

# Identification of some Novel Racetams as Potential Anti-Convulsant Agents: Virtual Screening, Molecular Docking, and ADMET Study

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**Abstract:** Synaptic vesicle protein 2A (SV2A) is an essential protein in the membrane and associated with epilepsy. SV2A is the molecular purpose of levetiracetam and its racetam analogs as anti-convulsant drugs or nootropic agents. Therefore, an in-silico study was performed to explore these issues. This study recognizes a possible small molecule as a racetam analog by the computational program. Files were downloaded in SDF format; 3D structures of about 2500 racetam and racetam-like ligands and 3D structures of SV2A protein were downloaded from the ZINC database in SDF and PDB format, respectively. Molegro Virtual Docker (MVD) and AutoDock vina were used for the docking study. The pharmacokinetics and toxicity prediction of selected racetam derivatives were performed. In this study, About 2500 compounds were used, and 26 novel compounds were identified as the most potent agent. after selecting these Chemical structures, ADME and toxicity and other parameters related to medicine chemical structures were evaluated, and 1 compound was identified as the best compound for *in vitro* and *in vivo* studies. A Phenyl piracetam derivative has a high affinity to SV2A with BBB permeability and without mutagenicity, tumorigenicity, and irritancy effect.

**Keywords:** SV2A; molecular docking; epilepsy; levetiracetam; virtual screening; ADME.

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## 1. Introduction

Epilepsy is the most prevalent chronic disorder related to the brain that influences people of any age. More than 50 million people globally have epilepsy presenting it as one of the most frequent neurological diseases worldwide [1]. Nearly four out of five people with epilepsy disorder live in low- and middle-income territories [2]. It is approximated that up to 70% of people living with epilepsy could live seizure-free if adequately diagnosed and also well-treated. However, the risk of premature mortality in people with epilepsy is three times greater than in the usual population [3]. Some symptoms happen, such as temporarily losing consciousness or awareness, movement or sensation disturbances, or other cognitive functions [1,4].

Unfortunately, a significant number of patients-especially in the context of medical-intractable epilepsies-may experience different degrees of memory or cognitive impairment, behavioral irregularities, or psychiatric symptoms, which restrict their daily functioning. Therefore, simultaneous treatment of epilepsy and improvement of cognitive disorders can be very helpful for these patients. As a result, epilepsy may resemble a neurodegenerative disorder in several patients. Synaptic vesicle protein 2A (SV2A) is an integral membrane protein

important for the proper function of the CNS and is connected to the pathophysiology of epilepsy [5].

Synaptic vesicle protein 2A (SV2A) is an integral membrane protein important for the proper function of the CNS and is connected to the pathophysiology of epilepsy [6-8]. SV2A act as the molecular target of the antiepileptic drug levetiracetam and its racetam analogs [9]. Levetiracetam is a multiple-action drug that primarily interacts with the synaptic vesicle protein 2A. Levetiracetam is the first approved drug of its kind for the treatment of epilepsy and is momentarily has been prescribed more than any other newer antiepileptic drug [10-13].

The racetam binding site in SV2A and the non-covalent interactions between racetams and SV2A are unknown; therefore, an in-silico study was performed to explore these issues. Furthermore, many studies have shown that levetiracetam [14-16] and brivaracetam [17] has the same efficacy rather than valproate sodium and carbamazepine, and phenytoin with fewer adverse reaction [18] or even are good choices as adjunctive agent for adjuvant therapy in refractory cases without a problem [18,19].

Levetiracetam, a novel AED is chemically unrelated to other currently available AEDs, Neuronal binding to synaptic vesicle protein 2A (SV2A). Levetiracetam looks to act as a modulator of synaptic vesicle exocytosis, managing to direct inhibition of presynaptic neurotransmitter release. It has shown a differential effect of the modulator on the interaction of LEV and BRV with the SV2A protein, suggesting that LEV and BRV act at different binding sites or interact with other conformational states of the SV2A protein. The magnified affinity of brivaracetam for SV2A compared to levetiracetam was guided by a superior antiseizure and antiepileptogenic action. So, finding a higher affinity racetam could help us to overcome this disorder. Furthermore, brivaracetam could be used as an adjunctive therapy for refractory partial epilepsy.

Beyond epilepsy, there is some evidence that pushes us to find rapid antidepressants [20] or nootropic agents for the treatment of polyneuropathy or memory impairment [21] and Alzheimer's disease [22,23] among racetams and racetam likes compounds [24-26]. Moreover, little evidence indicates anxiety-like properties by sv2a modulation [27].

However, there is no study to identify a new racetam with an in-silico model, and the lack of research in this field is completely felt. In general, and considering the cases mentioned above and the severe lack of information, it can be concluded that a computer study is necessary to identify compounds that affect the X-receptor with a potential anti-convulsant effect. Therefore, further research to discover some novel racetam derivatives seems necessary.

## 2. Materials and Methods

### 2.1. Ligand library.

The physicochemical and chemical structure of some known racetam came in table 1. these 22 ligands were selected, and with the help of a zinc database, compounds with at least 40% Similarity were downloaded in SDF format; After selecting and downloading all Desired ligands, 2464 ligands saved.

### 2.2. Ligand preparation.

As it was pointed out above, 3D structures of about 2500 racetam and racetam-like ligands were downloaded from the ZINC database [28] in structure-data file (SDF) format.

PyMol version 1 and then Open Babel (version 2.3.1) [29] were utilized in order to convert SDF to PDB format.

### 2.3. Protein preparation.

The crystal structure of the Synaptic vesicle protein 2A (SV2A) was downloaded as a PDB format using the protein data bank website (<http://www.rcsb.org>) with PDB ID: 4v11 [30]. It should be mentioned that in addition to water molecules deletion in the crystal structure, RMSD cut-off 0.30 °A was applied during minimization.

### 2.4. Molecular docking.

There are many methods for molecular docking each has its own mechanism with Different methods, equations, and results.

#### 2.4.1. Molegro virtual docker.

Molegro Virtual Docker (MVD) 2013.6.0.1 was used for the docking study. MVD requires a 3D structure of both protein and ligand and performs flexible ligand docking. Also, the optimal geometry of the ligand was ascertained for molecular docking. The candidates with the best conformational and energetic results were selected. MVD was used to calculate the interaction energies between ligands and macromolecular systems from the 3D structures of the protein and ligands. The algorithm used was the Mol-Dock Score, an adaptation of the Differential Evolution (DE) algorithm [31].

#### 2.4.2. AutoDock Vina.

First of all, using AutoDock 4.2, rotatable bonds were characterized to the ligands and were saved in PDBQT for the docking process. The protein with PDB format is saved as a PDBQT file. Later 90×90× 90Å (x, y, and z) grid box was centered on the protease binding pocket with 0.375 nm spacing for each dimension and a grid center at dimensions (x, y, and z, respectively): 1.186, -2.301, and 63.375 was designated. Energy range and exhaustiveness were set 8 and 10, respectively. Auto Grid 4.2 was used to create grid maps. Docking parameters were set as follows: energy range was set 4 and exhaustiveness was set 20, and finally, docking was performed by AutoDock 4.2. The procedure of docking was done automatically by codes and scripts written in-house.

#### 2.4.3. Visualizing 2D pharmacophore.

In the next step, 2D interaction SV2A protein with all molecules including levetiracetam and brivaracetam and novel racetams, was visualized by discovery studio client 2020 as well as LigPlus.

### 2.5. ADME and drug-likeness study.

The Swiss database performed the ADME (i.e., absorption, distribution, metabolism, and excretion) and toxicity prediction of selected racetam derivatives [32] and OSIRIS Property Explorer Osiris property explorer software available at <http://www.organic-chemistry.org/prog/peo/>.

2.5.1. ADME.

The SMILE codes of each ligand were input into the ADME Swiss database, and information such as chemical Formula, number of hydrogen bond acceptors; the number of hydrogen bond donors; the number of rotatable bonds; total polar surface area, and synthetic accessibility were obtained.

2.5.2. Drug likeness study.

OSIRIS Property Explorer Osiris property explorer software is used for the Assessment of Molecular weight, C Log P, Solubility prediction, Drug likeness, and potential Mutagenic, Tumorigenic Irritant, and Reproductive effectiveness.

2.6. Analyzing and sorting data.

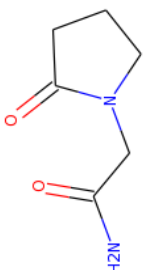
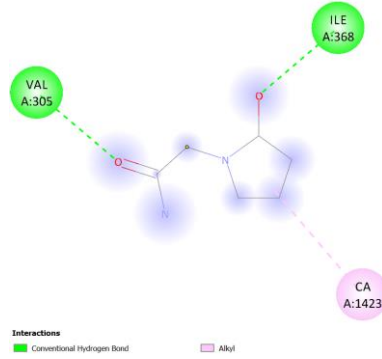
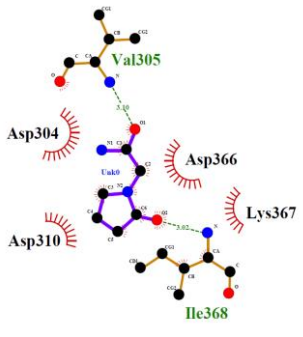
As a final step, molecular docking results and ADMET predictions were integrated, and the best ligand was introduced.

