Synthesis and Acivity of Chiral Amino Alcohols and Amides as New Cholinesterase Inhibitors for Alzheimer's Disease

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Abstract

A series of biological active compounds **1–14** have been synthesized and used as potential inhibitors for AChE and BuChE. Potential inhibitor efficacy of these molecules to the target enzymes have been searched in vitro and theoretical by dock and molecular dynamic calculations. The results show that chiral amino alcohol compounds **6**, **7** and **9** exhibited good value for medication scores. Among the tested compounds the best inhibition activities have been obtained with compounds **6** for AChE and BuChE by 87.68 and 92.46 % values, respectively at 50μg/mL concentration. The anticipated value of 6 also justified superb correlation with *invitro* statistics and it could be taken into consideration as drug candidate molecule for designing of novel drug.

Potential inhibitory outcomes of those molecules on the right track proteins were investigated the use of Docking and Molecular Dynamics calculations. Dock score evaluation and Lipinski parameters have been proven those ligands are ability inhibitors against applicable enzymes. Our findings endorse that related compounds can be applied as a capacity supply of anti-alzheimer active molecules for designing novel products.

Introduction

Among highly progressive neurodegenerative diseases, Alzheimer's disease (AD), that is a type of dementia frequently seen in elderly [1]. Every year, about 5 million people worldwide have been diagnosed with AD where it is expected that by 2050 this number might be raised remarkably to one among every eightyfive individuals [2,3]. The etiology of AD is enormously complex and yet not completely understood. But, there are some biomarkers which includes low ranges of acetylcholine, oxidative stress, neurofibrillary tangles (NFTs) β-amyloid (Aβ) deposits, tau-protein aggregation and dyshomeostasis of bio-metals are used for the analysis of AD [4-6].

One of the widely prevalent pathogenesis hypothesis of AD indicates that the low stage of acetylcholine in specific regions of the brain is the fundamental reason main to getting to know and reminiscence dysfunctions. The extent of acetylcholine via the use of reversible inhibitors to inhibit cholinesterases that encompass acetylcholinesterase (AChE) and butylcholinesterase (BuChE) is the primary goal for remedy of AD [7]. Beside their vascular results pain are commonly given to improve neuron-to-neuron transmission and as a consequence enhance cognitive functioning [8-10]. Presently, 4 pain inhibitors had been accepted by way of EU and US business enterprise: galantamine rivastigmine, donepezil, and tacrine (Fig. 1). These agents are vital for the palliative remedy of AD, but their clinically efficacy is confined, specially due to their poor selectivity, bioavailability and unfavorable aspect effects on peripheral worried gadget and liver.

The crystal shape of pain in complex with inhibitors disclosed that there are two binding sites, a peripheral anionic site (PAS) and catalytic energetic web site (CAS). Dual binding inhibitors bind to each binding site; which designed through assembling a heterocyclic ring to N-benzylpiperidine or to tacrine

moiety via a linker of suitable period. Heterocyclic ring interacts with the peripheral anionic web page whilst the opposite moiety binds to the gorge and the catalytic active site [11].

According to the shape characteristics of ache, numerous new compounds were synthesized as anti-ad marketers [12-16]. Currently no remedies are available to prevent, gradual, or reverse the development of the disorder procedure [17]. Available scientific therapeutics show handiest partial effectiveness in ameliorating advert symptoms and cognitive capability. Accordingly, designing an operative healing agent to treat ad is a right away and huge challenge in present time.

Frequently, the drug improvement is onerous, time consuming and overpriced manner with specifically low achievement and relatively high enfeebling costs. To aid this productivity gaps and time eating burden, multiple computational approaches consisting of drug repositioning are being utilized in recent era [18,19]. Drug repositioning technique assistances in to reduce the cost and time in drug development method due to their known efficacy and healing ability against different illnesses [20,21].

Here we record a new series of chiral β-amino alcohol and amides as cholinesterase inhibitors, which may be of exceptional significance to treat advert with potent and selective inhibitors of AChE and BuChE (Fig. 2).

Results And Discussions

Synthesis

Chiral amino alcohols and amides were synthesized and used for potential cholinesterease inhibitors. Chiral amino alcohol **1** has been prepared by regioselective ring opening of epoxides by chiral amine in a good yield as described [22]. Among the tested compounds **2, 3, 4, 10** and **5, 6, 9** have been reported previously [22,23]. Regioselective ring opening reaction of (*R*)-styrene oxide gave a new chiral amino alcohol **7** in low yield as a side product of chiral amino alcohols **9**. Bis-(*L*)-proline amide of chiral 1,2-diaminocyclohexane was prepared by using DCC to give the major product of **11** and by product **13** [24]. Compound **12** having imidazol-1,3-dion ring has been isolated by deprotection of compound **11** as a product.

Cholinesterases inhibition and structure-activity relationships

The synthesized compounds **1-14** were evaluated for their inhibitory potentials against AChE and BuChE enzymes. Compound **1** has a powerful activity against BuChE (79.83 % Inhibition) but not good for AChE at 50µg/mL concentration. Amino alcohol and phatalimid functionality of epimers **2** and **3** showed better activity against BuChE than the AChE whereas compound **3** showed no inhibition activity against AChE. This indicates that different orientations of epimers are important at recognition process of chiral macromolecules significantly. However, high selectivities against AChE rather than BuChE were obtained by *p*-phthalimidobenzenesulfonamide derivatives [7].

Chiral amino alcohols **6-8** exhibit high activity towards both AChE and BuChE. Among them amino alcohol **6** showed the highest inhibitory activity towards both AChE and BuChE with values of 87.68 and 92.46 % respectively. Whereas the activity values for galantamine are 76.08 and 57.52 %. Satisfactory activity of these compounds was attributed to their *β*-amino alcohol structures as many types of drugs such as adrenaline, noradrenaline, (+)-tembamide, etambutol, galantamine and β-blokers etc. Amino alcohol **9** exhibit somewhat lower activity than the compounds **6-8** was attributed its lower polarity. Compounds **3, 9, 11-14** displayed no activity against AChE, but high selectivity against BuChE. Regarding with BuChE results, compound **7, 8, 1,** and **9** exhibit satisfactory activity with <1, 1.69, 2.18 and 2.82 IC_{50} values respectively. However, the better inhibition activity of compounds **11-14** against BuChE has not been confirmed with respect to the IC_{50} values. Among them compounds **6** and 7 respectivelly, shows a good performance against AChE and BChE when compared with standard value.

