

Table of Contents

Contents	Page No.
Abstract	i-vi
List of Figures	vii-xiv
List of Tables	xv
List of Schemes	xvi
List of Abbreviations	xvii-xx
Chapter 1: Introduction	1.1–1.57
1.1 Introduction	1.1
1.2 A brief introduction to different classes of pesticides	1.1
1.2.1 Organochlorine pesticides	1.1
1.2.2 Organophosphate pesticides	1.3
1.2.3 Carbamate pesticides	1.4
1.2.4 Pyrethroid pesticides	1.5
1.2.5 Benzimidazole pesticides	1.6
1.2.6 Phenolic pesticides	1.7
1.3 Conventional methods of pesticide detection	1.8
1.3.1 Gas chromatography (GC)	1.8
1.3.1.1 Electron capture detector (ECD)	1.8
1.3.1.2 Nitrogen and phosphorus detector (NPD)	1.8
1.3.1.3 Flame photometric detector (FPD)	1.9
1.3.1.4 Mass spectrometry detector (MSD)	1.9
1.3.1.5 Gas chromatography tandem mass spectrometry (GC/MS/MS)	1.10
1.3.2 High performance liquid chromatography (HPLC)	1.10
1.3.2.1 UV, DAD, fluorescence and chemiluminescence detector	1.11
1.3.2.2 Mass spectrometry detector	1.11

1.3.3	Immunoassay (IA)	1.12
1.4	Biosensors	1.14
1.5	Components of Biosensor	1.14
1.5.1	The Transducer element	1.14
1.5.2	The sensing element	1.15
1.5.2.1	Advantages of using enzyme as sensor element	1.15
1.5.2.2	Disadvantages of using enzyme as sensor element	1.16
1.6	Enzyme Immobilization matrix	1.16
1.6.1	Advantages of immobilized enzyme	1.16
1.6.2	Disadvantages	1.17
1.6.3	Conducting polymers (CPs)	1.17
1.6.4	Silica	1.18
1.6.5	Nanoparticles	1.19
1.7	Different methods of immobilization of enzyme	1.19
1.8	Enzyme kinetics	1.22
1.8.1	The catalytic efficiency of enzymes	1.24
1.9	Types of Enzyme Inhibition	1.24
1.9.1	Competitive Inhibitors	1.25
1.9.2	Non-competitive Inhibitors	1.26
1.9.3	Mixed type of inhibition	1.26
1.9.4	Uncompetitive Inhibition	1.27
1.10	Electrochemical biosensor	1.28
1.10.1	Amperometry	1.29
1.10.2	Potentiometry	1.30
1.10.3	Conductometry	1.30
1.11	Acetylcholinesterase based biosensor	1.31
1.12	Organophosphorus Hydrolase (OPH) based biosensor	1.33

1.13	Tyrosinase based biosensor	1.34
1.14	Alkaline Phosphatase (ALP) based biosensor	1.36
1.15	Acid Phosphatase (AP) based biosensor	1.36
1.16	Glutathione S-transferase based biosensor	1.37
1.16.1	Key features of GST enzyme	1.38
1.16.2	Literature review on biosensors based on GST	1.39
1.17	Scopes, aim and objectives of the present investigation	1.40
1.17.1	Aim and Objectives	1.41
1.18	Plan of work	1.42
	References	1.43-1.57

Chapter 2: Materials and Methods	2.1–2.9	
2.1	Reagents and materials	2.1
2.2	Chemical structure of some comounds used in this study	2.1
2.3	Instruments	2.3
2.3.1	Electrode surface preparation	2.4
2.4	Details of the techniques used	2.4
2.4.1	Cyclic voltammetry (CV)	2.4
2.4.2	Ultraviolet-visible spectroscopy (UV-VIS)	2.5
2.4.3	Fourier transformed infrared spectroscopy (FTIR)	2.5
2.4.4	Scanning electron microscope (SEM)	2.6
2.4.5	Chronoamperometry (CA)	2.6
2.4.6	Gas chromatography-mass spectrometry (GC-MS)	2.7
2.4.7	Electrochemical impedance spectroscopy (EIS)	2.7
2.4.8	X-ray diffraction (XRD)	2.8
	References	2.9

Chapter 3: Electrochemical study of GSH-CDNB reaction in methanol and ethanol and the influence of GST and pesticides on it **3.1–3.27**

