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Bio-sensing of organophosphorus pesticides: A review

C.S. Pundir[a,*,1], Ashish Malik[b], Preety[a,1]

[a] Department of Biochemistry, M D University, Rohtak, Haryana, India
[b] Department of Botany, M D University, Rohtak, Haryana, India

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ABSTRACT

Organophosphorus (OP) pesticides have been used widely as agricultural and household pest control agents for almost five decades and persist in our water resources, fruits, vegetables and processed food as health and environmental hazardous compounds. Thus, detection of these harmful OP pesticides at an ease with high sensitivity and selectivity is the need of hour. Bio-sensing technology meet these requirements and has been employed at a large scale for detection. The present review is aimed mainly to provide the overview of the past and recent advances occurred in the field of biosensor technology employed for the detection of these OP compounds. The review describes the principle and strategy of various OP biosensors including electrochemical (amperometric, potentiometric), thermal, piezoelectric, optical (fluorescence, Surface Plasmon Resonance (SPR)), microbial and DNA biosensors in detail. The electrochemical biosensors are generally, based on inhibition of enzyme, acetyl cholinesterase (AChE), butyryl cholinesterase (BChE), tyrosinase and alkaline phosphatase or enzyme (organophosphorus hydrolase, OPH)) catalyzed reaction. The detection limits and linearity range of various OP biosensors have also been compared. AChE inhibition based amperometric OP biosensors exhibited the lowest detection limit of 1×10^{-11} μM with a linearity range of $1.0 \times 10^{-11} - 1.0 \times 10^{-2}$ μM.

1. Introduction

The term "pesticide" is defined as any chemical entity, which has the ability to kill the various kinds of pests including rodents, insects, fungi, weeds etc. and henceforth categorized accordingly as rodenticides, insecticides, fungicides and herbicides. However, on the basis of chemical composition, pesticides can be classified into five main groups as organochlorines, organophosphorus (OP), carbamates, pyrethrin and pyrethroids compound. OP compounds, due to their wide utility as insecticides, helminthicides, nematocides, fungicides and herbicides have been in continuous use for the five decades in agricultural pest control throughout the world with their alone contribution of about 38% of the total pesticides used worldwide (Singh, 2009). The extensive use of these OP compounds in contaminated water resources, fruits, vegetables and processed foods adversely affect the health of various non-target organisms such as birds, fishes and humans beings. The primary acute toxicity caused due to these OP compounds exposure is the inhibition of the activity of an enzyme acetylcholinesterase (AChE), essentially needed for the appropriate functioning of the central nervous system (CNS). Nearly all OP compounds are anti acetylcholinesterase and act via a common mechanism of phosphorylation of AChE, which disabled AChE to catalyze acetylcholine and result into over-accumulation of acetylcholine leading into cholinergic toxicity (Chapalamadugu and Chaudhry, 1992).

In view of their hazardous effect on human health (both cholinergic and non-cholinergic) and environment, the prime concern should be of their rapid and reliable detection by a convenient method. Although various laboratory based analytical methods like colorimetry, capillary electrophoresis (CE), thin layer chromatography (TLC), gas liquid chromatography(GLC), high performance liquid chromatography (HPLC), nuclear magnetic resonance (NMR) spectroscopy, mass spectrometry (MS) and enzyme-linked immunosorbant assays (ELISA) have been employed so far, but these suffer from one and the other drawback such as use of expensive instrumentation, time consuming process and requirement of trained personnel (Yao et al., 1991).

The need of rapid, cost effective and reliable method for field analysis of OP compounds has been fulfilled by the rapidly developing biosensor technology. Biosensors, as a small analytical device (having bio-recognition element and signal transducers) seem to be perfect for in site detection of these OP compounds, with quite high sensitivity and specificity (Kaur and Prabhakar, 2017). Hence, the present review describes the various biosensors employed for detection of OP compounds

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[*] Corresponding author. Department of Biochemistry Maharshi Dayanand University, Rohtak, India.
E-mail addresses: chandraspundir@gmail.com, pundircs@rediffmail.com (C.S. Pundir), ashishmalik31@gmail.com (A. Malik), preetymalik31@gmail.com (Preety).
[1] Postal address – Department of Biochemistry, M. D. University, Rohtak.

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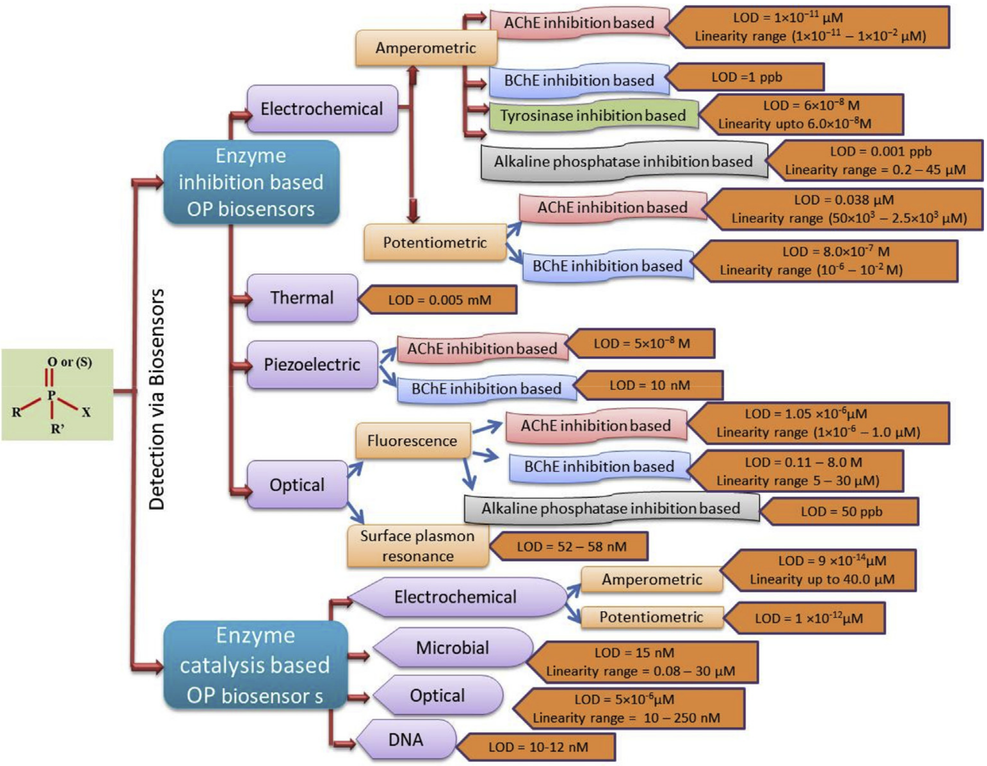

Scheme 1. Overview of bio-sensing techniques employed for the detection of organophosphorus (OP) compounds.

so far. A detailed study of detection limit, linearity range and other properties of these biosensors have also been explored as represented in Scheme 1.

Broadly, bio-sensing of OP compounds can be done either by assaying the residual activity of the enzyme, such as AChE, BChE, tyrosinase and alkaline phosphatase, which get inhibited by OP compound or by measuring the hydrolytic ability of enzyme, OPH, which catalyzes the hydrolysis of OP compounds, at ease. However, based on the transducer used, OP biosensors can be further categorized as Electrochemical (Amperometric and Potentiometric), Thermal, Piezoelectric, Optical (Fluorescence and Surface plasmon resonance (SPR)), Microbial and DNA biosensor.

2. Organophosphorus (OP) compounds

2.1. Chemical structure and role

OP compounds are basically esters of phosphoric acid with varying combinations of oxygen, nitrogen, carbon and sulphur attached to them. All the OP compounds shared a common structural pattern, a phosphorus atom present in the center, which is double bonded with either oxygen or sulfur atom and single bonded with alkoxy/aryloxy/thioalkoxy groups (R and R') and X the any leaving group.

As OP compounds showed low bioaccumulation, fast biodegradation, high toxicity and broad target range, they are widely used as pesticides in agriculture for protection of agricultural crops and in household for parasitic control in domestic animals and as pest repellant. Based on their structural diversity the most commonly used OP pesticides are chlorpyrifos, paraoxon, malathion, parathion, coumaphos, diazinon, methyl parathion, fenitrothion and cyanophos.

2.2. Mode of action

The basic mechanism of the OP pesticides action is to kill the pests by inhibiting the activity of their essential enzyme, AChE that impairs the regulated functioning of CNS. AChE is the prime enzyme of CNS which catalyzes hydrolysis of neurotransmitter, acetylcholine present in the synaptic membrane into choline along with generation of acetylated enzyme. Choline so formed further helps in regeneration of acetylcholine and regulated level of which conduct transmission of nerve impulse at its pace. However, organophosphates inhibit the enzyme irreversibly as they undergoes covalent bond formation with the serine residue (Ser_{200}) present at the active site of AChE via nucleophilic attack and generated phosphorylated enzyme, which is unable to catalyze acetylcholine as shown in Fig. 1. This results into accumulation of acetylcholine and develops cholinergic toxicity involving a deceasement in respiratory center of brain followed by paralysis and sometimes death (Pohanka et al., 2009).

3. Biosensors

Biosensor is the analytical device, which detects any target analyte of interest with the help of (i) bio recognition element that recognizes the physical, chemical or biological response produced by the analyte and (ii) transducer, which converts response generated into the measurable signal. As these biosensors provide solutions for analytical measurement of any kind of analyte in both laboratory and field testing, so have wide utility in safety measurements of food, monitoring of environmental issues, as biological and chemical warfare agents, agricultural product safety, clinical diagnostics, biomedical research to process control. Similarly, biosensors employed for the detection of OP pesticides/compounds proved to be more specific, reliable, reproducible and convenient as compared to other conventional methods available so far such as colorimetry, ELISA, CE, MS and chromatographic techniques such as TLC, GLC and HPLC (Pundir and Chauhan, 2012).

4. Enzymes based OP biosensors

Enzymatic biosensors employ enzymes (either inhibiting or

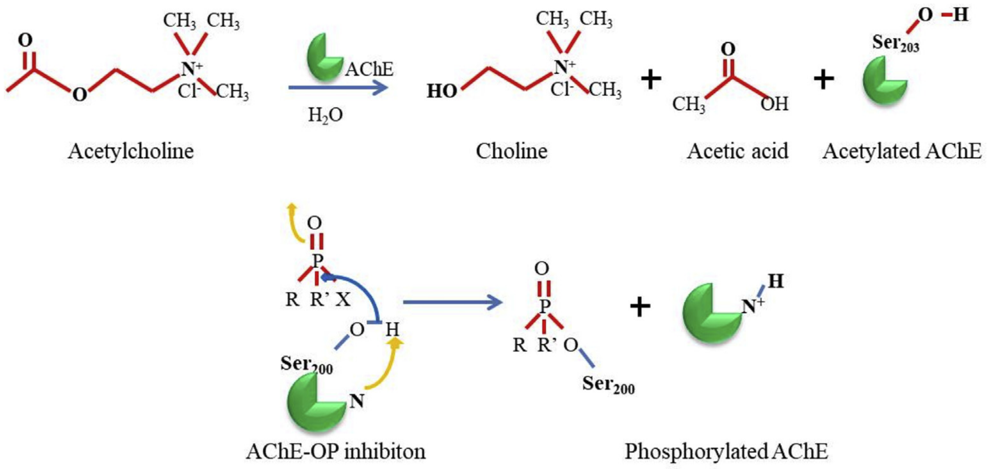

Fig. 1. Mode of action of OP pesticides on the activity of the enzyme, acetylcholinesterase (AChE).

hydrolytic) in close proximity with the transducer element facilitating high and specific reactivity with their substrate, at an ease. In OP biosensors, the target analyte can be detected by means of i) an inhibition mechanism, where, the OP pesticide inhibits the enzymatic activity of enzymes like AChE, BChE, tyrosinase and alkaline phosphatase and thus measuring the decrease in its response ii) a catalytic mechanism, where the hydrolytic ability of enzyme, OPH to catalyzes hydrolysis of OP compounds is directly measured, which is increased in accordance to OP compound. Thus, in the field of bio-sensing for OP compounds, enzymatic biosensors represent the most attractive area of research due to their fast response, high robustness and easiness of immobilization (Rassaei et al., 2011).

