



Molecularly Imprinted Polymers for the Detection of Chlorpyrifos (an Organophosphate Pesticide)

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ABSTRACT

Chlorpyrifos belongs to an organophosphate pesticides group that is frequently used to increase the yield of crops by controlling plant and pests-related diseases. Chlorpyrifos contaminates the environment and cause diseases to the human population. Molecularly imprinting technology lead to the development of molecularly imprinted polymers having templated oriented cavities with high selectivity, sensitivity, stability, and portability. Our review article aims to provide a collective study related to pesticides detection through molecularly imprinted polymers with existed constraints and necessary potential facets are discussed.

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1. INTRODUCTION

Pesticides are the chemical compounds or mixture of chemical compounds used for destroying any group of plant enemies like pests, insects, and weeds (Samsidar et al., 2018). Chemical pesticides significantly increase and boost crop yield by controlling pests and plant-related diseases and other microbes that disturb the growth and productivity of plants (Chawla et al., 2018). During the last few decades, pesticides are extensively used all over the world for food production progress to cope with the increasing demand of the rising human population. Almost 5.6 billion pounds' pesticides are utilized all over the globe. Pesticides are considered as a quick, easy, and less expensive solution for controlling plants disorders, weeds and insects, pests in the urban landscape of developing countries (Ghorab & Khalil, 2015).

Different types of pesticides are in practice to control various types of pests like insecticide, herbicide, fungicide, bactericide, and rodenticide. Formulation of pesticides involves the complex mixtures of the active ingredient and a wide variety of other substances that are added to increase the efficacy of the product (Hassaan & El Nemr, 2020; KANKAM, 2021).

Pesticides are categorized into 4 classes based on their chemical nature that is organophosphorus, organochlorine, carbamates, and pyrethroids. Organophosphorus pesticides and carbamates are widely used because of their high insecticidal activity. Contaminations caused by pesticides pose important hazards to the environment and non-targeted organisms. In humans, it may cause cancer, fetuses distortions, sterility, acute intoxications allergies, and even death (Costa et al., 2008).

Chlorpyrifos belongs to an organophosphate pesticides group that is frequently used to increase the yield of crops by controlling plant and pests-related diseases. Chlorpyrifos contaminates the environment and cause diseases to the human population. Molecularly imprinting technology lead to the development of molecularly imprinted polymers having templated oriented cavities with high selectivity, sensitivity, stability, and portability. Our review article aims to provide a collective study related to pesticides detection through molecularly imprinted polymers with existed constraints and necessary potential facets are discussed.

2. METHODS

The present review was done by collecting and reviewing data from papers from 1990 to recent publications. We analyzed the data and summarized the results. The way how to collect the papers can be found in the literature (Azizah et al., 2021).

3. RESULTS AND DISCUSSION

3.1. Classification of Pesticides

Pesticides are divided into different groups according to their chemical structures, their targeted uses, and their requirement of applications (Copplesstone, 1988; Jayaraj et al., 2016; Yadav & Devi, 2017; Kaur et al., 2019) (Figure 1). The classifications can be divided into:

- (i) Based on the target organism or intended use like herbicide, insecticide, fungicides, and rodenticides, etc.
- (ii) Based on the mode of action by which the pesticide effect the body of the pest, like stomach poisoning pesticide which causes poisoning when swallowed and digested with food. The second one is contact poisoning insecticides which affect the pest when in contact with the skin. The third action mechanism is fumigants that penetrate the organism by respiratory tracts in the form of gas and vapors.

- (iii) Based on the chemical composition of the pesticide. This category contains synthetic organic compounds like organochlorine, organophosphorus, organocarbamate, and synthetic pyrethroids.

