MOLECULAR DOCKING STUDY OF PHYTOCONSTITUENTS IDENTIFIED IN *ZINGIBER OFFICINALE & OCIMUM BASILICUM* ON BUTYRYLCHOLINESTERASE – AN ENZYME TARGET FOR ALZHEIMER'S DISEASE

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Abstract: Alzheimer's disease is a neurological condition that gradually impairs thinking and memory abilities as well as the capacity to do even the most basic tasks. The purpose of this study is to conduct an *in silico* molecular docking analysis of the chosen essential Phytoconstituents from two medicinal plants, namely Ginger (*Zingiber officinale*) and Basil (*Ocimum basilicum*) on target protein, Butyrylcholinesterase (PDB ID: 3DJY). 3DJY is responsible for Alzheimer's disease and is selected as the target protein for anti-Alzheimer activity. The essential phytoconstituents were obtained from PubChem chemical database. The Protein Data Bank was used to retrieve the protein structures in PDB format. *In silico* docking analysis was performed with Molegro Virtual Docker (MVD) software. Hydrogen bond interactions, Rerank score, and MolDock score are the parameters utilised in docking studies. Standard drug was utilised to compare against the docking scores of the phytoconstituents. Nerolidol, Rhamnetin showed the best MolDock scores. As possible binding to the enzyme is directly represented by the MolDock score, it was discovered that the examined phytoconstituents demonstrated potent inhibitory action when compared to that of the standard treatments. The investigated phytoconstituents support the anti-Alzheimer claims of their source plants and exhibit promise as anti-Alzheimer leads.

Keywords: Alzheimer's disease, *Zingiber officinale*, *Ocimum basilicum*, Molegro Virtual Docker, Neurodegenerative disease.

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INTRODUCTION

Alzheimer's disease (AD) is a degenerative neurological illness that causes brain atrophy and the death of brain cells. It is the most prevalent kind of dementia, which causes a person's capacity to operate independently to continuously deteriorate in mental, thinking, memory, reasoning skills, behavioural, and social abilities [1]. Other than environmental and genetic factors, the main risk factor for Alzheimer's disease is still ageing.

Amyloid plaques are more prevalent in AD patients and a high density of neurofibrillary

tangles causing cortical atrophy and substantial neuronal loss. Microtubules, which are collections of coupled helical filaments, are found inside neurons, required for cell's normal function. Tau protein, which connects these microtubules, is disrupted in AD patients, allowing neurons to twist into filaments that impair normal cell activity [2,3].

Amyloid plaques are dense insoluble deposits which contain A*β* protein. A*β* protein is the core of neuritic plaques that form around neurons and disrupt cell function and is a fragment of the protein known as amyloid precursor (APP). APP in AD patient's cleavage results in excessive A*β*. Apolipoprotein E **(**APOE) along with mutation of APP gene on chromosomes 21 and 14 has been linked to pathogenesis of AD. APOE testing has been considered as possible screening methods for asymptomatic AD [4].

In Alzheimer's patients accelerated cognitive and functional deterioration results in hallucinations, delusions, and higher mortality. Agitation is a common symptom of AD. Degeneration developing in various regions of the brain may result in the overestimation of threat and/or affective deregulation that produces hyper vigilance [5].