4.1. Enzyme inhibition based OP biosensors

4.1.1. Principle of enzyme inhibition based OP biosensor

In these OP biosensors, in the presence of the OP compounds, carbamates, neural toxins and other drugs, enzyme AChE or BChE is unable to convert the substrate acetyl thio-choline or butyryl thio-choline into thiocholine and acetic acid/butyric acid. Thio-choline if generated (in absence of OP compounds) undergoes oxidation/dimerization under applied voltage, but in presence of OP compounds, the current produced due to oxidation at anode have inverse relation to the amount of OP pesticides in the given sample (Rajangam et al., 2018). A schematic representation for the functioning of AChE/BChE biosensors for detection of OP compounds is shown in Fig. 2.

Other enzymes like tyrosinase (EC 1.14.18.1), a copper containing enzyme, which oxidizes easily the phenols (such as tyrosine) to o-quinonesand a Mg^{2+} and Zn^{2+} containing alkaline phosphatase enzyme known to catalyze the hydrolysis of phosphorylated compounds with broad substrate specificity, get inhibited due to the presence of various drugs, inorganic salts, heavy metals, toxic environmental pollutants including carbamates and OP pesticides. The inhibition in the activity of these enzymes results in the decrease of biosensor response measurement and can be quantified as the amount of OP compounds present in the given sample.

4.1.2. Classification of enzyme inhibition based OP biosensor

The enzyme inhibition based OP biosensors can be classified on the basis of various matrices used for immobilization of enzymes, on the physical and chemical interactions, between them. But on the basis of transducer used, enzyme inhibition based OP biosensor can be classified into 4 types (i) Electrochemical (Amperometric and Potentiometric) (ii) Thermal (iii) Piezoelectric (iv) Optical (Fluorescence and Surface plasmon resonance (SPR)).

4.1.3. Electrochemical enzyme inhibition based OP biosensor

Electrochemical OP biosensors analyzes the electro active species generated by the biological component or bio-receptor (enzymes – AChE, BChE, tyrosinase and alkaline phosphatase) with the help of electrochemically active transducer element i.e. electrode. The signals produced so can be easily monitored by either change in current generated by redox reaction as in amperometric biosensors or by change in the pH of the reaction medium as in potentiometric biosensors (Andreescu and Marty, 2006).

4.1.3.1. Amperometric enzyme inhibition based OP biosensor. **Principle:** In enzyme based amperometric biosensors, net current is generated due to the redox reactions catalyzed by biological recognition element (enzymes) under a constant potential applied between a working

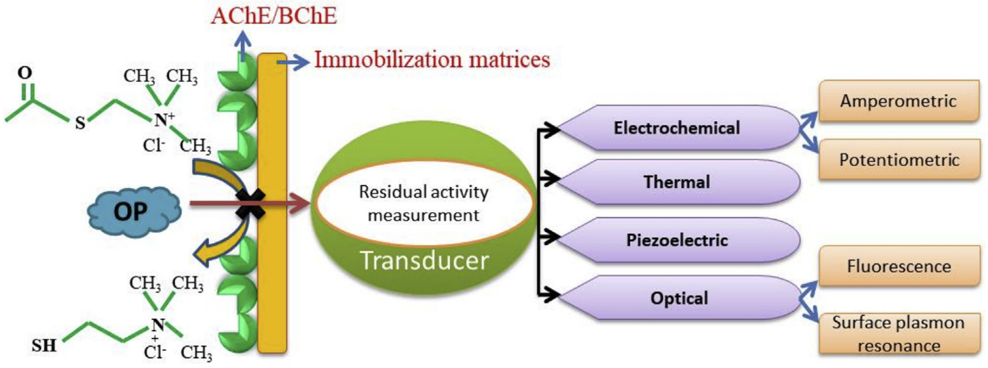

Fig. 2. A representation of AChE/BChE inhibition based OP biosensors.

Table 1
Various AChE inhibition based amperometric biosensors fabricated for OP detection.

Sr No.	Electrode material/immobilization matrix	Immobilization method	Detection limit (μM)	Linearity (μM)	Application/Analyte/Inhibitors	Incubation time (min)	Storage stability (days)	References
1.	SPE	Cross-linking	0.18	0.18–54.0	Paraoxon	10	NR	Li et al. (1999)
2.	Graphite–epoxy composite/SPE	Cross-linking	1.0×10^{-4} and 1.0×10^{-5}	NR	Paraoxon, carbofuran	15	5	Albareda-Sirvent et al. (2001)
3.	SPE (TCNQ mediator in the graphite electrode)	Adsorption	3.0×10^{-6}	$5 \times 10^{-2} - 0.2$	Chlorpyrifos ethyl oxon	10	50	Bonnet et al. (2003)
4.	o-phenylenediamine onto carbon/CoPC SPE	Entrapment	1×10^{-11}, 1×10^{-10} and 1×10^{-10}	$1.0 \times 10^{-11} - 1.0 \times 10^{-2}$	Dichlorvos, Parathion,			
Azinphos	10	92	Law and Higson (2005)					
5.	Al_2O_3 sol–gel matrix SPE	Adsorption	0.01	0.1–80	Dichlorvos	15	5	Shi et al. (2006)
6.	Sol–gel matrix on TCNQ-modified SPE	Entrapment	1×10^{-2}, 8×10^{-4} and 2×10^{-2}	NR	Carbaryl, carbofuran pirimicard	20	45	Bucur et al. (2006)
7.	PB-modified SPE	NR	0.126, 0.124, 7.2×10^{-3} and 1.6×10^{-3}	0.063–0.315, 0.124–0.497, 7.2×10^{-3}–18.1×10^{-3} and 1.6×10^{-3} –6.5×10^{-3}	Aldicarb, Carbaryl, Paraoxon, Chlorpyrifos-methyl oxon	30	21	Arduini et al. (2006)
8.	CoPC/SPCEs	Cross-linking	4.9×10^{-4}	10^{-5} – 1.0	Carbofuran	15	NR	Laschi et al. (2007)
9.	SWCNT—CoPC/SPE	Covalent	0.01 and 6.3×10^{-3}	0.018–0.181 and 6.36×10^{-3} –0.159	Paraoxon, malaoxon	15	3	Ivanov et al. (2011)
10.	CdTeQDs/AuNPs/CHIT/GCE	Covalent	1.34	4.4×10^{-3} – 4.48 and 8.96–67.20	Monocrotophos	8	30	Du et al., 2008a
11.	CdTe QDs/Au electrode	Covalent	2.98×10^{-3}	4.96×10^{-3}–2.48	Carbaryl	10	30	Du et al. (2008c)
12.	MWCNT-Au nanocomposites/GCE	Hydrophilic surface for biomolecule adhesion	1.81×10^{-3}	3.0×10^{-3} – 3.027	Malathion	8	30	Du et al. (2010a,b,c)
13.	AuNPs/PB/GCE	Adsorption	3.5×10^{-9}	4.48×10^{-3} –4.48×10^{-2}	Monocrotophos	10	30	Wu et al. (2011a,b)
14.	PB–CHIT/GCE	Covalent	3.0×10^{-3}	0.01–0.4 and 1.0–5.0	Carbaryl	10	NR	Song et al. (2011)
15.	CdS–decorated graphene nanocomposite	Adsorption	3.4×10^{-3}	9.9×10^{-3} – 9.93	Carbaryl	2	20	Wang et al. (2011a,b)
16.	TiO_2-decorated graphene/GCE	Adsorption	1.4×10^{-3}	4.9–74.5 and 74.5–9.9×10^{3}	Carbaryl	3	20	Wang et al. (2011a,b)
17.	AuNPs–$CaCO_3$ bioconjugate/Au electrode	Adsorption	0.1×10^{-3}	0.1×10^{-3} –100×10^{-3} and 0.1×10^{-3} –70×10^{-3}	Malathion, Chlorpyrifos	10	90	Chauhan et al. (2011a,b)
18.	Fe_3O_4NPs/MWCNT/Au electrode	Covalent	0.1×10^{-3}	0.1–40×10^{-3}, 0.1–50×10^{-3}, 1–50×10^{-3} and 10–100×10^{-3}	Malathion, Chlorpyrifos, Monocrotophos and Endosulfan	10	60	Chauhan and Pundir (2011)
19.	Fe_3O_4NPs/MWCNT/ITO electrode	Covalent	0.1×10^{-3}	0.1–70×10^{-3}, 0.1–50×10^{-3}, 0.1–70×10^{-3} and 0.1–100×10^{-3}	Malathion, Chlorpyrifos, Monocrotophos and Endosulfan	10	90	Chauhan and Pundir (2012)
20.	AuNPs -MWCNT/GCE	Adsorption	1.0×10^{-3}	0.1×10^{-3} - 7.0×10^{-3}	NR	30	NR	Jha and Ramaprabhu (2010)
21.	PVA-SbQ membrane/Pt electrode	Entrapment	7.2×10^{-5}, 1.88 and 0.049	NR	Paraoxon, Maneb and Thifensulfuronmethyl	30	30	Marty et al. (1993)
22.	Poly(2-hydroxyethyl methacrylate) membrane/oxygen electrode	Entrapment	0.119	0.05–2.62	Aldicarb	5	2	Kok et al. (2002)
23.	Cellophane membrane/Au electrode	Cross-linking	1.45	1.45–7.26	Paraoxon	15	NR	Rekha and Murthy (2008)
24.	Hybrid mesoporous silica membrane/Pt electrode	Entrapment	1.2×10^{-3}	1.0×10^{-3}–0.3	DZN-oxon	15	80	Shimomura et al. (2009)
25.	PVA-SbQ polymer/SPE	Entrapment	1.91×10^{-2} 1.24×10^{-3}	NR	Paraoxon and Chlorpyrifos Ethyl oxon	10	NR	Andreescu et al. (2002a)
26..	MWCNT/PAN membrane/Pt electrode	Affinity bonds using Concanavalin A	5.0×10^{-9}	$3.6 \times 10^{-8} - 3.6 \times 10^{-5}$	Paraoxon	20	120	Ivanov et al. (2010)
27.	PAN/AuNPs/Pt electrode	Covalent	0.026×10^{-5}	$3.6 \times 10^{-7} - 3.6 \times 10^{-4}$	Paraoxon	20	30	Marinov et al. (2010)

