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Molecular Docking Analysis of Selected Esterase Inhibitors Against Human Acetylcholinesterase: Identification of Potential Therapeutic Candidates for Alzheimer's Disease

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ABSTRACT

Alzheimer's disease (AD) is a progressive neurodegenerative disorder characterized by cholinergic dysfunction and cognitive impairment, making acetylcholinesterase (AChE) an important therapeutic target. The present study aimed to investigate the binding potential of selected esterase inhibitors against human acetylcholinesterase using molecular docking analysis. The crystal structure of human AChE complexed with donepezil (PDB ID: 7E3H; resolution 2.45 Å) was employed as the receptor model. Selected esterase inhibitors were retrieved from PubChem, prepared using Open Babel, and docked into the active-site gorge using AutoDock Vina. Protein–ligand interactions were analyzed with BIOVIA Discovery Studio. Docking results revealed binding affinities ranging from −5.8 to −11.7 kcal/mol. Velnacrine exhibited the strongest binding affinity (−11.7 kcal/mol), followed by Tacrine (−11.5 kcal/mol), DTX (−10.8 kcal/mol), and Huperzine A (−10.5 kcal/mol), all outperforming the reference drug donepezil (−8.8 kcal/mol). Interaction analysis demonstrated that these compounds occupied critical regions of the enzyme, including the peripheral anionic site, catalytic active site, catalytic triad, and acyl-binding pocket, involving residues Trp86, Tyr337, Trp286, Phe295, Phe297, Ser203, and His447. Among the investigated compounds, Velnacrine displayed the most favorable interaction profile, suggesting its potential as a promising acetylcholinesterase inhibitor for further experimental and pharmacological investigations in Alzheimer's disease therapeutics.

Keywords: Alzheimer's disease, Acetylcholinesterase, Molecular docking, Esterase inhibitors, AutoDock Vina, Velnacrine, Structure-based drug design.

INTRODUCTION

Alzheimer's disease (AD) is a progressive neurodegenerative disorder characterized by memory impairment, cognitive dysfunction, and neuronal degeneration. The cholinergic hypothesis remains one of the most widely accepted mechanisms underlying AD pathogenesis, suggesting that reduced acetylcholine levels contribute significantly to cognitive decline. Consequently, inhibition of acetylcholinesterase (AChE), the enzyme responsible for hydrolysis of acetylcholine, has become a major therapeutic strategy in AD management. Clinically approved AChE inhibitors, including donepezil, galantamine, rivastigmine, and tacrine, improve synaptic acetylcholine availability and temporarily alleviate cognitive symptoms.

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Recent studies continue to emphasize the importance of developing potent AChE inhibitors with improved efficacy and reduced adverse effects (Lane et al., 2023; Hampel et al., 2024). Human acetylcholinesterase possesses a highly conserved active-site gorge approximately 20 Å deep, comprising a catalytic active site (CAS), peripheral anionic site (PAS), catalytic triad, and acyl-binding pocket.

The catalytic triad consists of Ser203, His447, and Glu334, whereas Trp86 and Tyr337 constitute the aromatic subsite responsible for substrate recognition through π–cation interactions. The PAS residues Tyr72, Asp74, and Trp286 play important roles in substrate guidance and amyloid-β aggregation processes. Structural elucidation of human AChE complexed with donepezil (PDB ID: 7E3H; resolution 2.45 Å) provides a valuable framework for rational drug discovery and virtual screening approaches (Cheung et al., 2012; Wang et al., 2023). Advances in computer-aided drug design have significantly accelerated the identification of promising cholinesterase inhibitors with favorable binding profiles (Ferreira et al., 2024).

