



## **Analysis of Potential *Cinnamomum zeylanicum* Blume Essential Oil Against Alzheimer's Disease: A Molecular Docking Study**

Muhammad Ja'far Shodiq<sup>1</sup>, Farmindo Hartono<sup>1</sup>, Siti Khaerunnisa<sup>2\*</sup>, Abdulloh Machin<sup>3</sup>

<sup>1</sup>Faculty of Medicine, Universitas Airlangga, Surabaya, Indonesia

<sup>2</sup>Departement of Physiology and Medical Biochemistry, Faculty of Medicine, Universitas Airlangga, Surabaya, Indonesia

<sup>3</sup>Departement of Neurology, Faculty of Medicine, Universitas Airlangga - Dr. Soetomo General Hospital, Surabaya, Indonesia

\*Corresponding author: [st.khaerunnisa@fk.unair.ac.id](mailto:st.khaerunnisa@fk.unair.ac.id)

Submitted: 19 November 2022

Accepted: 3 March 2023

Published: 30 April 2023

### **Abstract**

**Background:** Alzheimer's Disease (AD) is a neurodegenerative disorder with progressive impairment of behavioural and cognitive functions and the most common cause of dementia. The pathophysiology of AD is associated with low acetylcholine, accumulation of amyloid beta plaque, and neurofibrillary tangles in the brain. *Cinnamomum zeylanicum* is known to have many medicinal properties, especially neuroprotective effects. **Objective:** This research was designed to determine the neuroprotective potential of the phytochemicals *C. zeylanicum* using an *in silico* study. **Methods:** There are 5 phytochemicals compounds of *C. zeylanicum* used in this study. It's qualified for Lipinski's rules of five and can cross blood brain barrier. The protein targets were AChE, BACE1, and GSK-3. Molecular docking and visualization were performed using Avogadro, AutoDock 4.2 PyMol and Biovia Discovery Studio 2019. **Results:** *In silico* results show that the main phytochemical compounds of *C. zeylanicum* Blume essential oil have great potency as an AD drug. The best interaction model of the compound was shown by *trans*-cinnamyl acetate and coumaric acid. Although the binding energy of the compounds is lower than AD drugs (donepezil, rivastigmine, galantamine), the binding energy is not much different from rivastigmine and galantamine. **Conclusion:** The phytochemical compounds of *C. zeylanicum* Blume essential oil have an effect as a neuroprotective agent for AD and should be investigated in future research.

**Keywords:** Alzheimer's Disease, *Cinnamomum zeylanicum*, Molecular Docking Study

### **How to cite this article:**

Shodiq, M. J., Hartono, F., Khaerunnisa, S. & Machin, A. (2023). Analysis of Potential *Cinnamomum zeylanicum* Blume Essential Oil Against Alzheimer's Disease: A Molecular Docking Study. *Jurnal Farmasi dan Ilmu Kefarmasian Indonesia*, 10(1), 111-125. <http://doi.org/10.20473/jfiki.v10i12023.111-125>

## INTRODUCTION

Alzheimer's Disease (AD) known as a neurodegenerative disorder that mostly attacks people over 65 years marked by insidious onset and progressive impairment of behavioral and cognitive functions including memory, comprehension, language, attention, reasoning, and judgment (Cassani *et al.*, 2018; Cortes-Canteli and Iadecola, 2020; Kumar *et al.*, 2022). AD is the supreme cause of dementia worldwide (60-80%) and also causes a decline in cognitive ability (Lucey, 2020; Litke *et al.*, 2021). In 2015, around 46.8 million people worldwide suffered from AD; it's predicted to be 82 million in 2030 and 152 million in 2050. About 10 million new cases of AD are reported yearly worldwide (Cassani *et al.*, 2018; Vinicius *et al.*, 2019). In the Asia-Pacific region, AD is estimated to increase from 23 million in 2015 to 71 million in 2050 (Alzheimer's Disease International and Alzheimer's Australia, 2014). In Indonesia, the prevalence of AD patients was about 1.2 million in 2013, and it was estimated to have a rapid increase to 1.9 million in 2030 and 3.9 million in 2050 (Ong *et al.*, 2021). The cause of this increase is due to increasing life expectancy worldwide. In 2015, life expectancy reached 72 years in Indonesia itself (Kumar *et al.*, 2022). This will have an impact on the health and socio-economic sectors (Ong *et al.*, 2021).