Table 1 AChE and BuChE inhibitory activities of chiral *β*-amino alcohol and amide derivatives **1-14.**

Compound	Înhibition (%), at 50µg/mL*		IC_{50}±SEM (µg/mL)*	
	AChE	BuChE	AChE	BuChE
1	5.97±0.02	79.83±4.02	115.75±6.16	2.18±0.02
2	61.05±2.67	60.78±2.98	19.89±0.34	14.15±0.38
3	NA	75.68±3.25	>1000	11.51±0.32
4	72.75±2.89	82.51±3.67	35.01±1.47	17.98±0.28
5	NA	76.84±2.76	>1000	8.21±0.25
6	87.68±3.05	92.46±4.03	0.20±0.001	18.65±0.39
7	75.82±3.12	91.25±3.78	16.29±0.35	<1
8	35.60±1.54	81.60±3.69	93.29±3.47	1.69±0.03
9	77.50±3.39	85.67±3.72	14.53±0.23	2.82±0.04
10	39.84±1.68	62.65±3.12	62.74±3.21	34.67±1.39
11 MK-5	NA	14.13±0.65	>1000	151.56±6.32
12	NA	23.53±0.86	>1000	175.37±9.21
13	NA	15.38±0.23	>1000	140.15±4.27
14	NA	56.34±2.60	>1000	41.34±2.54
Galantamine	76.08 ± 0.39	57.52 ± 0.41	5.13±0.02	6.19±0.12

*: Data are means ± standard error of the main of triplicate independent experiments.
NA: Non active

Molecular docking experiment is the useful method to take a look at the binding compatibility of ligands in the active region of goal proteins [25]. To assess the nice drug, all of the screened tablets have been docked towards ache and BuChE one by one and complexes had been analysed on the premise of the lowest binding energy values (kcal/mol). Table 2 consequences confirmed that most of medicine exhibited similar electricity cost as compared to galantamine docking electricity value. whilst in comparison to all other screened drugs, compound 6 showed lowest energy value (-85.26 and -93.01 kcal/mol) for both ache and BuChE, which is likewise supported by using experimental outcomes.

Similarly, compound **9, 4, 10** and **7** also displayed good docking energy values (-59.28, -57.46, -57.01, -54.04 kca/mol) for AChE respectively. Compound **3-10** exhibit better activity towards BuChE with the binding energy values of -62.24 – (-93.01) kcal/mol. To better compare our docking results galantamine was docked against AChE having same parameters and determined that it possessed -71.19 kcal/mol.

Table 2 Calculated thermodynamic parameters for complexation of ligands by Docking Method.

Comp. No	AChE			BuChE		
	Electrostatic Energy	VdW	Dock Score	Electrostatic Energy	VdW	Dock Score
1	-1.46	-29.74	-31.20	-5.46	-45.68	-51.14
2	-0.79	-45.39	-46.18	-5.75	-40.25	-44.99
3	-0.22	-24.21	-34.43	-5.81	-67.52	-73.32
4	-0.85	-56.61	-57.46	-5.11	-73.52	-78.63
5	-5.79	-27.60	-33.39	-3.83	-61.96	-65.78
6	-4.23	-81.02	-85.26	-8.38	-84.63	-93.01
7	-4.37	-49.68	-54.04	-4.61	-77.81	-82.41
8	-3.53	-38.90	-42.44	-4.63	-75.40	-80.03
9	-3.35	-55.94	-59.28	-2.73	-86.07	-88.80
10	-1.85	-55.17	-57.01	-2.65	-59.60	-62.24
11	-2.59	-41.01	-43.60	-1.11	-46.36	-47.25
12	-0.60	-28.41	-29.01	-0.68	-30.29	-33.97

Binding pocket evaluation of ache and BuChE

The ache binding pocket identity was confirmed by using protein crystal data form PDB (2X8B) (www.rcsb.org) [26]. This co-crystal form turned into used because the fasciculin-II became bound inside lively vicinity of goal protein which enables in identity of binding pocket residues. Moreover, giant ligand

conformation with respect to its goal molecule facilitates in comparisons with our selected ligands by using superimposition. The active web page of this enzyme is buried inside an arrow gorge of 20 Å deep, which lets in a couple of enzyme-substrate interplay thereby facilitating the formation of the transition state of pain [27,28]. Figure 3 confirmed the binding pocket and focused residues worried in fasciculin-II binding interaction. here eight residues such as Glu202, Tyr72, Tyr124, Phe338, Trp286, Tyr341, Tyr337, and Trp86are probably the counterpart of binding site. The drugs having ability to binds within this lively vicinity and interact with those residues can also remember as an amazing candidate against pain.

Power of complexation observed in range from -23.66 kcal/mol to -85.26 kcal/mol respectively. The calculated interplay of compound 6 with the binding site of ache displayed that entrenched in a awesome institution of amino acid with aromatic ring which includes Trp 86. The compound become dock with pain with its energetic site available whereas ligand placed parallel to Trp 86 constitutes π-π stacking (Fig. 3).

Strong hydrogen bonding consist via -OH group of compound 6 with -OH group of Ser203 and other hydrogen bonds also formed with -OH group of Gly120, Gly122, Ser229. Similarly molecule 7 and 9 also making hydrogen bonding via -OH group with Thr 83, Trp 86 Glu 202 with the active site of AChE (Fig. 3). Similarly molecules also showed strong hydrogen bonding interaction with Trp86, Ala 204, Ala 206, Gly120 and of AChE via -OH group with binding site residues.

BuChE binding pocket identification turned into showed by means of protein crystal data form PDB (4BBZ) [29]. This co-crystal structure was used because the CBDP was bound inside binding region of goal protein. Significant ligand conformation with appreciate to its goal molecule facilitates in comparisons with our decided on compounds by superimposition. The active sites are frequently vast components of the purposeful characterization of enzymes. Furthermore, in molecular docking, the binding of ligands within the binding pocket of target protein can predict proper effects compared to binding of different protein websites. Fig. 4 confirmed the binding pocket and centered residues related with CBDP binding interaction. In commonly, seven residues together with Thr84, Trp231, Gly117, Trp82, His438, Phe118, and Gly116 are possibly the counterpart of binding site. The drugs having potential to binds inside this active area and interact with these residues may contemplate as a very good candidate against BuChE.