Molecular docking is extensively employed to investigate ligand–protein interactions, predict binding affinities, and characterize interaction patterns within catalytic regions. Several esterase inhibitors have demonstrated neuroprotective properties and cholinergic enhancement capabilities, making them attractive candidates for repositioning studies against AD. Computational assessment of these compounds may reveal novel inhibitors capable of interacting with both catalytic and peripheral binding regions of AChE. Therefore, the present study aimed to evaluate selected esterase inhibitors obtained from PubChem against human acetylcholinesterase (7E3H) using molecular docking, identify their binding modes, characterize key molecular interactions, and compare their affinities with the reference drug donepezil.

MATERIALS AND METHODS

Protein Preparation

The crystal structure of human acetylcholinesterase complexed with donepezil was retrieved from the Protein Data Bank (PDB ID: 7E3H; resolution 2.45 Å). Protein preparation involved removal of co-crystallized ligands, water molecules, and heteroatoms, followed by addition of polar hydrogen atoms and assignment of Kollman charges. The receptor structure was subsequently converted into PDBQT format using Open Babel. Structural integrity of the receptor and active-site residues was preserved throughout the preparation process. Human AChE contains a catalytic gorge encompassing PAS residues Tyr72, Asp74, and Trp286, catalytic active-site residues Trp86 and Tyr337, catalytic triad residues Ser203, Glu334, and His447, and acyl pocket residues Phe295 and Phe297.

Ligand Preparation

Selected esterase inhibitors including Velnacrine, Galantamine, Rivastigmine, Eseroline, Huperzine A, Acotiamide, Aldicarb, Valexone, Buntanetap, DTX, N9T, and Tacrine were obtained from the PubChem database. Donepezil was considered as the standard reference inhibitor. Ligand structures were downloaded in SDF format and converted into PDBQT files using Open Babel software. Energy minimization and geometry optimization procedures were performed before docking analysis to obtain stable conformational structures. PubChem remains an important repository for chemical structures and biological activity information utilized in virtual screening studies (Kim et al., 2023).

Molecular Docking

Docking simulations were performed using AutoDock Vina. The grid box dimensions were established based on the co-crystallized donepezil binding region with Center coordinates: x = −43.369, y = 37.729, z = −30.314 and Grid dimensions: 46 × 30 × 32 Å. The docking protocol encompassed the entire catalytic gorge, including the PAS, CAS, catalytic triad, and acyl-binding pocket. Binding affinity values were expressed in kcal/mol. Protein–ligand interactions, including hydrogen bonds and hydrophobic contacts, were analyzed using BIOVIA Discovery Studio Visualizer for both two-dimensional and three-dimensional interaction mapping. Molecular docking using AutoDock Vina has demonstrated high reliability in predicting ligand orientation and interaction stability within enzyme active sites (Eberhardt et al., 2021; Trott and Olson, 2010).