One of the pathophysiologies of AD is cholinergic neuron dysfunction caused by cholinergic toxicity. Overactivity of AChE will cause a significant decrease in ACh levels in the synaptic cleft in the cortex, hippocampus, and amygdala. It will disturb cholinergic neurons involved in brain functions such as learning, memory, attention, response, sleep, and the delivery of sensory information (Kumar *et al.*, 2022; Samanta, Ramesh and Govindaraju, 2022). Accumulation of A $\beta$  plaques is also found in AD. This accumulation results in amyloid aggregation that promotes microglial activation and local inflammatory responses and leads to neurotoxicity. Amyloid deposition occurs around the meningeal and cerebral blood vessels and grey matter in AD (Dá Mesquita *et al.*, 2016; Tiwari *et al.*, 2019; Fan *et al.*, 2020; Kumar *et al.*, 2022). The presence of neurofibrillary tangles has been proven to have a relationship with AD. Hyperphosphorylation of the tau protein will form these tangles. The main function of tau protein is to stabilize axonal microtubules in the brain. Neurofibrillary tangles are stored in neurons and occur first in the hippocampus, then they are seen throughout the cerebral cortex (Long and Holtzman, 2019; Tiwari *et al.*, 2019; Kumar *et al.*, 2022). Some drugs that the FDA approves for AD are donepezil, galantamine, and rivastigmine (Yiannopoulou and Papageorgiou, 2020). However, these drugs have side effects such as gastrointestinal disturbances, dyspepsia, nausea, vomiting, loss of appetite, diarrhea, muscle aches, loss of balance, headache, and hepatotoxicity (Kumar, Chowdhury and Kumar, 2017; Kallel *et al.*, 2019). In recent years, there is many studies to develop new anti-AD drugs with lesser side effects (Kareti and Pharm, 2020).

*C. zeylanicum* Blume is an aromatic plant from Sri Lanka and the Malabar coasts of India. It was usually

used as seasoning in Asian traditional food and herbal medicine in different cultures and systems (Fahad *et al.*, 2018; Kallel *et al.*, 2019). It has many medicinal properties such as anti-inflammatory, anti-oxidant, analgesic, anti-cancer, anti-diabetic, anti-microbial, cardiovascular protective, cytoprotective, and neuroprotective (Momtaz *et al.*, 2017; Wang *et al.*, 2020). The herbal plants used to treat several diseases have developed rapidly in recent years (Kareti and Pharm, 2020). Plants are considered to be one of the sources of therapeutic active compounds, so this research was conducted to find new anti-AD drugs from herbal plants that might later have fewer side effects. *C. zeylanicum* Blume essential oil was chosen because it is believed to have a useful neuroprotective effect on AD. In previous studies, *C. zeylanicum* had anti-alzheimer's activity by inhibiting the nucleation process and the formation of tau filaments (Dhage *et al.*, 2021). Tepe and Ozaslan (2020) reported that *C. zeylanicum* had AChE inhibitory activity, which plays a role in the pathophysiology of AD. Frydman-Marom *et al.* (2011) also reported that *C. zeylanicum* can inhibit A $\beta$  plaque accumulation in AD.

## MATERIAL AND METHODS

### System Configuration

This molecular docking study used operating Windows 10 OS 32 bit laptop with an Intel Core i3 processor and 2 GB RAM. Applications in silico research include Avogadro, AutoDock 4.2, Biovia Discovery Studio 2019, and PyMol.

### Ligand and Protein Selection

*C. zeylanicum* Blume essential oil has the main phytochemical compounds namely cinnamaldehyde, *trans*-cinnamyl acetate, 1,4-benzene dicarboxylic acid, 1,8-cineole,  $\alpha$ -pinene, coumaric acid, and 9-octadecenoic acid (Kallel *et al.*, 2019). The compound that is qualified for Lipinski's rule of five and can cross blood-brain barrier (BBB) is used in this molecular docking. The terms of Lipinski's rule of five can be checked through the website <http://www.swissadme.ch/index.php>. The qualified compounds will be compared with donepezil, galantamine, and rivastigmine, which are some of the FDA-approved drugs for AD (Yiannopoulou and Papageorgiou, 2020). The qualified compounds are cinnamaldehyde, *trans*-cinnamyl acetate, 1,8-cineole,  $\alpha$ -pinene, and coumaric acid (Table 1). The 3D structure of the phytochemicals is downloaded from the website <https://pubchem.ncbi.nlm.nih.gov> and will be used as a ligand. The ligands were docked with the acetylcholinesterase/AChE receptor (PDB ID: 7E3H),  $\beta$ -site APP cleaving enzyme 1/BACE1 (GDP ID: 4DJU), and glycogen synthase kinase-3/GSK-3 (PDB ID: 1Q5K). These receptors play an important role in the pathophysiology of AD (Jagust, 2018). The protein structure was downloaded from the protein data bank (PDB) website <http://www.rcsb.org>.

### Ligand Structure Optimization

The compounds downloaded in ".sdf" format are optimized using Avogadro software and saved in ".mol2" format. After that, the compounds in ".mol2"

format was converted to “.pdbqt” format using AutoDock 4.2 software.

**Protein Structure Preparation**

The macromolecular structure of the protein downloaded in “.pdb” format from Protein Data Bank (PDB) was searched for the active site using the BIOVIA Discovery Studio 2019 software. In AChE, BACE1, and GSK-3 proteins, there are two chains, Chain A and B. Then we choose chain A, which is used for docking using AutoDock 4.2 software. Proteins were optimized by adding polar hydrogens, merging non-polar, adding Kollman charges in proteins, and computing gasteiger in native ligands. After that, the receptor is separated from the native ligand and saved in “.pdbqt” format.