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Table 1 (*continued*)

Sr No.	Electrode material/immobilization matrix	Immobilization method	Detection limit (μM)	Linearity (μM)	Application/Analyte/Inhibitors	Incubation time (min)	Storage stability (days)	References
28.	PAMAM-Au/CNTs/GCE	Electrostatic interaction	4.0×10^{-3}	$4.8 \times 10^{-3} - 9.0 \times 10^{-2}$	Carbofuran	9	21	Qu et al. (2010)
29.	MSF-PVA/GCE	Entrapment	0.2×10^{-3}	0.2×10^{-3} –44.8×10^{-3}	Monocrotophos	10	30	Wu et al., (2011a)
30.	Polyacrylamide membrane/pH electrode	Cross-linking	3.62×10^{3}	NR	Dichlorvos	30	50	Stein and Schwedt (1993)
31.	PEI/SPE	Non-covalent	1.0×10^{-4}	NR	Dichlorvos	2 days	NR	Vakurov et al. (2004)
32.	Mercaptobenzothia-zole/PANI/Au electrode	Adsorption	0.48×10^{-3} 0.61×10^{-3}	NR	Diazinon Fenthion	20	NR	Somerset et al. (2007)
33.	PPy and PANI copolymer doped with MWCNTs/GCE	Adsorption	3.02×10^{-3}	0.030–1.51 and 3.027–75.67	Malathion	15	30	Du et al. (2010a)
34.	ZnS and poly indole-5-carboxylic acid/Au electrode	Covalent	0.1×10^{-3} and 1.5×10^{-3}	0.1–50 $\times 10^{-3}$ and 1.5–40 $\times 10^{-3}$	Malathion and Chlorpyrifos	10	60	Chauhan et al. (2011a)
35.	Silica sol–gel/SPE	Encapsulation	0.024, 0.015 and 0.012	0.01–0.001	Paraoxon, Dichlorvos and Chlorpyrifos Ethyl oxon	20	6	Andreescu et al. (2002b)
36.	TEOS sol–gel/GCE	Encapsulation	0.008	0.008–0.81	Oxydemeton methyl	20	21	Anitha et al. (2004)
37.	TMOS sol gel film/SPE	Encapsulation	1.0×10^{-3}	1.0 and 3.0×10^{3}	Dichlorvos	15	NR	Sotiropoulou, and Chaniotakis (2005)
38.	Alumina sol–gel/sonogel-carbon electrode	Encapsulation	2.5×10^{-4}	0.5	Chlorpyriphos-ethyl-oxon	10	50	Zejli et al. (2008)
39.	Zinc oxide sol–gel/SPE	Electrostatic interactions	0.127	0.127–5.01	Paraoxon	10	90	Sinha et al. (2010)
40.	Silica sol–gel film/carbon paste electrode	Encapsulation	3.0×10^{-4} and 0.47	3.7×10^{-4} –1.8×10^{-3} and 0.27–4.09	Methyl parathion and acephate	20 and 4	30	Raghu et al. (2011)
41.	ZrO_2/CHIT composite film/GCE	Adsorption	1.3, 5.0×10^{-3} and 1.7	6.6–440, 0.01–0.59 and 8.6–520	Phoxim, Malathion and Dimethoate	15	30	Yang et al. (2005)
42.	MWCNT-CHIT composite (MC)/GCE	Covalent	NR	NR	Carbaryl, Malathion, Dimethoate Monocrotophos	8	30	Du et al. (2007a)
43.	AuNPs/Au electrode	Adsorption	33×10^{-3}	10×10^{-3}–135×10^{-3}	Carbofuran	20	7	Shulga and Kirchhoff (2007)
44.	CHIT-GNPs/Au electrode	Chemisorption/ Esorption	0.1×10^{-3}	$0.3 \times 10^{-3} - 60.5 \times 10^{-3}$	Malathion	15	NR	Du et al. (2008d)
45.	Gold–platinum bimetallic NPs/GCE	Crosslinking with gluteraldehyde	50×10^{-3}, 40×10^{-3} and 40	50–200 $\times 10^{-3}$, 1.40–50 $\times 10^{-3}$ and 40–60	Paraoxon ethy, Sarin and Aldicarb	25	NR	Upadhyay et al. (2009)
46.	PbO_2/TiO_2/Ti	Adsorption	0.1×10^{-3}	0.01–20	Trichlorfon	10	5	Wei et al. (2009)
47.	PB and CHIT/GCE	Glutaraldehyde crosslinking	0.113×10^{-4} 0.703×10^{-4} 0.194×10^{-4} and 0.33×10^{-4}	$0.45 \times 10^{-4} - 0.045$, $0.234 \times 10^{-3} - 0.046$, $0.116 \times 10^{-3} - 0.0194$ and 0.167×10^{-3} - 0.0335	Dichlorvos, Omethoate, Trichlorfon and Phoxim	10	NR	Sun and Wang (2010)
48.	AuNPs/GCE	Adsorption	7.0×10^{-3}	28×10^{-3} - 170×10^{-3}	Methamidophos	10	7 days	Yan et al. (2010)
49.	Calcium carbonate–CHIT composite film/GCE	Entrapment	3.7×10^{-3}	0.018–0.759 to 2.84–14.24	Methyl parathion	10	NR	Gong et al. (2009a)
50.	PANI/CNT wrapped with ssDNA/Au electrode	Covalent	$1.0 \times 10-6$	$1.0 \times 10-5$ and 1.0	Methyl Parathion, Chlorpyrifos	15	5 days	Viswanathan et al. (2009)
51.	AuNPs–PPy nanowires composite film modified GCE	Entrapment	$7.5 \times 10-3$	0.018–0.45 and 1.89–17.0	Methyl parathion	12	30	Gong et al. (2009b)
52.	AuNPs-SiSG/GCE	Hydrogen bonds	0.44	NR	Monocrotophos	10	30	Du et al. (2008b)
53.	xGnPs–CHIT composite/GCE	Covalent	1.58×10^{-4}	1×10^{-4}–1.0	Chloropyrifos	10	10	Ion et al. (2010)
54.	MWCNTs/AuNPs-CHIT/GCE	Adsorption	0.01	0.1 to 10	Monocrotophos	NR	50	Norouzi et al. (2010)
55.	Recombinant AChE B394/CoPC/PVA-AWP	Entrapment	5×10^{-18}, 5×10^{-15}, 5×10^{-16}	NR	Chlorpyriphos-oxon, ethyl paraoxon and malaoxon	15	NR	Rupesh et al. (2012)
56.	MWCNTs/SnO_2/CHIT	Adsorption	0.05	0.05 to 1.0×10^{5}	Chlorpyriphos	NR	NR	Chen et al. (2015a,b)

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Table 1 (*continued*)

Sr No.	Electrode material/immobilization matrix	Immobilization method	Detection limit (μM)	Linearity (μM)	Application/Analyte/Inhibitors	Incubation time (min)	Storage stability (days)	References
57.	MWCNTs-SnO_2-CHIT-SPE	Adsorption	0.05	0.05 to 1.0 × 10^3	Chlorpyriphos	NR	30	Chen et al. (2015a,b)
58.	CH–MTclay/ITO/GE	Entrapment	0.448	0.5 to 1000	Chlorpyriphos	NR	NR	Sarkar et al. (2017)
59.	IL-GR/Gelatin gel modified GCE	Entrapment	4.6 × 10^{-20}	1.0 × 10^{-16} to 5.0 × 10^{-14}	Monocrotophos	5	15	Zheng et al. (2015)
60.	CdSe@ZnS QDs/graphene/ITO/GE	Covalent	10^{-20} 10^{-18}	10^{-18} - 10^{-12}	Paraoxon Dichlorvos	NR	NR	Li et al. (2015)
61.	Unsubstitutedpillar[5]arene/Carbodiimide/Carbon black/GCE	Covalent	4 × 10^{-18} 5 × 10^{-15} 2 × 10^{-17}	1 × 10^{-17}-1 × 10^{-12} 1 × 10^{-14} – 7 × 10^{-12} 1 × 10^{-16}-2 × 10^{-12}	Malaoxon Methyl-paraoxon Carbofuran	10	NR	Shamagsumova et al. (2015)
62.	Platinum–carbon aerogels (Pt–CAs) composite/Boron-doped diamond(BDD) electrode	Entrapment	3.1 × 10^{-19} 2.7 × 10^{-18}	10^{-11} –10^{-6}	Methamidophosmonocrotophos	NR	NR	Liu and Wei, 2014
63.	AuNP/RGO/GCE	Adsorption	0.35	0.3–300	Triazophos	15	7	Ju et al. (2015)
64.	([BSmim]HSO_4)-AuNPs-porous carbon composite modified BDD electrode	Entrapment	2.99 × 10^{-19}	4.5 × 10^{-19}–4.5 × 10^{-15}	Dichlorvos	NR	NR	Wei and Wang (2015)
65.	ERGO/AuNPs-β-cyclodextrin/Prussian blue-chitosan/GCE	Cross linking	4.14 × 10^{-6}	7.98 × 10^{-6}-2.00 × 10^{-3}	Malathion	NR	NR	Haiyan et al. (2015)
66.	Fe_3O_4/GR/SPE	Covalent	0.02	0.05–100	Chloropyrifos	15	NR	Wang et al. (2016)
67.	MWCNTs/IL/SPE	Adsorption	0.05	0.05 to 1.0 × 10^5	Chloropyrifos	14	30	Chen et al. (2017)
68.	AChE + ChO/CHIT/Fe@AuNPs/Au electrode	Adsorption and covalent binding	0.005	0.005–400	Acetylcholine	NR	90	Chauhan and Pundir (2014)
69.	3Dgraphene oxide network/MWCNTs	Entrapment	0.025 × 10^{-3}	0.05–1 × 10^{-3}	Paraoxon	10	20	Li et al. (2017a,b)
70.	SnO_2-cMWCNTs/Cu	Glutaraldehyde cross-linking	0.1	1.0–160.0	Methyl parathion	NR	40	Dhull (2018)
71.	MWCNT modified film strip/SPE	Physical adsorption	0.5 × 10^{-3}	0.5–6.9 × 10^{-3}	Paraoxon	NR	NR	Joshi et al. (2005)
72.	CNT-NH_2/GC electrode	Physical adsorption	0.08 × 10^{-3}	0.2–1.0 × 10^{-3}	Paraoxon	NR	NR	Yu et al. (2015)
73.	Poly(4-(2,5-di(thiophen-2-yl)-1H-pyrrol-1-yl) benzenamine) (poly(SNS-NH_2))	Entrapment	0.09 × 10^3	0.05–8.00 × 10^3	Paraoxon, parathion and chlorfenvinphos	NR	NR	Kesik et al. (2014)
74.	Polypyrrole	Cross linking	1.1	NR	Paraoxon	NR	120	Dutta and Puzari (2014)
75.	poly (p-aminothiophenol) PATP/AuNPs/SPCE	Physical adsorption	1.0 × 10^{-15}	NR	Paraoxon	NR	NR	Li et al. (2017a,b)
76.	Bi/Gr/GCE	Physical adsorption	2.0 × 10^{-3}	NR	Paraoxon	NR	NR	Stoytcheva et al. (2017)

NR – Not reported.

electrode and a reference electrode and the magnitude of this current is directly proportional to the electro active species concentration present in the sample. AChE activity is inhibited by the presence of OP pesticides and by the use of different kind of immobilization matrices for AChE, various amperometric biosensors with different detection limit and linearity range for OP pesticides have been fabricated so far as summarized in Table 1.