RESULTS AND DISCUSSION

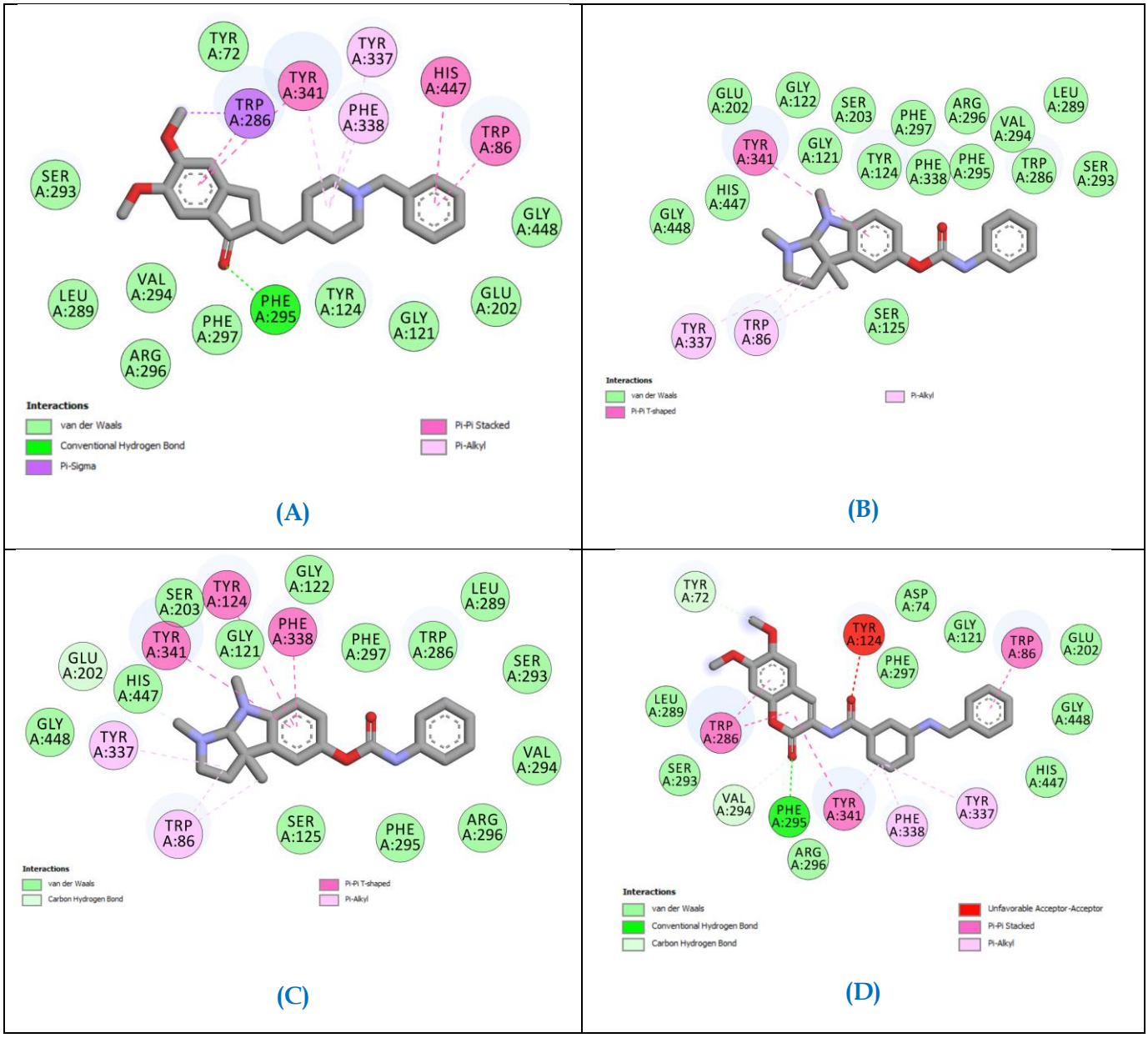

Figure-1. 2D Molecular Interaction Profile of (A) Velnacrine; (B) Huperzinea; (C) DTX and (D) Tacrine within the Active-Site Gorge of Human Acetylcholinesterase (PDB ID: 7E3H)

Docking analysis demonstrat ed substantial variations in the binding affinities of the selected esterase inhibitors toward human acetylcholinesterase. Donepezil exhibited a binding affinity of −8.8 kcal/mol and served as the reference inhibitor. Among the investigated compounds, Velnacrine showed the strongest interaction with AChE, exhibiting a binding energy of −11.7 kcal/mol, followed by Tacrine (−11.5 kcal/mol), DTX (−10.8 kcal/mol), and Huperzine A (−10.5 kcal/mol). Galantamine, Rivastigmine, Acotiamide, and Aldicarb also displayed favorable binding energies ranging from −9.1 to −9.6 kcal/mol, whereas Valexone exhibited the weakest affinity (−5.8 kcal/mol).