**Grid Box Determination**

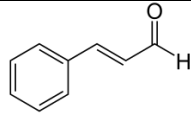
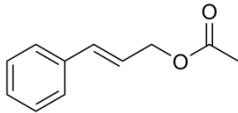
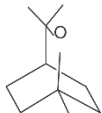
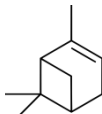
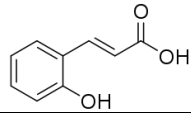
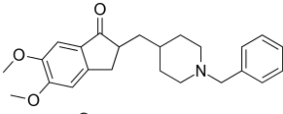
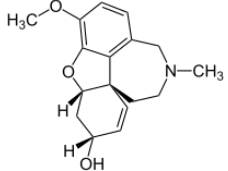
The size, centre coordinates, and spacing of the grid box are determined by the position of the native ligand using AutoDock 4.2 software. In AChE protein, the native ligand E20 grid was set in 40x40x40 (XYZ) point size, -43.74, 37.597, -30.039 centre coordinates, and 0.375 A spacing. In the BACE1 protein, the native

ligand OKK grid was set in 40x40x40 (XYZ) point size, 21,029, 10,689, 22,069 centre coordinates, and 0.375 A spacing. In the GSK-3 protein, the native ligand TMU grid was set in 40x40x40 (XYZ) point size, 23,148, 22.189, 8,978 centre coordinates, and 0.375 A spacing.

**Molecular Docking**

Proteins and ligands that have been prepared and grid boxes that have been determined are saved in “.gpf” format, and molecular docking is carried out with AutoDock 4.2 software. The ligand conformation output was analyzed using the Lamarckian Genetic Algorithm. Compounds that have the lowest bond energy ( $\Delta G$ ) show strong bonds and favourable conformations for ligand and protein interactions. After that, the results obtained in “.dlg” format are reopened using AutoDock 4.2 software to view the ligand bonds with amino acids and saved in “.pdb” format. The molecular docking results were visualized using the PyMol and BIOVIA Discover Studio 2019 software (Khaerunnisa, Suhartati and Awaluddin, 2020).

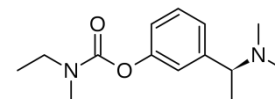
**Table 1. Ligand Structure of Molecular Docking**

Ligand	Molecular Formula	PubChem CID	Molecular Structure
<i>C. zeylanicum</i> blume essential oil			
Cinnamaldehyde	C <sub>9</sub> H <sub>8</sub> O	637511	
Trans-cinnamyl acetate	C <sub>11</sub> H <sub>12</sub> O <sub>2</sub>	5282110	
1,8-cineole	C <sub>10</sub> H <sub>18</sub> O	2758	
$\alpha$ -pinene	C <sub>10</sub> H <sub>16</sub>	6654	
Coumaric acid	C <sub>9</sub> H <sub>8</sub> O <sub>3</sub>	637542	
Alzheimer's disease drug			
Donepezil	C <sub>24</sub> H <sub>29</sub> NO <sub>3</sub>	3152	
Galantamine	C <sub>17</sub> H <sub>21</sub> NO <sub>3</sub>	9651	

Rivastigmine

C<sub>14</sub>H<sub>22</sub>N<sub>2</sub>O<sub>2</sub>

77991



**RESULTS AND DISCUSSION**

*C. zeylanicum* Blume essential oil has 23 phytochemical compounds which represent at 99.39% of total essential oil. The main phytochemical compounds were cinnamaldehyde, *trans*-cinnamyl acetate, 1,4-benzenedicarboxylic acid, 1,8-cineole,  $\alpha$ -pinene, coumaric acid, and 9-octadecenoic acid (Kallel *et al.*, 2019). Phytochemicals were screened before docking. Five phytochemical compounds were selected for molecular docking analysis. Due to Lipinski's rule of five, all of the ligands have zero violations, but 1,4-

benzene dicarboxylic acid and 9-Octadecenoic acid can't cross the BBB, so the ligands are excluded (Table 2). Lipinski's rule of five terms consists of molecular weight <500 Da, log P <5, H-bond donor <5, and H-bond acceptor <10. Lipinski's rule of five can be used to classify the phytochemicals that may be effective in being used as drugs (Lipinski *et al.*, 2001). The compound must cross the BBB because the target receptor being inhibited is located in the brain (Pardridge, 2020).

**Table 2. ADME Analysis of *C. zeylanicum* Blume Essential Oil Compound**

Phytochemical Compound	Lipinski's Rule of Five					Violations	BBB Permeant
	Molecular Weight	Log P	H-bond donor	H-bond acceptor			
Cinnamaldehyde	132,16	1,97	0	1	0	Yes	
<i>Trans</i> -cinnamyl acetate	176,21	2,33	0	2	0	Yes	
1,4-benzenedicarboxylic acid	166,13	1,13	2	4	0	No	
1,8-cineole	154,25	2,67	0	1	0	Yes	
$\alpha$ -pinene	136,23	3,44	0	0	0	Yes	
Coumaric acid	164,16	1,26	2	3	0	Yes	
9-Octadecenoic acid	282,46	5,71	1	2	0	No	

