

# QSAR and pharmacophore analysis on pyridazinone derivatives as acetylcholinesterase inhibitors

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## Abstract

**Aim:** The aim of this study is to provide a qualitative and quantitative explanation of the structure activity relationships with pharmacophore analysis and Quantitative Structure Activity Relationship (QSAR) studies of the compounds synthesized as acetylcholinesterase enzyme inhibitor by our research group.

**Material and Methods:** Maestro 11.9 (Schrödinger, New York) was used for pharmacophore model studies. Pharmacophore analysis was performed for all compounds showing acetylcholine esterase inhibitory effect. For QSAR studies, various physicochemical parameters of these compounds were calculated using GaussView 5.0 and ChemDraw 15.0 programs. Regression analysis was performed by using these parameters and QSAR equation was obtained.

**Results:** All compounds overlapped and hypotheses generated. The most appropriate pharmacophore model was created by comparing the hypothesis and activity results of the compounds. The analyses were performed using 8 different parameters for all compounds. R<sup>2</sup> value of equation was found 1.

**Conclusion:** The pharmacophore analysis and the QSAR equation are applicable for all compounds synthesized as acetylcholinesterase inhibitory and containing pyridazinon-2-ylacetohydrazide structure. Also, the estimated IC<sub>50</sub> values can be calculated before the compounds are synthesized using the QSAR equation.

**Keywords:** Acetylcholinesterase; QSAR; pharmacophore analysis; pyridazinone

## INTRODUCTION

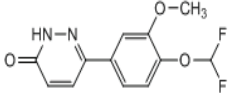
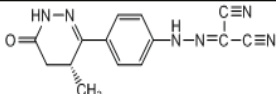
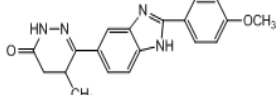
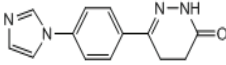
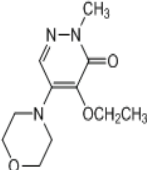
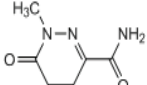
Alzheimer's disease is a progressive fatal neurodegenerative disease. In the later stages of the disease, decreased ability to perform daily activities, cognitive dysfunctions and various neuropsychiatric symptoms are appeared (1-3). While close memory loss is the first clinical symptom of the disease, distant memory is relatively protected during illness. As the disease progresses, visual tests, the ability to use objects and objects and the ability to calculate are reduced. The level of alertness and motor strength persists until further periods of the disease. Although muscle contractions are common in later stages of the disease, motor strength is not damaged even in these stages (3). Although the pathophysiology of Alzheimer's disease is not yet known, it is thought to be the result of the disappearance of

brain cells for an undetermined reason. Three main hypotheses regarding the formation of the disease are proposed. According to the cholinergic hypothesis, which is the oldest hypothesis proposed, Alzheimer's disease is thought to be caused by the decrease of acetylcholine which is an important neurotransmitter (4,5). Degeneration of cholinergic neurons in the basal forebrain in Alzheimer's patients; There is a significant decrease in cholinergic receptors and choline acetyltransferase (ChAT) levels in cerebral cortex. Although many of the previous treatment approaches have been based on this hypothesis, clinical research has shown that treatment strategies for increasing acetylcholine (ACh) levels provide symptomatic improvement (6). Recent studies on cholinergic hypothesis have shown that the use of cholinesterase inhibitors may affect the formation of amyloid fibrillation (7,8). Acetylcholinesterase (AChE)

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which is known as one of cholinesterase enzymes which take place in serine hydrolase enzyme series (9). The main role of AChE is hydrolysis of ACh, which is involved in cholinergic synapses and plays a role in the regulation of cognitive functions in humans. (10-13). ACh level rises in the cholinergic synapses when these enzymes are inhibited. Thus, cholinesterase inhibitors are used in the treatment of various neuromuscular disorders such as Alzheimer's Disease (AD), which the level of ACh is low. (14-16).