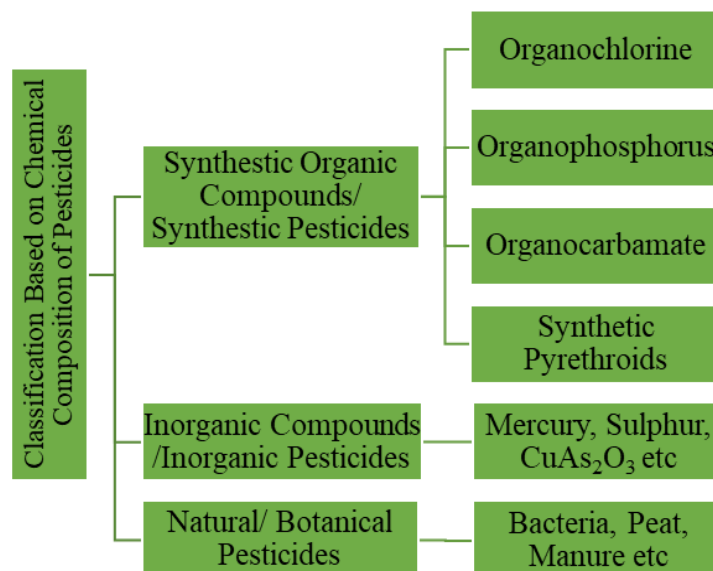


Figure 1. Schematic illustration of various pesticides on the basis of chemical nature.

3.2. Routes of Exposure

There are several sources of exposure to pesticides. Three main routes for the exposure of pesticides are inhalation, dermal absorption, and oral ingestion. Air, water, and soil are the means for pesticide exposure. Inhalation and dermal absorption mostly occur in the people that work in the field or living near the area where these pesticides are continually used. Pesticide residues persist in the ecosystem for a long period due to their persistent nature and enter into the food chain that facilitates the oral ingestion of pesticides (Lewis *et al.*, 1994; So *et al.*, 2014).

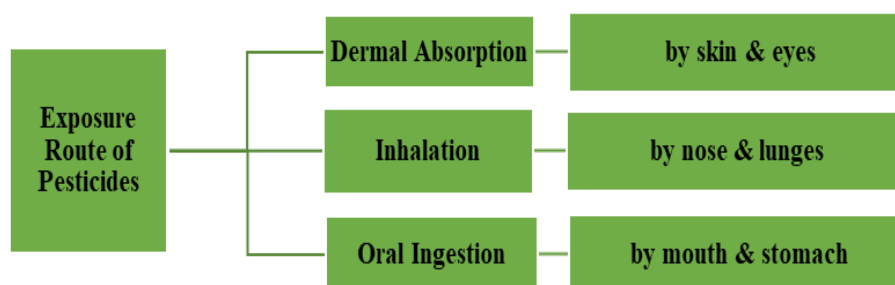


Figure 2. Pesticides roots of contact.

3.3. Mechanism of Action

Most pesticides affect the nervous system or nerves with slight changes in their mechanism of action (Sharma *et al.*, 2020). Organophosphorus pesticides affect the transmission of the nerve impulse by preventing acetylcholinesterase enzyme to produce acetylcholine (Umar & Aisami, 2020). The improper transmission of the nerve impulse causes muscle contraction or paralysis and eventually death. The most commonly used organophosphorus-based pesticide is chlorpyrifos. It is reported that contamination of chlorpyrifos is found up to 24 km away from the place of application (Zhu *et al.*, 2018; Arya *et al.*, 2019). There are many potential molecular targets of chlorpyrifos including cytotoxicity that affect macromolecule syntheses

like DNA and RNA interactions with enzymes, signal transduction pathway, and neurotransmitter receptors (Zhao *et al.*, 2006; Farag *et al.*, 2010; Ridano *et al.*, 2017; Sheppard *et al.*, 2020). Due to the large number of hazards of chlorpyrifos, it is necessary to detect this pesticide in the water and foodstuff for the detection of these pesticides there are conventional as well as modern methods are in practice (Mogha *et al.*, 2016; Jiao *et al.*, 2017; Talan *et al.*, 2018; Islam *et al.*, 2019). Some conventional techniques are being used for the detection of chlorpyrifos are discussed below.