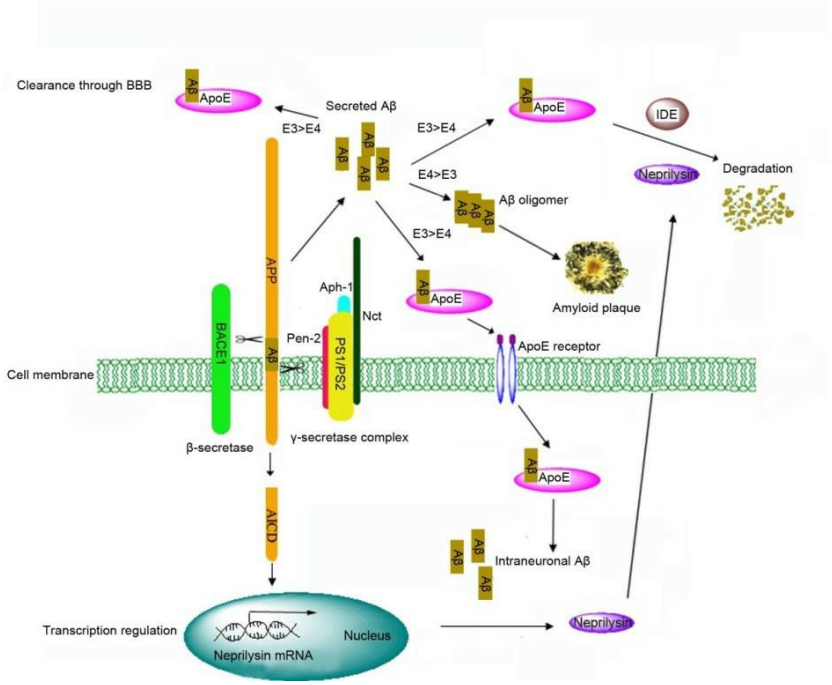

Figure 1. Progression of Alzheimer Disease [6]

The probability of getting AD is approximately 3% at age 65 and increases to nearly 30% by age 85 [7]. In adults over 60 years old, the estimated global prevalence of dementia is 3.9%. Around 5 million new cases of dementia are diagnosed each year, affecting more than 25 million individuals worldwide, the majority of whom have Alzheimer's disease [4,8,9].

Therapeutic intervention is based on understanding the pathogenic factors of AD. Among these is a decline in cholinergic function (cholinergic replacement therapy and neurotrophies), the amyloid cascade (A*β* vaccination, *β*- and *γ*-secretase effectors, statins), oxidative stress (antioxidant treatment), and inflammatory mediators (NSAIDs), deficits in steroid hormones (hormone replacement therapy), excitotoxicity (memantine), and the impact of dietary factors (low saturated fat diets, moderate alcohol intake). Estrogen replacement treatment (ERT) can assist to minimize A*β* formation, improve cerebral blood flow in women, prevent cholinergic neuron atrophy, reduce oxidative stress, and modify the actions of nerve growth factors [10]. Medicinal plants chosen for this study were Ginger (*Zingiber officinale*), and Basil (*Ocimum basilicum*).

For more than 2000 years, people have used the rhizome of Ginger (*Zingiber officinale* Roscoe) is a member of the zingiberaceae family, which is thought to provide a variety of medical benefits. The antioxidant [11,12] and anti-inflammatory [13] characteristics that are important for the treatment of AD are particularly significant.

Butyrylcholinesterase (BChE) is a gene that spans about 73 kb on chromosome 3q26 and has four exons and three sizable introns. BChE is a protein that hydrolyzes acetylcholine. BChE has been linked to NFTs and Aβ in the pathogenesis of AD and is thought to be pertinent to the dementia and progressive memory loss in AD. BChE has also been shown to be crucial for the maturation of AD plaques. These facts make BChE an ideal target for molecular docking studies of Alzheimer's disease [14].

Ocimum basilicum L. (Lamiaceae family) popularly known as sweet basil, has historically been used to cure respiratory issues, warts, diarrhoea, worms, headaches, constipation, coughs, and kidney disorders. According to some studies, it can also be used to treat cancer, diabetes, menstrual cramps, cardiovascular disease, and neurological illnesses [14,15]. *O. basilicum* ethanol extracts are high in phenolic components and have potent antioxidant properties [16,17].

Studies validating the potential of these phytoconstituents for neuroprotection against Aβ toxicity assisted in identifying the molecular target of action contributing to its anti-Alzheimer.

MATERIALS AND METHOD

DruLiTo and Swiss ADME

DruLiTo is an open-source virtual screening tool. DruLiTo is based on various drug likeness rules like Lipinski's rule, MDDR-like rule, Veber's rule, Ghose filter, BBB rule, CMC-50 like rule and Quantitative Estimate of Drug-likeness (QED). Seven major phytoconstituents used in the treatment of Alzheimer were selected as ligands. The 3D structures of ligands were downloaded from PubChem in the form of SDF format. All the prepared ligands were tested in DruLiTo software [9]. Swiss ADME is a web tool used to determine pharmacokinetic properties like Absorption, distribution, metabolism, and excretion. Two important chemical descriptors correlate with PK properties, the 2D polar surface area and the lipophilicity levels in the form of atom-based LogP. Smiles notation of a ligand was imported in a workspace, the results were obtained and analysed in the project table.

T.E.S.T (Toxicity Estimation Software Tool)

Toxicity Estimation Software Tool (TEST) was used to estimate toxicity present in chemicals using Quantitative Structure Activity Relationships (QSARs) methodologies. LC_{50} threshold based on each model's predication as well as a consensus average of the component model was determined by using TEST software. The QSARs methodologies employed in this research work are hierarchical method, single-model method, group contribution method, consensus method and nearest neighbour method.