AChE (PDB ID: 7E3H) is a cholinergic enzyme primarily found at postsynaptic neuromuscular junctions (NMJ). AChE immediately breaks down acetylcholine into acetic acid and choline in NMJ. AChE plays a critical role in AD (Trang and Khandhar, 2022). 7E3H has a total structure Weight of 120.37 kDa, an Atom Count of 8437, a Modeled Residue Count of 1054, a Deposited Residue Count of 1080, and one Unique protein chain. This receptor has a native ligand E20 that binds to 14 active site amino acids namely TYR072, ASP074, GLU202, TRP286, VAL294, PHE295, PHE338, TYR341, SER293, TRP086, GLY121, TYR124, TYR337, and HIS447. BACE1 (PDB ID: 4DJU) is a  $\beta$ -secretase involved in the  $\beta$ -amyloid peptide, a dominant component in AD (Gnanaraj *et al.*, 2022). 4DJU has a total structure Weight of 93.33 kDa, an Atom Count of 7068, a Modeled Residue Count of 779, a Deposited Residue Count of 828, and one Unique protein chain. This receptor has a native ligand 0KK that binds to 13 active site amino acids namely LEU091, ASP093, GLY095, SER096, VAL130, TYR132, TRP137, PHE169, TRP176, ILE179, ASP289, GLY291, and THR292 (Cumming *et al.*, 2012). GSK3 (PDB ID: 1Q5K) is a serine/threonine protein kinase that phosphorylates Tau protein, whose expression is associated with AD (Gnanaraj *et al.*, 2022). 1Q5K has a total structure Weight of 92.85 kDa, an Atom Count of 5930, a Modeled Residue Count of 689, a Deposited Residue Count of 828, and one Unique protein chain. This receptor has a native ligand TMU that binds to 12 active

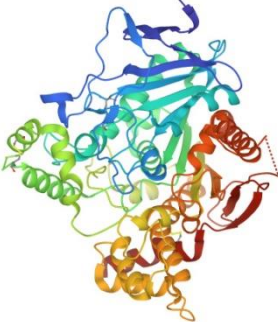

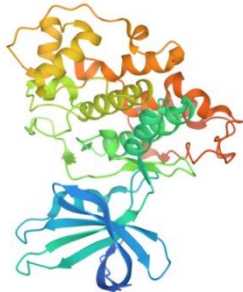
site amino acids, namely VAL061, ILE062, VAL070, ALA083, VAL135, PRO136, LEU132, LEU188, ASP133, TYR134, ARG141, and CYS199 (Bhat *et al.*, 2003). Table 3 also visualizes the 3D structure of each receptor at 2.45, 1.80, and 1.94 Å X-ray diffraction resolutions.

Based on the results of molecular docking, the phytochemical of *C. zeylanicum* Blume essential oil that has the strongest binding energy to the AChE receptor is *trans*-cinnamyl acetate ( $\Delta G$ : -6.21 kcal/mol) with an inhibition constant of 27.84  $\mu$ M. The other compounds,  $\alpha$ -pinene, 1,8-cineole, coumaric acid, and cinnamaldehyde had binding energies of -5.96, -5.86, -5.76, and -5.50 kcal/mol with inhibition constants of 43.49, 50.56, 60.95, and 92.75  $\mu$ M. In the AD drug, donepezil, galantamine, and rivastigmine had binding energies of -11.04, -8.05, and -7.29 kcal/mol with inhibition constants of 8.15 nM, 1.26  $\mu$ M, and 4.52  $\mu$ M. At the BACE1 receptor, the phytochemical of *C. zeylanicum* Blume essential oil with the strongest binding energy is *trans*-cinnamyl acetate ( $\Delta G$ : -5.16 kcal/mol) with an inhibitory constant of 164.04  $\mu$ M. The other compounds, cinnamaldehyde, coumaric acid,  $\alpha$ -pinene, and 1,8-cineole have binding energies of -4.62, -4.51, -4.43, and -4.41 kcal/mol with inhibition constants of 408.29, 496.02, 567.62, and 583.24  $\mu$ M. . Meanwhile, donepezil, galantamine, and rivastigmine had binding energies of -8.50, -6.08, and -5.89 kcal/mol with inhibition constants of 588.62 nM, 34.99  $\mu$ M, and 47.78  $\mu$ M. At the GSK-3 receptor, the phytochemical of *C. zeylanicum* Blume essential oil with the strongest

binding energy is coumaric acid ( $\Delta G$ : -5.61 kcal/mol) with an inhibition constant of 77.41  $\mu\text{M}$ . Other compounds, *trans*-cinnamyl acetate, 1,8-cineole, cinnamaldehyde, and  $\alpha$ -pinene have binding energies of -5.47, -4.93, -4.92, and -4.83 kcal/mol with inhibition

constants of 97.51, 245.32, 248.03, and 287.66  $\mu\text{M}$ . Meanwhile, donepezil, galantamine, and rivastigmine had binding energies of -8.53, -6.29, and -6.22 kcal/mol with inhibition constants of 562.75 nM, 24.71  $\mu\text{M}$ , and 27.74  $\mu\text{M}$  (Table 4).