3.4. Conventional Methods for the Detection of Chlorpyrifos

It is necessary to monitor the pesticides residues in food, water, soil, and environment to ensure their acceptance within legislation limits. Analytical methods like gas and liquid chromatography, HPLC, enzyme-linked immunosorbent assay, capillary electrophoresis, and mass spectrometry are practiced for organophosphorus compounds detection in agriculture samples as well as in the environment. These techniques have good sensitivity, selectivity, and are reproducible but these methods also have some limitations as they are time-consuming, laborious, expensive, and non-portable (Jiang *et al.*, 2010; Van Dyk & Pletschke, 2011; Nsibande & Forbes, 2016).

Therefore, there is a need to develop new technologies that are real-time, inexpensive, and easy to handle. Molecularly imprinting technology is one of the convenient techniques to detect harmful chlorpyrifos due to template-oriented cavities in the polymer matrix (Fang *et al.*, 2021a). We presented a short review from recent past work to summarize the work related to the chemistry of molecularly imprinted polymers (MIPs) and MIP application for the detection of chlorpyrifos.

3.5. Molecular Imprinting Technology

A molecular imprinting polymer is the imprinting technique in which cavities are generated into a polymer with an affinity for template substances. Template and monomers interact with each other to form a polymer by the addition of Initiators. Specific porogenic solvents are used to wash the polymeric matrix for the development of template-oriented cavities for the selective recognition of the molecules. Imprinted polymers have an affinity for original molecules and exhibit high selectivity (Jenkins *et al.*, 2001; Uzun & Turner, 2016; Ayankojo *et al.*, 2018; Saylan *et al.*, 2019). MIT is the viable alternative to the natural antibodies compatible simulated receptor for the rapid, selective, and economical detection of the analytes without any sample preparation steps. Two types of interactions are found in the MIT namely; covalent and non-covalent imprinting approaches (Fang *et al.*, 2021a; Fang *et al.*, 2021b; Li *et al.*, 2021).

3.5.1. Covalent Imprinting

This approach is considered more suitable for separation or catalytic purposes because of bonding. In this imprinting approach monomer and analyte are bound by a covalent bond. Template and monomer in solution interact through reversible covalent bonds before polymerization and recognition sites are obtained as a result of cleavage and breakage of these bonds (Mosbach, 1994; Cieplak *et al.*, 2015).

3.5.2. Non-covalent Imprinting

The development of non-covalent interactions like hydrogen bonding, van der Waal forces, metal coordination, electrostatic forces in the process of MIP synthesis, and binding unbinding of the analyte. When a molecular imprint is prepared by non-covalent interactions,

there will be weak forces between functional monomer and template. A high monomer amount is used for the development of a stable polymerization complex (Mosbach, 1994; Yan & Row, 2006).

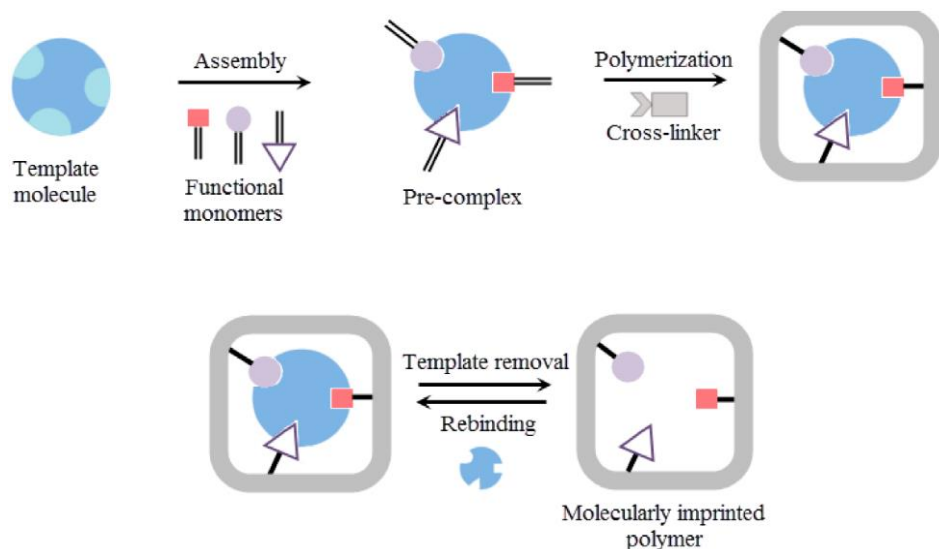


Figure 3. Fabrication mechanism of MIPs (Saylan *et al.*, 2019).