Molegro Virtual Docker

Molegro Virtual Docker (MVD) is a protein-ligand docking simulation program. Forecasting the binding conformation of ligands to appropriate tar- get binding site was done using MVD. It is used in estimation of MolDock score of the ligand.

Molecular Docking

Ligand Preparation:

In this study, docking of the selected ligands molecules against the AD protein target was carried out by MVD software. The major phytoconstituents were selected as the study ligands. The activity of ligands was predicted using web tool, Way2Drug (http://www.way2drug.com). 6-Shagaol, Nerolidol, Maslinic acid, Rhamnetin, Geraniol, Kaempferol and Betulin were selected from the medicinal plants for the study. The selected ligands were downloaded from

PubChem database (https://pubchem.ncbi.nlm.nih.gov) in SDF format. All the ligands were imported in MVD workspace. The docking scores of the phytoconstituents were compared against the standard drugs (Donepezil, Aducanumab and Memantine). For precise docking, it is important that the imported structures have proper atom connectivity and bond order with partial atomic charges assigned.

Target Preparation

The target protein which is responsible for Alzheimer disease was selected and downloaded from Protein Data Bank (https://www.rcsb.org) in PDB format. The PDB id of the target molecule, Butyrylcholinesterase (BChE) was 3DJY [18].

Molecular Import and Preparation

Molecular structure files were parsed into relevant components (ligands, cofactors, water molecules and proteins) and were automatically prepared. MVD assisted in detecting bonds, aromaticity, assigns charges, and adds explicit hydrogen. The built-in cavity detector identified promising binding locations (cavities), making it possible to restrict the search space to the most interesting regions.

Docking

Molecular docking programs simulate how a target protein (enzyme or receptor) interacts with a small molecule (ligands). Here we have used the Molegro Virtual Docker software. The docking scoring function of MVD is based on Piecewise Linear Potential **(**PLP).

For each ligand docking, the best orientation for the ligand-protein complex was analyzed and hydrogen bonds were identified and labelled. The ligand energy was inspected and analyzed using MVD score, a linear combination of hydrogen bonding and electrostatic interactions. All necessary valence checks and H atom addition was done.

The cavities found by the cavity detection algorithm were actively used by the search algorithm guided differential evolution to focus the search, to that specific area during the docking stimulation. Warning arising from unlikely preparations and missing structural information (e.g. unknown residues) were given attention and attempts were made to rectify the errors. Reranking procedure was applied to the highest ranked poses to further increase docking accuracy

Analysis

The returned poses from the docking engine were observed with the help of pose organizer. Pose organizer was capable of loading poses from a docking run dynamically, making it possible to browse thousands of ligands. Various energy terms and interactions were inspected simultaneously and more advanced reranking along with binding affinity measures were calculated. Hydrogen bonds and electrostatic interactions were updated dynamically when switching between poses.

The MolDock score of selected ligands obtained were compared with the standard drug. Ligands showing the best MolDock scores were designated as the best ligands and as a possible lead molecule for cure against Alzheimer.

RESULTS

The foundation of MolDock is a novel heuristic search technique that combines cavity prediction and differential evolution. A modification of the piecewise linear potential (PLP), the docking scoring function of MolDock incorporates new electrostatic and hydrogen bonding

terms. The MolDock Score is a key parameter for analysing the docking results. MolDock score rerank score and hydrogen bond interactions were used for evaluating the ligand molecules. The ligand with the least MolDock score shows a strong affinity towards its enzyme target.

In silico docking analysis of the selected ligands against Butyrylcholinesterase based on MolDock score is shown in table 2. Ranking of ligand possess against BChE protein based on rerank score is shown in table 3 and ranking of ligands based on H-bond is shown in table 4. Ligand energy inspector tool is utilised for capturing the binding pattern of poses.