Formula / Chemical Name	Generic Name	Activity
 6-(4-(difluoromethoxy)-3-methoxyphenyl)pyridazin-3(2H)-one	Zardaverine	Cardiotonic
 (R)-N-(4-(4-methyl-6-oxo-1,4,5,6-tetrahydropyridazin-3-yl)phenyl)carbonohydrazonyl dicyanide	Levosimendan	Calcium sensitizer
 6-(2-(4-methoxyphenyl)-1H-benzo[d]imidazol-5-yl)-5-methyl-4,5-dihydropyridazin-3(2H)-one	Pimobendan	Positive inotropic, Vasodilator
 6-(4-(1H-imidazol-1-yl)phenyl)-4,5-dihydropyridazin-3(2H)-one	Imazodan	Cardiotonic
 4-ethoxy-2-methyl-5-morpholinopyridazin-3(2H)-one	Emorfazon	Analgesic
 1-methyl-6-oxo-1,4,5,6-tetrahydropyridazine-3-carboxamide	Medazonamid	Antitussive

**Figure 1.** Some drugs carrying pyridazinone ring used in treatment

The compounds containing the pyridazinone ring are involved in the structure of different drug molecules because of their various biological activities. Some of the present day drugs such as emorfazone (analgesic), pimobendan (positive inotropic, vasodilator), levosimendan (calcium sensitizer), imazodan (cardiotonic), zardaverin (cardiotonic) medazonamide (antitussif) are the best

examples for potent molecules possessing pyridazinone nucleus (Figure 1) (9-12).

Pharmacophore analysis is based on the interpretation of the receptor structure by taking advantage of an impressive technology structure in design according to the ligand structure. If the three-dimensional (3D) structure of the receptor is not known, pharmacophore analysis is performed. In this way, the conformation of the biological effect may play a role. The structure of the receptor may then be come on view or mapped from such ligands. In the way, it is possible to design new drug candidates by using existing structure-effect relationships. Pharmacophore is defined as the basic functional groups required for the biological activity of ligands. In other words, it is the spatial editing of the structural elements necessary for certain biological activity (17).

Pharmacophore model; hydrophobic groups comprising the 3D structure of the molecule, the charged or ionizable groups, shows the molecular properties of hydrogen bonds such as donors and receptors. The compounds are superimposed to form a pharmacophore model and their common properties are determined. Compounds compatible with the pharmacophore model can be used as a 3D data model (18).

Absorption, distribution, metabolism and elimination properties of drugs are very important for the successful use of drugs in the clinic. It is estimated that approximately 50% of drugs are unsuccessful due to decreased bioavailability as a result of low intestinal absorption and unexpected drug metabolisms. For this reason, ADME screening is performed beforehand in order to eliminate the ADME properties that are not suitable for the drug design process (17,18).

QSAR is a quantitative study of the relationship between the physical and chemical properties of chemical compounds and their biological activities by using mathematical methods. With these studies, it is ensured that the new pioneer compounds are designed in a rational manner and that the data that can contribute to the development of these are obtained (19).

Various constants indicating the physicochemical properties of the compounds are used as independent variable parameters in QSAR analyses. These physicochemical properties enable the identification of the factors involved in the interaction of the active substance and the target (receptor, enzyme, etc.). The pharmacokinetic process involving the distribution of the active substance after its absorption and its transport to the site of action will also be associated with physicochemical properties (17,18). With these studies, it is possible to design new pioneer compounds in a rational way and to obtain data that can contribute to their development.

According to this information, in this study, QSAR and

pharmacophore analyses of a series of N'-[(substituted phenyl) sulfonyl]-2-(6-(substituted phenyl)-3(2H)-pyridazinone-2-yl) derivatives were performed (compound 1-10). These compounds were previously synthesized and cholinesterase inhibitor effects were determined (20). In addition, the ADME properties of the compounds were investigated as in silico (Figure 2).