**Table 3. Protein Target Associated with Alzheimer’s Disease**

Protein Target	PDB ID	Active Site	3D Structure
<b>AChE</b> <b>(Acetylcholinesterase)</b> Crystal structure of human acetylcholinesterase in complex with donepezil	7E3H	TYR072, ASP074, TRP086, GLY121, TYR124, GLU202, TRP286, SER293, VAL294, PHE295, TYR337, PHE338, TYR341, HIS447	
<b>BACE1</b> <b>(<math>\beta</math>-site APP Cleaving Enzyme 1)</b> Structure of BACE bound to 2-imino-3-methyl-5,5-diphenylimidazolidin-4-one	4DJU	LEU091, ASP093, GLY095, SER096, VAL130, TYR132, TRP137, PHE169, TRP176, ILE179, ASP289, GLY291, THR292	
<b>GSK-3</b> <b>(Glycogen Synthase Kinase-3)</b> crystal structure of glycogen synthase kinase 3 in complexed with inhibitor	1Q5K	VAL061, ILE062, VAL070, ALA083, LEU132, ASP133, TYR134, VAL135, PRO136, ARG141, LEU188, CYS199	

The molecular docking results are then visualized in Table 5. At the AChE receptor, the phytochemical of *C. zeylanicum* Blume essential oil, cinnamaldehyde binds to 9 amino acids with 7 van der Waals bonds, *trans*-cinnamyl acetate binds to 8 amino acids with 4 van der Waals bonds, 1,8-cineole binds to 8 amino acids with 2 van der Waals bonds,  $\alpha$ -pinene binds to 8 amino acids with 4 van der Waals bonds, and coumaric acid binds to 12 amino acids with 9 van der Waals bonds. In the AD drug, donepezil binds to 14 amino acids with 6 van der Waals bonds, galantamine binds to 10 amino acids with 6 van der Waals bonds, and rivastigmine binds to 11 amino acids with 5 van der Waals bonds. At the BACE1 receptor, it was found that cinnamaldehyde binds to 11

amino acids with 5 van der Waals bonds, *trans*-cinnamyl acetate binds to 10 amino acids with 6 van der Waals bonds, 1,8-cineole binds to 7 amino acids with 2 van der Waals bonds,  $\alpha$ -pinene binds to 7 amino acids with 3 van der Waals bonds, and coumaric acid binds to 10 amino acids with 7 van der Waals bonds. Meanwhile, donepezil binds to 17 amino acids with 11 van der Waals bonds, galantamine binds to 12 amino acids with 6 van der Waals bonds, and rivastigmine binds to 11 amino acids with 1 van der Waals bond. At the GSK-3 receptor, it was found that cinnamaldehyde binds to 12 amino acids with 8 van der Waals bonds, *trans*-cinnamyl acetate binds to 14 amino acids with 9 van der Waals bonds, 1,8-cineole binds to 9 amino acids with 3

van der Waals bonds,  $\alpha$ -pinene binds to 11 amino acids with 4 van der Waals bonds, and coumaric acid binds to 11 amino acids with 5 van der Waals bonds. Meanwhile, donepezil binds to 12 amino acids with 6 van der Waals

bonds, galantamine binds to 12 amino acids with 7 van der Waals bonds, and rivastigmine binds to 11 amino acids with 5 van der Waals bonds.

**Table 4. Molecular Docking Results of Ligands with The Protein Target**

Ligand	Protein Targets Associated With Alzheimer's Disease					
	AChE		BACE1		GSK-3	
	$\Delta G$ (kcal/mol)	$K_i$	$\Delta G$ (kcal/mol)	$K_i$	$\Delta G$ (kal/mol)	$K_i$
<i>C. zeylanicum</i> blume essential oil						
Cinnamaldehyde	-5.50	92.75 $\mu M$	-4.62	408.29 $\mu M$	-4.92	248.03 $\mu M$
<i>Trans</i> -cinnamyl acetate	-6.21	27.84 $\mu M$	-5.16	164.04 $\mu M$	-5.47	97.51 $\mu M$
1,8-cineole	-5.86	50.56 $\mu M$	-4.41	583.24 $\mu M$	-4.93	245.32 $\mu M$
$\alpha$ -pinene	-5.96	43.49 $\mu M$	-4.43	567.62 $\mu M$	-4.83	287.66 $\mu M$
Coumaric acid	-5.76	60.95 $\mu M$	-4.51	496.02 $\mu M$	-5.61	77.41 $\mu M$
Alzheimer's disease drug						
Donepezil	-11.04	8.15 nM	-8.50	588.62 nM	-8.53	562.75 nM
Galantamine	-8.05	1.26 $\mu M$	-6.08	34.99 $\mu M$	-6.29	24.71 $\mu M$
Rivastigmine	-7.29	4.52 $\mu M$	-5.89	47.78 $\mu M$	-6.22	27.74 $\mu M$