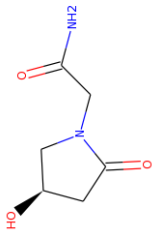
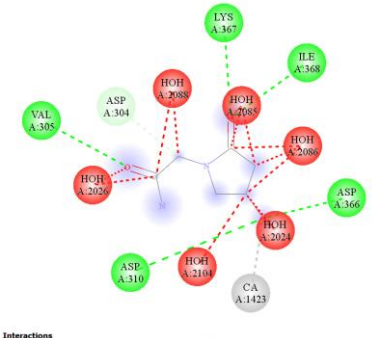
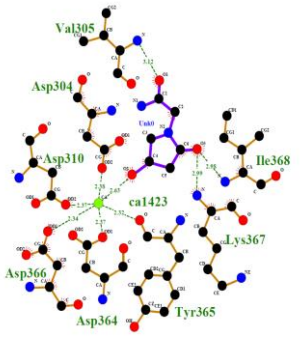
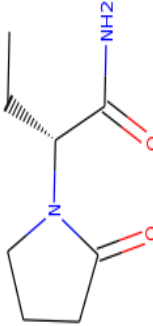
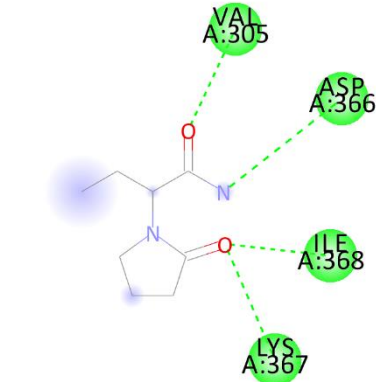
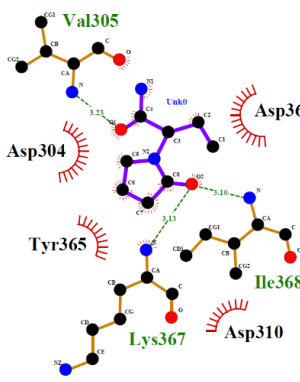
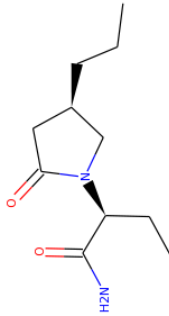
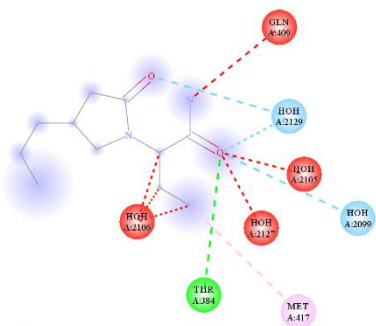
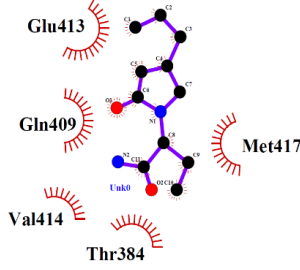
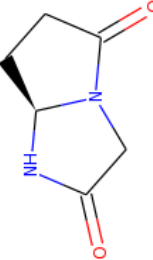
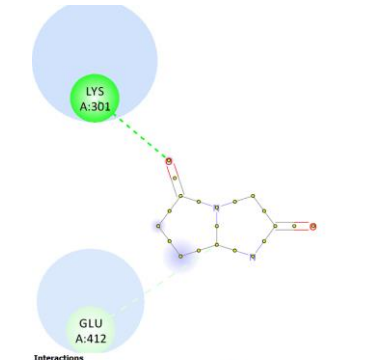
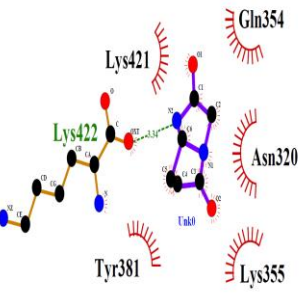
3. Results and Discussion

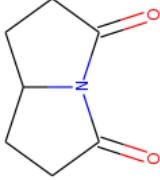
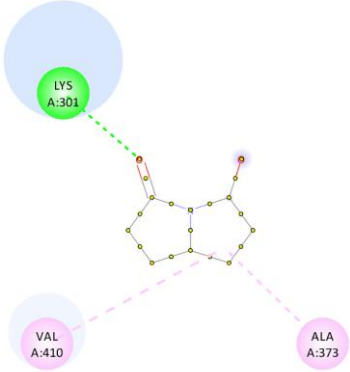
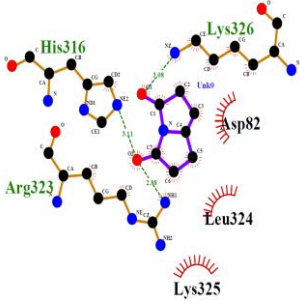
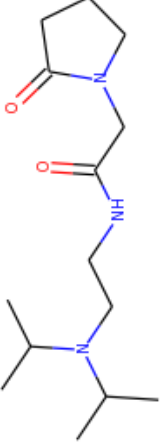
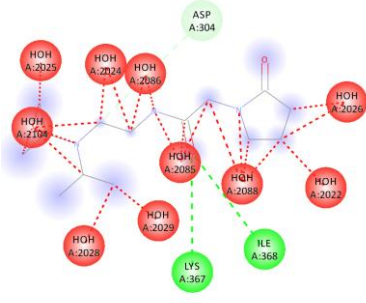
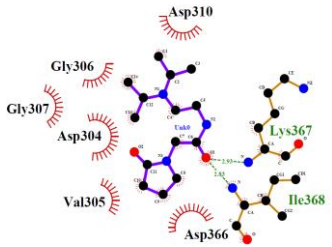
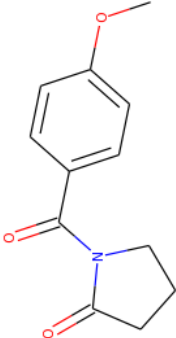
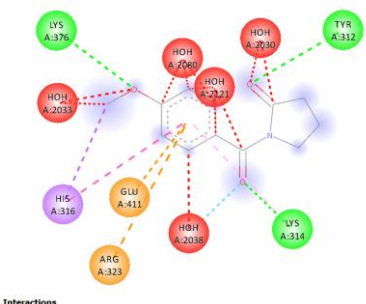
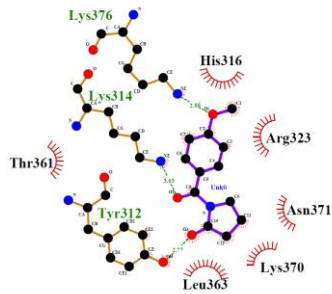
3.1. Finding ligands by some known racetams with the help of the ZINC database.

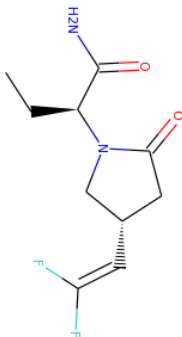
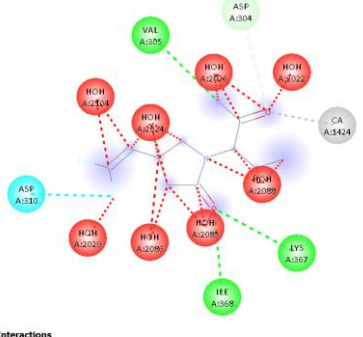
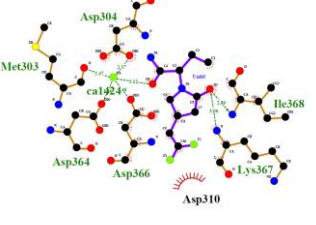
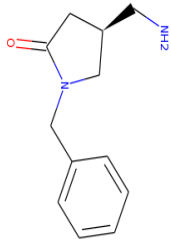
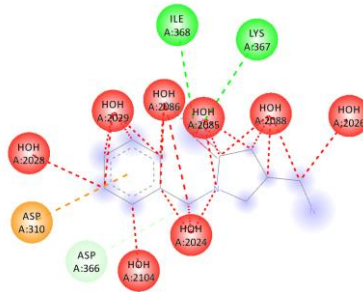
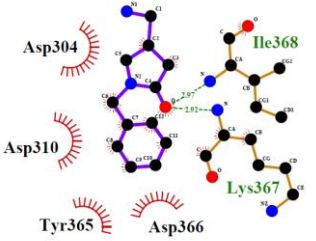
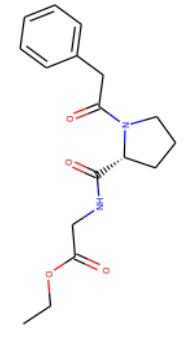
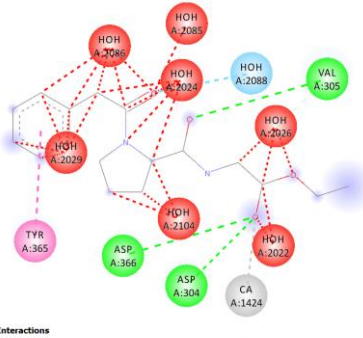
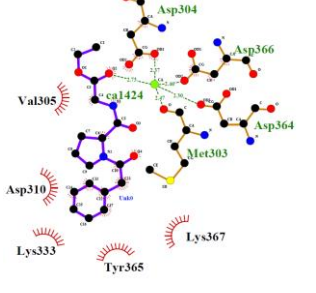
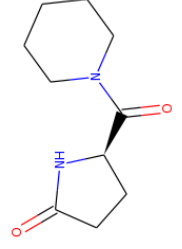
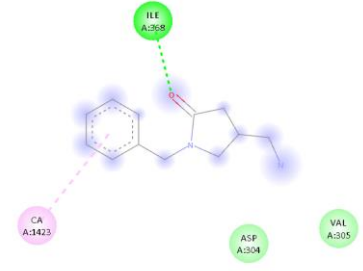

First of all, 22 known Racetams were selected, and Molecular Weight, Log P, Mol Dock Score, Re Rank Score, H bond based on Molegro Virtual Docker, and Affinity (kcal/mol) based on AutoDock Vina were calculated.