The molecular docking investigation on BuChE active site indicate that well created polar interactions and hydrogen bonding. Observed energy and complexlation range are from -33.97 kcal/mol to -93.01 kcal/mol, respectively. When compared AChE with BuChE different type of binding interaction have been obtained due to the difference of amino acid chain in active site of BuChE (Table 2). Molecular docking results indicate some important interactions of aromatic π-π stacking occurred between Trp82, Trp231, , Phe118 and hydrogen bonding with His 438, Gly116, Thr 284, and Gly117 (Fig. 4).

Bio-chemical properties and Lipinski's parameters

The potential inhibitor effect of compounds **1–14** has also been investigated by using calculated Lipinski parameter which was thought as an important physicochemical parameter to evaluate the ability to cross blood-brain barrier as shown in Table 3. It was reported that logP with the optimum central nervous system penetration was around 2±0.7 [7]. The results indicate that the most of the log P values of synthesized ligands ranged from 1.72–6.46, indicate that all the ligannds are sufficiently lipophilic to pass the blood-brain barrier. Moreover, the ligans should possess no greater than 10 nON and 5 nOHNH, respectively. Literature data exposed that the exceed values of nON and nOHNH results in poor permeation [30].

The hydrogen-bonding potential has been taken into consideration as giant parameter for drug permeability. Our results justified that the all synthesized compounds own < 10 nON and < five nOHNH values which were similar with popular values. of the most compounds had been also having appropriate log P values (<5).

The number of rotatable bonds inside chemical structure is likewise an big topological parameter to measure the molecular flexibility. Earlier research confirmed that extended quantity of rotatable bonds (≥10) has been related to bad oral bioavailability, specifically whilst associated with a high polar floor area (Studio Discovery, 2008). Our anticipated results justified the most of compounds exhibited < 10 rotatable bonds which ensured the importance of our chemicals have some drug like behaviour.

Polar surface areas also are considered as appropriate descriptor for characterizing the drug absorption, such as intestinal absorption, bioavailability and blood-brain barrier penetration. Our predicted consequences showed that most of compounds possess < 78.95, whereas compound 10 and 11 possessed >100˚A2 values. The eager residences are hydrophobicity, digital distribution, hydrogen bonding characteristics, molecule size and flexibility and of course presence of various pharmacophoric functions affect the behavior of molecule in a dwelling organism, consisting of bioavailability, delivery houses, affinity to proteins, reactivity, toxicity, metabolic balance.

Table 3 Complexation compounds of Calculated Lipinski parameters **1-14** by Molinspiration method.

Comp. No	miLogP	Mw	nON	nOHNH	nRotb	PSA
1	3,53	247.38	2	2	5	32.26
2	3.28	330.43	5	2	6	71.33
3	3.28	330.43	5	2	6	71.33
4	4.27	414.50	5	1	8	62.54
5	4.02	410.60	4	2	10	46.93
6	6.63	594.80	6	2	12	65.40
7	6.46	534.74	4	2	12	46.93
8	3.06	348.45	4	0	4	40.62
9	6.46	534.74	4	2	12	46.93
10	5.67	672.78	10	2	12	127.48
11	2.82	508.66	10	2	8	117.28
12	1.72	222.29	4	0	1	40.62
13	5.13	421.58	7	1	5	78.95
14	4.28	340.50	3	0	3	30.94
Galantamine	1.74	275.35	4	1	1	41.93

LogP: Octanol/water partition coefficient, Mw: Molecular weight, nON: Number of hydrogen bond acceptors, nOHNH: Number of hydrogen bond donors, nRotb: Number of rotatable Bonds. [31]

Materials And Methods

General Information

All chemical compounds have been reagent grade until in any other case certain. five,five-dithiobis- (2-nitrobenzoic acid) (DTNB), acetylcholinesterase, butylcholinesterase, galantamine hydrobromide, were obtained from Sigma-Aldrich (Steinheim, Germany). Acetylthiocholine iodide reagent was from Applichem (Germany). Butylcholine iodide (Fluka), sodium carbonate, ammonium acetate, sodium hydrogen phosphate and sodium dihydrogen phosphate had been bought from Riedel-de-Haen (Germany). (R)-styrene oxide and (S)-propylene oxide and (S)-glycidyl phenyl ether had been purchased from Sigma-Aldrich. Silica Gel 60 (Merck, 0.040–0.063 mm) and silica gel/TLC-playing cards (F254) had been used for column chromatography and TLC. All reactions were completed under an N2 environment with a dry solvent beneath anhydrous conditions, until in any other case stated. Melting factors have been decided with Gallenkamp model equipment with open capillaries. Optical rotations had been taken on a Perkin Elmer 341 model polarimeter. IR spectra have been recorded on Mattson a thousand ATI Unicam toes-IR spectrophotometer. 1H NMR (400 MHz) and 13C NMR (100 MHz) spectra were recorded on Bruker Avance-400 MHz high overall performance virtual ft-NMR spectrometer, with tetramethylsilane because the inner popular solutions in deuteriochloroform. Elemental analyses have been received with CARLO-ERBA model 1108 device.

Synthesis

Compounds **5, 6, 9** and **2-4**, **10** have been synthesized as our previous reports respectively [22, 23].