Cinnamaldehyde is a natural flavonoid and derivative compound of the volatile chemical aldehyde that gives cinnamon its flavour and aroma. Cinnamaldehyde is the main phytochemical compound of *C. zeylanicum* Blume essential oil (about 90%) (National Center for Biotechnology Information, 2022c). It has many pharmacological activities, including anti-inflammatory, antimicrobial, anti-cancer, anti-hyperglycemic, and neuroprotection (Zhang *et al.*, 2015). *Trans*-cinnamyl acetate is an acetate ester produced from the condensation of cinnamyl alcohol and acetic acid (National Center for Biotechnology Information, 2022b). Its pharmacological properties include anti-inflammatory, anticancer, antioxidant, antimicrobial, antidiabetic, anti-anxiety, antidepressant, and neuroprotection (Rao and Gan, 2014). Cinnamaldehyde and *trans*-cinnamyl acetate have neuroprotection functions that can be used for preventive and therapeutic nervous system diseases (Zhang *et al.*, 2015). 1,8-Cineole (eucalyptol) is an achiral aromatic component of many plants, including *C. Zeylanicum* (National Center for Biotechnology Information, 2022a). 1,8-Cineole has potential pharmacological properties such as anti-inflammatory, antioxidative, anti-cancer, and neuroprotection. 1,8-Cineole was used to treat nervous system disease (Cai *et al.*, 2020).  $\alpha$ -pinene is an organic compound of the polyphenolic group terpene and a component of many aromatic, dietary plants, such as *C. Zeylanicum* (National Center for Biotechnology Information, 2022e).  $\alpha$ -pinene has pharmacological properties such as antinociceptive and antioxidant.  $\alpha$ -pinene has a strong anti-inflammatory effect in some pathological

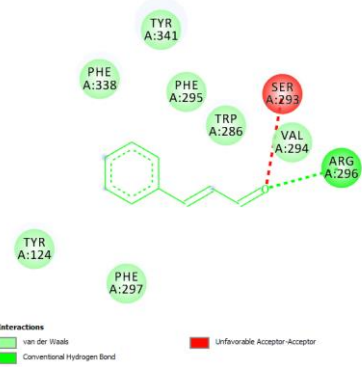
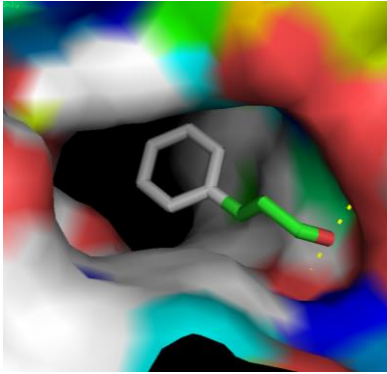
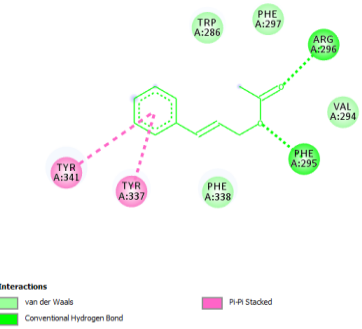
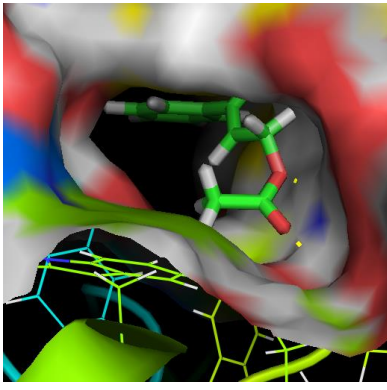
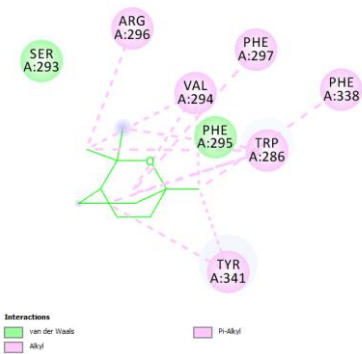
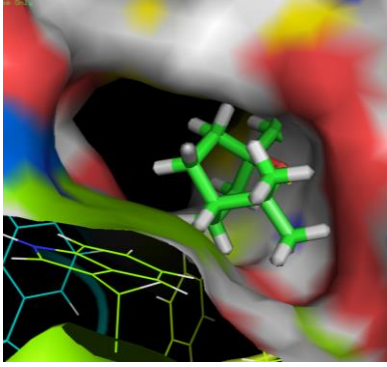
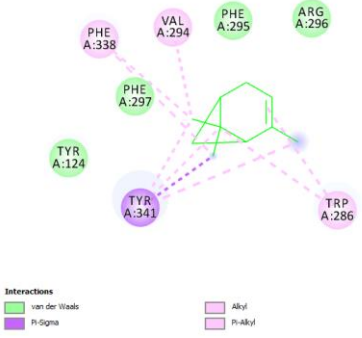
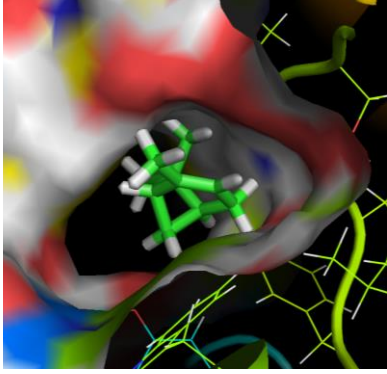
conditions.  $\alpha$ -pinene also has neuroprotective effect, and it's capable of restoring BBB function and attenuating sensorimotor dysfunctions (Khoshnazar, Parvardeh and Bigdeli, 2020). Coumaric acid is derivative of cinnamic acid mono-hydroxylated at the phenyl group. It is the most abundant isoform and is found at significant levels in many plants, including *C. Zeylanicum* (National Center for Biotechnology Information, 2022d). Coumaric acid has many pharmacological activities, including anti-inflammatory, antidiabetes, antibacterial, hepatoprotective, nephronprotective, and neuroprotective. It has high free radical scavenging. Coumaric acid has potential preventive and therapeutic value for memory-impaired individuals, especially age-related memory-impaired in older people (Ferreira *et al.*, 2018).