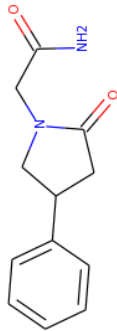
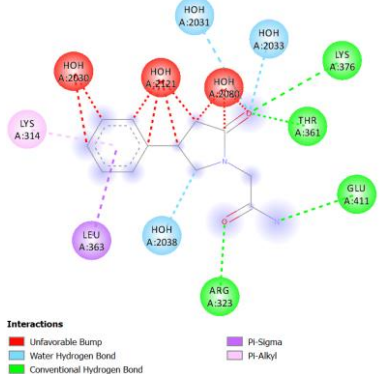
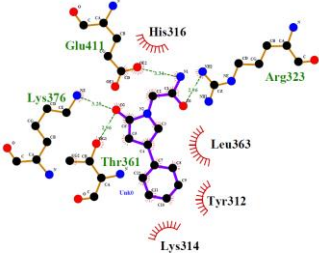
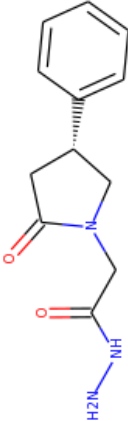
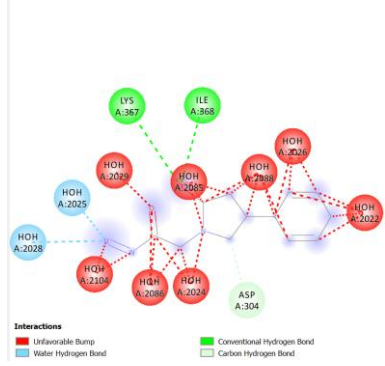
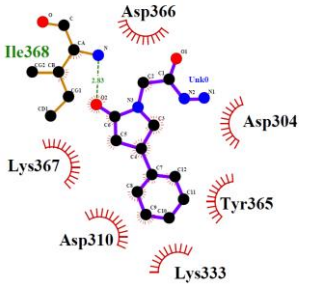
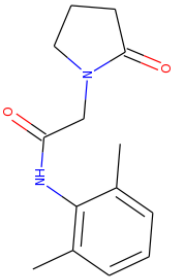
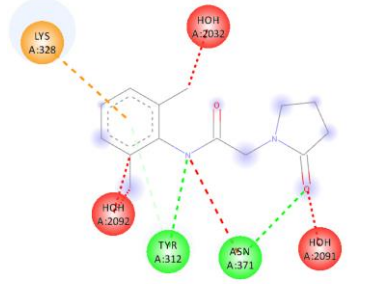
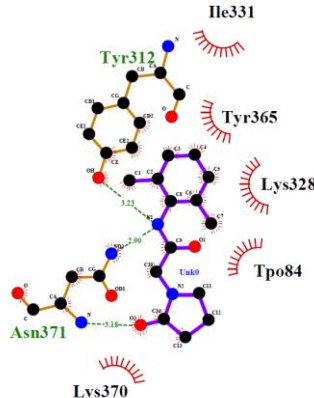
**Table 1.** Some known ligands sort by Molegro virtual docker, with 2D interaction of known Racetams with Synaptic vesicle protein 2A (SV2A), by Visual Discovery Studio and LigPlus.

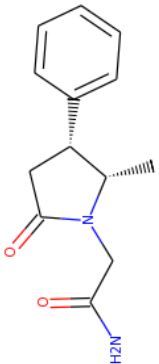
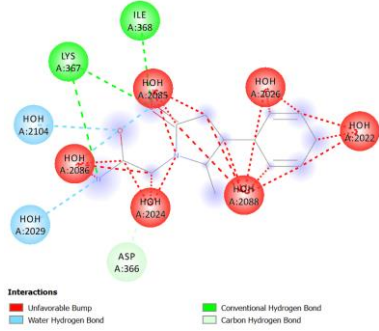
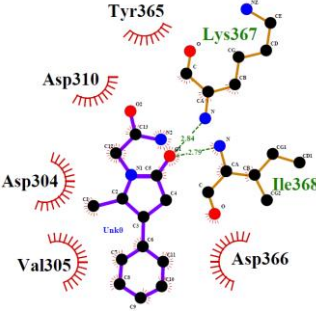
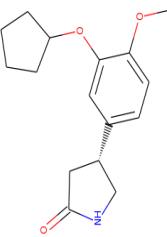
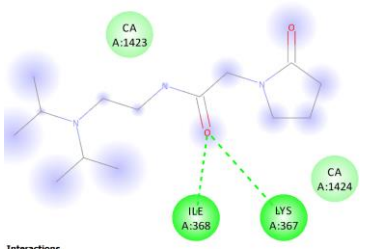
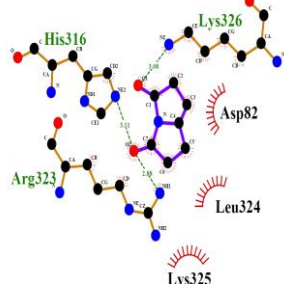
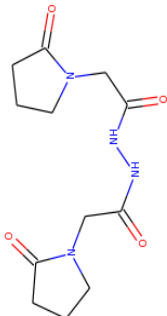
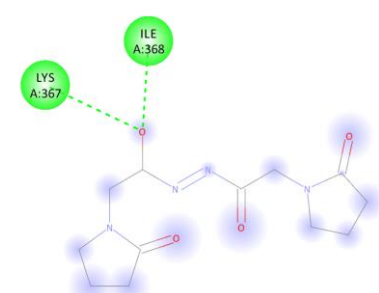
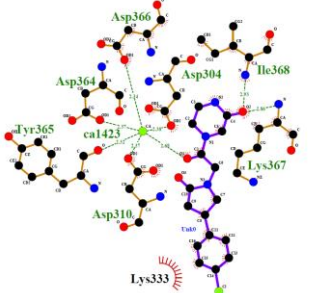
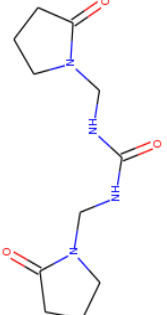
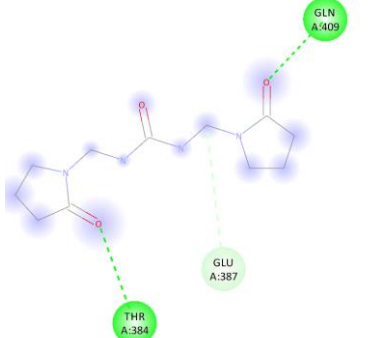
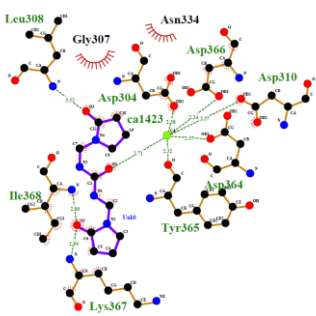
N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
1	Piracetam				-69.564 / -50.915 / -3.1311	-3.8