Table 1. Drug likeness properties of phytoconstituents

S.No	Ligand	Lipinski			Veber's			Blood Brain Barrier			CMC50	
		MW	LogP	HBA	HBD	TPSA	nRB	n-acidic group	nHB	Log P	AMR	nAtom
1	6-Shogoal	276.17	3.77	3	1	46.53	9	0	0	3.77	82.07	44
2	Nerolidol	222.2	4.29	1	1	20.23	7	0	2	4.29	73.67	42
3	Maslinic acid*	472.36	8.24	4	3	77.76	1	1	7	8.24	137.08	82
4	Rhamnetin	316.06	2.15	7	4	116.45	2	0	11	2.15	88.48	35
5	Geraniol	154.14	2.52	1	1	20.23	4	0	2	2.52	51.3	29
6	Kaempferol	286.05	1.486	6	4	107.22	1	0	10	1.48	81.83	31
7	Betulin*	442.38	9.714	2	2	40.46	2	0	4	9.71	131.81	82
8	Donepezil	379.2	2.633	4	0	38.77	6	0	4	2.63	115.79	57
9	Memantine	179.2	3.524	1	1	26.02	0	0	2	3.52	54.06	34
10	Aducanumab	755.3	-1.86	12	9	169.7	29	0	21	-1.8	124.48	99

* Fails the Lipinski rule of 5 as LogP value was greater than 5
MW – Molecular weight
LogP – Partition coefficient
HBA – Hydrogen Bond Acceptor
HBD – Hydrogen Bond Donor
nRB – Number of Rotatable Bonds
TPSA – Topological Surface Area

The Drug likeness analysis clearly shows that all of the phytoconstituents assessed in the study have drug-like properties, with the exception of Maslinic acid, Carnosol and Betulin which have a LogP value larger than 5.

Table 2. Ranking of Ligands and poses against BChE based on MolDock score

S.No	Ligand	MolDock score	Rerank score	H bond
1	Maslinic acid	-106.63	-91.04	-4.99
2	Nerolidol	-87.27	-58.93	-2.5
3	Rhamnetin	-83.71	-74.79	-10.41
4	Betulin	-77.87	-18.17	0
5	Donepezil	-77.59	207.97	0
6	Kaempferol	-77.46	-57.07	-6.94
7	Geraniol	-76.07	-60.49	-2.5
8	Memantine	-65.97	-57.62	-2.28
9	6- Shogaol	-64.52	-95.79	-1.97
10	Aducanumab	119.36	1688.66	-4.76

Table 3. Ranking of ligands and poses against BChE based on rerank score

S.No	Ligand	MolDock score	Rerank score	H bond
1	6- Shogaol	-64.52	-95.79	-1.97
2	Maslinic acid	-106.63	-91.04	-4.99
3	Rhamnetin	-83.71	-74.79	-10.41
4	Geraniol	-76.07	-60.49	-2.5
5	Nerolidol	-87.27	-58.93	-2.5
6	Memantine	-65.97	-57.62	-2.28
7	Kaempferol	-77.46	-57.07	-6.94
8	Betulin	-77.87	-18.17	0
9	Donepezil	-77.59	207.97	0
10	Aducanumab	119.36	1688.66	-4.76

Table 4. Ranking of ligands and poses against BChE based on H-bond score

S.No	Ligand	MolDock score	Rerank score	H bond
1	Rhamnetin	-83.71	-74.79	-10.41
2	Kaempferol	-77.46	-57.07	-6.94
3	Maslinic acid	-106.63	-91.04	-4.99
4	Aducanumab	119.36	1688.66	-4.76
5	Nerolidol	-87.27	-58.93	-2.5
6	Geraniol	-76.07	-60.49	-2.5
7	Memantine	-65.97	-57.62	-2.28
8	6- Shogaol	-64.52	-95.79	-1.97
9	Betulin	-77.87	-18.17	0
10	Donepezil	-77.59	207.97	0

