Computer-Aided Design of Novel Active Components in Plant Protection

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Abstract: - The production demands highly specific environmentally and toxicologically acceptable plant protection products are increasing. Computer-aided molecular design of new active components has a great deal in developing plant protection products to avoid that long-lasting and expensive process. Computational design of future compounds and their synthesis, evaluation of their effectiveness on harmful and beneficial organisms in the soil, as well as detailed research mechanism of action at the molecular level, represents an initial stage in the long-lasting and expensive process of plant protection products. In this paper, the recent advances in quantitative structure-activity relationship (QSAR) studies, molecular docking, and calculation of "Pesticide-likeness properties ", as well, have been reviewed. QSAR models for antifungal activities against phytopathological fungi were obtained for the thiazoline and coumarine derivatives, coumarinyl Schiff bases, and coumarin-1,2,4-triazoles. A molecular docking study revealed that antifungal activities of fluorinated pyrazole aldehydes are related to the inhibition of proteinase K, coumarinyl Schiff bases with endoglucanase and pectinase, hybrids of coumarins and 1,2,4-triazoles with sterol 14α-demethylase inhibition, 3-gydroxycoumarin chitin synthase, while γ-thionins strongly binds to fungal membrane moieties.

Key-Words: - plant protection, QSAR, molecular docking, pesticide-likness, antifungal, insect pests

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1 Introduction

Organic compounds have always been and still are, of vital importance for the protection of crops. Since the world population continues to grow, there is a requirement for increased crop yields and better-quality food.

Although the present use of synthetic compounds in plant protection has limited the occurrence and development of plant diseases and pests, they have shown environmental and health hazards. That indicates an urgent need for new, safe active ingredients of plant protection products. Therefore, during the last decades, the development of new and selective active ingredients of plant protection products has taken place with special emphasis on the assessment of the behavior of these chemicals in the environment. Although the use of synthetic compounds in plant protection has reduced the occurrence and development of plant diseases and pests, great problems arise because of the resistance to pesticides and their environmental and health hazards. These compounds must be effective at extremely low doses, easily degradable, having the least side effects on human health, non-target organisms, and the environment. Non-target organisms are mainly non-vertebrates that plays an essential role in ecosystems as pest controllers (predators), pollinators, detrivores and saprophages, [1].

Computer-aided molecular design (CAMD) is a modern strategy for the development of plant protection substances for its high efficiency in the design of new compounds. Applying CAMD reduces the economic costs and development time of new plant protection products reducing the number of chemical synthesis and biological tests. The REACH (Registration, Evaluation, and Authorization of Chemicals) regulation from 2011 promotes non-animal test methods in testing chemicals' impact on human health and the environment. Therefore, the European Chemicals Agency (ECHA) suggested that animal tests can be avoided by calculating hazardous

properties using computer models of the quantitative structure-activity relationship (QSAR) approach, [2]. QSAR technique relates chemical structure and biological activity providing information on structural features relevant to the observed activity. Quality predictive QSAR model allows the design and development of new molecules with improved activity, [3]. Molecular docking is a molecular modeling technique used to elucidate the mode of action of active compounds that interact with receptors (enzyme, protein) related to the observed biological activity. The molecular docking allows the screening of the binding affinity of the ligand according to the scoring function (binding energy), [4].

This study aims to review a recent advance in the use of CAMD techniques for the development of a novel active component of the future plant protection product.

2 Methodology

2.1 QSAR Method

QSAR development process includes several steps: 1. Drawing chemical structures. 2. Generation of the 3D chemical structure using different software, such as Avogadro, [5], and PyMol, [6] 3. Optimization of molecular structures using different methods: molecular mechanics force field (MM+), [7], semiempirical methods, [8], density functional theory (DFT), [9]. 4. Calculation of molecular descriptor calculation performed using different web platforme, such as, Parameter Client, [10] and OCHEM, [11]. 5. Reducing the number of descriptors from the initial set; generation of the QSAR models using the descriptor sets; splitting molecules in training and test set; generation of QSAR models (multiple linear regression) could be performed by QSARINS-Chem 2.2.1. (University of Insubria, Varese, Italy), [12] 6. Generation of QSAR models. 7. Obtained QSAR models are assessed by fitting criteria; internal cross-validation using the leave-one-out (LOO) method and external validation. The robustness of QSAR models is tested by the Y-randomisation test. The applicability of the obtained models is checked by the residual plots and Williams plots using QSARINS. 8. Interpretation of the QSAR model relieves elucidation of important physicochemical and structural requirements for the biological activity of heterocyclic compounds. 9. Drawing structures of the future molecules. 10. Predict the activity for the future molecules using obtained QSAR models.

