling and filtration system should be developed for better on-field detection suitability and antiinterference ability. The simulated model for studying electrode kinetics at unmodified planar and porous electrodes can be extended to other specie undergoing multi-electron multistep electrode processes. The formulated model can open the field of electrochemistry simulation to more complex multi-electron and multi-step processes.

The sensors fabricated in this dissertation work utilized carbon nanotubes for their excellent electrocatalytic properties. However, the synthesis and utilization of carbon nanotubes-based nanomaterials in sensors come with an environmental and human risk assessment due to their toxicity to human health and the environment. To make the fabricated sensors sustainable and eco-friendly in the future, green synthesis of carbon

nanotubes is suggested that will reduce the release of toxic by-products. Furthermore, efforts can be made to reduce the temperature of the carbonization process to limit energy consumption. Moreover, it is suggested to use silver or gold nanoparticles from green synthesis to replace the toxic carbon nanotubes and make the sensors more sustainable. To improve the reproducibility of the sensors, efforts can be made to improve the dispersion of electrode modifying ink to ensure reproducible deposition of films on the electrode surfaces. This will limit the variability in the electrode properties and analyte diffusion, thereby making the sensor more reproducible. Better ink dispersion can be achieved with the utilization of suitable surfactants during ink synthesis. Since the sensor's excellent electrochemical performance was majorly due to the analyte capture ability from the adsorption mechanisms, the sensor may have low reusability. This is due to the adsorbed analyte that will passivate the electrode surface and reduce the sensing performance. To improve the reusability of the fabricated sensors, it is suggested to replace the Square Wave voltammetry (SWV) detection technique with Square Wave-Anodic Stripping Voltammetry (SW-ASV) that detects the analyte and then strips out the adsorbed analyte at the appropriate potential. This suggested SW-ASV will greatly improve the reusability of the fabricated sensors without comprising the sensing performances.