Among the large number of fabricated amperometric AChE based OP biosensors, the lowest detection limit (1×10^{-11} μM) and linearity range ($1.0 \times 10^{-11} - 1.0 \times 10^{-2}$ μM) was observed by the biosensor having immobilized AChE on o-phenylenediamine/carbon/cobalt pthalocyanin (CoPc) screen printed electrode for detection of OP compounds viz. dichlorvos, parathion and azinphos. The enzyme was entrapped in the immobilization matrix and showed a good storage stability of 92 days with an incubation time of only 10 min (Pundir and Chauhan, 2012). However, AChE immobilized onto cellophane membrane/Au electrode via crosslinking showed the highest limit of detection (LOD) (1.45 μM) and linearity range (1.45–7.26 μM) with 15 min of incubation time for detection of paraoxon (Pundir and Chauhan, 2012).

To detect paraoxon in water samples and standard pesticides samples, amperometric biosensors werefabricated by entrapping BChE alone and BChE in combination with choline oxidase into kappa carrageenangel. The biosensors showed LOD of 4.5 μg/L and 4.8 μg/L respectively (Luigi et al., 1991, 1999). Amperometric biosensor having BChE immobilized over prussian blue modified screen printed electrodes showed a LOD of 4 ppb for paraoxon and 1 ppb for chlorpyrifos-methyl oxon (Pundir and Chauhan, 2012). In another work, Arduini et al. (2015) proposed a BChE based amperometric biosensor embedded in a flow system by cross-linking BChE onto a screen-printed electrode having prussian blue nanoparticles. The LOD for detection of paraoxon was 1 ppb with storage stability of 60 days. BChE tends to show quite higher storage stability and hence could be used easily for future commercialization in the agro food biosensor market.

In another amperometric studies, biosensor fabricated with tyrosinase cross linked onto CoPc showed a linearity range of $2.28 \times 10^{-8} - 3.8 \times 10^{-7}$ M for methyl parathion and $6.24 \times 10^{-8} - 1.64 \times 10^{-7}$ M for diazinon (Vidal et al., 2006). However, tyrosinase entrapped and cross-linked to 1,2-naphthoquinone-4-sulfonate (NQS) and prussian blue revealed a LOD of 6×10^{-8} M and 10^{-7} M and linearity up to 8×10^{-6} M and $10^{-7} - 10^{-6}$ M for dichlorvos and paraoxon OP pesticides respectively (Albuquerque and Ferreira, 2007; Sajjadi et al., 2009). A newer approach of organic phase enzyme sensor by immobilizing tyrosinase onto kappa carrageenan gel was also utilized for the detection of dimethoate, paraoxon and malathion OP pesticides at a LOD of 10^{-6} M, 5×10^{-6} M, 5×10^{-6} M and linearity range of $2 \times 10^{-6} - 0.2$ M, $10^{-5} - 10^{-2}$ M, $10^{-5} - 10^{-2}$ M respectively (Campanella et al., 2007). To detect OP compound, malathion amperometric biosensor based on immobilization of the enzyme alkaline phosphatase obtained from thylakoids of spinach via a new method called as laser-induced forward transfer (LIFT) for achieving fast and reproducible detection with a lower value of 0.001 ppb and linearity range of 0.2–45.0 μg/L (Touloupakis et al., 2012). In other biosensor, alkaline phosphatase cross-linked with algae-bovine serum albumin (BSA)/ZnO nanoparticles/glassy carbon electrode was used for voltametric detection of OP pesticide, chloropyrifos without any kind of interferences from other pesticides (acephate, malathion, triazophos) as well as alkali metals (Pabbi et al., 2018).

Merits: These inhibition based amperometric OP biosensors showed rapid response and high sensitivity with quite wide linear range and LOD. **Demerits:** Low specificity due to interfering substances such as heavy metals, carbamate pesticides and long incubation steps were the major limitations for these inhibition based biosensors.

4.1.3.2. Potentiometric enzyme inhibition based OP biosensor. **Principle:** The basic principle of potentiometric biosensors lies on capability of conversion of hydrogen (H^+) ions either generated or absorbed by ion-selective electrodes (transducing element) into an electrical signal. A change in pH directly depends on the target analyte present in the given sample (Andreescu and Marty, 2006). To detect OP pesticides dichlorvos, Lvnitskii and Rishpon (1994) fabricated a potentiometric biosensor by linking covalently AChE on PEI-coated GCE, which showed a good LOD of 1.0 μM with an incubation of 10 min. AChE was also cross linked with glutaraldehyde onto nylon and cellulose nitrate membrane/pH electrode by Ivanov et al. (2000), which exhibited LOD of 0.038 μM and linearity range of 50×10^3–2.5×10^3μM for trichlorfon detection at an incubation time of 15 min and good storage stability of 30 days.

A potentiometric BChE inhibition based OP biosensor having entrapped BChE onto plasticized poly vinyl chloride in the pH in the range of 2.0–10.5 showed an improved selectivity with linearity range of $10^{-6} - 10^{-8}$ M for profenofos and $10^{-5} - 10^{-6}$ M for 2, 2 dichloro vinyl dimethyl phosphate(DDVP) (Toshihiko and Nobuhiko, 1995). The potentiometric biosensor having BChE coupled onto the film with glutaraldehyde revealed a LOD of 10^{-7} M for trichlorofon with a quick response time (Karine et al., 2002). The BChE based potentiometric biosensor with highly sensitive disposable screen-printed heptakis (2,3,6-tri-o-methyl)-β-cyclodextrin (β-CD) as ionophore was constructed for detection of malathion OP pesticide in human serum with a LOD of 8×10^{-7} mol L^{-1} and linear range of 10^{-6} to 10^{-2} mol L^{-1} (Khaled et al., 2010).

Merits: The LOD and enhanced reproducibility achieved by these potentiometric OP biosensors were adequate to monitor contaminations in food and other agro-products. **Demerits:** Requirement of high potential and fouling problems affects the thiol and OP compounds detection.

4.1.4. Thermal enzyme inhibition based OP biosensor

Principle: The measurement of heat evolved during the enzyme or microorganism catalyzed reactions is used by a heat sensing transducer (thermistor)and employed for fabrication of thermal OP biosensor. For detection of OP pesticide, malathion, an AChE based thermal biosensor measures the decrease in heat evolution as the concentration of malathionin the sample (Verma and Eneyew, 2001). Cholinesterase (AChE/BChE) cross-linked with controlled pores glass beads detected parathion in vegetables (onion, lettuce, salad) at a LOD of 0.005–0.013 mg/kg via phothermal biosensing technique (Lea and Mladen, 2003).

Merits: A major merit of thermal OP biosensors lies in the universal detection principle of specificity of biological reactions, as these all are basically exothermic. **Demerits:** Non-specific heat generated during the reaction, changes in pH value, viscosity and ionic strength can also produce signals, which can interfere in the OP compound detection.

4.1.5. Piezoelectric enzyme inhibition based OP biosensors

Principle: A change in the mass of any piezoelectric crystal occurs, when they are immersed partially or completely in a liquid. This property of piezoelectric crystals has been utilized for fabrication of OP biosensors with implication of biological molecules (antigens, antibodies and enzymes) as the active coatings of these crystals (Marrazza, 2014).

In piezoelectric AChE inhibition based OP biosensors, AChE is immobilized onto the one of the face of QCM crystal exposing it to the reaction mixture. The change in the mass of QCM surface is measured by the changed frequency induced due to enzymatic reaction taking place in the solution. As OP pesticides inhibit the activity of AChE, a diminution in the signal from its original level is observed as a measure of their concentration viz. Abad et al. (1998) noted a LOD of 5.0×10^{-8} M for paraoxon. Karousos et al. (2002) employed two enzymes, AChE and choline oxidase todetermine dichlorvos up to a level of 1 ppm. By using quartz crystal microbalance-precipitation sensor, Kim et al. (2007) measured the amount of OP pesticide, p-nitrophenyl thionobenzenephosphonate, while Halamek et al. (2005) detected diisopropyl fluorophosphate in river water.A novel methodology of coupling the human tetrameric enzyme, BChE to the inhibitor, which is immobilized as ligand on a piezoelectric quartz crystal, was used to avoid complex irreversible cholinesterase OP compounds interactions. This competitive affinity assay showed a LODof 10 nmol/L for diisopropyl fluorophosphate. The biosensor offered a real-time monitoring with regeneration of the sensing surface by destroying the chelate complex formed onto the sensing layer with EDTA (Makower et al., 2003). The highly sensitive and selective quartz crystal microbalance sensors with high analytical performance were obtained by Funari et al. (2013) and Ozkutuk et al., 2013 using photonic and molecular imprinted film for the detection of parathion and paraoxon, respectively.

Merits: Relatively good results are obtained without any need of expensive or hazardous labeled compound. Real time output, simplicity of their use and high sensitivity are the other advantages of these QCM based OP biosensors. **Demerits:** High cost of devices, lesser implementation of new technical protocols, longer time to establish a stable base line and more sensitivity of devices towards high molecular mass analytes limit their use in daily routine analysis.

4.1.6. *Optical enzyme inhibition based OP biosensor*

With the measurement of optical parameters like absorbance, fluorescence and chemiluminescence various biosensors were fabricated by employing fibre optics and optoelectronic transducers. However for detection of OP compounds, only fluorescence-based and surface plasmon resonance (SPR) biosensors have been fabricated so far.

4.1.6.1. Fluorescence enzyme inhibition based OP biosensor. **Principle:**The basic principle of the fluorescence based OP biosensor/chemo sensor is the specific interaction that occurs in between the bio-recognition molecule and target analyte. An optical signal in the form of change in absorption or emission band generated by the chemical interaction is easily detected by the fluorophore and which can be co-related with the concentration of analyte.

Optical biosensors based on AChE electrostatically bonded to poly (allylamine hydrochloride)/CdTe quantum dots/glass resulted into a LOD of 1.05×10^{-5} μM and 4.47×10^{-6} μM and linearity range of $1.0 \times 10^{-6} - 1.0$ μM and 1.0–0.1 μM for paraoxon and parathion respectively (Pundir and Chauhan, 2012), while AChE encapsulated onto chromoionophore (ETH5294) (CM) doped sol–gel film yielded a highest LOD of 2.26 μM and high linear range of 2.26–31.67 μM for dichlorvos (Pundir and Chauhan, 2012). The nanoparticles associated fluorescence biosensor with quite low LOD has been fabricated previously by various researchers as shown in Table 2.