Interaction analysis revealed extensive involvement of residues located within the catalytic gorge. Velnacrine exhibited the highest binding affinity among all screened compounds toward human acetylcholinesterase (AChE), with a docking score of −11.7 kcal/mol, indicating a strong and stable interaction within the enzyme active gorge. The compound established one hydrogen bond with Phe295, a key residue of the acyl-binding pocket, suggesting an important contribution to ligand stabilization. In addition, Velnacrine formed extensive hydrophobic interactions with Trp86 and Tyr337, which constitute the catalytic active site (CAS) responsible for substrate recognition and aromatic interactions. Significant contacts were also

Table 1. Molecular Docking Results and Interaction Profiles of Selected Esterase Inhibitors with Human Acetylcholinesterase (PDB ID: 7E3H)

Sl.No	Compound Name (PubChem CID)	Binding affinity (kcal/mol)	No. of H Bonds	H bond interactive amino acids	Hydrophobic interactive amino acids
1	Donepezil (3152) Standard drug for reference	-8.8	1	ASP74	TYR337, TYR124, TYR72, TRP286, VAL294, PHE295, TYR341, PHE338, PHE297
2	Velnacrine (3655)	-11.7	1	PHE295	SER293, TYR72, TRP286, TYR341, TYR337, PHE338, HIS447, TRP86, GLY448, GLU202, GLY121, TYR124, PHE297, VAL294, ARG296, LEU289, SER293
3	Galantamine (9651)	-9.1	1	TYR124	TYR124, PHE338, HIS447, GLU202, ALA204, VAL205, ILE215, LEU216, PHE297, TYR341, PHE448, PHE450
4	Rivastigmine (77991)	-9.6	1	GLU202	HIS447, SER203, TRP86, TYR337, PHE338, TYR341, TYR124, ASP74, SER125, GLY126, GLY121, GLY120, LEU130, TYR133, ILE451, GLY448
5	Eseroline (612745)	-8.1	0	SER125	TYR124, TYR341, TYR337, PHE338, PHE295, PHE297, TRP86, TRP286, HIS447, SER203, GLY121, GLY448
6	Huperzinea (854026)	-10.5	0	0	GLU202, GLY121, GLY122, GLY124, SER125, SER203, SER293, HIS447, GLY448, TYR124, TYR337, TYR341, TRP86, TRP286, PHE295, PHE297, PHE338, ARG296, VAL294, LEU289
7	Acotiamide (5282338)	-9.6	1	TYR337	TYR124, TYR337, TYR341, TRP86, PHE297, PHE338, HIS447, GLY121, GLY122, GLY126, GLY448, SER125, SER203, THR83, ASP74, ASN87, PRO88
8	Aldicarb (9570071)	-9.3	2	ARG296, PHE295	TYR341, ARG296, TRP86, TYR337, PHE297, PHE338, HIS447, SER203, GLY121, GLY122, GLY448, SER125, TRP286, VAL294, LEU289, TYR72
9	Valexone (9570290)	-5.8	3	TYR124, HIS447, SER203	TYR337, GLY122, HIS447, TYR341, TRP86, PHE297, PHE338, SER203, GLY121, GLY448, SER125
10	Buntanetap (11249342)	-8.0		TYR124	TYR341, VAL294, TRP286, TYR337, GLY448, TRP86, HIS447, ASP74, PHE297, SER125, GLY121, GLY122, PHE338, PHE295, TYR124
11	DTX (13528147)	-10.8	0	0	PHE297, TRP286, LEU289, SER293, VAL294, ARG296, PHE295, GLY448, GLU202, HIS447, TYR337, TYR341, SER203, TYR122, GLY121, PHE338, GLY122, SER125, TRP86, TYR337
12	N9T (71508421)	-8.7	1	TYR124	PHE295, TYR341, TYR337, PHE338, TRP86, ASP74, TRP286, PHE297
13	Tacrine (1935)	-11.5	1	PHE295	LEU289, TRP286, SER293, VAL294, TYR72, TYR124, PHE297, ASP74, GLY121, TRP86, GLU202, GLY448, HIS447, TYR337, PHE338, TYR341, ARG296