(*R*)-1-Phenyl-2-[(*R*)-α-cyclohexylethyl)amine]ethanol (1): Compounds **1** have been synthesized using a procedure reported [23]. (***R***)-α-cyclohexylethylamine (10 mmol, 1.21 g (1.2 equiv.)) in 5 mL *i*-propanol was added to the solution of (*R*)-styrene oxide (990 mg, 8.33 mmol) in 5 mL of *i*-propanol at in an ice bath and stirred for 1 h. then refluxed for 12 hours. The reaction monitored by TLC using H:EA:TEA (4:1:0.1). Solvent was evaporated then the crude product was purified by crystallization from *n*-hexane to give 1.48 g (72 %) of pure product as a white crystal. M.p.: 72-73 ^{o}C. ^{1}H NMR($CDCl_3$): 7.41-7.28(m, 5H), 4.78-4.74(dd, 4Hz, 1H), 2.93-2.89(m, 1H), 2.722.64(m, 1H); 2.51-2.47(p, 1H); 1.79-1.68(m, 5H), 1.34-0.9(m, 10H). ^{13}C NMR ($CDCl_3$): 142.84, 127.55, 127.35, 125.82, 71.55, 57.28, 54.60, 43.22, 29.71, 28.25, 26.75, 17.05. Anal. Calcd. for $C_{16}H_{25}NO$ C: 77.73, H: 10.12, N: 5.67. Found: C: 77.94, H: 9.79, N: 6.03.

(*1R,2R*)-Dibenzyl-bis[(*R*)-2-hydroxy(1-phenyl)ethyl]-1,2-diaminocyclohexane (7): This (*R*)-Styrene oxide (1.63 g, 13.6 mmol) was added to a solution of (*R,R*)-*N,N*′-dibenzyl-1,2-diaminocyclohexane (2 g, 6.8 mmol) in methanol (3 mL) and stirred at 40, 50 and 60^{o}C, 24 h for each temperature. Solvent was evaporated and then unreacted epoxide and amine was removed by Kugelrohr distillation apparatus. Crude product was purified by column chromatograph using petroleum ether (60-80): ethylacetate: triethyl amine (85/10/5) to give **7** as a regioisomer of compound 10 (0.87 g, 24 %) as a colourless solid. M.p.: 80-81 ^{o}C; ^{1}H (400 MHz, $CDCl_3$) 7.56-7.25 (20H, m), 4.66-4.63(d, 9.6 Hz, 2H); 4.15(bs, 3H); 3.749-3.715(d, 13.6Hz, 2H); 3.6(bs, 2H); 2.64-2.61(d, 13.6Hz, 3H); 1.63- 1.14(m, 8H). ^{13}C (100 MHz, $CDCl_3$): 142.18, 130.42, 130.03, 129.77, 129.55, 129.02, 128.85, 127.35, 126.01, 71.09, 57.44, 48.00, 42.97, 27.50, 25.29, 25.03. Anal. Cald. for $C_{36}H_{42}N_2O_2$: C, 80.89; H, 7.86; N, 5.24, Found: C, 80.25; H, 7.46; N, 5.30.

***N,N*′-Bis-[(*L*)-*N*-Boc-proline]-(*1R,2R*)-1,2-cyclohexanediamide (11) :** To an ice cold (*S*)-*N*-(*tert*-butyloxycarbonyl)proline (3.77g, 17.54 mmol), (1*R*,2*R*)-1,2-diaminocyclohexane (1g, 8.77 mole) in dicholorometane (20 mL) a solution of dicyclohexylcarbodiimide (3.7g, 18 mmole) in 10 mL DCM was added dropwise. The resulting mixture was stirred and the temperature of the reaction mixture was allowed to raise the room temperature. The progress of the reaction was monitored by TLC *n*-hexane:ethyl acetate (R_f 0.25). After completition of reaction dicyclohexylurea and solvent were separated by filtration and evaporation respectively. Remain mixture was separated by column chromatograph by using *n*-hexane:ethyl acetate (3:1) with silica gel packed column to give 2.8 g (62.85 %) of compound **11** (R_f=0.25) as a white solid. M.p.: 87-88 ^{o}C. $[\alpha]_D^{24}$= -55.5 (c=1, MeOH). ^{1}H NMR ($CDCl_3$): 6.67(m, 1H, -C=ONH-), 4.136 (bs, 1H), 3.56(bs, 1H); 3.42(m, 2H, -CH2-); 1.99(m, 3H, -CH2-); 1.76(m, 3H, 1.41(s, 9H,

$(CH_3)_3C$-) ; 1.27-1.17(m, 2H). ^{13}C NMR($CDCl_3$): 172.98, 154.63, 80.36, 61.54, 53.22, 47.07, 32.45, 31.14, 28.35, 24.32, 23.83. Anal. Calcd for $C_{26}H_{44}N_4O_6$ C: 61.42, H: 8.66, N: 11.02. Found: C: 60.94, H: 8.39, N: 11.18.

Cyclohexyltetrahydro-1*H*-Pyrolo-[1,2-c]imidazole-1,3-(2*H*)-dione, (12): Compound **12** is a by product of compound **13.** Thus compound **13** (1.12g, 2.56 mmol) was deprotected by using trifloroacetic acid (2.55 mL) in DCM (3 mL) according to the literature [29]. Solvent evaporated and remain mixture was separated by column chromatograph by using *n*-hexane: ethyl acetate (4:1) with silica gel packed column to give 250 mg of compound **12** (R_f = 0.2) as a white solid. M.p.: 102-104 ^oC. $[\alpha]_D^{22}$= -62. 0 (c=1, MeOH) for compound **12**. ^1H NMR($CDCl_3$): 6.67(m, 1H, -C=ONH-), 4.136 (bs, 1H), 3.56(bs, 1H); 3.42(m, 2H, -CH_2-); 1.99(bm, 3H, -CH_2-); 1.76(bm, 3H, 1.41(s, 9H, $(CH_3)_3C$-) ; 1.27-1.17(m, 2H). ^{13}C NMR($CDCl_3$): 172.98, 154.63, 80.36, 61.54, 53.22, 47.07, 32.45, 31.14, 28.35, 24.32, 23.83 Anal. Calcd for $C_{12}H_{18}N_2O_2$ C: 64.86, H: 8.11, N: 12.61. Found: C: 65.12, H: 8.03, N: 12.87.