In comparison to BChE, inhibitors are more selective for AChE and showed higher affinity for AChE due to presence of more frequent aromatic amino acids in it. So far various optical biosensors have been fabricated that employed AChE but only one optical biosensors having butyryl cholinesterase entrapped in sol gel directly detecteddichlorovos compounds optically at a LOD of 0.11–8.0 mg/L and working range of 5–30 μg/L with an accuracy of 94.9% and reproducibility of 3–4% (Vangelis and Yannis, 2002). Alkaline phosphatase activity is inhibited by the presence of OP compounds, which is easily utilized for fabrication of optical biosensors with the help of specific hydrophobic biotin–streptavidin interactions as done by Ayyagari et al. (1995). A biotinylated copolymer immobilized onto silanized glass surface was attached to the streptavidin conjugate of alkaline phosphatase and the decrease in the chemiluminescent signal strength was directly measure of inhibited enzyme activity due to the presence of paraoxon. A quite low LOD value of 50 ppb for paraoxon was observed by the developed biosensor with the advantage of a number of times its reusability.

Merits: These are very fast, highly sensitive with 106 times lower the detection limits of the other absorbance techniques (Andreescu and Marty, 2006).The application of quantum dots has many advantages

Table 2
Enzyme Inhibition based optical biosensors fabricated for detection of organophosphorus pesticides.

Sr.No	Enzyme used	Electrode material/immobilization matrix	Immobilization method	Detection limit (μM)	Linearity (μM)	Application/Analyte/ Inhibitors	Incubation time (min)	Storage stability (days)	Reference
1.	AChE	Poly(allylamine hydrochloride)/CdTe quantum dots/glass	Electrostatic interaction	1.05×10^{-5} and 4.47×10^{-6}	$1.0 \times 10^{-6} - 1.0$ and 1.0–0.1	Paraoxon Parathion	15	35	Pundir and Chauhan (2012)
2.	AChE	Glass/sol–gel indicator/ polyvinylidenefluoride membrane	Crosslinking with glutaraldehyde	0.53 and 0.023	0.54–39.8 and 0.022–0.13	Carbaryl Dichlorvos	10	21	Pundir and Chauhan (2012)
3.	AChE	Sol-gel crystals derived from TMOS	Encapsulation	0.94 42.19	3.17–31.48 14.89–998.4	Naled Mecarbam	5	30	Pundir and Chauhan (2012)
4.	AChE	Sol–gel film on a glass cap	Encapsulation	0.098	0.098–0.55	Paraoxon	30	NR	Pundir and Chauhan (2012)
5.	AChE	Chromoionophore (ETH5294) (CM) doped sol–gel film	Encapsulation	2.26	2.26–31.67	Dichlorvos	15	NR	Pundir and Chauhan (2012)
6.	AChE	Bromothymol blue (BTB) doped sol–gel film	Encapsulation	0.11	0.14–5.70	Chlorpyrifos	8	60	Pundir and Chauhan (2012)
7.	BChE	Sol-gel	Encapsulation	0.11×10^{-3} -8.0×10^{-3}	5.0–30.0	Dichlorvos	NR	NR	Vangelis and Yannis (2002)
8.	Alkaline phosphatase	Silanized glass surface	Covalent binding	NR	NR	Paraoxon	NR	NR	Ayyagari et al. (1995)
9.	AChE	Alkanethiol self-assembled monolayer	Covalent binding	$52–58 \times 10^{-12}$	NR	Chloropyrifos	NR	NR	Mauriz et al. (2006)

NR – Not reported.

over other fluorescence reagents, as their high extinction coefficients with size-tunable light emission have narrow and symmetric emission spectra and simultaneous excitation of multiple fluorescence colors. **Demerits:** More prone to compounds having similar chemiluminescent properties.

4.1.6.2. Surface plasmon resonance (SPR) enzyme inhibition based OP biosensors. **Principle:** SPR biosensors measure the concentration of analyte with the help of analysis of binding kinetics between the two interacting molecules. Recently, with the help of alkane thiol self-assembled monolayer, SPR-sensor system and directly immobilized AChE on the surface of SPR chip has been developed for the detection of chloropyrifos at a LOD of 52–58 ng/L (Mauriz et al., 2006; Lin et al., 2006). A change in intensity of surface plasmon resonance angle was recorded in the presence of target analyte.

Merits: SPR based OP biosensors showed quite good reliability even in the presence of low molecular mass analytes. These devices have singularity and offer the real-time monitoring of the analyte without requirement of any labeled molecules. **Demerits:** Although SPR biosensors are mainly employed in the direction of the affinity measuring biosensors but with enzyme inhibition based OP biosensors fewer or none devices has been constructed.

4.2. Enzyme catalysis based OP biosensors

Various limitations associated with enzyme inhibition based biosensors were easily overcome by furnishing the catalytic activity of enzymes, which hydrolyzes these OP compounds into simpler one. Mainly two enzymes (i) organophosphorus acid anhydrolase (OPAA) (E.C. 3.1.8.2) and (ii) organophosphorus hydrolase (OPH) (E.C.3.1.8.1) are capable of effectively cleaving the P–F bond of OP compounds but OPAA is not effective towards other class of OP compounds which contains P–O, P–S and P–CN bonds (Singh, 2009). However, OPH showed a broad range catalytic activity towards P–O, P–S, P–F and P–CN bonds of OP compounds, which makes it the most appropriate alternative for fabrication of biosensors used for OP detection.

4.2.1. Principle

Organophosphorus hydrolase (OPH) is a bacterial enzyme of the amido hydrolase family withbroad specificity towards a range of OP compounds having P–O, P–S, P–F and P–CN bonds as – paraoxon, parathion, coumaphos, diazinon, dursban, methyl parathion etc. In OPH biosensor, OPH catalyzes the hydrolysis of OP compounds as shown in Fig. 3 andyields 4-nitrophenol, which upon recognition by different transducer can be directly proportional to OP pesticides concentration (Singh, 2009).

This hydrolysis either involves a change in pH or generation of electro active species (oxidation of 4-nitrophenol), which is used for quantification by potentiometric and amperometric sensors respectively as represented in Fig. 4.

4.2.2. Classification

Based on the type of transducer used for detection of OP pesticides, these OPH biosensors can be classified into four categories i.e. electrochemical, optical, microbial and DNA biosensors.

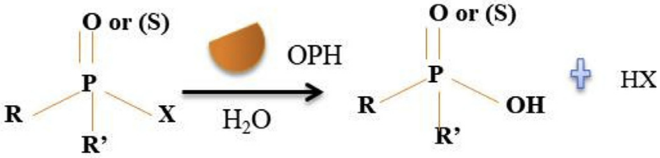

Fig. 3. Mode of action of OP compounds on the activity of the enzyme, organophosphorus hydrolase (OPH).

4.2.3. Electrochemical OPH based OP biosensors

Different OP pesticides like paraoxon, parathion, methyl parathion, fenitrothion are easily hydrolyzed by OPH to generate an electro active species, 4-nitrophenol which upon oxidation produces a net current under applied voltage. The current generation is directly recorded as a biosensor response and proportional to the concentration of OP pesticides in the sample.

4.2.3.1. Amperometric OPH based OP biosensors. A lot of amperometric biosensors have been fabricated so far by various scientists for detection of one and other OP pesticides. These biosensors differ from each other in immobilization matrices but their LOD and linearity range are quite similar. The LOD (9×10^{-14} and 7×10^{-14} μM) and linearity up to 40 and 5 μM was obtained for paraoxon and methyl parathion respectively by the amperometric biosensor having OPH enzyme entrapped onto carbon electrode (Mulchandani et al., 1999a). In Table 3 the characteristics of various OPH based amperometric biosensors for OP pesticides detection are also summarized.

Merits: They are not sensitive towards interference from any other coloured compounds. No requirement of any other enzyme or reagent is needed. **Demerits:** Some compounds like low-molecular mass antioxidants, thiol-containing compounds, glutathione get oxidized by the applied voltage and interfere in the assay.

4.2.3.2. Potentiometric OPH based OP biosensors. Different potentiometric OPH enzyme electrode were modified by immobilizing the enzyme via physical adsorption, cross linking with glutaraldehyde, entrapment and covalent linkage with immobilization matrices such as membrane, screen printed electrode, microsphere, nanoparticles for the direct detection of OP compounds (Table 3) (Weiying et al., 2014). The LOD of 1×10^{-12} μM was obtained for detection of paraoxon by covalently linking the OPH enzyme onto the aqueous amino propyl triethoxysilane (APTS)/glutaraldehyde with a good storage stability of 70 days (Flounders et al., 1999). However, OPH cross linked onto glass membrane showed highest LOD of 5.0 μM for ethyl parathion, methyl parathion and diazinon OP compounds with a storage stability of 30 days only (Mulchandani et al., 1998).

Merits: The required potentiometric devices for this mode of transduction are readily available in market and also simple to construct. **Demerits:** The major problem associated with this pH-sensitive electrode is the strong dependence of the sensor response on the buffer capacity of the containing sample solution. Even the small change in the pH value can lead to a narrow dynamic range with loss of biosensor sensitivity in some cases.

4.2.4. Optical OPH based OP biosensors

In an OPH based optical biosensor, OPH is covalently linked to sulfo-N-hydroxy succinimide modified gold nanoparticles via its lysine residue. A variation in the fluorescence with the change in distance between fluorophore and gold nanoparticle directly detect paraoxon with high sensitivity (Simonian et al., 2005). In another optical biosensors, *Sphingomonas sp.* with OPH is immobilized onto microplate and onion membrane and both of these detect methyl parathion at a detection limit of 4–80 μM (Kumar and D'Souza, 2010; Kumar and D'Souza, 2011a,b). However, a further improvement in sensitivity and stability is done by Mishra et al. (2017) with the help of polyethyleneimine (PEI) functionalized silica nanoparticles (Si NP) integration with *Sphingomonas sp.* cells. A quite low LOD value of 0.1–1.0 ppm was obtained for methyl parathion with enhanced stability of 180 days.OPH conjugated to nanomagnet-silica core-shell with Coumarin 1 as a fluorescence-generating molecule designed as optical biosensor and useful for detection of paraoxon in a working range of 10–250 nM and LOD of 5×10^{-6} μM (Khaksarinejad et al., 2015).