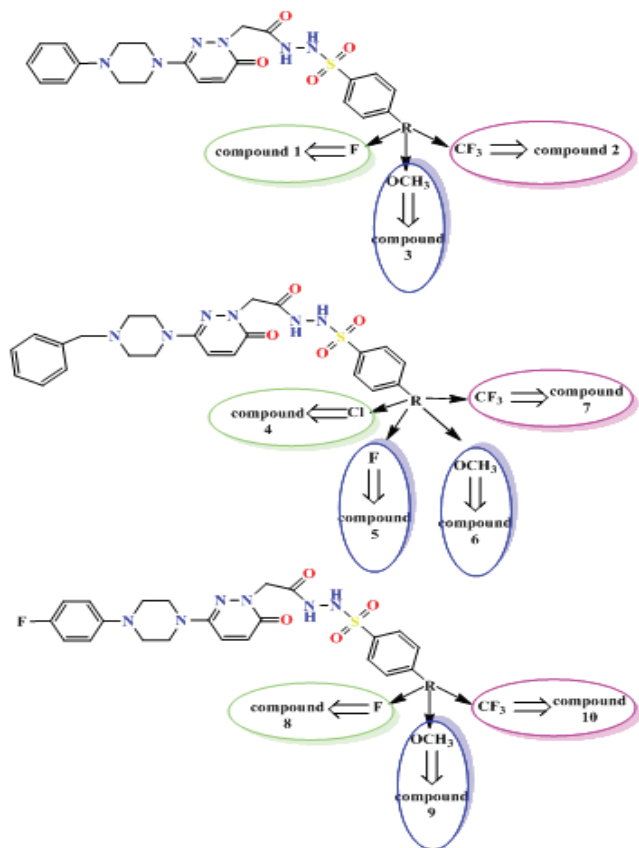


Figure 2 . Structure of compounds 1-10 (20)

## MATERIAL and METHODS

### Pharmacophore Analysis

Maestro 11.9 (Schrödinger, New York) was used for pharmacophore model studies. Ten compounds with have activity were identified and the pharmacophore hypothesis was formed. A (acceptor), D (frost), H (hydrophobic), N (negative ionic), P (positive ionic), R (aromatic ring) properties were selected for hypothesis. After the hypothesis was created, the compatibility of ligands with the hypothesis was investigated in Phase Ligand Screening (Schrödinger, New York). The results are evaluated as Alignment score (is root mean squared deviation (RMSD) between hypothesis sites and the corresponding matches in ligand sites).

### QSAR Analysis

IC<sub>50</sub> values of compounds were calculated online at [www.aatbio.com/tools/ic50-calculator](http://www.aatbio.com/tools/ic50-calculator) using % inhibition values at 50 μM, 100 μM and 200 μM concentrations.

In QSAR analysis, physicochemical parameters were

calculated. Parameter value of the compounds and their steric parameter values, were homo, lumo, band gap, dipole moment and length were calculated using gauss view 5.0 program. logP and molar refractivity were calculated with chemdraw professional 15.0. The synthesized compounds were prepared in LigPrep (maestro, schrodinger 11.9). And also 227 parameters are calculated using the qikprop (maestro, schrodinger 11.9) software.

As a result of the study, 8 parameters legenth, SASA, PISA, WPSA, Volume, polar surface area (PSA), dipole moment (DM), and QPlogS which related to activity were determined and QSAR equation was formed.

## RESULTS

### QSAR Analysis

The analyses were performed using 8 different parameters for ten compounds. R<sup>2</sup> values in both equations were found 1 and the obtained QSAR equations are very similar.