(*S*)-1-Pyrrolidinecarboxylicacid,2-[[cyclohexyl[(cyclohexylamino)-carbonyl]-amino]carbonyl]-1,1-dimethylethyl ester (13): 1.2 g (% 32.78) of compound **13** (*Rf* = 0.7)

was obtained as a by product of compound **11** as above description. M.p.: 73-74 ^oC. $[\alpha]_D^{24}$= +11.9 (c=1, MeOH) for compound **13**. ^1H NMR ($CDCl_3$): 8.08-8.06(d, 8Hz, 1H); 4.49-4.47(m, 1H); 4.46-4.45(m, 1H); 3.57-3.55(m, 1H); 3.54-3.52(m, 1H); 3.41-3.40(m, 1H); 1.97-1.60(m, 18H); 1.45(s, 9H); 1.37-1.11(m, 6H). ^{13}C NMR ($CDCl_3$): 171.6, 154.9, 154.2, 58.43, 54.44, 49.99, 47.45, 32.67, 31.79, 31.62, 31.28, 29.12, 28.49, 26.12, 25.95, 25.57, 25.40, 24.57. Anal. Calcd. for $C_{23}H_{39}N_3O_4$: C: 65.55, H: 9.26, N: 9.97. Found: C: 64.96, H: 9.32, N: 9.83.

(*R,R*)-Methoxy-{*N,N',N''*-1,2',3-[(1-phenylethanolato)-(2-phenylethanolato)propyl]amino}silane (14): 3-Aminopropyltrimetoxysilan (0.755g, 5 mmol) and (*R*)-phenyloxyrane (1.3g, 10.5 mmol) refluxed in methanol for 24 h. Crude product was purified by column chromatograph using H:EA (6:1) after evaporation of solvent. M.p.: 178-149 ^oC. IR (cm^{-1}): 3033, 2914, 2874, 2821, 1490, 1452, 1268, 1205, 1124, 1077, 937, 811, 756, 639. ^1H NMR ($CDCl_3$): 7.47-7.28(m, 10H); 4.99-4.91(m, 2H); 3.60(s, 3H); 3.58-3.12(m,1H); 3.0-2.9(m, 2H); 2.67-2.49(m, 3H); 1.84-1.67(m, 2H); 1.06-1.02(m,1H); 0.76-0.71(m, 1H). ^{13}C NMR ($CDCl_3$): 141.66, 141.57, 128.44, 128.39, 127.55, 125.42, 125.30, 70.76, 70.16, 60.41, 60.15, 52.87, 50.17, 20.69, 8.14. Anal. Calculated for $C_{20}H_{37}NO_3Si$: C: 65.50, H: 10.08, N: 3.82. Found: C: 64.96, H: 9.78, N: 3.69.

Anticholinesterase inhibition activity

All samples were dissolved in ethanol to prepare their stock solution at 100 and 1000 g/mL concentration. Aliquots of 140, 150, 155, 158 µL, of 100 mM sodium phosphate buffer (pH 8.0), 20, 10, 5, 2 µL of sample solution (10, 100 and 1000 µg/mL) and 20 µL BuChE (or AChE) solution were mixed and incubated for 15 min at 25 ºC, and DTNB (10 µL) is added. The reaction was then initiated by the addition of butyrylthiocholine iodide (or acetylthiocholine iodide) (10 µL). Final concentration of the tested solutions was 100, 50, 25, 10, 5, 2.5, 1, 0.5, 0.25 and 0.1 µg/mL [24]. The hydrolysis of these substrates were monitored using a BioTek Power Wave XS at 412 nm.

Computational methodology

Resumption of goal proteins from PDB the 3D structure of AcHE and BuChE with PDB IDs 2X8B and 4BBZ were accessed from Protein information bank (PDB) (www.rcsb.org). Crystallographic water molecules were eliminated from all of the systems and the lacking coordinates of the atoms were modelled the usage of xLeAP and an ff99SB force field. Atoms on proteins were assigned the PARM99 expenses, and all ionizable residues were set at their default protonation states at impartial pH. All structures were in addition processed with the aid of the xLeAP module of AMBER. The molecular structures were neutralized through the addition of counterions. the chosen goal proteins were minimized with Amber force field [32-35] by employing conjugate gradient algorithm in UCSF Chimera 1.10.1 [36]. The discovery Studio 4.0 a visualizing tool became used to generate the hydrophobicity graphs and graphical depiction of target proteins. The protein Ramachandran graph become accessed thru PDB. the discovery Studio 2.1 customer [37] become used to view 3D conformation of goal proteins.

Docking Studies

Dock 6.5 [38] module permits all tiers of a docking procedure to be done with the technology of ligand conformations, ligand docking, and the scoring of the binding modes. As in this situation, in which a inflexible receptor approximation changed into used, it is predicted that one hundred thirty the extraordinary receptors taken into consideration will result in one-of-a-kind ligand binding modes relying at the preliminary size of the enzyme-binding hollow space. for that reason, 14 designed molecules were docked onto available receptors following a multistep technique. To be able to describe receptor-binding residences, a grid of potential strength became calculated for atoms taking part within the binding pocket. these atoms had been acquired from the evaluation of each protein-ligand complicated. on this step, default parameters have been used. The ligand changed into then docked using the calculated grid to vicinity it into the binding site and score the proposed binding mode.

Conclusions

In the present work, a series of new chiral amino alcohols were designed and synthesized. Their AChE and BuChE inhibition activity were evaluated in vitro and molecular docking studies. Promising inhibitors were revealed to be compounds 6, 7, 9 which were further progressed to in silico studies using molecular docking study. According to our findings, all of the compounds tested could be good precursor models for BuChE-inhibiting molecules and among them 6,7 and 9 in particular could be more promising since it can display dual inhibition on AChE and BuChE enzymes effectively. Furthermore, moleculer modeling data well-matched with the in vitro outcomes of this study. As a conclusion designed and synthesis these specific compounds have potential inhibitör effects of AChE and BuChE enzymes.

Declarations

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Author contribution All authors equally contributed to this manuscript.

Code availability The calculations have been carried out using AMBER 11

provided by AMBER Software Company, Dock 6.0 have been used publicly available source code which are freely available on the internet.

Declarations

Conflict of interest The authors declare no competing interests.

Data availability We confirm the availability of all the data and materials in this manuscript.

References

1. Ferri CP, Prince M, Brayne C, Brodaty H, Fratiglioni L, Ganguli M, Hall K, Hasegawa K, Hendrie H, Huang Y (2006) Global prevalence of dementia: A Delphi consensus study. Lancet 366: 2112–17.