Merits: Good market availability of optical devices made them suitable for the easy and quick biosensor constructions. However, the direct ability to check the optical reaction with the naked eye is further

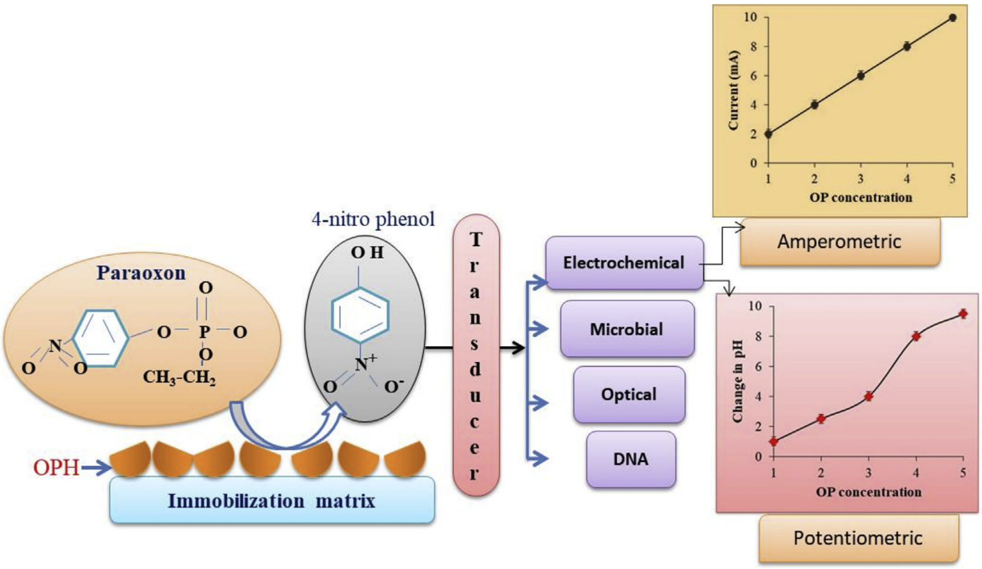

Fig. 4. An overview of organophosphorus hydrolase (OPH) based OP biosensors.

advantageous over the other biosensors. **Demerits:** Optical biosensors can suffer from interference of other coloured compounds such as food colorants and hemoglobin.

4.2.5. Microbial OPH based OP biosensors

Principle: In a microbial or whole cell biosensor, microorganisms are immobilized onto a transducer surface using different chemical and physical techniques. The changes occur due to the activation and inactivation of microbial respiration in the form of generation of electro-active metabolites can be detected using a dissolved oxygen electrode or amperometrically (Weiying et al., 2014). For detection of OP compounds viz. paraoxon, methyl parathion, parathion, fenitrothion and ethyl p-nitrophenol thiobenzenephosphonate, enzyme OPH has been co-immobilized with micro-organisms *Pseudomonas putida* JS444, and the p-nitrophenol released were detected by amperometric biosensor (Mulchandani et al., 2005, 2006). Along with this, a potentiometric and colorimetric microbial biosensor for the direct detection of paraoxon and methyl parathion was also constructed by immobilizing recombinant *E. coli* containing *opd* gene on a glass pH electrode and *Flavobacterium* sp. on a glass fibre filter paper respectively (Rainina et al., 1996; Kumar et al., 2006). A whole cell biosensor was also fabricated by co-immobilizing the enzymes OPH and methyl parathion hydrolase on the cell surface of *Escherichia coli* tagged with green fluorescent protein (GFP), truncated ice nucleation protein (INPNC) and Lpp-OmpA as the anchoring motifs. OP compounds concentration was detected rapidly by evaluating fluorescence of surface displayed GFP (Liu et al., 2013). For on-site detection of p-nitrophenyl-substituted OP, a very rapid microbial biosensor has been developed using genetically engineered *Escherichia coli* strain with surface displayed mutant OPH. The OPH-bacteria on glass carbon electrode (GCE) modified with ordered mesopore carbons (OMCs) showed a linear range of 0.05–25 μM for paraoxon and parathion, and 0.08–30 μM for methyl parathion with LOD of 9.0 nM, 10 nM and 15 nM respectively(Tang et al., 2014; Kumar and Melo, 2015).

Merits: As microbial biosensors employ whole cells as bio recognition element, these provide simple and direct detection of OP compounds, avoiding the expensive process of enzyme purification. **Demerits:** These biosensors have poor sensitivity and longer response time for detection of OP pesticides, as mass transport of substrate and product across the cytoplasmic and periplasmic membranes does not occur easily.

4.2.6. DNA based OP biosensors

Principle: DNA based OPbiosensors exploit the hybridization between complementary sequences of the nucleic acid. In these biosensors, a single-stranded DNA probe complementary to the target DNA is immobilized onto the surface and generate signals, when bind to its complementary sequence by hybridization (Nowicka et al., 2010). OP pesticide, paraoxon-ethyl, difluorobenzuron cause DNA damage, which can be easily detected electrochemically by DNA biosensor (Nowicka et al., 2010). A reversible electrochemical response has been measured by cyclic voltametric studies of covalently bound amino-functionalized DNA (with SWCNTs the carboxylic groups) adsorbed onto the gold electrode hybridization with ferrocene-labelled complementary oligonucleotide (He and Dai, 2004).Recently, an electrochemical DNA apta sensor for the detection of OP compound, profenofos, was fabricated by immobilizing thiol-tethered DNA capture probe, complementary to the chosen aptamer sequence onto gold nanoparticles/polyaniline composite film-modified electrodes (AuNPs/PANI/SPE). The aptasensor showed a linear reponse of 0.10–10.0 μM and LOD of 0.27 μM (Selvolini et al., 2018).

Merits: High affinity, accuracy, specificity and portability with real life sample analysis are the main advantages of using these DNA based OP biosensors. However, apta-sensors can be used broadlytowards a wide range of target analytes, as these are easy to prepare and require very low amount of reagents. **Demerits:** There is strong urge towards commercial production of these apta-sensors devices and their extensive useby improving basic research in developing the sensing strategies, analytical instrumentations and procedures.

5. Conclusion

In the present review, attempts were made to summarize the salient features of various enzyme inhibition or catalysis based OP biosensors reported so far. On one hand, enzymes (AChE, BChE, tyrosinase, alkaline phosphatase and OPH) have been used as bio-recognition elements in these OP biosensors due to their high selectivity and specificity but on other hand, their purification is quite costly, time-consuming and these possess a poor thermal stability, less efficiency at all pH and temperature ranges. Specifically, AChE based OP biosensors seems to be the most fascinating one, due to their simplicity, specificity in terms of irreversible inhibition, cost-effectiveness, disposability and rapidity with complete automation in real time sample analysis. Apart from this, affinity-based bio-sensing with the advantageous choice of different

Table 3
Characteristics of different organophosphorus hydrolase based electrochemical (amperometric and potentiometric) biosensors for OP pesticides detection.

Sr. No	Electrode material/immobilization matrix	Immobilization method	Detection limit (μM)	Linearity range (μM)	Target analyte	Storage stability (days)	References
			Amperometric				
1.	Carbon electrode	Entrapment	9×10^{-14} 7×10^{-14}	up to 40.0 up to 5.0	Paraoxon Methyl parathion	NR	Mulchandani et al. (1999a)
2.	Carbon paste electrode	Entrapment	4×10^{-13} 9×10^{-13}	$4.6–46 \times 10^{-12}$	Methyl parathion Paraoxon	NR	Wang et al. (1999)
3.	Carbon electrode	Entrapment	0.001	NR	Parathion	NR	Sacks et al. (2000)
4.	Carbon Paste Electrode	Covalent binding	0.02	Up to 140 Up to 120	Methyl parathion Paraoxon	30	Mulchandani et al. (2001b)
5.	Carbon Paste Electrode	Cross-linking	0.015 0.020	0.02–0.18	Parathion Paraoxon	60	Chough et al. (2002)
6.	Gold electrode	Cross-linking	0.1	1–10	Paraoxon Methyl parathion	28	Wang et al. (2003)
7.	MWCNT modified SPCE	Physical adsorption	NR	NR	Paraoxon	NR	Trojanowicz et al. (2004)
8.	Carbon nanotube (CNT) modified glassy carbon electrode	Entrapment	0.15 0.8	Up to 4	Paraoxon Methyl parathion	NR	Deo et al. (2005)
9.	MWCNTs modified glassy carbon electrode	Cross-linking	0.314	0.5–2	Paraoxon	90	Laothanachareon et al. (2008)
10.	Mesoporous carbon and carbon black modified glass carbon electrode	Entrapment	0.12	0.2–8	Paraoxon	NR	Lee et al. (2010)
11.	Single-walled carbon nanotubes (SWCNT)	Covalent binding	0.01	0.5–8.5	Paraoxon	210	Pedrosa et al. (2010)
12.	AuNPs-MWCNTs-QDs glassy carbon electrode	Covalent binding	0.001	1.9×10^{-14}-7.6×10^{-13}	Methyl parathion	NR	Du et al. (2010a,b,c)
13.	Disposable strip	Physical adsorption	0.5	0.5–100	Paraoxon	NR	Hossain et al. (2011)
14.	Screen printed carbon electrode (SPCE)	Covalent binding	2.0	2–80.0	Methyl parathion	NR	Kumar and D'Souza (2011a,b)
15.	Methyl parathion hydrolase/AuNP/Silica/MWCNT	Covalent binding	0.3×10^{-6}	Up to 1×10^{-6}	Methyl parathion	NR	Chen et al. (2011)
15.	Screen printed carbon electrode (SPCE)	Covalent binding	1.8×10^{-5}	NR	Paraoxon, Parathion	NR	Mulyasuryani and Dofir, 2014
16.	CNT electrode	Gluteraldehyde coupling	0.1	0.1–200	Methyl parathion	50	Gothwal et al. (2014)
17.	ELP-OPH/BSA/TiO2NFs/c-MWCNTs	Covalent binding	12×10^{-9} 10×10^{-9}	10×10^{-9} -12×10^{-9}	Methyl parathion Parathion	NR	Bao et al. (2016)
18.	CS-CNT-ZrO2-modified GCE	Physical adsorption	20.0×10^{-3}	20.0×10^{-3} – 40.0	Paraoxon	NR	Cabarcas et al. (2018)
			Potentiometric				
1.	Glass membrane	Cross-linking	2.0	$0.002–0.4 \times 10^{-3}$	Paraoxon Parathion	30	Mulchandani et al. (1998)
2.	Glass membrane	Cross-linking	2.0–5.0	NR	Paraoxon, ethyl parathion, methyl parathion, diazinon	30	Mulchandani et al. (1999c)
3.	Silica microspheres	Covalent binding	1×10^{-12}	NR	Paraoxon	70	Flounders et al. (1999)
4.	Glass membrane	Entrapment	NR	0.01 to 0.47×10^{-3} 0.025 to 0.4×10^{-3}	Paraoxon	23 6	Gaberlein et al. (2000)
5.	Al/p-Si/SiO2	Cross-linking	2.0	NR	Paraoxon	60	Schoning et al. (2003)
6.	Paraoxonase 1 (PON1)/copper grid/pH-sensitive LC/4-cyano-4′-pentylbiphenyl (5CB) doped with 0.3% of 4′-pentyl- biphenyl-4-carboxylic acid (PBA)	Entrapment	1.0	NR	Paraoxon	NR	Chen and Yang, 2013

NR- Not reported.

aptamers as bio-recognition element can contribute as a valid and innovative analytical tool for OP pesticides detection. Aptasensors possess high thermal stability, undergoes *in vivo* synthesis with the possibility of designing their structure, denaturalization, re-hybridization and distinguishable targets with different functional groups that results first time into a simple, easy to prepare portable devices for the analysis of OP pesticides in real life samples. Hence, only the adequate combination of bio-recognition elements, nanomaterials and efficient electronic signal transducer system can only result in best biosensors in terms of improved analytical performances, high sensitivity, reproducibility and specific determination of OP compounds.