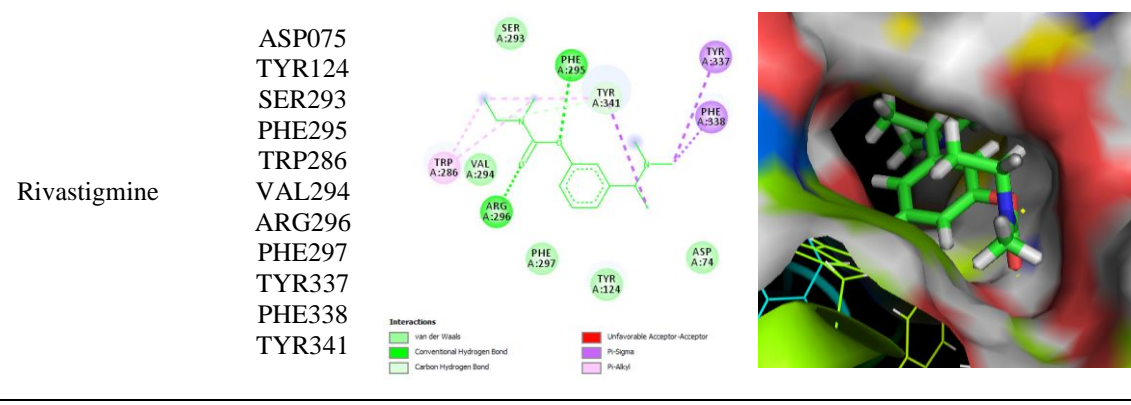
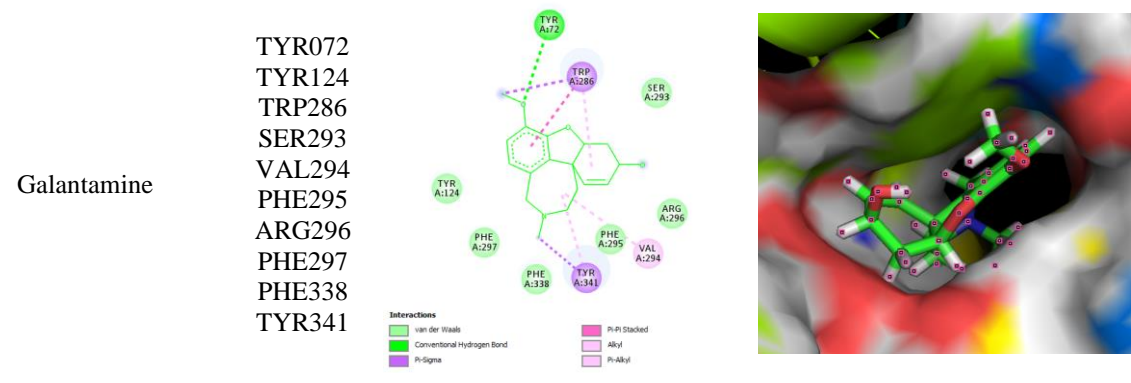
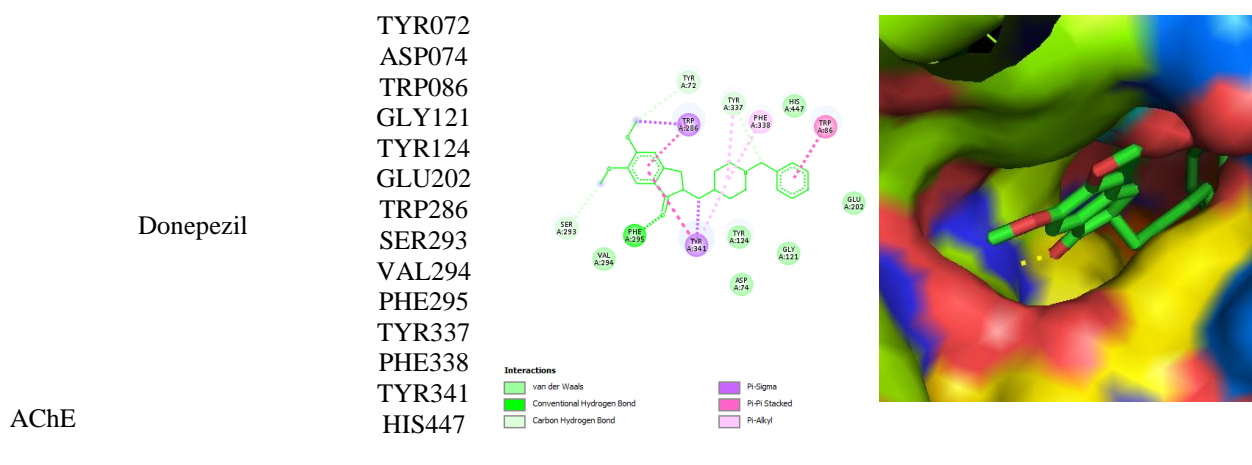
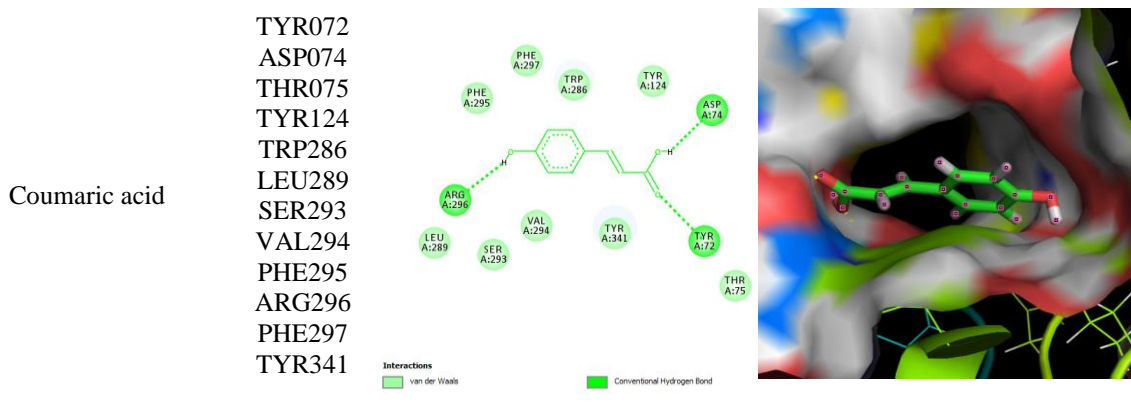
*C. zeylanicum* Blume essential oil, with its main phytochemical compound cinnamaldehyde, *trans*-cinnamyl acetate, 1,8-cineole,  $\alpha$ -pinene, and coumaric acid, has a neuroprotective potential function. It was shown from molecular docking that it can inhibit the receptors AChE, BACE, and GSK-3. It prevents acetylcholinesterase from breaking down acetylcholine. The higher concentration of acetylcholine leads to better communication between nerve cells in the brain and may ease some symptoms of AD (Kumar *et al.*, 2022). *C. zeylanicum* Blume essential oil also prevents the accumulation and formation of A $\beta$  plaques and neurofibrillary tangles in neurons from preventing neurotoxicity and worsening AD. It has an antioxidant, neurostimulator function, and prevents neuronal loss (Hamidpour *et al.*, 2015; Momtaz *et al.*, 2017; Hajinejad *et al.*, 2020).