2. Brookmeyer R, Johnson E, Ziegler-Graham K, Arrighi H M (2007) Forecasting the global burden of Alzheimer's disease. Alzheimers Dement 3: 186–91.

3. Association AS (2012) Alzheimer's disease facts and figures. Alzheimers Dement 8: 131–168.

4. Ashe KH, Zahs KR (2010) Probing the biology of alzheimer’s disease in mice. Neuron 6: 631–645.

5. Huang Y, Mucke L (2012) Alzheimer mechanisms and therapeutic strategies. Cell 148: 1204–22.

6. Querfurth HW, LaFerla FM (2010) Alzheimer’s disease, New England J Med Surg and Collat Branches Sci. 362:329–44.

7. Soyer Z, Uysal S, Parlar S Tarıkoğulları D, Alptuzun AH (2017) Synthesis and molecular docking studies of some 4 phthalimidobenzenesulfonamide derivatives as acetylcholinesterase and butyrylcholinesterase inhibitors Journal Enzyme Inhibition and Medicinal Chemistry 32: 13-19.

8. Bartus RT, Dean RL, Beer B, Lippa AS (1982) The cholinergic hypothesis of geriatric memory dysfunction. Science 217: 408–417.

9. Lawrence AD, Sahakian BJ (1995) Alzheimer disease, attention, and the cholinergic system. Alzheimer Dis Assoc Disorders 943–49.

10. Jeffrey LC (2000) Cholinesterase inhibitors: a new class of psychotropic compounds. Am J Psychiatry 157:4–15.

11. Silva D, Chioua M, Samadi A (2013) Synthesis, Pharmacological Assessment, and Molecular modeling of acetylcholinesterase/butyrylcholinesterase inhibitors: effect against amyloid-β-induced neurotoxicity ACS Chem Neurosci 4: 547-65.

12. Shaikh S, Verma A, Siddiqui S (2014) Current acetylcholinesterase-inhibitors: A neuroinformatics perspective. CNS Neurol Disord Drug Targets 13: 391–401.

13. Toda N, Kaneko T, Kogen H (2010) Development of an efficient therapeutic agent for Alzheimer's disease: design and synthesis of dual inhibitors of acetylcholinesterase and serotonin transporter. Chem Pharm Bull 58: 273–87.

14. Farina R, Pisani L, Catto M (2015) Structure-based design and optimization of multitarget-directed 2H-chromen-2-one derivatives as potent inhibitors of monoamine oxidase B and cholinesterases. J Med Chem 58: 5561–78.

15. Bajda M, Więckowska A, Hebda M (2013) Structure-based search for new inhibitors of cholinesterases. Int J Mol Sci 14: 5608–5632.

16. Pohanka M (2014) Inhibitors of acetylcholinesterase and butyrylcholinesterase meet immunity. Int J Mol Sci 15: 9809-9825.

17. Masters CL, Bateman R, Blennow K, Rowe CC, Sperling RA, Cummings JL (2015) Alzheimer’s disease. Nat Rev Dis Primers 1: 15056.

18. Yella JK (2018) Changing trends in computational drug repositioning, Pharmaceuticals 11: 57.

19. Nagaraj AB (2018) Using a novel computational drug-repositioning approach (Drug Predict) to rapidly identify potent drug candidates for cancer treatment, Oncogene 37: 403-414.

20. Ghofrani HA (2006) Sildenafil: from angina to erectile dysfunction to pulmonary hypertension and beyond. Nat Rev Drug Discov 5: 689-702.

21. Joglekar S, Levin M (2004) The promise of thalidomide: evolving indications. Drugs Today 40: 197-204.

22. Karakaplan M, Ak D, Colak M, Ozhan Kocakaya S, Pirinccioglu N (2013) Synthesis of new diaza-18-crown-6 ethers derived from trans- (R,R)-1,2-diaminocyclohexane and investigation of their enantiomeric discrimination ability with amino acid ester salts. Tetrahedron 69: 349-358.

23. Karakaplan M, Jameel B (2016) Synthesis and application of amino alcohol imides as NMR solvating agents for chiral discrimination of carboxylic acids. Tetrahedron Asymmetry 27:597-602.

24. Ellman GL, Courtney KD, Jr. Andres V, Featherstone RM (1961) A new and rapid colorimetric determination of acetylcholinesterase activity. Biochem Pharmacol 7: 88-95.

25. Hassan M, Raza H, Abbasi MA, Moustafa AA, Seo SY (2019) The exploration of novel Alzheimer's therapeutic agents from the pool of FDA approved medicines using drug repositioning, enzyme inhibition and kinetic mechanism approaches. Biomedicine and Pharmacotherapy 109: 2513–26.

26. Carletti E, Colletier JP, Dupeux F, Trovaslet M, Masson P, Nachon F (2010) Structural evidence that human acetylcholinesterase inhibited by tabun ages through O-dealkylation. J Med Chem 53: 4002-4008.

27. Silman I, Sussman JL (2008) Acetylcholinesterase: how is structure related to function? Chem Biol Interact 175: 3–10.

28. Zhou Y, Catalytic reaction mechanism of acetylcholinesterase determined by Born-Openheimer ab initio QM/MM molecular dynamics simulations (2010) J Phys Chem B 114: 8817–8825.

29. Carletti E, Colletier JP, Schopfer LM, Santoni G, Masson P, Lockridge O, Nachon F, Weik M (2013) Inhibition Pathways of the Potent Organophosphate Cbdp with Cholinesterases Revealed by X-Ray Crystallographic Snapshots and Mass Spectrometry Chem.ResToxicol; 26: 280.

30. Kadam RU, Roy N (2007) Recent trends in drug-likeness prediction: A comprehensive review of In silico methods. Indian J Pharm Sci 69: 609-615.

31. Lipinski CA, Lombardo F, Dominy BW, Feeney PJ (1997) Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings Adv Drug Deliver Rev 23: 4-25.