6. Future perspectives

In context of the harmful effects of OP pesticides on human health, there should be an increase in the monitoring activities for detection of high levels of OP pesticides in agro and food products including development of simpler extraction and cleanup procedures for faster OP pesticides analysis process so that the need of sophisticated instruments can be omitted. On the other hand, spectrophoto-, metric detection and aptasensors must be developed for quick monitoring of these residual OP pesticides in the real samples. With the development of more reliable and efficient immobilization matrices and microelectronics techniques, efforts should be made in future for construction of portable instruments. Sooner low cost technology that reaches to general public interest particularly to agriculture sector is essentially needed for giving this wide research field a fruitful direction.

Declaration of interests

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Conflict of interest

The authors indicate no potential conflicts of interest.

References

Abad, J.M., Pariente, F., Hernandez, L., Abruna, H.D., Lorenzo, E., 1998. Anal. Chem. 70, 2848–2855.
Albareda-Sirvent, M., Merkoçi, A., Alegret, S., 2001. Anal. Chim. Acta 442, 35–44.
Albuquerque, Y.D., Ferreira, L.F., 2007. Anal. Chim. Acta 596, 210–221.
Andreescu, S., Barthelmebs, L., Barthelmebs, J.L., 2002a. Anal. Chim. Acta 464, 171–180.
Andreescu, S., Marty, J.L., 2006. Biomol. Eng. 23, 1–15.
Andreescu, S., Noguer, T., Magearu, V., Marty, J.L., 2002b. Talanta 57, 169–176.
Anitha, K., Venkata, S.M., Reddy, S.J., 2004. Biosens. Bioelectron. 20, 848–856.
Arduini, F., Forchielli, M., Amine, A., Neagu, D., Cacciotti, I., Nanni, F., Moscone, D., Palleschi, G., 2015. Microchim. Acta. 182, 643–651.
Arduini, F., Ricci, F., Tuta, C.S., Moscone, D., Amine, A., Palleschi, G., 2006. Anal. Chim. Acta 580, 155–162.
Ayyagari, M.S., Kamtekar, S., Pande, R., Marx, K.A., Kumar, J., Tripathy, S.K., Kaplan, D.L., 1995. Mater. Sci. Eng. C 2, 191–196.
Bao, J., Hou, C., Dong, Q., Ma, X., Chen, J., 2016. Biosens. Bioelectron. 85, 935–942.
Bonnet, C., Andreescu, S., Andreescu, J.L., 2003. Anal. Chim. Acta 481, 209–211.
Bucur, B., Fournier, D., Danet, A., Marty, J.L., 2006. Anal. Chim. Acta 562, 115–121.
Cabarcas, B., Stoytcheva, M.T., Zlatev, M., Montero, R., Velkova, G., Gochev, Z., Veliza, G., 2018. Curr. Anal. Chem. 14, 75–82.
Campanella, L., Lelo, D., Martini, E., Tomassetti, M., 2007. Anal. Chim. Acta 587, 22–32.
Chapalamadugu, S., Chaudhry, G.S., 1992. Crit. Rev. Biotechnol. 12, 357–389.
Chauhan, N., Narang, J., Pundir, C.S., 2011a. Biosens. Bioelectron. 29, 82–88.
Chauhan, N., Narang, J., Pundir, C.S., 2011b. Int. J. Biol. Macromol. 27, 82–87.
Chauhan, N., Pundir, C.S., 2014. Biosens. Bioelectron. 61, 1–8.
Chauhan, N., Pundir, C.S., 2011. Anal. Chim. Acta 701, 66–74.
Chauhan, N., Pundir, C.S., 2012. Electrochim. Acta 67, 79–86.
Chen, C.H., Yang, K.L., 2013. Sensor. Actuator. B Chem. 181, 368–374.
Chen, D., Fu, J., Liu, Z., Guo, Y., Sun, X., Wang, X., Wang, Z., 2017. Int. J. Electrochem. Sci. 12, 9465–9477.
Chen, D., Jiao, Y., Jia, H., Guo, Y., Sun, X., Wang, X., Xu, J., 2015a. Int. J. Electrochem. Sci. 10, 10491–10501.
Chen, D., Sun, X., Guo, Y., 2015b. Bioproc. Biosyst. Eng. 38, 315–321.
Chen, S., Huang, J., Du, D., Li, J., Tu, H., Liu, D., Zhang, A., 2011. Biosens. Bioelectron. 26, 4320–4325.
Chough, S.H., Mulchandani, A., Mulchandani, P., Chen, W., Wang, J., Rogers, K.R., 2002. Electroanalysis 14, 273–276.
Deo, R.P., Wang, J., Block, I., Mulchandani, A., Joshi, K.A., Trojanowicz, M., Scholz, W., Chen, Y., Lin, F., 2005. Anal. Chim. Acta 530, 185–189.
Dhull, V., 2018. Int. J. of Anal. Chem. 1–7.
Du, D., Chen, S., Cai, J., Zhang, A., 2008a. Talanta. pp. 766–772.
Du, D., Chen, S., Song, D., Li, H., Chen, X., 2008b. Biosens. Bioelectron. 24, 475–479.
Du, D., Chen, W., Cai, J., Zhang, J., Qu, F., Li, H., 2008c. J. Electroanal. Chem. 623, 81–85.
Du, D., Chen, W., Zhang, W., Liu, D., Li, H., 2010a. Biosens. Bioelectron. 25, 1370–1375.
Du, D., Ding, J., Tao, Y., Chen, X., 2008d. Sens. Actuators, B 134, 908–912.
Du, D., Huang, X., Cai, J., Zhang, A.D., 2007. Biosens. Bioelectron. 23, 285–289.
Du, D., Wang, M., Cai, J., Qin, Y., Zhang, A., 2010b. Sens. Actuators, B 143, 524–529.
Du, D., Ye, X., Cai, J., Liu, J., Zhang, A., 2010c. Biosens. Bioelectron. 25, 2503–2508.
Dutta, R., Puzari, P., 2014. Biosens. Bioelectron. 52, 166–172.
Flounders, A.W., Singh, A.K., Volponi, J.V., Carichner, S.C., Wally, K., Simonian, A.S., Wild, J.R., Schoeniger, J.S., 1999. Biosens. Bioelectron. 14, 715–722.
Funari, R., Ventura, B.D., Schiavo, L., Esposito, R., Altucci, C., Velotta, R., 2013. Anal. Chem. 85, 6392–6397.
Gaberlein, S., Spener, F., Zaborosch, C., 2000. Appl. Microbiol. Biotechnol. 5, 652–658.
Gong, J., Liu, T., Song, D., Zhang, X., Zhang, L., 2009. Electrochem. Commun. 11, 1873–1876.
Gong, L., Wang, L., Zhang, L., 2009. Biosens. Bioelectron. 24, 2285–2288.
Gothwal, A., Beniwal, P., Dhull, V., Hooda, V., 2014. Int. J. of Anal. Chem. 1–8.
Haiyan, Z., Xueping, J., Beibei, W., Na, W., 2015. Biosens. Bioelectron. 65, 23–30.
Halamek, J., Pribyl, J., Makower, A., Skladal, P., Scheller, F.W., 2005. Anal. Bioanal. Chem. 382, 1904–1911.
He, P.G., Dai, L.M., 2004. Chem. Commun. 348–349.
Hossain, M.M., Faisal, S.N., Kim, C.S., Cha, H.J., Nam, S.C., Lee, H.J., 2011. Electr. Commun. 13 (6), 611–614.
Ion, A.C., Ion, I., Culetu, A., Gherase, D., Moldovan, C.A., Iosub, R., Dinescu, A., 2010. Mater. Sci. Eng. C 30, 817–821.
Ivanov, A.N., Evtugyn, G.A., Gyurcsányi, R.E., Tóth, K., Budnikov, H.C., 2000. Anal. Chim. Acta 404, 55–65.
Ivanov, A.N., Younusov, R.R., Evtugyn, G.A., Arduini, F., Moscone, D., Palleschi, G., 2011. Talanta 85, 216–221.
Ivanov, Y., Marinov, I., Gabrovska, K., Dimcheva, N., Godjevargova, T., 2010. J. Mol. Catal. B. 63, 141–148.
Jha, N., Ramaprabhu, S., 2010. Nanoscale 2, 806–810.
Joshi, K.A., Tang, J., Haddon, R., Wang, J., Chen, W., Mulchandani, A., 2005. Electroanalysis 17, 54–58.
Ju, K.J., Feng, J.X., Feng, J.J., Zhang, Q.L., Xu, T.Q., Wei, J., Wang, A.J., 2015. Microchim. Acta. 182, 2427–2434.
Karine, R., Sofiane, Z., Nicole, J.R., Bernard, F., 2002. Talanta 56, 1015–1020.
Karousos, N.G., Aouabdi, S., Way, A.S., Reddy, S.M., 2002. Anal. Chim. Acta 469, 189–196.
Kaur, N., Prabhakar, N., 2017. Trends Anal. Chem. 92, 62–85.
Kesik, M., Kanik, F.E., Turan, J., 2014. Sensor. Actuator. B Chem. 205, 39–49.
Khaksarinejad, R., Mohsenifar, A., Rahmani-Cherati, T., Karami, R., Tabatabaei, M., 2015. Appl. Biochem. Biotechnol. 176, 359–371.
Khaled, E., Hassan, H.N.A., Mohamed, G.G., 2010. Talanta 83, 357–363.
Kim, N., Park, I.S., Kim, D.K., 2007. Biosens. Bioelectron. 22, 1593–1599.
Kok, F.N., Bozoglu, F., Hasirci, V., 2002. Biosens. Bioelectron. 17, 531–539.
Kumar, J., D'Souza, S.F., 2010. Biosens. Bioelectron. 26, 1292–1296.
Kumar, J., D'Souza, S.F., 2011a. Biosens. Bioelectron. 26, 4399–4404.
Kumar, J., D'Souza, S.F., 2011b. Biosens. Bioelectron. 26, 4289–4293.
Kumar, J., Jha, S.K., D'Souza, S.F., 2006. Biosens. Bioelectron. 21, 2100–2105.
Kumar, J., Melo, J.S., 2015. Adv. in Biosens. Res. 89–111.
Laothanachareon, T., Champreda, V., Sritongkham, P., Somasundrum, M., Surareungchai, W., 2008. World J. Microbiol. Biotechnol. 24, 3049–3055.
Laschi, S., Ogonczyk, D., Palchetti, I., Mascini, M., 2007. Enzym. Microb. Technol. 40, 485–489.
Law, K.A., Higson, S.P.J., 2005. Biosens. Bioelectron. 20, 1914–1924.
Lea, P., Mladen, F., 2003. Biosens. Bioelectron. 18, 1–9.
Lee, J.H., Park, J.Y., Min, K., Chac, H.J., Choid, S.S., Yoo, Y.J., 2010. Biosens. Bioelectron. 25, 1566–1570.
Li, S., Luo, Q., Liu, Y., Zhang, Z., Shen, G., Wu, H., Chen, A., Liu, X., Zhang, A., 2017a. Polymers 9, 359.
Li, X., Zheng, Z., Liu, X., Zhao, S., Liu, S., 2015. Biosens. Bioelectron. 64, 1–5.
Li, Y., Zhao, R., Shi, L., Han, G., Xiao, Y., 2017b. RSC Adv. 7, 53570.
Li, Y.G., Zhou, Y.X., Feng, J.L., Jiang, Z.H., Ma, L.R., 1999. Anal. Chim. Acta 382, 277–282.
Lin, T.J., Huang, K.T., Liu, C.Y., 2006. Biosens. Bioelectron. 22, 513–518.
Liu, R., Yang, C., Xu, Y., Xu, P., Jiang, H., 2013. J. Agric. Food Chem. 61, 7810–7816.
Liu, Y., Wei, M., 2014. Food Control 36, 49–54.
Luigi, C., Achilli, M., Sammartino, M.P., Mauro, T., 1991. Bioelectrochem. Bioenerg. 26, 237–249.
Luigi, C., DeLuca, S., Sammartino, M.P., Mauro, T., 1999. Anal. Chim. Acta 385, 59–71.
Lvnitskii, D.M., Rishpon, J., 1994. Biosens. Bioelectron. 9, 569–576.
Makower, A., Halamek, J., Skladal, P., Kernchen, F., Scheller, F.W., 2003. Biosens. Bioelectron. 18, 1329–1337.
Marinov, I., Ivanov, Y., Gabrovska, K., Godjevargova, T., 2010. J. Mol. Catal. B. 62, 67–75.
Marrazza, G., 2014. Biosensors 4, 301–317.
Marty, J.L., Mionetto, N., Noguer, T., Ortega, F., Roux, C., 1993. Biosens. Bioelectron. 8,