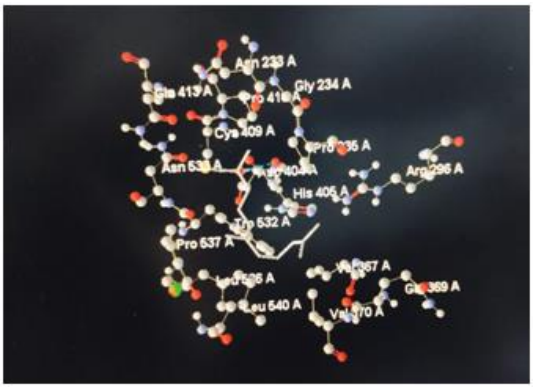

Figure 2. Docking view of Nerolidol against BChE

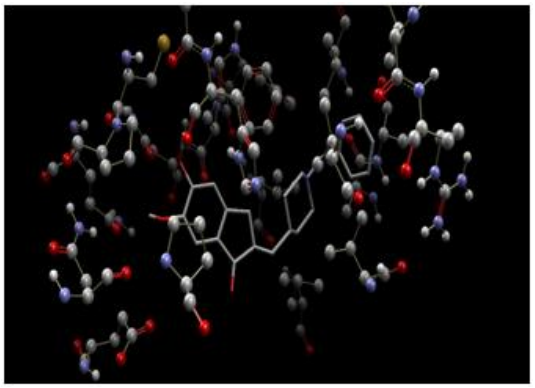

Figure 3. Docking view of Donepezil against BChE

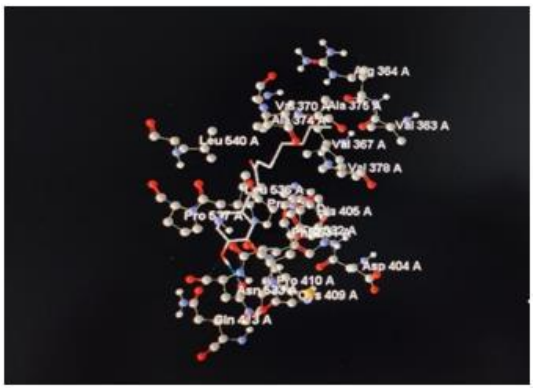

Figure 4. Docking view of 6-Shogaol against BChE

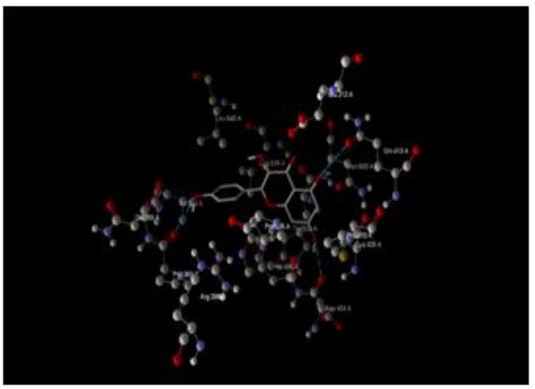

Figure 5. Docking view of Kaempferol against BChE

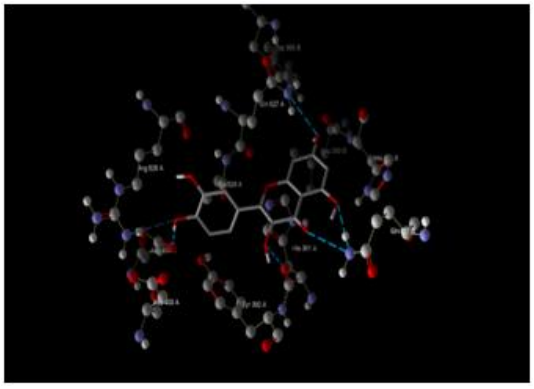

Figure 6. Docking view of Rhamnetin against BChE

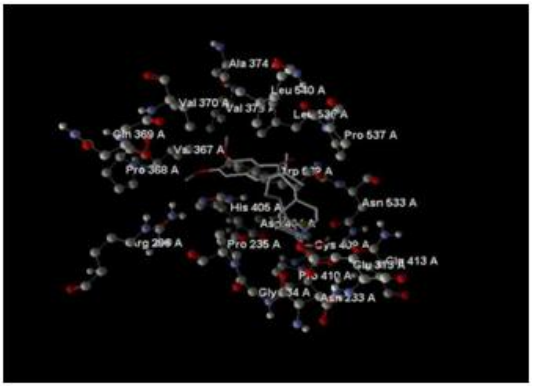

Figure 7. Docking view of Geraniol against BChE

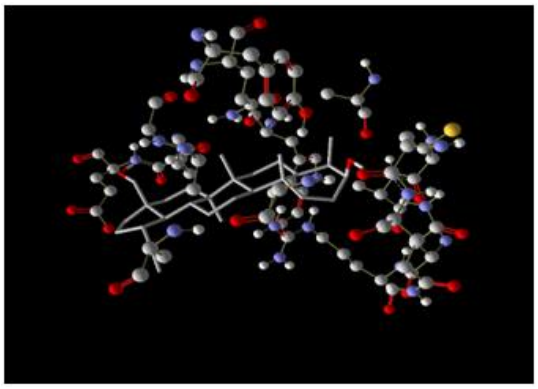

Figure 8. Docking view of Betulin against BChE

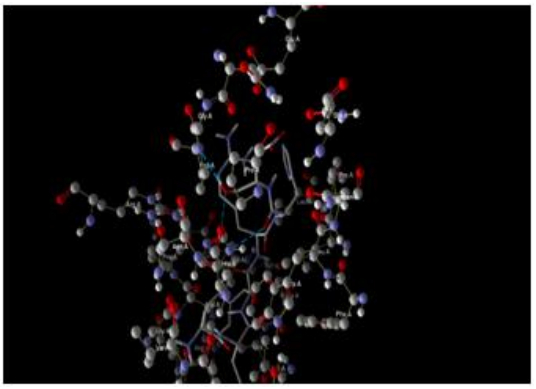

Figure 9. Docking view of Aducanumab against BChE

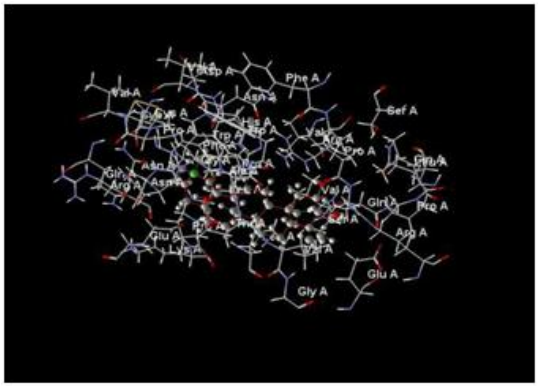

Figure 10. Docking view of Maslinic acid against BChE

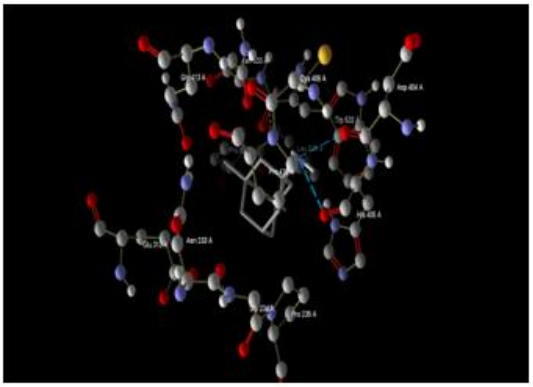

Figure 11. Docking view of Memantine against BChE

DISCUSSION

Donepezil is a cholinesterase inhibitor, used to improve cognitive function. Research has shown Donepezil to cause bradycardia, heart block and GI disturbances. Given that the visual association cortex is more active when a person is in REM sleep, cholinesterase inhibitors like Donepezil can make people have nightmares [19,20].

Aducanumab belongs to the class of monoclonal antibodies. It is used in the treatment and management of Alzheimer disease. There is a risk of developing Amyloid-related imaging abnormalities (ARIA) during Aducanumab medication. Other side effects like delirium, altered mental status, disorientation, GI disturbances and the most common being headache (13%) are related to Aducanumab treatment [21].