32. Case DA, Darden T.A., Cheatham III T.E., Simmerling CL, Wang J, Duke RE, Luo R, Walker RC, Zhang W, Merz KM, Roberts BP, Wang B, Hayik S, Roitberg A, Seabra G, Kolossvary I, Wong KF, Paesani F, Vanicek J, Liu J, Wu X, Brozell SR, Steinbrecher T, Gohlke CQ, Ye X, Wang J, Hsieh MJ, Cui G, Roe DR, Mathews DH, Seetin MG, Sagui CH, Babin V, Luchko T (2010) Gusarov S, Kovalenko A, Kollman PA, AMBER 11, University of California: San Francisco.

33. Duan Y, Wu C, Chowdhury S, Lee MC, Xiong GM, Zhang W, Yang R, Cieplak P, Luo R, Lee T, Caldwell J, Wang MJ, Kollman P (2003) Point-charge force field for molecular mechanics simulations of proteins based on condensed-phase quantum mechanical calculations. J Comput Chem 1999-2012.

34. Case DA, Cheatham TE, Darden T, Gohlke H, Luo R, Merz KM, Onufriev A, Simmerling C, Wang, Woods RJ (2005) The Amber biomolecular simulation programs. J Comput Chem 1668-1688.

35. Bayly CI, Cieplak P, Cornell WD, Kollman PA (1993) A Well- Behaved Electrostatic Potential Based Method Using harge Restraints for Deriving Atomic Charges - the Resp Model. J Phys Chem 10269-10280.

36. Pettersen EF, Goddard TD, Huang CC, Couch GS, GreenBlatt DM, Mang EC, Ferrin TE (2004) UCSF Chimera—A visualization system for exploratory research and analysis. J Comput Chem 1605-1612.

37. Accelrys Software Inc. Discovery Studio Modeling Environment, Release 3.5 San Diego, Accelrys Software Inc. 2012

38. Lang PT, Moustakas D, Brozell S, Carrascal N, Mukherjee S, Pegg S, Raha K, Shivakumar D, Rizzo R, Case D, Shoichet B, Kuntz I (2007) University of California San Francisco, http://dock.compbio.ucsf. edu/.

Figures

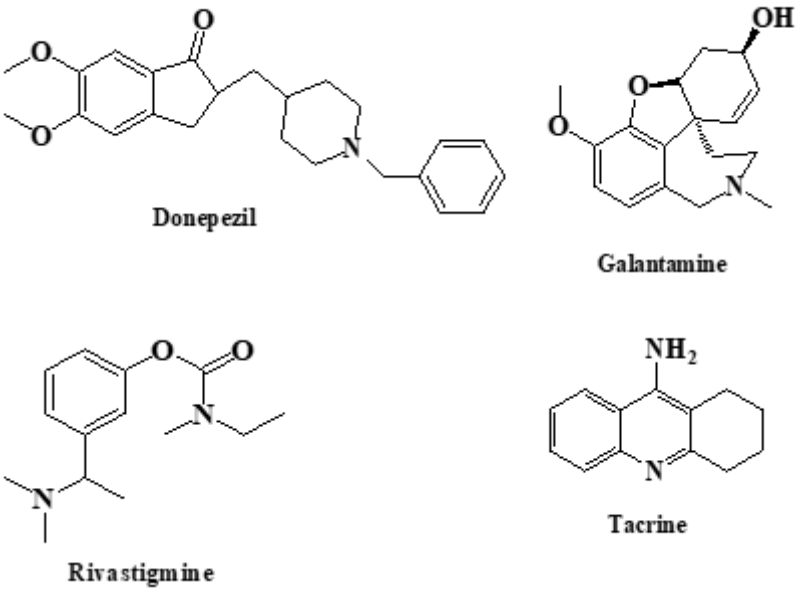

Figure 1

Chemical structure of FDA approved AChE inhibitors.

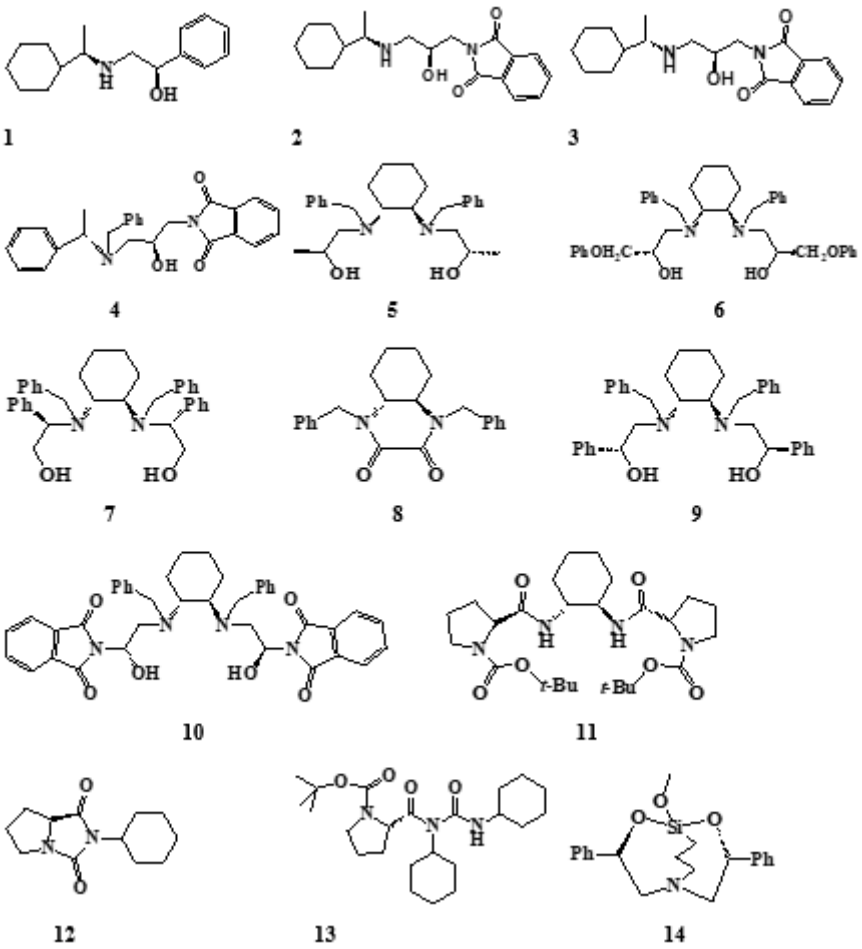

Figure 2

3D Graphical depiction of chiral β-amino alcohols and amides tested for AD disease.