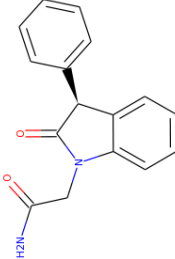
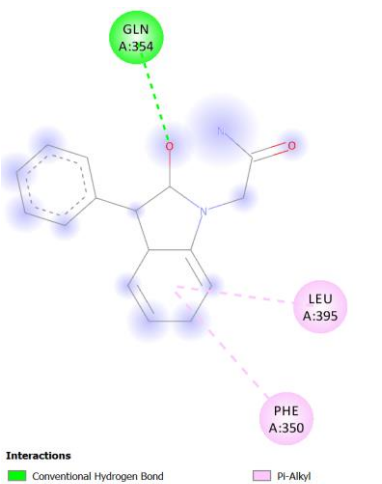
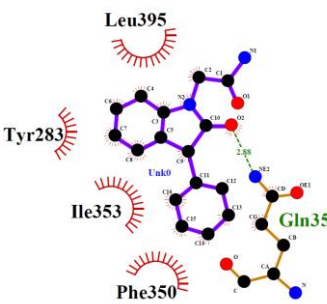
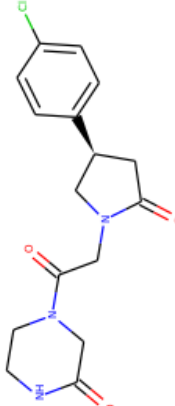
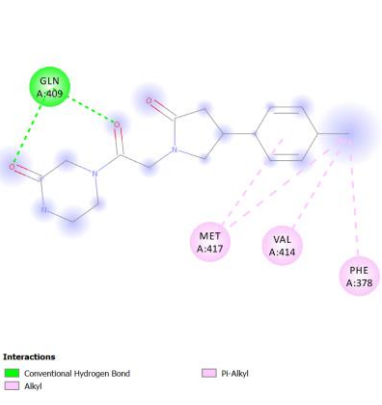
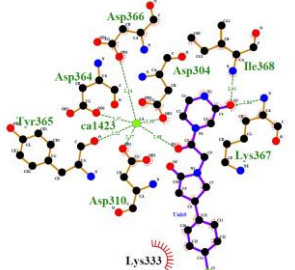
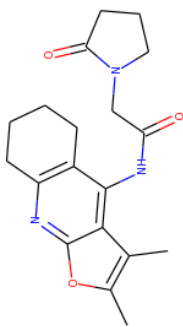
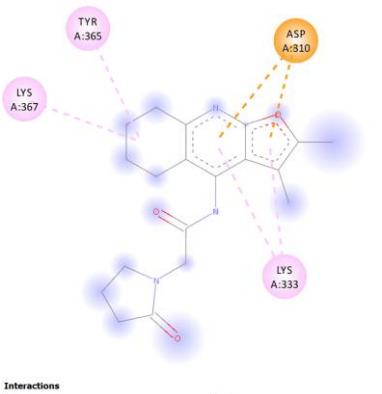
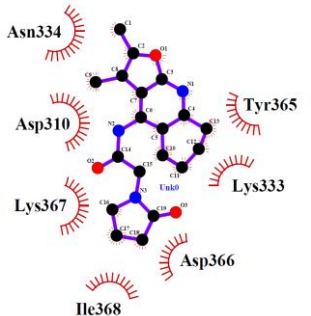
N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
2	Oxiracetam		 <p>Interactions</p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>Metal Acceptor</li> </ul>		-56.479 / -45.532 / -2.7513	-4.0
3	Levetiracetam		 <p>Interactions</p> <ul style="list-style-type: none"> <li>Conventional Hydrogen Bond</li> </ul>		-65.617 / -47.632 / -2.5563	-4.0
4	Brivaracetam		 <p>Interactions</p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Unfavorable Donor-Donor</li> <li>Alkyl</li> </ul>		-64.107 / -43.985 / -5.0486	-4.2
5	Dimiracetam		 <p>Interactions</p> <ul style="list-style-type: none"> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> </ul>		-64.516 / -52.842 / -4.8037	-4.2

N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
6	Rolziracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Conventional Hydrogen Bond</li> <li>Alkyl</li> </ul>		-74.615 / -47.825 / -2.1522	-4.7
7	Pramiracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> </ul>		-80.181 / -56.106 / -3.1919	-4.7
8	Aniracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>PI-Cation</li> <li>PI-Anion</li> <li>PI-Sigma</li> <li>PI-PI T-shaped</li> <li>PI-Alkyl</li> </ul>		-78.622 / -48.290 / -1.8793	-4.8

N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
9	Seletracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>Metal-Acceptor</li> <li>Halogen (Fluorine)</li> </ul>		-81.635 / -63.011 / -4.7802	-4.9
10	Nebracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>Pi-Anion</li> </ul>		-88.291 / -66.559 / -4.8182	-4.9
11	Ombracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>Metal-Acceptor</li> <li>PI-T-shaped</li> </ul>		-66.716 / -51.401 / -5.9137	-5.1
12	Fasoracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>van der Waals</li> <li>Conventional Hydrogen Bond</li> <li>PI-Alkyl</li> </ul>		-60.898 / -55.02 / -4.0995	-5.2

N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
13	Phenyl piracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>PI-Sigma</li> <li>PI-Alkyl</li> </ul>		-78.316 / -36.054 / -3.7065	-5.2
14	Phenyl piracetam hydrazide		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> </ul>		-79.91 / -36.118 / -3.298	-5.4
15	Nefiracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Conventional Hydrogen Bond</li> <li>Unfavorable Donor-Donor</li> <li>PI-Cation</li> <li>PI-Donor Hydrogen Bond</li> <li>PI-PI Stacked</li> </ul>		-85.618 / -14.223 / -1.6552	-5.5

N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
16	Methyl phenyl piracetam		 <p><b>Interactions</b>  <span style="color:red">■</span> Unfavorable Bump  <span style="color:blue">■</span> Water Hydrogen Bond  <span style="color:green">■</span> Conventional Hydrogen Bond  <span style="color:lightgreen">■</span> Carbon Hydrogen Bond</p>		-58.375 / -23.498 / -3.1172	-5.6
17	Rolipram		 <p><b>Interactions</b>  <span style="color:lightgreen">■</span> van der Waals  <span style="color:green">■</span> Conventional Hydrogen Bond</p>		-79.143 / -68.69 / -4.599	-5.6
18	Dupracetam		 <p><b>Interactions</b>  <span style="color:green">■</span> Conventional Hydrogen Bond</p>		-90.246 / -72.594 / -2.512	-5.6
19	Imuracetam		 <p><b>Interactions</b>  <span style="color:green">■</span> Conventional Hydrogen Bond  <span style="color:lightgreen">■</span> Carbon Hydrogen Bond</p>		-97.297 / -72.951 / -4.000	-5.7

N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
20	Doliracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Conventional Hydrogen Bond</li> <li>Alkyl</li> <li>Pi-Alkyl</li> </ul>		-64.27 / 10.054 / -1.926	-6.2
21	Cebaracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Conventional Hydrogen Bond</li> <li>Alkyl</li> <li>Pi-Alkyl</li> </ul>		-72.72 / -45.64 / -5.84	-6.3
22	Coluracetam		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Pi-Anion</li> <li>Alkyl</li> <li>Pi-Alkyl</li> </ul>		-91.557 / -65.486 / -3.9218	-6.3

All data was recovered from Online Databases such as Drug Bank Database <http://zinc15.docking.org/substances/home/> and <https://pubchem.ncbi.nlm.nih.gov/>.

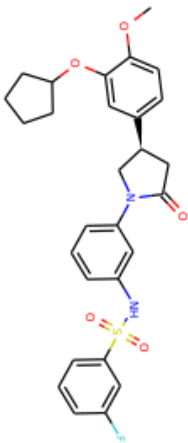
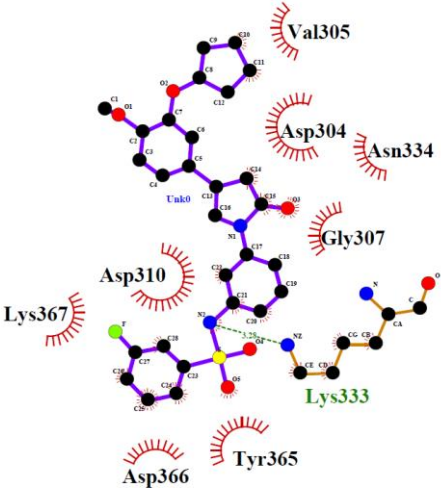
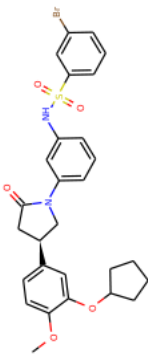
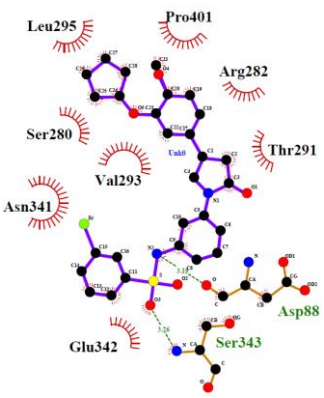
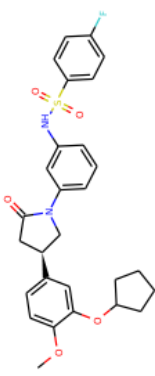
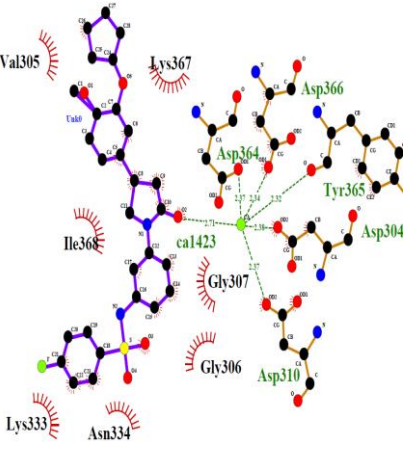
### 3.2. Finding ligands with virtual screening.