3.5.3. Molecular Imprinting Techniques

There are two molecularly imprinting techniques namely as bulk and surface imprinting:

- (i) Bulk imprinting. Template molecules are embedded into the polymer matrix as a whole to generate template-oriented cavities and templates are eliminated after the polymerization. The bulk imprinting technique is successful for compounds with low molecular weight.
- (ii) Surface imprinting. Surface imprinting is the solid-phase imprinting in which solid substrate is required for the imprinting of the MIP. The only surface that has the binding sites as compared to bulk imprinting technique. The number of templates is less in surface imprinting and cavities generated are highly accessible. Surface imprinted polymers are extensively used for several types of analytes, including proteins. This technique has drawbacks due to its low selectivity as compared to bulk imprinting (Xing *et al.*, 2017).

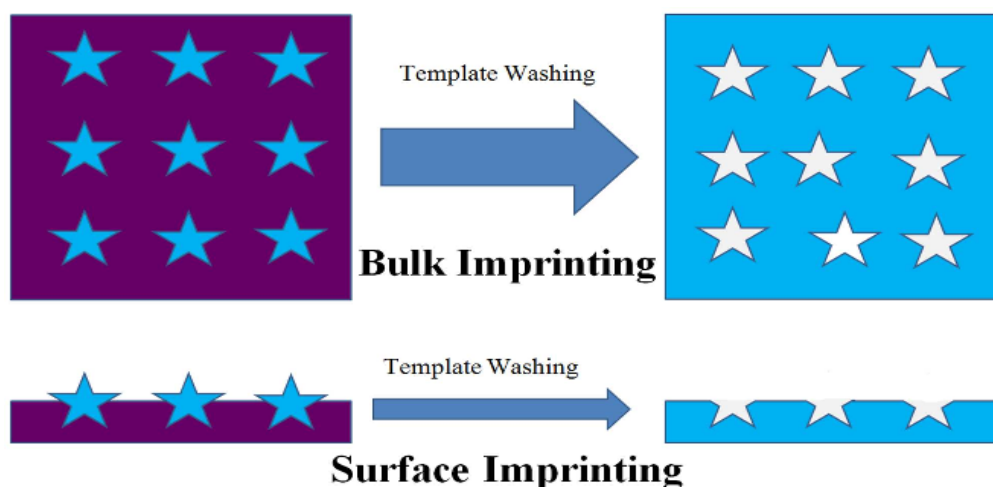


Figure 4. Bulk and surface imprinting techniques.

3.5.3. Essential Elements in Molecular Imprinting

Template, monomer, free radical initiator, crosslinker, and porogenic solvent are required for molecular imprinting polymer. Several essential elements are:

- (i) **Template.** The template molecule is the functional group of the specific analyte. Its role in molecular imprinting is the ordering of the functional groups to monomers. The template should be chemically inert under polymerization condition (Yan & Row, 2006).
- (ii) **Initiators.** Initiators are the free radical that are used in polymer synthesis to start the polymerization process and produce the free radicals during polymerization. Initiators are added in a very low concentration as compare to monomer (Wackerlig & Schirhagl, 2016).
- (iii) **Monomer.** The functional monomer is used for the binding interaction on imprinted sight. The role of the monomer is to create specific cavities for target molecule. e.g., MAA, polystyrene (Nicholls et al., 2009).
- (iv) **Cross linker.** Cross linker is the material that help in maintaining the shape of polymer matrix. It plays essential role in imprinted polymer synthesis e.g. EGDMA (Kim et al., 2005).
- (v) **Solvent.** The nature of the solvent help in determining the forces between non-covalent interactions. Porogenic solvent effect the morphology of polymer, which influence the proficiency of MIP as it is important for the formation of pores in molecular imprinted polymer (Foroughirad et al., 2021).