2.2 The Molecular Docking Method

Protein crystal structures in complex with docked ligands are downloaded from the Protein Data Bank (PDB, https://www.rcsb.org/). Various programs are used for the molecular docking of compounds, such as AutoDock Vina, [13], Glide, [14], or iGEMDOCK, [15]. For the target enzymes without a known three-dimensional structure, the homology modeling method is applied and carried out using MODELLER program, [16]. The screening compounds are ranked based on the energy of interactions with amino acid residues of the binding site.

2.3 Calculation of Pesticide-Likeness, Environmental and Health Hazards Properties of Compounds

"Pesticide-likeness" are physical-chemical properties that characterize compounds as potentially successful plant protection agents, [17]. According to this role leading candidate for the development of pesticide candidates should have: molecular weight (MW ≤ 435 Da); lipophilicity (ClogP ≤ 6), number of H-bond acceptors (HBA ≤ 6), the number of H-bond donors (HBD ≤2), number of rotatable bonds (ROB ≤9), and the number of aromatic bonds (ARB≤ 17). For the calculation of pesticide-like properties, and environmental and health hazards properties of compounds, as well, these are several free-available software: ADMETlab 2.0, [18], SwissADME, [19], and (T.E.S.T. (Toxicity Estimation Software Tool), [20]. Software T.E.S.T. estimates toxicity using several advanced QSAR methodologies (hierarchical clustering; multiple linear regression model; group contribution approach; nearest neighbor; mode of action (MOA) method; consensus method): T.E.S.T estimates the value for several endpoints such toxicity for: *Pimephales promelas*, *Daphnia magna*, *Tetrahymena pyriformis*; oral rat toxicity; bioaccumulation factor; developmental toxicity; mutagenicity (Ames test), [20]. Estimation of blood-brain barrier penetration, carcinogenicity in rodents; and mutagenicity in *Salmonella typhimurium* could be calculated by Lazar online toxicity predictions software, [21].

3 Recent advances in computer-aided molecular design of novel active components of plant protection

Song et al. (2008), [22] developed QSAR models for fungicidal activities of thiazoline derivatives to identify important structural factors for fungicidal activities. The five-descriptor MLR model showed lower quality (R^2 = 0.829), than non-linear relationships obtained by NN (R^2 = 0.966). Comparative molecular field analysis (CoMFA) and comparative molecular similarity indices analysis (CoMSIA) have been used to establish QSAR models for the fungicidal activity of 38 *N*-nitrourea derivatives. Both models shown good prediction capability (R^2 = 0.959; and R^2 = 0.936, respectively), [23]. Also, CoMFA and CoMSIA models were developed for the antifungal activities of coumarin derivatives against phytopathogenic fungi *Valsa mali* (correlation coefficients were 0.918 and 0.949, respectively). The models have shown that electropositivity of substituents are favorable for the antifungal activity of coumarins, [24].

The nonlinear methods least-squares support vector machine (LS-SVM) and project pursuit regression (PPR) have been shown as good modeling tools for the modeling of fungicidal activities against the rice blast disease of thiazoline derivatives, [25]. Coumarine derivatives showed notable antifungal effects against phytopathogenic fungi *Macrophomina phaseolina* and *Sclerotinia sclerotiorum*. QSAR analysis was performed for the activities against fungi, and the predictive QSAR model were obtained for *M. phaseolina* (R^2_{tr} = 0.78; R^2_{ext} = 0.67). Generated QSAR models indicated the importance of multiple electron-withdrawal groups, especially at position C-3, which enhanced antifungal activity against *M. phaseolina*. The increased antifungal activity against *S. sclerotiourum* contributes hydrophobic benzoyl group at the pyrone ring, as well as the methoxy group at the benzene ring. To elucidate the mechanism of antifungal action, molecular docking was performed on the enzymes responsible for the fungal growth and the plant cell wall-degrading. The results have shown that coumarine derivatives possibly act inhibitory against proteinase K, the plant wall-degrading enzyme (Fig. 1), [26].