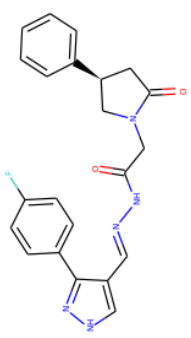
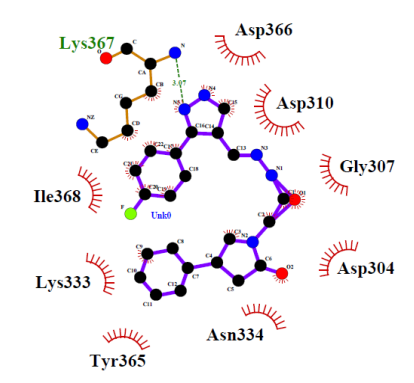
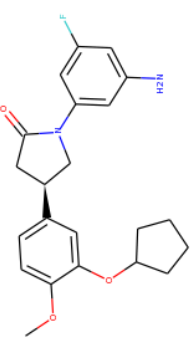
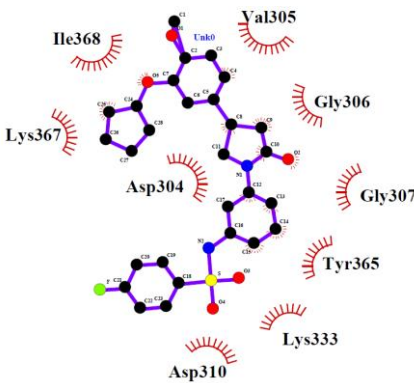
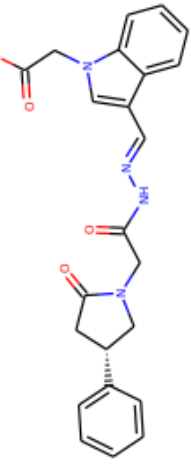
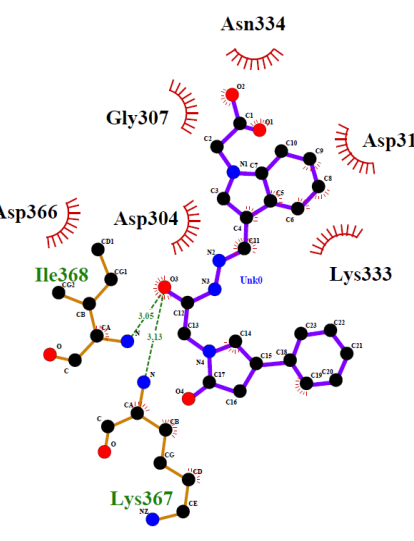
To increase accuracy and efficiency and the simultaneous use of several molecular docking mechanisms, screening of ligands was done with AutoDock Vina; 14 of the best of them were chosen, and after that, docking with Molegro Virtual Docker was done for those.

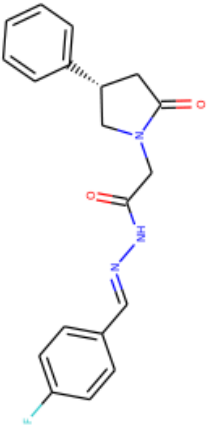
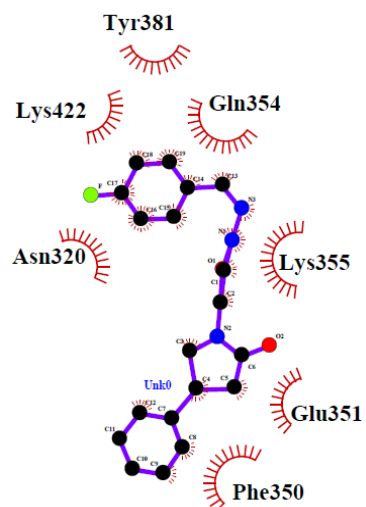
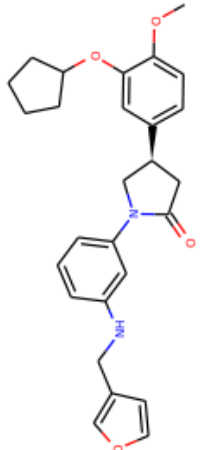
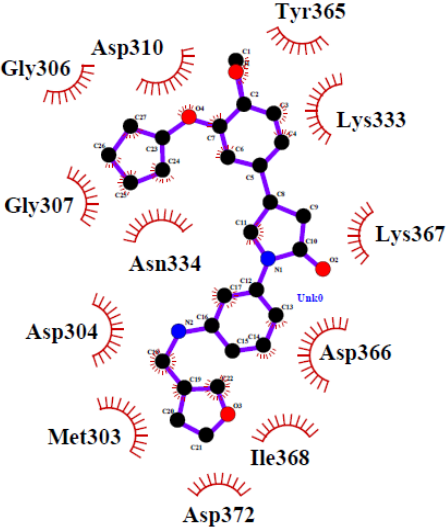
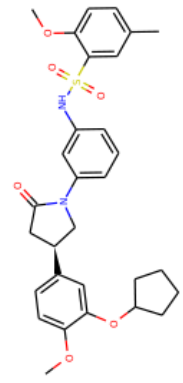
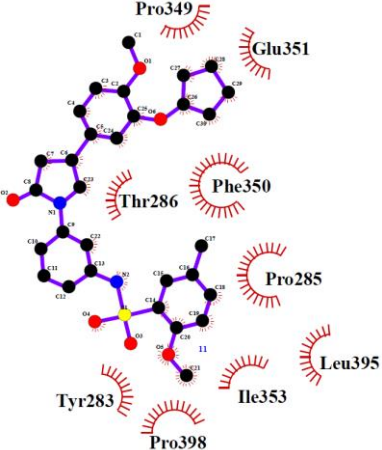
In table 1, 22 known racetam has been shown with their interaction with SV2A receptor.

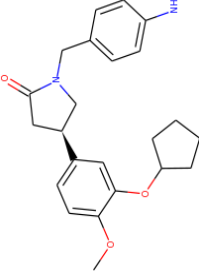
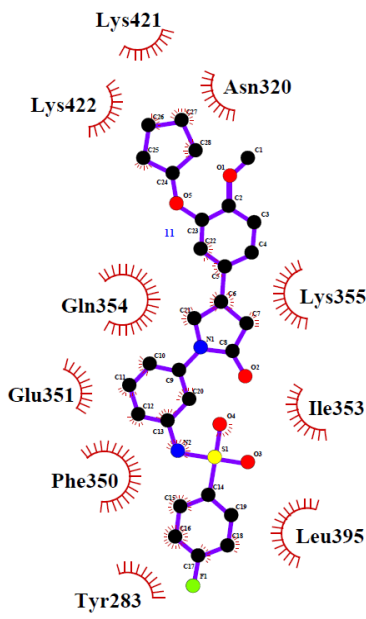
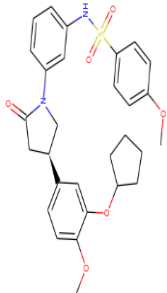
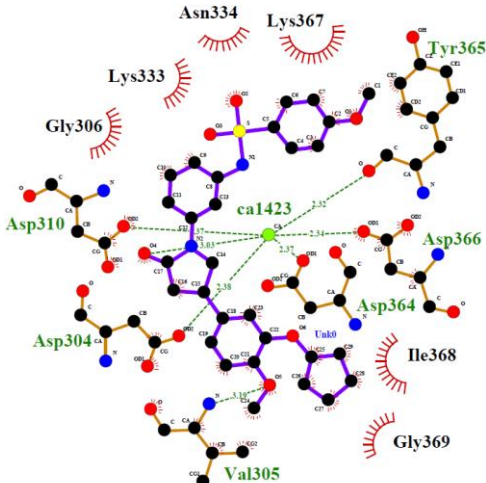
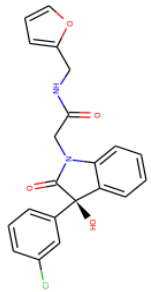
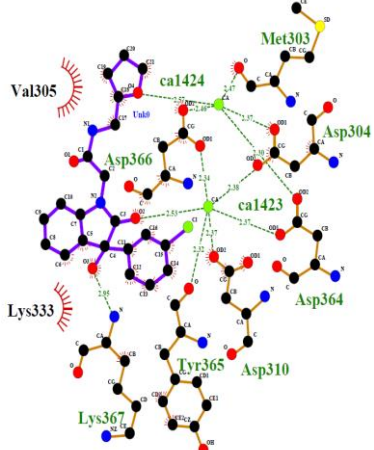
3.2.1. Molecular docking with Molegro virtual docker.