Memantine is a non-competitive N-methyl-D-aspartate (NMDA) receptor antagonist used to treat Alzheimer disease. Memantine is associated with some serious side effects like hypertension, agitation, somnolence, constipation and headache. It is challenging to balance memantine's effectiveness and safety in this situation [22,23].

The drawback for these medications encourages the development of herbal medications in alternative medicine and suggests using herbs in daily life to prevent disorders like Alzheimer disease and other mental abnormalities that are lifestyle-modifying.

The phytoconstituents Nerolidol present in ginger, showed the least MolDock score (-87.27) against the BChE as compared to the standard drug, Donepezil (-77.59). Nerolidol is a naturally occurring sesquiterpene alcohol that is present in essential oils with a floral odour. Nerolidol is extracted by hydro distillation method using Clevenger-type apparatus [24]. Nerolidol is a natural organic compound that has two geometric isomers, trans and cis.

Nerolidol is used anti-microbial, anti-oxidant, anti-inflammatory and anti-cancer. Nerolidol also occurs in jasmine, lavender, tea tree, *Cannabis sativa* and *Cymbopogon citrates* [25].

Rhamnetin is an O-methylated flavonol. It is present in ginger. Additionally, cloves, sweet wormwood, and green vegetables like coriander seeds and leaves contain rhamnetin. Cloves and coriander are staples in a variety of cuisines. It has shown to possess anti-oxidant activity. Molegro software has shown Rhamnetin with the least MolDock score (-83.71) against BChE. Rhamnetin is known for its anti-oxidant, anti-inflammatory, anti-cancer and anti-bacterial activity [26,27].

The standard drug, Aducanumab showed a MolDock score of 119.36; Memantine showed MolDock score of -65.97 and Donepezil showed a MolDock score of -77.59.