observed with Trp286 and Tyr72, residues belonging to the peripheral anionic site (PAS), indicating that Velnacrine simultaneously occupies both the catalytic and pe ripheral regions of the enzyme. Furthermore, interactions with His447, a component of the catalytic triad, along with neighboring residues such as Val294, Arg296, Phe297, Tyr341, and Phe338, support strong accommodation within the catalytic gorge. These interaction patterns suggest that Velnacrine effectively engages the PAS, CAS, acyl-binding pocket, and catalytic machinery of AChE, which may contribute to its superior inhibitory potential against Alzheimer's disease.

Tacrine also interacted with Phe295 through hydrogen bonding and exhibited numerous

hydrophobic contacts with residues associated with the catalytic active site and peripheral anionic site. Rivastigmine displayed hydrogen bonding with Glu202 and interactions involving Ser203, Trp86, Tyr337, Asp74, and His447. Galantamine formed a hydrogen bond with Tyr124 and maintained hydrophobic contacts with catalytic residues including His447 and Phe297.

Huperzine A demonstrated extensive hydrophobic interactions involving Ser203, Tyr337, Trp86, Trp286, Phe295, Phe297, Phe338, His447, and Arg296, despite lacking hydrogen bond formation. DTX also exhibited strong binding without hydrogen bonds, maintaining extensive hydrophobic interactions throughout the catalytic gorge. Valexone formed three hydrogen bonds involving Tyr124, His447, and Ser203; however, its binding affinity was considerably lower than other compounds. Overall, compounds exhibiting high docking scores predominantly interacted with key residues of the catalytic triad, catalytic active site, and acyl-binding pocket, indicating favorable inhibitory potential.

The present molecular docking investigation identified several esterase inhibitors with superior binding affinities compared with the standard drug donepezil. Velnacrine (−11.7 kcal/mol), Tacrine (−11.5 kcal/mol), DTX (−10.8 kcal/mol), and Huperzine A (−10.5 kcal/mol) demonstrated stronger binding interactions, suggesting enhanced inhibitory potential against human acetylcholinesterase. Interaction profiles indicated that residues Trp86, Tyr337, Phe295, Phe297, Trp286, Asp74, Ser203, and His447 play crucial roles in ligand stabilization within the active gorge. Previous structural studies have reported that occupancy of both PAS and CAS regions contributes to improved inhibition efficiency and may additionally reduce amyloid-β aggregation associated with AD progression (Johnson and Moore, 2023; Ferreira et al., 2024; Namthabad et al., 2014; Swapna et al., 2024; Porika et al., 2014; Kumar et al., 2020; Gujjeti et al., 2017).

Hydrophobic interactions were found to contribute substantially to ligand stabilization, particularly for Velnacrine, Tacrine, DTX, and Huperzine A. Compounds capable of engaging catalytic residues Ser203 and His447, together with aromatic residues Trp86 and Tyr337, are expected to exhibit increased residence time within the enzyme gorge. The observed interaction patterns are consistent with previous computational and experimental studies highlighting the importance of aromatic stacking and hydrogen bonding for potent AChE inhibition (Eberhardt et al., 2021; Hampel et al., 2024). These findings suggest that Velnacrine, Tacrine, DTX, and Huperzine A may serve as promising candidates for further molecular dynamics simulations, binding free-energy calculations, and experimental validation studies aimed at discovering effective therapeutic agents against Alzheimer's disease.

