Molecular modelling of 2-iminothiazoles as insecticidal activity

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Insecticides are used in agriculture, medicine, industry and by consumers, indoor. Insecticides are also claimed to be a major factor behind the increase in agricultural 20th century’s productivity. On the other hand, modes of their action is important in understanding whether an insecticide will be toxic to unrelated species, such as fish, birds and mammals. On the other hand, molecular docking, a new way of illuminating the effect mechanisms of biologically active chemicals offer a new green chemistry field. As provided herein, although 2-iminothiazoles are designed, synthesized and tested as protein tyrosine phosphatase 1B inhibitors, cannabinoid receptor ligands, pifithrin-α p53 inactivators, etc. there is not any remarkably record on their insecticidal docking study. Present work introduce the molecular modelling and mapping of active site of previously synthesized by us insecticidal 2-iminothiazole derivatives, by using classical docking techniques i.e. MOE, etc. and discuss their result.

Key words: molecular modelling; docking; insecticide; 2-iminothiazole; acetyl CoA carboxylase

1. INTRODUCTION

Insecticides are essential tools for preventing or minimizing insect damage to, and significantly increasing the quality and quantity of crops, as well as for improving the quality of life for humans, domestic animals and livestock. There are currently more than 20 different mechanisms, or modes of action, by which various commercial insecticides control insects by disrupting specific vital biological processes, but not all of these can be used against any particular pest insect. Despite the best efforts of the entire crop protection industry, a new insecticide mode of action comes to market only every 5 or 10 years, the last being in 2007 [1].

Neonicotinoid insecticides, i.e. imidacloprid (IMI) and clothianidin (CTD), which act on nicotinic acetilcholine receptors (nAChRs), yet the molecular basis of such action is poorly understood. The crystal structures in complex with acetylcholine binding protein from Lymnaea stagnalis (Ls-AChBP) which are deposited to Protein Data Bank (PDB) suggested that the guanidine moiety of IMI and CTD stacks with Tyr185, while the nitro group of IMI but not of CTD makes a hydrogen bond with Gln55 [2].

[Fig. 1. Neonicotinoid insecticides]

IMI showed higher binding affinity for Ls-AChBP than that of CTD, consistent with weaker CH–p interactions in the Ls-AChBP–CTD complex than in the Ls-AChBP–IMI complex and the lack of the nitro group-Gln55 hydrogen bond in CTD. Yet, the NH at position 1 of CTD makes a hydrogen bond with the backbone carbonyl of Trp143, offering an explanation for the diverse actions of neonicotinoids on nAChRs [2].

With an aim to explore the binding sites, the complementary applications of molecular docking were employed to understand the interaction between bovine serum albumin (BSA) and the organophosphate insecticides monocrotophos and phosphamidon [3], cf. Fig. 4.
Fig. 2. Imidacloprid (IMI) and clothianidin (CTD) binding to *Lymnaea stagnalis* AChBP (Ls-AChBP). (a) IMI–Ls-AChBP complex. (b) CTD–Ls-AChBP complex [2].

Fig. 3. Electron density maps of bound ligands and their interactions with the loop E region [2].

Fig. 4. Organophosphate insecticides monocrotophos and phosphamidon.

Fig. 5. The molecular docking results suggested that the insecticides bind to BSA into the hydrophobic cavity of subdomain IIA [3].
Among several classes of pesticides, carbamate compounds are widely used insecticides and acaricides. Formetanate (FMT) is one of them [4].

**Laccases** which are found in many plants, fungi and microorganisms are copper-containing oxidase enzymes. The latest research regarding modeling of laccase inhibition by formetanate suggested that the calculations identified Asp206 as the most relevant moiety in the interaction of FMT with the laccase enzymatic ligand binding domain. The amino acid residue Cys453 was important, because the Cys453-FMT interaction energy was not affected by the dielectric constant, although it was not a very close residue [4].

![Fig. 6. Formetanate (FMT).](image)

**Fig. 7.** Schematic representation of the binding pocket of the Lac showing the most important residues involved in the binding interaction in two different views. The only relevant residue that forms a hydrogen bond with FMT is Asp206, as shown in B. Cys453 residue is important for interaction [4].

QSAR analyses of organophosphates for insecticidal activity and its *in-silico* validation using molecular docking study have been revealed that selected OP analogues from combinatorial library were docked against the generated model of AChE of *M. domestica* as receptor. The binding pockets of these ligands were observed to be very similar to that of other OPs including TCVP as reported earlier, thus hinting about the insecticidal nature of the selected OP ligands. The binding sites (Fig. 8) analyses reflected that four amino acid residues (Ser-235, Try-160, Trp-83, Phe-368 and His-477) were commonly present as the interacting residues with docked OP ligands. Also, the binding efficiency was elevated when other aromatic side chains (viz., Tyr-367, Tyr-367, etc.) were involved in interaction with the ligands [5].