Thus the phytoconstituents responsible for maximum MolDock score namely Nerolidol, Rhamnetin, as compared with the standard drugs may possess anti-Alzheimer property.

A total number of 7 phytoconstituents were filtered using the DruLiTo software and the total number of phytoconstituents violated the rule (i.e., LogP > 5) is 3 i.e., Maslinic acid, Carnosol and Betulin, which showed a LogP value of 8.24, 5.1 and 9.71 respectively.

The 7 phytoconstituents of the plants were docked against the Alzheimer protein, BChE. Nerolidol and Rhamnetin showing the least MolDock scores, represent a strong binding affinity towards BChE, as evident from docking score. Followed by Shogaol, Geraniol and Kaempferol exhibited best MolDock scores, as evident from the docking scores. All the phytoconstituents had drug likeness property except Maslinic acid, Carnosol and Betulin, which had a LogP value greater than 5. Hence these phytoconstituents may act as a potential drug lead for the treatment and management of Alzheimer disease.

CONCLUSION

From this research it can be concluded that phytoconstituents from *Zingiber officinale* and *Ocimum basilicum* can have potential pharmacological activity for therapeutic use against Alzheimer disease. Natural herbs can be used for a wide range of population from different age groups, as they exert least adverse effects. Further these herb supplements can be used on a daily basis for preventing Alzheimer's disease and other mental abnormalities, as they provide feasibility to acquire and use from the local market. These herbs could be a better alternative to conventional allopathic treatments for Alzheimer disease without adverse effects. Further, *ex vivo* and pre-clinical studies may be performed to better understand the mechanism of action involved, through which these phytoconstituents exhibit anti-Alzheimer activity.

Ethical approval: Not Applicable

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Conflicts of Interest - The authors report no conflicts of interest in this work.

Author contributions: Idea and planning of work was supervised by SJ and VAR. Experimental work was carried out by AS, SS, VA and HH. CA and SJ wrote the manuscript. VAR and CA re-evaluated the paper and made changes. CA and SJ reviewed the manuscript at the end. All authors read and approved the final manuscript.

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REFERENCE

1. Roth MT, Tym E, Mountjoy CQ, Huppert FA, Hendrie H, Verma S, Goddard R. CAMDEX: a standardised instrument for the diagnosis of mental disorder in the elderly with special reference to the early detection of dementia. The British journal of psychiatry. 1986 Dec;149(6):698-709.
2. Geldmacher DS. Differential diagnosis of dementia syndromes. Clinics in geriatric medicine. 2004 Feb 1;20(1):27-43.
3. Dorland M, Van Kooij RJ, Te Velde ER. General ageing and ovarian ageing. Maturitas. 1998 Oct 12;30(2):113-8.
4. Liddell M, Williams J, Bayer A, Kaiser F, Owen M. Confirmation of association between the e4 allele of apolipoprotein E and Alzheimer's disease. Journal of medical genetics. 1994 Mar 1;31(3):197-200.
5. Lanctôt KL, Amatniek J, Ancoli-Israel S, Arnold SE, Ballard C, Cohen-Mansfield J, Ismail Z, Lyketsos C, Miller DS, Musiek E, Osorio RS. Neuropsychiatric signs and symptoms of Alzheimer's disease: New treatment paradigms. Alzheimer's & Dementia: Translational Research & Clinical Interventions. 2017 Sep 1;3(3):440-9.
6. Dong S, Duan Y, Hu Y, Zhao Z. Advances in the pathogenesis of Alzheimer's disease: a re-evaluation of amyloid cascade hypothesis. Translational Neurodegeneration. 2012 Dec;1(1):1-2.
7. Alzheimer's Association. 2011 Alzheimer's disease facts and figures. Alzheimer's & dementia. 2011 Mar;7(2):208-44.
8. Qiu C, Kivipelto M, Von Strauss E. Epidemiology of Alzheimer's disease: occurrence, determinants, and strategies toward intervention. Dialogues in clinical neuroscience. 2022 Apr 1.
9. Ekins S, Mestres J, Testa B. *In silico* pharmacology for drug discovery: methods for virtual ligand screening and profiling. British journal of pharmacology. 2007 Sep;152(1):9-20. Ekins S, Mestres J, Testa B. *In silico* pharmacology for drug discovery: methods for virtual ligand screening and profiling. British journal of pharmacology. 2007 Sep;152(1):9-20.
10. Shah RS, Lee HG, Xiongwei Z, Perry G, Smith MA, Castellani RJ. Current approaches in the treatment of Alzheimer's disease. Biomedicine & Pharmacotherapy. 2008 Apr 1;62(4):199-207.
11. Stoilova I, Krastanov A, Stoyanova A, Denev P, Gargova S. Antioxidant activity of a ginger extract (*Zingiber officinale*). Food chemistry. 2007 Jan 1;102(3):764-70.
12. Shirin AP, Jamuna P. Chemical composition and antioxidant properties of ginger root (*Zingiber officinale*). Journal of Medicinal Plants Research. 2010 Dec 18; 4(24):2674-9.
13. Grzanna R, Lindmark L, Frondoza CG. Ginger—an herbal medicinal product with broad anti-inflammatory actions. Journal of medicinal food. 2005 Jun 1;8(2):125-32.
14. Ji H, Dai D, Wang Y, Jiang D, Zhou X, Lin P, Ji X, Li J, Zhang Y, Yin H, Chen R. Association of BDNF and BCHE with Alzheimer's disease: Meta-analysis based on 56 genetic case control studies of 12,563 cases and 12,622 controls. Experimental and therapeutic medicine. 2015 May 1;9(5):1831-40.
15. Balakrishnan P, Ramalingam P, Nagarasan S, Ranganathan B, Gimbun J, Shanmugam K. A comprehensive review on *Ocimum basilicum*. Journal of Natural Remedies. 2018;8(3):71-85.
16. Joshi RK. Chemical composition and antimicrobial activity of the essential oil of *Ocimum basilicum* L.(sweet basil) from Western Ghats of North West Karnataka, India. Ancient science of life. 2014 Jan;33(3):151.
17. Gülçin I, Elmastaş M, Aboul-Enein HY. Determination of antioxidant and radical scavenging activity of Basil (*Ocimum basilicum* L. Family Lamiaceae) assayed by different methodologies. Phytotherapy Research: An International Journal Devoted to Pharmacological and Toxicological Evaluation of Natural Product Derivatives. 2007