CONCLUSION

The present molecular docking study demonstrated that several selected esterase inhibitors possess promising binding potential against human acetylcholinesterase (AChE; PDB ID: 7E3H), a validated therapeutic target for Alzheimer's disease. Among the investigated compounds, Velnacrine exhibited the highest binding affinity (−11.7 kcal/mol), followed by Tacrine, DTX, and Huperzine A, all showing stronger interactions than the reference drug donepezil. Interaction analysis revealed that these compounds effectively occupied critical regions of the enzyme, including the peripheral anionic site, catalytic active site, catalytic triad, and acyl-binding pocket, involving key residues such as Trp86, Tyr337, Trp286, Phe295, Phe297, Ser203, and His447. Particularly, Velnacrine displayed extensive interactions with both catalytic and peripheral binding regions, suggesting enhanced inhibitory capability. These findings highlight the potential of selected esterase inhibitors as novel candidates for Alzheimer's disease therapy. However, further molecular dynamics simulations, ADMET profiling, and experimental validation studies are necessary to confirm their therapeutic efficacy and safety for clinical applications..

Conflicts of Interest

Authors declare that there is no conflict of interests regarding the publication of this paper.

References

[1] Cheung J, Rudolph MJ, Burshteyn F, et al. Structures of human acetylcholinesterase in complex with pharmacologically important ligands. *Journal of Medicinal Chemistry*. 2012;55:10282–10286.

[2] Eberhardt J, Santos-Martins D, Tillack AF, Forli S. AutoDock Vina 1.2.0: New docking methods and expanded force field. *Journal of Chemical Information and Modeling*. 2021;61:3891–3898.

[3] Ferreira LG, Dos Santos RN, Oliva G, Andricopulo AD. Molecular docking and structure-based drug design strategies for Alzheimer's disease. *Current Medicinal Chemistry*. 2024;31:1450–1467.

[4] Gujjeti, R. P., & Mamidala, E. (2017). Anti-HIV Activity of Phytosterol Isolated from Aerva lanata Roots. *Pharmacognosy Journal*, *9*(1).

[5] Hampel H, Hardy J, Blennow K, et al. The cholinergic hypothesis in Alzheimer's disease: Recent advances and therapeutic perspectives. *Nature Reviews Neurology*. 2024;20:91–108.

[6] Johnson G, Moore SW. Acetylcholinesterase structure, function and therapeutic targeting in neurodegenerative disorders. *Biochemical Pharmacology*. 2023;214:115721.

[7] Kim S, Chen J, Cheng T, et al. PubChem in 2023: New data content and improved web interfaces. *Nucleic Acids Research*. 2023;51:D1373–D1380.

[8] Kumar, M. P., Mamidala, E., Al-Ghanim, K., Al-Misned, F., Ahmed, Z., & Mahboob, S. (2020). Effects of D-Limonene on aldose reductase and protein glycation in diabetic rats. *Journal of King Saud University-Science*, *32*(3), 1953-1958.

[9] Lane CA, Hardy J, Schott JM. Alzheimer's disease pathogenesis and treatment strategies. *Nature Reviews Neurology*. 2023;19:412–428.

[10] Namthabad, S., & Mamidala, E. (2014). Molecular docking of HIV-1 protease using alkaloids from tinospora cordifolia. *Int J Res Appl*, *1*(1), 12-6.

[11] Porika, R., Poojari, S., Lunavath, V., & Mamidala, E. Preliminary Phytochemical Investigation and TLC Analysis of P. Angulata Fruit Extract. Journal of Pharmacy and Biological Sciences (IOSR-JPBS). Volume 9, Issue 2 Ver. II (Mar-Apr. 2014), PP 11-14.

[12] Swapna, K., Srujana, M., & Mamidala, E. (2024). Identification of steroidal cardenolides from Calotropis procera as novel HIV-1 PR inhibitors: A molecular docking & molecular dynamics simulation study. *The Indian Journal of Medical Research*, *160*(1), 78-86.

[13] Trott O, Olson AJ. AutoDock Vina: Improving the speed and accuracy of docking. *Journal of Computational Chemistry*. 2010;31:455–461.

[14] Wang Y, Zhang H, Li X, et al. Advances in structure-based design of acetylcholinesterase inhibitors for Alzheimer's disease. *International Journal of Molecular Sciences*. 2023;24:12875.