**Table 2.** Some novel racetams found by Molegro virtual docker.

N	Ligand	Structure	LigPlus	Mol Dock Score	Re Rank Score	H bond	Affinity (kcal/mol)
1	ZINC000348 20611			-130.507	-84.64	-1.73	-6.9
2	ZINC000440 32178			-129.355	-58.25	-3.96	-6.5
3	ZINC000348 20614			-126.615	-48.97	-7.30	-7.3

N	Ligand	Structure	LigPlus	Mol Dock Score	Re Rank Score	H bond	Affinity (kcal/mol)
4	ZINC000007 19283			-119.594	-80.96	-9.03	-6.3
5	ZINC000387 95668			-117.87	-76.21	-3.03	-6.8
6	ZINC000057 41149			-116.586	-21.35	-7.79	-5.5

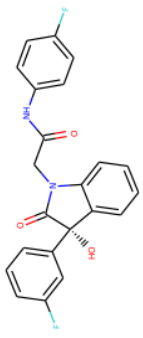
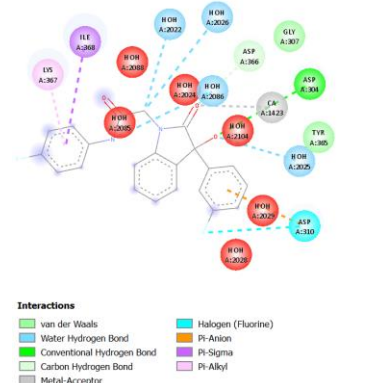
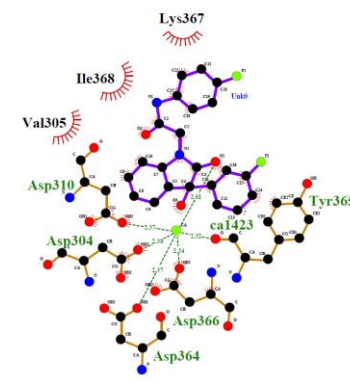
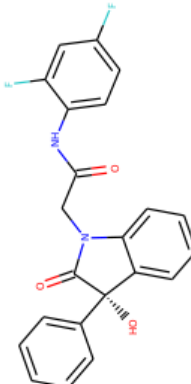
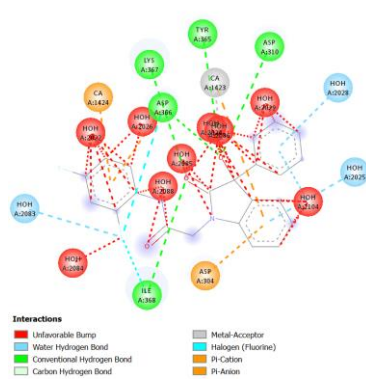
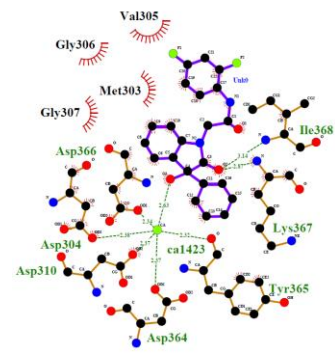
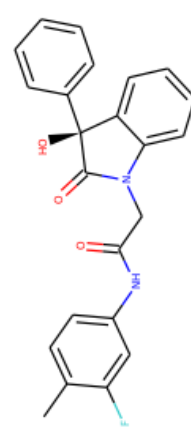
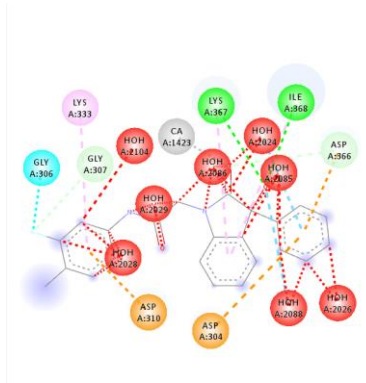
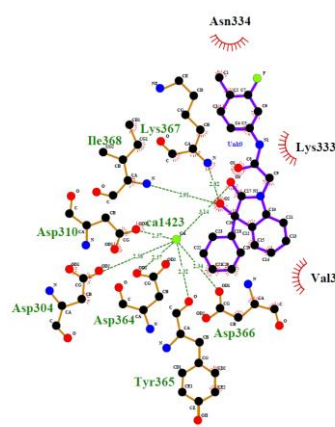
N	Ligand	Structure	LigPlus	Mol Dock Score	Re Rank Score	H bond	Affinity (kcal/mol)
7	ZINC000408972527			-115.297	-76.90	-0.39	-7.4
8	ZINC000034820597			-114.878	-76.16	-2.26	-7.1
9	ZINC000044052513			-106.409	-26.19	-4.0	-7.1

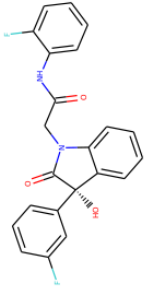
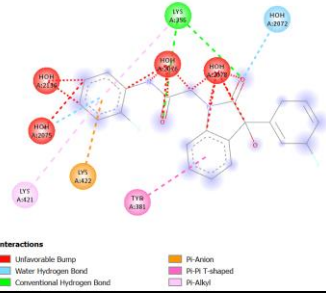
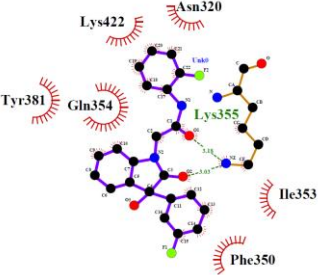
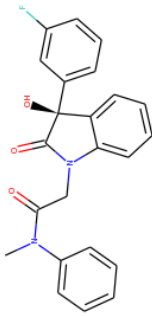
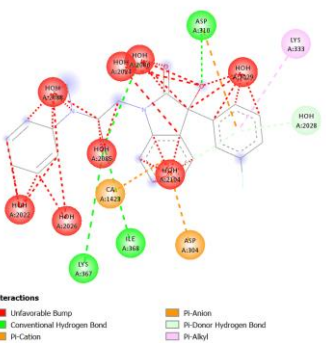
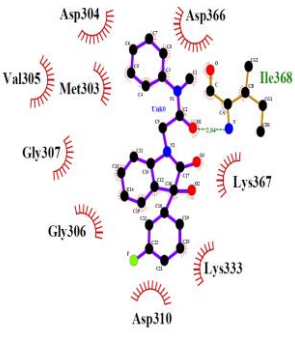
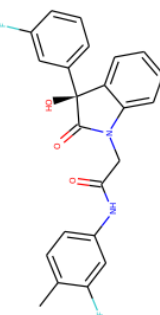
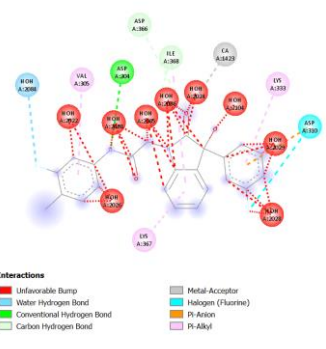
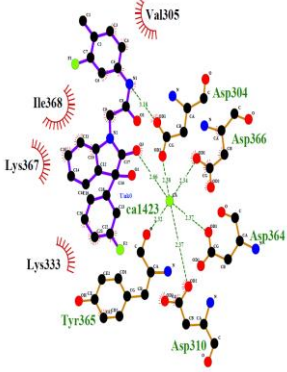
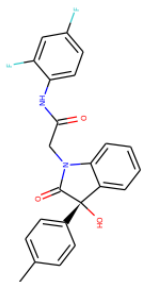
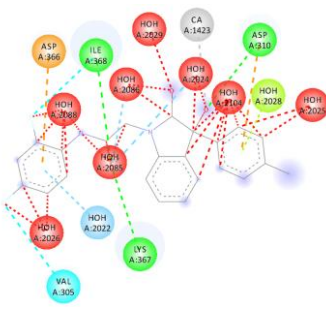
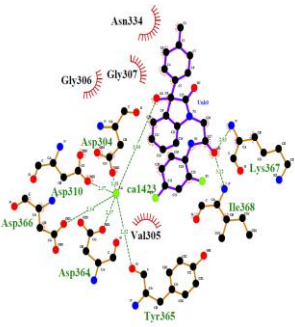
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11	ZINC000442 65623			-103.094	-33.61	-8.1	-6.5
12	ZINC000651 55459			-102.627	-80.70	-3.9	-7.3