Apr;21(4):354-61.
18. Mlakić M, Čadež T, Barić D, Puček I, Ratković A, Marinić Ž, Lasić K, Kovarik Z, Škorić I. New Uncharged 2-Thienostilbene Oximes as Reactivators of Organophosphate-Inhibited Cholinesterases. Pharmaceuticals. 2021 Nov 11;14(11):1147.
19. Seltzer B. Donepezil: an update. Expert opinion on pharmacotherapy. 2007 May 1;8(7):1011-23.
20. Bryson HM, Benfield P. Donepezil. Drugs & aging. 1997 Mar 1;10(3):234-9.
21. Ferrero J, Williams L, Stella H, Leitermann K, Mikulskis A, O'Gorman J, Sevigny J. First-in-human, double-blind, placebo-controlled, single-dose escalation study of aducanumab (BIIB037) in mild-to-moderate Alzheimer's disease. Alzheimer's & Dementia: Translational Research & Clinical Interventions. 2016 Sep 1;2(3):169-76.
22. Taipale H, Tanskanen A, Koponen M, Tolppanen AM, Tiihonen J, Hartikainen S. Antidementia drug use among community-dwelling individuals with Alzheimer's disease in Finland: a nationwide register-based study. International clinical psychopharmacology. 2014 Jul;29(4):216-223.
23. Blanco-Silvente L, Capellà D, Garre-Olmo J, Vilalta-Franch J, Castells X. Predictors of discontinuation, efficacy, and safety of memantine treatment for Alzheimer's disease: meta-analysis and meta-regression of 18 randomized clinical trials involving 5004 patients. BMC geriatrics. 2018 Dec;18(1):1-6.
24. Bakkali F, Averbeck S, Averbeck D, Idaomar M. Biological effects of essential oils–a review. Food and chemical toxicology. 2008 Feb 1;46(2):446-75.
25. Kaiser R. The scent of orchids: olfactory and chemical investigations. Elsevier Science Publishers BV; 1993.
26. Medeiros DL, Lima ET, Silva JC, Medeiros MA, Pinheiro EB. Rhamnetin: a review of its pharmacology and toxicity. Journal of Pharmacy and Pharmacology. 2022 Jun;74(6):793-9.
27. Jnawali HN, Lee E, Jeong KW, Shin A, Heo YS, Kim Y. Anti-inflammatory activity of rhamnetin and a model of its binding to c-Jun NH2-terminal kinase 1 and p38 MAPK. Journal of natural products. 2014 Feb 28;77(2):258-63.