273–280.
Mauriz, E.A., Calle, J.J., Manclus, A., Montoya, A.M., Escuela, J., Sendra, R., Lechuga, L.M., 2006. Sensor. Actuator. B. 118, 399–407.
Mishra, A., Kumar, J., Melo, J.S., 2017. Biosens. Bioelectron. 87, 332–338.
Mulchandani, A., Mulchandani, P., Chen, W., 1998. Field Anal. Chem. Technol. 2, 363–369.
Mulchandani, A., Mulchandani, P., Chen, W., Wang, J., Chen, L., 1999a. Anal. Chem. 71, 2246–2249.
Mulchandani, P., Chen, W., Mulchandani, A., 2001. Environ. Sci. Technol. 35, 2562–2565.
Mulchandani, P., Chen, W., Mulchandani, A., 2006. Anal. Chim. Acta 568, 217–221.
Mulchandani, P., Hangarter, C.M., Lei, Y., Chen, W., Mulchandani, A., 2005. Biosens. Bioelectron. 21, 523–527.
Mulchandani, P., Mulchandani, A., Kaneva, I., Chen, W., 1999c. Biosens. Bioelectron. 14, 77–85.
Mulyasuryani, A., Dofir, M., 2014. Engineering 6, 230–235.
Norouzi, P., Pirali-Hamedani, M., Ganjali, M.R., Faridbod, F.A., 2010. Int. J. Electrochem. Sci. 5, 1434–1446.
Nowicka, A.M., Kowalczyk, A., Stojek, Z., Hepel, M., 2010. Biophysical 1, 42–53.
Ozkutuk, E.B., Dilterniz, S.E., Ozalp, E., Say, R., Ersoz, A., 2013. Mater. Sci. Eng. C 33, 938–942.
Pabbi, M., Kaur, A., Mittal, S.K., Jindal, R., 2018. Sensor. Actuator. B Chem. 258, 215–227.
Pedrosa, V.A., Paliwal, S., Balasubramanian, S., Nepal, D., Davis, V., Wild, J., Ramanculov, E., Simonian, A., 2010. Colloids and Sur. B Biointer. 77, 69–74.
Pohanka, M., Musilek, K., Kuca, K., 2009. Curr. Med. Chem. 16, 1790–1798.
Pundir, C.S., Chauhan, N., 2012. Anal. Biochem. 429, 19–31.
Qu, Y., Sun, Q., Xiao, F., Shi, G., Jin, L., 2010. Bioelectrochemistry 77, 139–144.
Raghu, P., Swamy, B.E.K., Reddy, T.M., Chandrashekar, B.N., Reddaiah, K., 2011. Bioelectrochemistry 83, 19–24.
Rainina, E.I., Efremenco, E.N., Varfolomeyev, S.D., Simonian, A.L., Wild, J.R., 1996. Biosens. Bioelectron. 11, 991–1000.
Rajangam, B., Daniel, D.K., Krastanov, A.I., 2018. Eng. Life Sci. 18, 4–19.
Rassaei, L., Marken, F., Sillanpää, M., Amiri, M., Cirtiu, C.M., 2011. Trac. Trends Anal. Chem. 30, 1704–1715.
Rekha, K., Murthy, B.N., 2008. Food Agric. Immunol. 19, 273–281.
Rupesh, K.M., Rocio, B.D., Sunil, B., Roberto, M., 2012. Biosens. Bioelectron. 32, 56–61.
Sacks, V., Eshkenazi, I., Neufeld, T., Dosoretz, C., Rishpon, J., 2000. Anal. Chem. 72, 2055–2058.
Sajjadi, S., Ghourchian, H., Tavakoli, H., 2009. Biosens. Bioelectron. 24, 2509–2514.
Sarkar, T., Narayanan, N., Solanki, P.R., 2017. Int. J. Environ. Res. 11, 5–6.
Schoning, M.J., Arzdorf, M., Mulchandani, P., Chen, W., Mulchandani, A., 2003. Sensor. Actuator. B. 91, 92–97.
Selvolini, G., Bajan, I., Hosu, O., Cristea, C., Sandulescu, R., Marrazza, G., 2018. Sensors 18, 2035–2047.
Shamagsumova, R.V., Shurpik, D.N., Padnya, P.L., Stoikov, I.I., Evtugyn, G.A., 2015. Talanta 144, 559–568.
Shi, M., Xu, J., Zhang, S., Liu, B., Kong, J., 2006. Talanta 68, 1089–1095.
Shimomura, T., Itoh, T., Sumiya, T., Mizukami, F., Ono, M., 2009. Enzym. Microb. Technol. 45, 443–448.
Shulga, O., Kirchhoff, J.R., 2007. Electrochem. Commun. 9, 935–940.
Simonian, A.L., Good, T.A., Wang, S.S., Wild, J.R., 2005. Anal. Chim. Acta 534, 69–77.
Singh, B.K., 2009. Nat. Rev. Microbiol. 7, 156–164.
Sinha, R., Ganesana, M., Andreescu, S., Stanciu, L., 2010. Anal. Chim. Acta 661, 195–199.
Somerset, V.S., Klink, M.J., Baker, P.G.L., Iwuoha, E.I., 2007. J. Environ. Sci. Health B. 42, 297–304.
Song, Y., Zhang, M., Wang, L., Wan, L., Xiao, X., Ye, S., Wang, J., 2011. Electrochim. Acta 56, 7267–7271.
Sotiropoulou, S., Chaniotakis, N.A., 2005. Biomaterials 26, 6771–6779.
Stein, K., Schwedt, G., 1993. Anal. Chim. Acta 272, 73–81.
Stoytcheva, M., Zlatev, R., Montero, G., Velkova, Z., Gochev, V., 2017. Microchim. Acta. 1–8.
Sun, X., Wang, X., 2010. Biosens. Bioelectron. 25, 2611–2614.
Tang, X., Zhang, T., Liang, B., Han, D., Zeng, L., Zheng, C., 2014. Biosens. Bioelectron. 60, 137–142.
Toshihiko, I., Nobuhiko, I., 1995. Biosens. Bioelectron. 10, 435–441.
Touloupakis, E., Boutopoulos, C., Buonasera, K., Zergioti, I., Giardi, M.T., 2012. Anal. Bioanal. Chem. 402, 3237–3244.
Trojanowicz, M., Mulchandani, A., Mascini, M., 2004. Anal. Lett. 37, 3185–3204.
Upadhyay, S., Rao, G.R., Sharma, M.K., Bhattacharya, B.K., Rao, V.K., Vijayaraghavan, R., 2009. Biosens. Bioelectron. 25, 832–838.
Vakurov, A., Simpson, C.E., Daly, C.L., Gibson, T.D., Millner, P.A., 2004. Biosens. Bioelectron. 20, 1118–1125.
Vangelis, G.A., Yannis, D.C., 2002. Biosens. Bioelectron. 17, 61–69.
Verma, N., Eneyew, T.M., 2001. In: Mann, A.P.S., Munshi, S.K., Gupta, A.K. (Eds.), Biochemistry – Environment and Agriculture. Kalyani Publishers, New Delhi, pp. 265–270.
Vidal, J.C., Esteban, S., Gil, J., Castillo, J.R., 2006. Talanta 68, 791–799.
Viswanathan, S., Radecka, H., Radecki, J., 2009. Biosens. Bioelectron. 24, 2772–2777.
Wang, H., Zhao, G., Chen, D., Wang, Z., Liu, G., 2016. Int. J. Electrochem. Sci. 11, 10906–10918.
Wang, J., Chen, L., Mulchandani, A., Mulchandani, P., Chen, W., 1999. Electroanalysis 11, 866–869.
Wang, J., Krause, R., Block, K., Musameh, M., Mulchandani, A., Schoning, M.J., 2003. Biosens. Bioelectron. 18, 255–260.
Wang, K., Li, H.N., Wu, J., Ju, C., Yan, J.J., Liu, Q., Qiu, B., 2011a. Analyst 136, 3349–3354.
Wang, K., Liu, Q., Dai, L., Yan, J., Ju, C., Qiu, B., Wu, X., 2011b. Anal. Chim. Acta 695, 84–88.
Wei, M., Wang, J., 2015. Sensor. Actuator. B. 211, 290–296.
Wei, Y., Li, Y., Qu, Y., Xiao, F., Shi, G., Jin, L., 2009. Anal. Chim. Acta 643, 13–18.
Weiying, Z., Abdullah, M.A., Deli, L., Dan, D., Lin, Y., 2014. Trends Anal. Chem. 54, 1–10.
Wu, S., Lan, X., Zhao, W., Li, Y., Zhang, L., Wang, H., Han, M., Tao, S., 2011a. Biosens. Bioelectron. 27, 82–87.
Wu, S., Zhang, L., Qib, L., Tao, S., Lan, X., Liu, Z., Meng, C., 2011b. Biosens. Bioelectron. 26, 2864–2869.
Yan, R.L., Zhi, Y.G., Yan, F.L., Qian, L., Jian, C.B., Zhi, H.D., Min, H., 2010. Sci. China Chem. 53, 820–825.
Yang, Y., Guo, M., Yang, M., Wang, Z., Shen, G., Yu, R., 2005. Int. J. Environ. Anal. Chem. 85, 163–175.
Yao, S., Meyer, A., Henze, G., 1991. Fresenius' J. Anal. Chem. 339, 207–211.
Yu, G., Wu, W., Zhao, Q., Wei, X., Lu, Q., 2015. Biosens. Bioelectron. 68, 288–294.
Zejli, H., H-H, J.L., Cisneros, D., Naranjo-Rodriguez, I., Liu, B., Temsamani, K.R., Marty, J.L., 2008. Talanta 77, 217–221.
Zheng, Y., Liu, Z., Jing, Y., Li, J., Zhan, H., 2015. Sensor. Actuator. B. 210, 389–397.