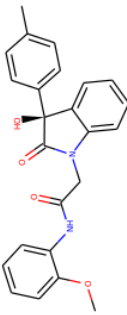
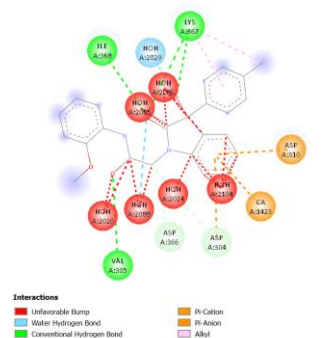
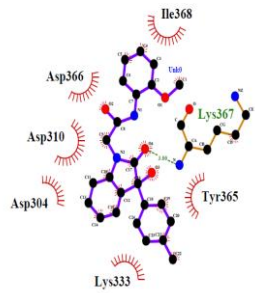
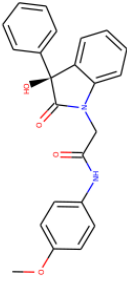
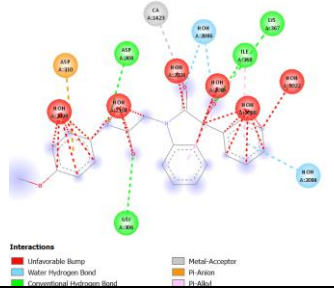
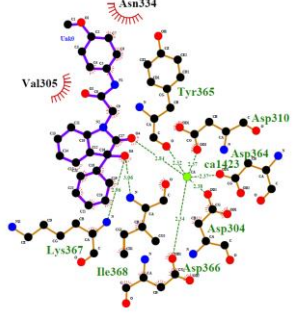
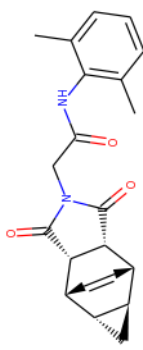
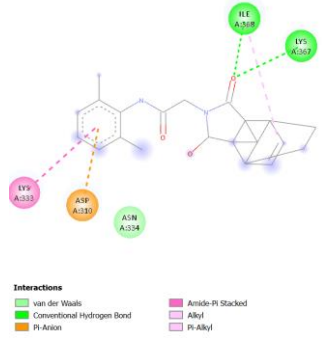
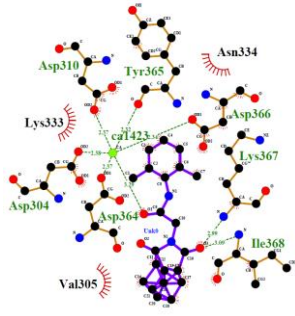
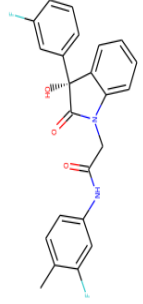
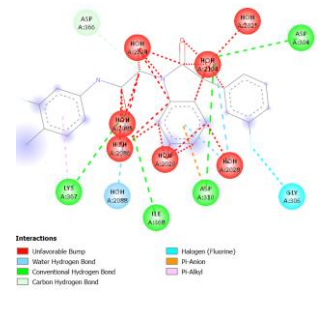
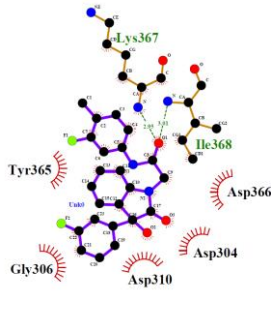
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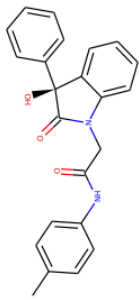
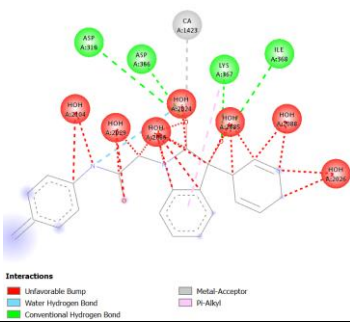
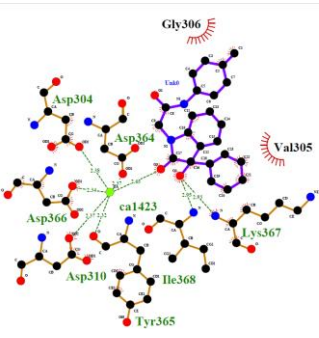
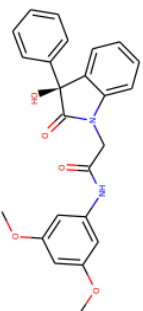
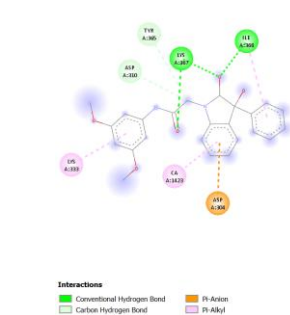
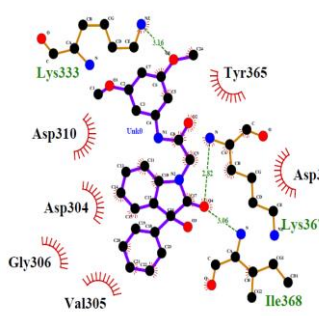
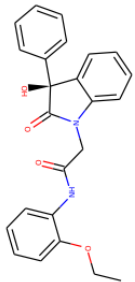
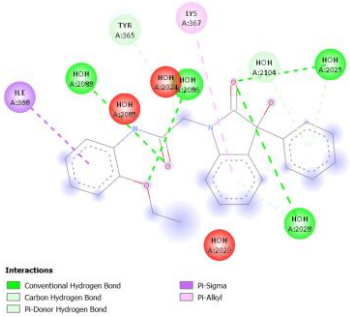
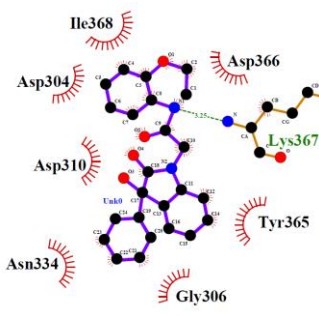
3.2.2. Virtual screening with Autodock vina 4.2.

**Table 3.** Best ligands found by Auto Dock Vina; with 2D interaction of novel Racetams with Synaptic vesicle protein 2A (SV2A), by Visual Discovery Studio and LigPlus.

N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
1	ZINC00006 5155437		 <b>Interactions</b> <ul style="list-style-type: none"> <li>van der Waals</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>Metal-Acceptor</li> <li>Halogen (Fluorine)</li> <li>Pi-Anion</li> <li>Pi-Sigma</li> <li>Pi-Alkyl</li> </ul>		-97.82 / -80.57 / -6.08	-7.4
2	ZINC00006 5155615		 <b>Interactions</b> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>Metal-Acceptor</li> <li>Halogen (Fluorine)</li> <li>Pi-Cation</li> <li>Pi-Anion</li> </ul>		-95.54 / -54.34 / -6.411	-7.4
3	ZINC00006 5155663		 <b>Interactions</b> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>Metal-Acceptor</li> <li>Halogen (Fluorine)</li> <li>Pi-Anion</li> <li>Pi-Alkyl</li> </ul>		-92.85 / -71.88 / -6.188	-7.4

N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Rank Score / H bond	Affinity (kcal/mol)
4	ZINC00006 5155386		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Pi-Anion</li> <li>Pi-Pi T-shaped</li> <li>Pi-Alkyl</li> </ul>		-88.57 / -69.45 / -4.2	-7.4
5	ZINC00006 5155328		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Pi-Cation</li> <li>Pi-Anion</li> <li>Pi-Donor Hydrogen Bond</li> <li>Pi-Alkyl</li> </ul>		-84.18 / -54.01 / -1.903	-7.3
6	ZINC00006 5155331		 <p><b>Interactions</b></p> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>Metal-Acceptor</li> <li>Halogen (Fluorine)</li> <li>Pi-Anion</li> <li>Pi-Alkyl</li> </ul>		-99.46 / -81.10 / -4.135	-7.0
7	ZINC00006 5155104				-96.91 / -66.15 / -1.124	-7.0

N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
8	ZINC00006 5155096		 <b>Interactions</b> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>PI-Cation</li> <li>PI-Anion</li> <li>Allyl</li> <li>PI-Allyl</li> </ul>		-86.18 / - 58.93 / - 4.479	-6.8
9	ZINC00006 5155705		 <b>Interactions</b> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Metal-Acceptor</li> <li>PI-Anion</li> <li>PI-Allyl</li> </ul>		-79.90 / - 62.84 / - 3.873	-6.8
10	ZINC00010 2593076		 <b>Interactions</b> <ul style="list-style-type: none"> <li>van der Waals</li> <li>Conventional Hydrogen Bond</li> <li>PI-Anion</li> <li>Amide-PI Stacked</li> <li>Allyl</li> <li>PI-Allyl</li> </ul>		+1939.92 / - 26.73 / - 6.351	-6.8
11	ZINC00006 5155329		 <b>Interactions</b> <ul style="list-style-type: none"> <li>Unfavorable Bump</li> <li>Water Hydrogen Bond</li> <li>Conventional Hydrogen Bond</li> <li>Carbon Hydrogen Bond</li> <li>Hydrogen (Fluorine)</li> <li>PI-Anion</li> <li>Allyl</li> </ul>		-95.87 / - 75.18 / - 5.385	-6.5

N	Ligand	Structure	Visual Discovery Studio	LigPlus	Mol Dock Score / Re Rank Score / H bond	Affinity (kcal/mol)
12	ZINC00006 5155745				-96.85 / - 69.47 / - 9.400	-6.4
13	ZINC00006 5155650				-95.96 / - 62.53 / - 5.424	-6.3
14	ZINC00006 5155753				-94.54 / - 77.75 / - 5.824	-6.2

All data recovered from Online Databases such as Drug Bank Database <http://zinc15.docking.org/substances/home/> and <https://pubchem.ncbi.nlm.nih.gov/>.

As shown in Table 2 and Table 3, some residual amino acids have the most contribution in the interaction of new racetams and SV2A binding sites, such as Asp 304, Val 305, Gly 306, Gly 307, Asp 310, Lys 333, Asn 334, Phe 350, Glu 351, Ile 353, Gln 354, 355, Tyr 365, Asp 366, Lys 367 and Ile 368. So, these residuals could be good options for drug discovery and finding new racetams.

### 3.3. ADMET study.

Although pharmacodynamic properties play an important role in medicine efficacy, we cannot expect medicine to have suitable effects without appropriate pharmacokinetics traits and toxicity properties. Therefore, in this study, not only ligand-protein interaction was evaluated but also Physicochemical properties, toxicity risks, and ADME prediction were estimated by *in silico* studies.

### 3.3.1. Physicochemical properties and toxicity risks.

Physicochemical properties, as well as toxicity risks of the molecules, were predicted using *OSIRIS Property Explorer* in table 4.

**Table 4.** Physicochemical properties and toxicity risks of compounds predicted by OSIRIS; Abbreviations: MW: Molecular weight; S P: Solubility prediction; D.L: Drug likeness; R E: Reproductive effective.