![Fig. 8. The docking site of selected OP analogues of combinatorial library and Tetrachlorvinphos (TCVP) showing interacting residues when docked against *M. Domestica* AChE (pdb ID: 1DX4) [5].](image)
Molecular modeling of insecticides have been studied for tetronic acid derivatives via the inhibition of acetyl-CoA carboxylase (ACCase) [6].

\[
\text{HO} \quad \text{Tetronic acid}
\]

\[
\text{Spirotetramat} \quad \text{Spirodiclofen} \quad \text{Spiromesifen}
\]

**Fig. 10.** The structure of representative tetronic acid derivatives and designed lead compound [6].

**Fig. 11.** Interaction mode of ACCase and active compounds 91b (A), 91j (B) and 91k (C) proposed by molecular docking and their molecular overlay (D). (pdb ID: 3PGQ) [6].
This application relates to thiazolylidene containing compounds, compositions comprising such compounds, and methods of treating conditions and disorders using such compounds and compositions.

[Chemical structure image]

**Fig. 12.** Thiazolylidene compounds.

(-) – Δ9-Tetrahydrocannabinol (Δ9-THC), the major psychoactive constituent of marijuana, exerts a broad range of effects through its interactions with two cannabinoid (CB) receptor subtypes, CB1 and CB2 [7].

Organophosphorus compounds (OPs) may irreversibly inhibit acetylcholinesterase (AChE), this enzyme is responsible for the hydrolysis of acetylcholine (ACh) which terminates nerve impulse transmission. A promising alternative route for OP detoxification and degradation is enzymatic catalysis. OP degrading enzymes are phosphotriesterase (PTE) (cleave different OP, breaking P-O, P-F, P-CN and Ps bonds), Human Serum Paraoxonase 1 (HssPON1) (degrade substrates such as esters, lactones, oxidized phospholipids and OPs), diisopropyl fluorophosphatase (DFPase) and Human Butyrylcholinesterase G117H Mutant (HssBuChE) [8].

[Chemical structure image]

**Fig. 13.** General structures of organophosphorus (OPs) and common used OP compounds.

This reported knowledge point out that, at the AChE active site, the catalytic triad (Ser203-Glu334-His447) is found at the bottom of the active site gorge, surrounded by three important structural features for catalytic activity: the acyl pocket (residues Phe295, Phe297 and Phe338), the oxyanion hole (main chain nitrogen from residues Gly121, Gly122 and Ala204), and the choline-binding site (Trp86 and Tyr337) (Fig A). The phosphoryl oxygen of tabun adduct interacts with the amino acid residues in the oxyanion hole (distance of 2.5, 2.7 and 3.0Å from Gly121, Gly122 and Ala204, respectively). The dimethylamino group is in the acyl pocket, interacting with Trp236 and Phe338 residues (Fig B). The two Zn²⁺ ions (α-Zn and β-Zn) in the PTE active site are found at a distance of ~3.4Å from each other and they are linked to the enzyme structure by means of the side chains of His55, His57, His201, His230, Asp301 and Lys169. (Fig C).

The proposed reaction mechanism for the PTE catalyzed reaction begins with a nucleophilic attack by an activated water molecule on the P center, resulting in an inversion of configuration. That is, the reaction takes place via bimolecular nucleophilic substitution (SN2) mechanism, where the Asp301 serves as a weak base, which removes an H atom of the water molecule, activating it. Afterwards, the resulting hydroxyl ion attacks the central P. A second reaction mechanism is possible...
for the OP degradation by PTE. In this case, there will be an attack to the P center directly by the carboxylic oxygen from the side chain of Asp301, promoting the expulsion of the leaving group and

![Diagram A](image1)

(A) 3D structure of *H. sapiens* AChE obtained from the PDB under the code 3LII, and detailed view of the catalytic site, with the catalytic triad (cyan) and the main amino acid residues of the acyl pocket (blue), the oxyanion hole (red), and the choline-binding site (green).

![Diagram B](image2)

(B) The main interactions of tabun in the active site of hAChE (PDB: 2X8B). Hydrogen bonds are represented by black dashes.

![Diagram C](image3)

(C) Crystallographic structure of PTE (PDB: 1DPM) and coordination environment of the two Zn²⁺ ions (α-Zn and β-Zn).

![Diagram D](image4)

(D) Crystallographic structure of *H. sapiens* PON1 (PDB: 1V04) and coordination environment of catalytic calcium ion (Ca²⁺ -2).

![Diagram E](image5)

(E) 3D structure of squid DFPase (PDB: 3BYC) and coordination environment of catalytic Ca²⁺ ion (Ca²⁺ -1).

![Diagram F](image6)

(F) 3D structure of *H. sapiens* BuChE (PDB: 1V0M) and detailed view of the catalytic site, with the catalytic triad (cyan) and the main amino acid residues of the acyl pocket (blue), the oxyanion hole (red), and the choline-binding site (green).