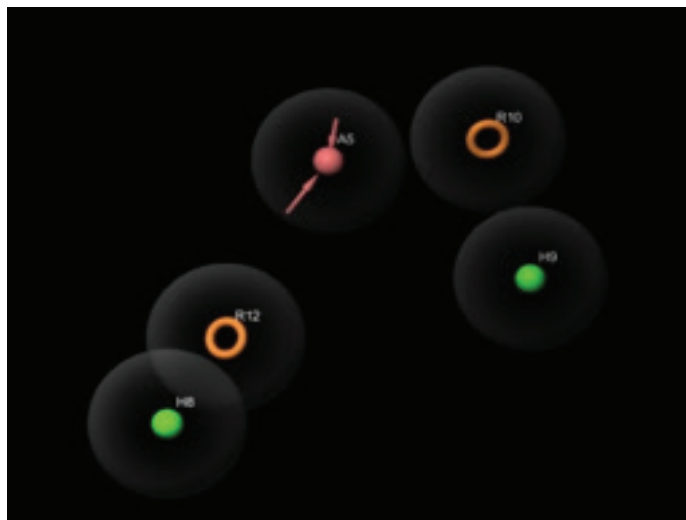


Figure 3. The most appropriate pharmacophore model

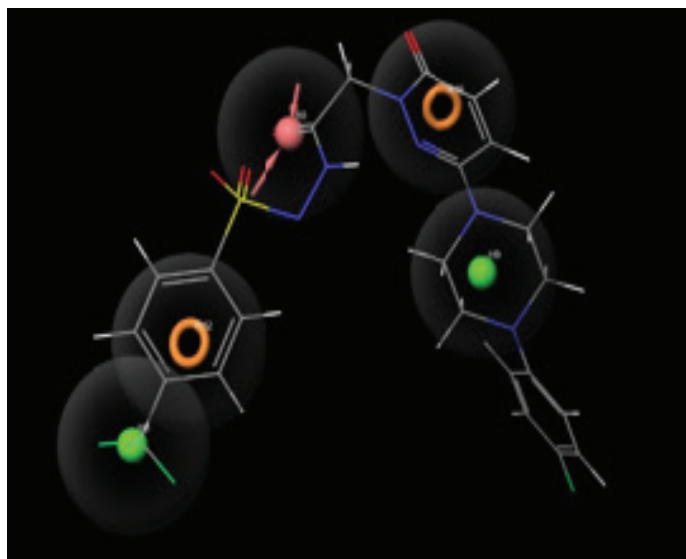


Figure 4. The matching of the compound 10 compound and the pharmacophore model (AHHRR)

QSAR equation for compounds 1-10 for IC<sub>50</sub> values;

$$IC_{50} = 11516.01 + 57.92(\text{dipole}) - 21.26(\text{SASA}) + 29.21(\text{FOSA}) + 27.36(\text{PISA}) + 35.58(\text{WPSA}) - 7.95(\text{volume}) + 277.30(\text{QPlogS}) - 1.81(\text{PSA}) - 8.38(\text{lenght})$$

### Pharmacophore Analysis

Active 10 compounds overlapped and hypotheses

generated. The most appropriate pharmacophore model was created by comparing the hypothesis and activity results of the compounds (Figure 3). The compatibility of the compound 10 with the best activity with the generated pharmacophore hypothesis is shown in Figure 4.

### ADME Prediction

In silico ADME results are given in Table 1.

**Table 1. Some physicochemical and pharmacokinetic properties of compounds.**

Compound	Dipole moment	SASA	FOSA	PISA	WPSA	Volume	QPlogS	PSA	Lenght
Comp. 1	1.48	740.02	179.60	361.74	46.7	1348.81	-5.49	119.07	12.73
Comp. 2	7.25	746.3	274.80	317.8	4.47	1390.2	-4.976	123.904	17.94
Comp. 3	7.04	793.27	181.53	340.8	116.18	1448.1	-6.708	127.38	14.52
Comp. 4	5.50	791.74	208.66	354.0	72.55	1455.6	-4.565	128.898	17.87
Comp. 5	7.43	792.6	192.08	390.5	49.61	1433.1	-4.479	127.089	18.51
Comp. 6	7.93	803.12	302.12	346.2	4.203	1476.5	-4.01	129.043	18.83
Comp. 7	4.88	715.2	175.02	297.7	82.4	1437.5	-3.32	121.039	15.35
Comp. 8	1.26	743.4	181.03	309.4	93.68	1364.2	-5.759	121.451	12.67
Comp. 9	8.99	781.9	274.95	300.3	50.88	1431.98	-5.781	129.601	18.08
Comp. 10	4.51	796.9	179.15	288.9	167.6	1452.92	-6.996	121.451	17.31