	Compounds	Physicochemical properties					Toxicity risks			
		M.W.	C Log P	S.P	D. L	Drug score	Mutagenic	Tumorigenic	Irritant	R. E
1	ZINC000034820611	524	4.8	-6.54	-3.45	0.17	No	No	No	No
2	ZINC000044032178	584	5.43	-7.06	-4.02	0.13	No	No	No	No
3	ZINC000034820614	524	4.8	-6.54	0.45	0.26	No	No	No	No
4	ZINC000000719283	405	2.77	-4.24	8.16	0.42	No	No	Yes	No
5	ZINC000038795668	384	3.83	-5.28	-4.17	0.17	Yes	No	No	No
6	ZINC000005741149	418	2.17	-3.24	3.03	0.46	No	No	Yes	No
7	ZINC000408972527	339	3.12	-3.89	7.3	0.46	No	No	Yes	No
8	ZINC000034820597	446	4.6	-5.93	1.8	0.39	No	No	No	No
9	ZINC000044052513	550	4.98	-6.59	1.39	0.21	No	No	No	No
10	ZINC000065155459	396	1.97	-4.23	5.7	0.73	No	No	No	No
11	ZINC000065155104	408	3.01	-4.97	3.28	0.61	No	No	No	No
12	ZINC000065155328	390	2.81	-4.29	3.76	0.7	No	No	No	No
13	ZINC000065155331	408	3.01	-4.97	0.77	0.52	No	No	No	No
14	ZINC000065155386	394	2.67	-4.63	2.81	0.66	No	No	No	No
15	ZINC000065155437	394	2.67	-4.63	3.25	0.67	No	No	No	No
16	ZINC000065155615	394	2.67	-4.63	6.94	0.68	No	No	No	No
17	ZINC000065155663	390	2.91	-4.66	4.74	0.67	No	No	No	No

### 3.3.2. ADME prediction.

ADME prediction of compounds, predicted by Swiss ADME has been shown in Table 5.

**Table 5.** ADME prediction of selected compounds, Abbreviations: ADME, absorption, distribution, metabolism, excretion; Log P, Log P (iLOGP); Log s, Log S (ESOL); TPSA, total polar surface area; SA, Synthetic accessibility.

	Compounds	TPSA	Log P	Log S	GI absorption	BBB permeant	P-gp substrate	Lipinski	S.A.
1	ZINC000034820611	93.32 Å <sup>2</sup>	3.83	-5.80	Low	No	Yes	Yes	4.08
2	ZINC000044032178	93.32 Å <sup>2</sup>	4.03	-6.55	Low	No	Yes	Yes	4.15
3	ZINC000034820614	93.32 Å <sup>2</sup>	4.10	-5.80	Low	No	Yes	Yes	4.09
4	ZINC000000719283	90.45 Å <sup>2</sup>	2.21	-3.70	High	No	Yes	Yes	3.57
5	ZINC000038795668	64.79 Å <sup>2</sup>	2.91	-4.33	High	Yes	Yes	Yes	3.37
6	ZINC000005741149	104.00 Å <sup>2</sup>	2.10	-3.48	High	No	Yes	Yes	3.63
7	ZINC000408972527	61.77 Å <sup>2</sup>	2.65	-3.33	High	Yes	No	Yes	3.18
8	ZINC000034820597	63.94 Å <sup>2</sup>	4.21	-5.29	High	Yes	Yes	Yes	4.01
9	ZINC000044052513	102.55 Å <sup>2</sup>	4.04	-6.03	Low	No	Yes	Yes	4.51
10	ZINC000065155459	82.78 Å <sup>2</sup>	2.98	-3.90	High	No	Yes	Yes	3.36
11	ZINC000065155104	69.64 Å <sup>2</sup>	3.14	-4.60	High	Yes	Yes	Yes	3.36
12	ZINC000065155328	60.85 Å <sup>2</sup>	3.27	-4.32	High	Yes	No	Yes	3.42
13	ZINC000065155331	69.64 Å <sup>2</sup>	2.67	-4.60	High	Yes	Yes	Yes	3.46
14	ZINC000065155386	69.64 Å <sup>2</sup>	2.86	-4.30	High	Yes	Yes	Yes	3.36
15	ZINC000065155437	69.64 Å <sup>2</sup>	2.54	-4.30	High	Yes	Yes	Yes	3.31
16	ZINC000065155615	69.64 Å <sup>2</sup>	2.62	-4.30	High	Yes	Yes	Yes	3.24
17	ZINC000065155663	69.64 Å <sup>2</sup>	2.81	4.44	High	Yes	Yes	Yes	3.32

## 4. Discussion

In this study, first of all, the interaction of known racetams with synaptic vesicle protein 2A, like levetiracetam and brivaracetam, was evaluated. Then, some new racetams have been identified using *in silico* methods and software such as molegro virtual docker and AutoDock tools. Furthermore, the interaction of these molecules with SV2A proteins was shown. In the next step, toxicity effects and ADME properties were estimated.

Levetiracetam, a pyrrolidone derivative, is a novel antiepileptic drug (AED) [33] that is irrelevant chemically to the first generation of AEDs Neuronal binding to synaptic vesicle protein 2A (SV2A). By binding to SV2A, levetiracetam seems to act as a synaptic vesicle exocytosis modulator [11], leading to direct inhibition of presynaptic neurotransmitter discharge in a use-dependent manner [34]. Decreasing the calcium release from intraneuronal stocks, which directs to shifts in the levels of intraneuronal calcium [35-39]; levetiracetam has not demonstrated any modulation of voltage-gated sodium and T-type calcium channels *in vitro* and *in vivo* studies [40,41].

It has revealed a differential effect of the modulator on the interaction of LEV and BRV with the SV2A protein, recommending that LEV and BRV act at different binding sites or interact with different conformational SV2A protein states [42,43]. The magnified affinity of brivaracetam for SV2A compared to levetiracetam was conducted by a better antiseizure and antiepileptogenic activity [44-47]. Brivaracetam could be an adjunctive treatment for refractory partial epilepsy in patients with epilepsy [48].

Piracetam, a modulator of the GABAergic system, has various physiological effects that may result, at least partially, from the restoration of cell membrane fluidity [49,50]. In combination with another drug, piracetam exerts promising effects on cognitive disabilities [24,50,51]. It also reported that phenylpiracetam, on the other hand, has beneficial effects for patients with cognitive deficits and depression comorbidity in encephalitis. Phenylpiracetam is more potent than piracetam and is used for a wider range of indications, and as mentioned above, highly potent Racetams could have a better effect in the case of antiseizure and other brain impairments also, phenylpiracetam exerts anti-convulsant effects in rodents [24] as well.

In a similar trial, asthenia and chronic fatigue syndrome patients treated with phenylpiracetam had better results [52]. Seletacetam is a pyrrolidone derivative with a one-log-unit higher affinity for the synaptic vesicle protein 2A (SV2A) than levetiracetam (Keppra) [53].

Dimiracetam is a nootropic drug of the racetam family, derivatives of which may have applications in the treatment of neuropathic pain. Our introduced compounds could be considered potent nootropic drugs, but more study needs to proof of this claim [54]. Unlike other racetams, nefiracetam shows a high affinity for the GABA<sub>A</sub> [55] receptor (IC<sub>50</sub>) = 8.5 nM, where it is supposed to be an agonist [56]. Aniracetam has been determined to AMPA receptor-positive modulate [57]. Coluracetam intensifies high-affinity choline uptake (HACU) [58]. Studies have shown that coluracetam improves learning impairment in rats, and similar compounds of coluracetam that have more potent than coluracetam could be considered an improver of learning impairment identified in this study. Following studies have explained that it may induce long-lasting pro-cognitive effects in cholinergic neurotoxin-treated rats by changing the choline transporter handling arrangement [59,60]. Moreover, fasoracetam could interact with all three subunits of glutamate receptors [61,62]. However, it was found in clinical trials that it does not have nootropic effects [63], which is why *in silico* and *in vitro* studies need clinical study [24,64].

Overall, this study identifies some novel compounds as potent SV2A receptor agonists to treat epilepsy or act as a nootropic agent.

## 5. Conclusions

In this study, About 2500 compounds were used with the help of the ZINC database from 22 known racetams were selected, and compounds with at least 40% Similarity were downloaded in SDF format. After selecting and downloading all Desired ligands, 2464 ligands were saved. 26 Compounds were identified based on their affinity to receptor: 16 compounds were found by AutoDock Vina, and Molegro Virtual Docker identified 12 molecules. The top 10 of them were selected, and their pharmacokinetics (such as absorption, distribution, metabolism, and elimination) and also their toxicity were evaluated. The best compound was chosen. After selecting these Chemical structures, ADME, toxicity, and other parameters related to medicines' chemical structures were evaluated, and 1 compound was identified as the best compound for *in vitro* and *in vivo* studies.

ZINC000408972527, a Phenylpiracetam derivative, has a high affinity to SV2A with BBB permeability and without mutagenicity, tumorigenicity, and irritancy effect and finally without has been P-gp substrate with a good score of Lipinski and synthetic accessibility.

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## Conflicts of Interest

The authors declare no conflict of interest.

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