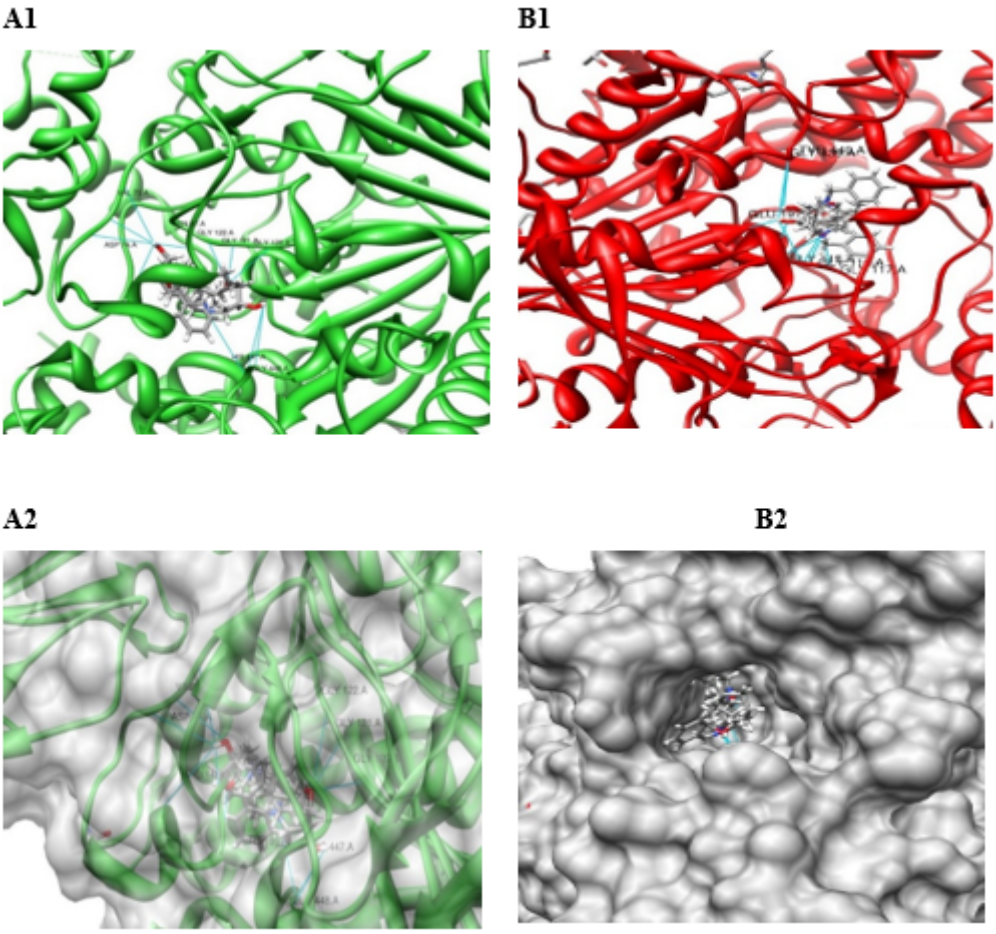

Figure 3

A1: Compound 6-AChE docking complex, AChE is highlighted in green color, A2: Binding pocket analysis of AChE. B1: Compound 6-BuChE docking complex, BuChE is highlighted in red colour, B2: Binding pocket analysis of BuChE.

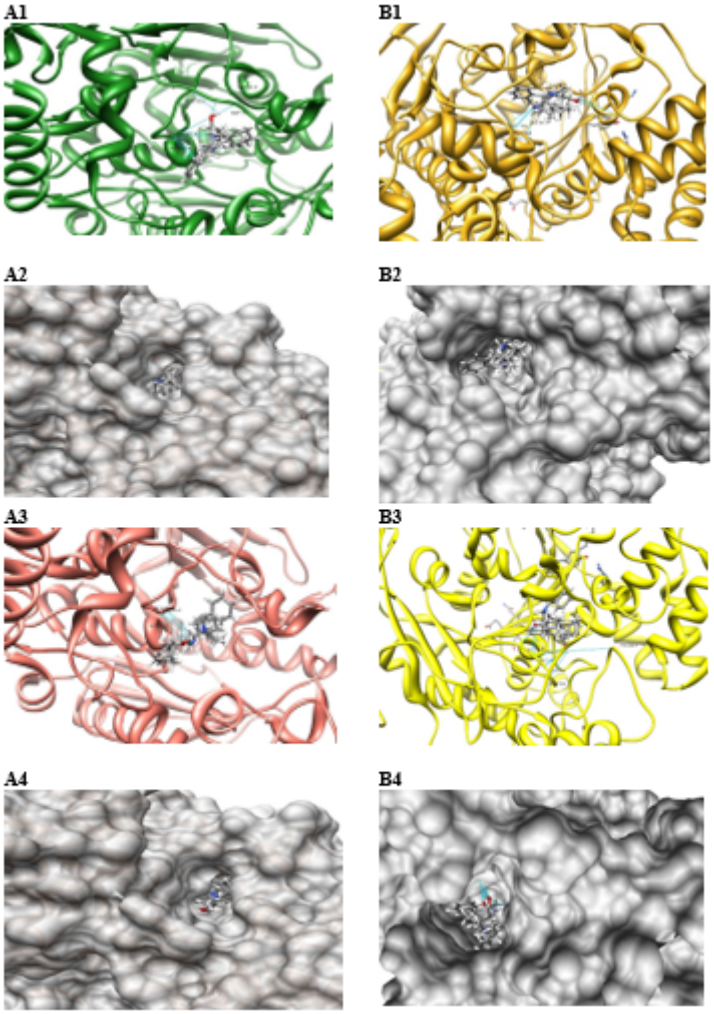

Figure 4

Binding pocket analysis of AChE and BuChE, A1 and B1: Docking complexes of 7 against AChE and BuChE, A2 and B2: 7 binding conformational position inside the active region of AChE and BuChE. A3 and B3: Docking complex of 9 against AChE and BuChE, A4 and B4: 9 binding conformational position inside the active region of AChE and BuChE.