Dipole: Computed dipole moment of the molecule. SASA: Total solvent accessible surface area in square angstroms using a probe with a 1.4 Å radius. FOSA: Hydrophobic component of the SASA (saturated carbon and attached hydrogen). PISA: π (carbon and attached hydrogen) component of the SASA. WPSA: Weakly polar component of the SASA (halogens, P, and S). Volume: Total solvent-accessible volume in cubic angstroms using a probe with a 1.4 Å radius. QPlogS: Predicted aqueous solubility, log S. S in mol dm<sup>-3</sup> is the concentration of the solute in a saturated solution that is in equilibrium with the crystalline solid. PSA: Van der Waals surface area of polar nitrogen and oxygen atoms. Lenght: Distance between two farthest atoms of molecule.

## DISCUSSION

The five-feature pharmacophore model was generated which has an acceptor group (A5), two hydrophobic groups (H8, H9) and two rings aromatic feature (R10, R12). Compounds were screened using the generated pharmacophore model (AHRRR) to search for potential acetylcholinesterase inhibitors. According to ADME properties (Table 1) and the Alignment scores (Table 2) and compound 10 with the lowest IC<sub>50</sub> provides the best fit with our pharmacophore model (Figure 4).

As a result, QSAR analysis and 3D-common feature pharmacophore hypothesis generation indicated that the physicochemical and conformational properties of the compounds are important in the inhibition of acetylcholinesterase. Compounds possessing a fluor and trifluoromethyl at R2 position moiety structure increase the activity. It can be considered that substituted aromatic ring at R1 is required for anticholinesterase activity. The estimated IC<sub>50</sub> values of the new compounds having similar skeleton can be calculated using this QSAR equations and the pharmacophore analysis.

**Table 2. IC<sub>50</sub> values and Alignment Score of molecules**

Molecule	IC <sub>50</sub>	Alignment Score
Comp. 1	99.78	0.776
Comp. 2	141.32	1.059
Comp. 3	93.32	0.442
Comp. 4	141.52	0.950
Comp. 5	133.42	1.237
Comp. 6	101.83	0.560
Comp. 7	85.37	0.603
Comp. 8	99.52	0.736
Comp. 9	95.66	0.628
Comp.10	79.84	0.487

## CONCLUSION

In this study, we examined the relationships between the structures of 10 compounds and AChE inhibitor activities in vitro using various computer programs. In order to design more effective, targeted compounds, pharmacophore analysis was performed and QSAR equation was formed.



The general skeleton which responsible for the activity of our compounds was determined with the help of acceptor group, hydrophobic groups and two rings aromatic properties determined in pharmacophore analysis (Figure 3). When the alignment score given in Table 2 was compared with  $IC_{50}$  values, a significant relationship was observed. As shown in the table, the  $IC_{50}$  values of the compounds with low alignment score are also low.

In addition to their three-dimensional structure, some physicochemical activities are important in the activity of the compounds. For this purpose, some physicochemical parameters of the compounds were found to be related to the calculated activity is shown in Table 1. These parameters were used in the QSAR equation. According to this analysis, the increases in the length, PSA and SASA values of the compounds decreases the  $IC_{50}$  value; increasing of FOSA, PISA, WPSA, QPLogS, volume and dipole moments increases the  $IC_{50}$  value.

Consequently the estimated  $IC_{50}$  values and anticholinesterase activity of the new compounds having similar skeleton can be calculated using this QSAR equations and the pharmacophore analysis.

*Competing interests: The authors declare that they have no competing interest.*

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