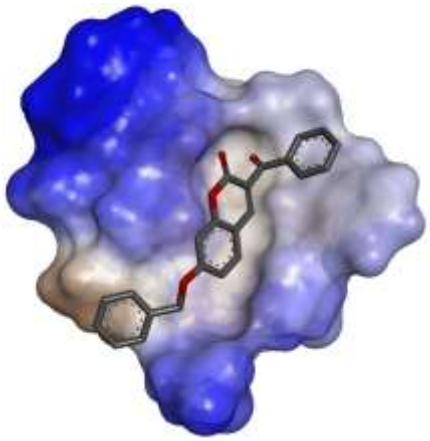

Fig. 1. Hydrophobic surface representation of proteinase K (pdb ID: 2pwb) active site with docked coumarin derivative.

Coumarinyl Schiff bases have been shown as promising antifungal effects against *M. phaseolina*. Molecular docking indicated that the tested compounds inhibit the growth of *M. phaseolina* by inhibiting enzymes that participate in the breakdown of plant cell walls, such as endoglucanase and pectinase, [27].

The fluorinated pyrazole aldehydes showed moderate antifungal properties against four phytopathogenic fungi: *Fusarium oxysporum*, *Fusarium culmorum*, *M. phaseolina,* and *S. sclerotiorum*. According to the molecular docking study, their antifungal activity is possibly related to the inhibition of proteinase K, [28].

A hybrid compounds, coumarin-1,2,4-triazoles successfully inhibited the growth of S. *sclerotiorum* and *F. oxysporum* and two predictive QSAR models were generated for both fungi (R^2 = 0.79; and R^2 = 0.977, respectively). QSAR models revealed that longer linkers between triazole and coumarin motifs, additional tertiary sp^2 carbon atom or ether group, and electronegative substituents could enhance the antifungal activity of the compounds. These compounds are possibly acting as sterol 14α-demethylase inhibitors, according to the molecular docking results, [29].

3-Hydroxycoumarin has been shown as inhibitors of the growth of *Moniliophthora perniciosa* fungus, the agent of witches' broom disease in *Theobroma cacao* L.. Results of molecular docking suggested that it inhibits the production of chitin synthase, [30].

Computational methods have also been used for the design of insecticides. CoMFA and CoMSIA models have been generated for the insecticidal activity of coumarine derivatives against carmine spider mite, *Tetranychus cinnabarinus*. The 3D-QSQAR models revealed that C-3, C-6, and C-7 positions in the

skeletal structure of the coumarins are the most suitable active sites. The molecular docking revealed the interaction between the scopoletin and Ca2+-ATPase 1 gene (TcPMCA1), which is involved in the mechanism of its detoxification, [31]. Bingchuan et al., [32] have applied the stepwise regression analysis method for building predictive QSAR) model for acaricidal bioactivity against the plant pest carmine spider (*Tetranychus cinnabarinus*) of coumarin compounds. Based on strong acaricidal activities of scopoletin phenolic ether derivatives against female adults of *T. cinnabarinus*, authors have developed statistically significant QSAR model (R^2 = 0.967) that reveals the importance of polarizability and bulkiness of substituents, hydrophobic groups, and electron positive groups at the specific position for their activity.

Series 4-amino-5-substituted aryl-3-mercapto-(4H)-1,2,4-triazoles have shown nematicidal activity against *Meloidogyne incognita* and *Rotylenchulus reniformis*. According to the multiple linear regression equation obtained by QSAR analysis, authors have concluded that nematicidal activity is influenced by bulkiness, width, and electronic effect of substituents on the benzene ring, [33].

γ-Thionins are antimicrobial peptides acting in plant defense against pathogenic fungi and insects that could be used as natural fungicides and insecticides. A molecular docking study revealed that γ-thionins bind on the fungal membrane moieties leading to cell death [34]. The k-nearest Neighbor (k-NN) method has been used to develop a QSAR model of acute contact toxicity of 256 pesticides on honey bees (*Apis mellifera*). The obtained model could be used for the prediction of toxicity of new pesticides, [34].

4 Conclusion

CAMD has great importance in the design of highly efficient agrochemicals, saving time and costs for unnecessary synthesis and biological tests on animals. QSAR models could help to elucidate the most important structural characteristics studied series of compounds for biological activities, which allows to design of the next series of series with improved activities. Statistically significant QSAR models should be implemented in existing or new applications for predicting the activity of compounds related to plant protection. The models should also be extended for the prediction of the effects on beneficial, non-target organisms to avoid the negative ecotoxicology impact of studied agrochemicals. Molecular docking is a promising tool for screening of untested compounds, elucidating pesticides target-site and their mechanism of action.

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