3.6. Application of MIPs in Chlorpyrifos Detection

MIPs are very selective due to template-oriented cavities and are being utilized for the qualitative and quantitative analysis of chlorpyrifos. Sun et al. (2017) developed a Photo-electrochemical sensor by surface molecular self-assembly strategy for anchoring MIP on the surface of branched titanium oxide nano-rods for selective detection of chlorpyrifos. Hydrothermal method was used to grow B-TiO₂ NRs on fluorine-doped tin oxide substrate and worked as a matrix for immobilization of MIP. Chlorpyrifos (CPS) and P-amino thiophenol were constructed on the shell of B-TiO₂ NRs by H-bonding interactions. Advantage of the fabricated sensor is its high selectivity and sensitivity that is due to the superior photoelectric transferring efficiency of B-TiO₂ NRs and excellent selectivity of MIP with the lowest detection limit (LOD) of 7.4 pg mL⁻¹ (Sun et al., 2017). Mn(II)-doped ZnS quantum dots coated with an acrylamide-based MIP was fabricated for chlorpyrifos fluorescent detection of. MIP-coated QDs exhibited high selectivity to chlorpyrifos with LOD of 17 nmol L⁻¹ (Ren et al., 2015).

Xie et al. (2011) reported a surface molecular imprinting strategy for producing core-shell particles for chlorpyrifos detection. These particles were prepared by copolymerization of methacryloyl groups on the surface of silica particles modified with 3-methacryloxypropyl trimethoxysilane a functional monomer and a cross-linking agent. The silica core was removed and the imprinted resonating particles exhibited rapid and selective identification of the template molecules. Imprinted hollow particles captured in a column with the aid of polyvinyl alcohol and chemiluminescence reaction for the chlorpyrifos assessment in the vegetable sample. The results showed that the MIP –based CL method improved the sensitivity and selectivity with the LOD of 0.92 nM. This method could possibly be used for online identification of the analytes (Xie et al., 2011). Mustaghfiroh et al. (2019) reported a MIP- polyvinyl Alcohol (PVA)-Fe₃O₄ based electrochemical sensor. Layer of MIP-Fe₃O₄ was generated on the surface of SPCE to determine the results. The sensitivity was enhanced by

adding citric acid at 24 mV/decade with the concentration range of 10⁻¹³ - 10⁻⁶ M (Mustaghfiroh *et al.*, 2019).

MWCNTs are being used to increase the sensing characteristics due to their Excellent conductivity. Huang *et al.* (2020) synthesized MIP layer on the surface of silica nano-spheres to overcome the problem of uneven shapes, pore size, and low imprinting capacity. The prepared sensor was best for the detection of chlorpyrifos in vegetables and fruits (Huang *et al.*, 2020). Nanoparticles has larger surface area and the increase the number of active sites for the template molecules in molecularly imprinting technology. MIP/(Co₃O₄) composite was coated on the SPCE and electrochemical properties of the developed sensor were characterized by cyclic voltammetry and differential pulse voltammetry. MIP/(Co₃O₄)/SPCE based sensor exhibited excellent sensitivity and selectivity with the LOD of 0.1 ppb.

4. CONCLUSION

The purpose of designing these MIP-based sensors is to detect harmful chlorpyrifos such as from the environment. Monitoring of such environmental pollutants has become necessary due to their toxic effects on human health. As the pesticide residues concerns are getting serious today, it is preferable among researchers to invent those devices that are highly sensitive, selective, economical, and portable. Our review summarizes the work related to chlorpyrifos detection by the MIP and also provides prospects to develop new commercial MIP to overcome the shortcomings. In conclusion, future research is focused on the development of MIP-based testing kits as a fast detection tool but it will take time due to advancement in a chain to chain.

6. AUTHORS' NOTE

The authors declare that there is no conflict of interest regarding the publication of this article. The authors confirmed that the paper was free of plagiarism.

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