![Diagram G](image7)

(G) The main interactions of VX in the active site of G117H mutant (PDB: 2XMG). Hydrogen bonds are depicted by black dashes.

**Fig. 14.** Detailed view of the catalytic site of OP degrading enzymes with their ligands [8].

Proposed hydrolysis mechanism for *H. sapiens* BuChE cover that as a first Ser198 is activated by His438 and Glu325 and then Ser198 attacks the OP center, forming a pentavalent TS, before the departure of the leaving group. The TS is stabilized by the amino acid residues in the oxyanion hole. Then, a phosphorylated enzyme intermediate is formed. This intermediate is very stable in the wild type *H. sapiens* BuChE but unstable in the G117H mutant. The following step is the nucleophilic attack on the P atom by a water molecule, which may be activated by His438. Lastly, the product is released and the enzyme regenerated (for Fig F).

In (Fig G), the main interactions of VX in the G117H active site are represented. The VX adduct is positioned in such way that methyl group is located near His438 and the ethoxy group in the acyl-binding pocket, pointing toward Val288. The phosphoryl oxygen is interacting with the main-chain nitrogen atoms of the residues in the oxyanion hole (distance of 2.7, 2.8 and 3.2Å from His117, Gly116 and Ala199, respectively).
As pointed out at the many manuscript, bioremediation presents the most promising technology today to destroy OPs have been shown to be very efficient in detoxification of several OPs but the lack of knowledge of their mechanisms of action have limited the development of this action of these enzymes and have already contributed to important advances in the engineering of more efficient OP degrading enzymes. It is apparent that the theoretical investigations will continue reveal much more and contribute for the development of more efficient ways of detoxifying OPs [8].

On the other hand, our focused on specific functional heterocycle, 2-iminothiazoles are designed, synthesized and tested as protein tyrosine phosphatase 1B inhibitors [9], cannabinoid receptor ligands [7], pifithrin-α p53 inactivators [10], etc. there is not any remarkably record on their insecticidal docking study.

The present work aimed to introduce the molecular modelling and mapping of active site of previously synthesized by us insectisidal 2-iminothiazole derivatives [11], by using classical docking techniques, i.e. MOE, etc. [12]

2. MATERIAL AND METHODS

Synthesis

Synthetic procedure of 2-iminothiazole compounds was described previously by us [11]. Promising insecticid derivatives were selected for docking data set and illustrated on Table 1.

![Fig. 15. General structures of 2-iminothiazoles](image)

**Table 1.** The structures of tested compounds:

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<td>26</td>
<td><img src="image" alt="Structure 26" /></td>
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<td><img src="image" alt="Structure ID 99" /></td>
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Materials

Literature search were performed by using SciFinder, Web of Science and Protein Data Bank databases. Chemical drawing programs: 2D Drawings were made using ChemDraw and ISIS Draw programs. 3D drawing of chemical molecules was carried out using the MOE 2016 program (AUBAP, 2017, 1701S009). All text writing and chemical drawing, application procedure were accomplished by Intel Core i7 processor (7700 HQ), Windows 2010 operating system workstation (AUBAP, 2017, 1701S009).

Preparation of ligands

The ligand files for molecular docking studies were prepared in Molecular Operating Environment (MOE-2016.0802) by chemical computing group (CCG) and were followed by energy optimization at standard MMFF94 force field level, with a 0.0001 kcal/mol energy gradient convergence criterion [12]. The optimized geometries were saved in molecular data base file for further studies.

Molecular docking studies

The optimized ligands were docked with the Ls-AChBP (PDBID 2ZJU) protein using the MOE-Dock program. For docking, default MOE docking parameters, i.e. Triangle Matcher Algorithm with two rescoring functions, London dG and GBVI/WSA dG were utilized to generate 30 poses of each compound. As a result of docking run, mdb output files were generated enclosing all docking results with scoring and multiple conformations of ligands. All the docked conformations were analyzed and the best scored pose for each compound was selected for further studies of interaction evaluation. The 2D ligand-protein interactions were visualized using the MOE ligand interactions program.

RESULTS

Activity on nicotinic acetilcholine receptors (nAChRs) has been checked by using Ls-AchBP crystal structure complexed with imidacloprid (PDBID 2ZJU) replaced by data set. Docked compounds on 2ZJU showed correspondence interaction with imidacloprid.
CONCLUSIONS

It is stand out that the alignment of data set is not able to accomplish because of non-symmetric property of imines. Their configuration of imines may result in different activity or ranging from more to less affinity to related enzyme binding. To identify clearance of activity, further purification and elucidation of structures i.e. X-ray crystallography may offer valuable activity results for non-symmetric imines. Based on the preliminary docking study of data set, we observed favourable interaction complexes of the designed compounds with target protein Ls-AchBP. Further biological testing will be carried out to verify the docking studies. In accordance with the in-silico docking and activity results would allow us to develop new strategies for novel active compounds.
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REFERENCES