3.1	Introduction	3.1
3.2	Objectives of this chapter	3.3
3.3	Experimental	3.3
3.3.1	Instrumentation	3.3
3.3.2	Analysis procedure	3.3
3.3.3	Interference study	3.4
3.3.4	Optimization	3.4
3.4	Results and discussion	3.4
3.4.1	Cyclic voltammetric study of GSH-CDNB reaction in methanol	3.4
3.4.2	Interference due to cross reactivity of the components under the applied electrochemical condition.	3.6
3.4.2.1	Cross reactivity of MeOH and GSH	3.6
3.4.2.2	Plausible mechanism	3.8
3.4.2.3	IR spectroscopic evidence of proposed mechanism	3.10
3.4.2.4	Cross reactivity of MeOH with PB	3.16
3.4.2.5	Cross reactivity of CDNB with MeOH	3.16
3.4.2.6	Cross reactivity of GSH with PB	3.17
3.4.3	Optimization	3.18
3.4.3.1	Effect of GST amount	3.18
3.4.3.2	Apparent Michaelis-Menten constant (K_m^{app})	3.18
3.4.3.3	Effect of incubation time	3.19
3.4.3.4	Effect of pH	3.20
3.4.3.5	Effect of organic solvent	3.20
3.4.3.6	Optimum methanol composition and effect of ethanol on the reaction	3.20

3.4.4 Pesticides interaction study	3.21
3.4.4.1 Quantification of cypermethrin	3.22
3.4.4.2 Method validation study	3.23
3.5 Conclusion	3.24
References	3.25-3.27

Chapter 4: Application of GST catalyzed GSH-CDNB reaction in methanol as a new method for detection of selected pesticides belonging to different classes **4.1-4.21**

4.1 Introduction	4.1
4.2 Objectives of this chapter	4.3
4.3 Experimental	4.3
4.3.1 Materials and reagents	4.3
4.3.2 Analysis procedure	4.4
4.3.2.1 Cyclic voltammetry measurements	4.4
4.3.2.2 Optimum pesticide incubation time	4.4
4.3.2.3 UV-Visible spectroscopic study	4.5
4.3.2.4 Validation study	4.5
4.4 Results and Discussion	4.5
4.4.1 Cyclic voltammetric study of GSH-CDNB reaction in presence of GST	4.5
4.4.2 Pesticides interaction study	4.6
4.4.3 UV-VIS study	4.7
4.4.4 Optimization of kinetic parameters	4.7
4.4.5 Quantification of fenobucarb, temephos and dimethoate	4.9
4.4.6 Method validation	4.11
4.4.7 Comparison of the method with other methods	4.11
4.5 Conclusion	4.13

References	4.14-4.21
Chapter 5: Biosensor fabrication by immobilizing GST enzyme on platinum electrode using graphene oxide mediator	5.1–5.22
5.1 Introduction	5.1
5.2 Objectives of this chapter	5.2
5.3 Experimental	5.3
5.3.1 Synthesis of graphene oxide	5.3
5.3.2 Biosensor preparation	5.4
5.3.3 Analytical procedure	5.5
5.3.3.1 Electrochemical measurements	5.5
5.3.3.2 Gas chromatographic measurements	5.5
5.4 Results and Discussion	5.5
5.4.1 Characterization	5.5
5.4.2 Electrochemical impedance measurements	5.6
5.4.3 Cyclic voltammetric behaviour	5.7
5.4.4 Chronoamperometric study	5.8
5.4.5 Sensor operational and kinetic parameters	5.9
5.4.5.1 Saturated substrate concentration and apparent Michaelis-Menten constant	5.9
5.4.5.2 Effect of pH	5.11
5.4.5.3 Incubation time	5.12
5.4.5.4 Enzyme reactivation studies	5.12
5.4.5.5 Enzyme leaching test	5.13
5.4.5.6 Precision measurement	5.13
5.4.5.7 Storage stability	5.13
5.4.6 Quantification of pesticides	5.14
5.4.7 Validation study	5.17

5.5 Conclusion	5.18
References	5.19-5.22

Chapter 6: Study of inhibition kinetics of selected pesticides belonging to different classes by using GST catalyzed GSH-CDNB reaction **6.1–6.11**

6.1 Introduction	6.1
6.2 Objectives of this chapter	6.2
6.3 Experimental	6.2
6.3.1 Cyclic voltammetry measurements	6.2
6.4 Results and Discussion	6.3
6.4.1 Kinetics of Inhibition	6.3
6.5 Conclusion	6.8
References	6.9-6.11

Chapter 7: Conclusions and future scope **7.1–7.5**

7.1 Overall conclusions	7.1
7.1.1 Chapter 1	7.1
7.1.2 Chapter 2	7.2
7.1.3 Chapter 3	7.2
7.1.4 Chapter 4	7.3
7.1.5 Chapter 5	7.3
7.1.6 Chapter 6	7.4
7.2 Significance of the work	7.5
7.3 Drawbacks	7.5
7.4 Future scope	7.5

Appendix A **A.i–A.xliv**

Appendix B **B.i–B.ii**