**Table 5. Visualization of Ligands with The Protein Target**

Protein	Ligand	Amino Acid Bond	2D Visualization	3D Visualization
---------	--------	-----------------	------------------	------------------



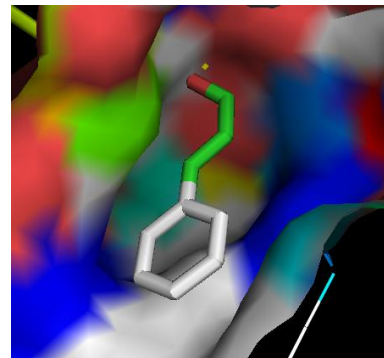
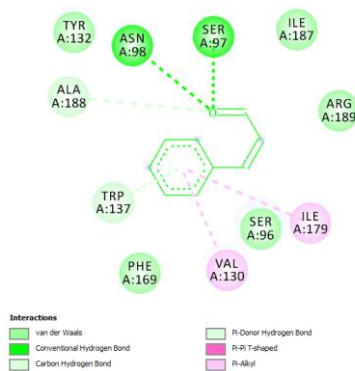
Cinnamaldehyde	TYR124 TRP286 SER293 VAL294 PHE295 ARG296 PHE297 PHE338 TYR341			
<i>Trans</i> -cinnamyl acetate	TRP286 VAL294 PHE295 PHE297 ARG296 TYR337 PHE338 TYR341			
AChE				
1,8-cineole	TRP286 SER293 VAL294 PHE295 ARG296 PHE297 PHE338 TYR341			
$\alpha$ -pinene	TYR124 TRP286 VAL294 PHE295 ARG296 PHE297 PHE338 TYR341			
Protein	Ligand	Amino Acid Bone	2D Visualization	3D Visualization





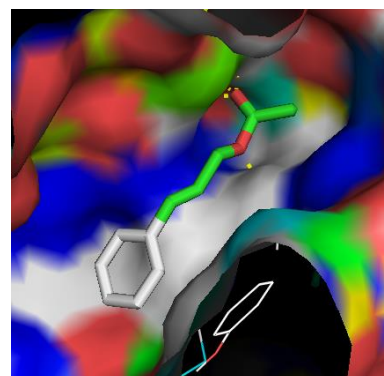
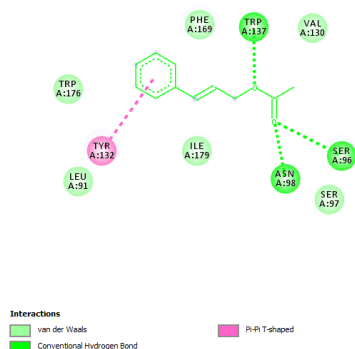
Cinnamaldehyde

SER096  
SER097  
ASN098  
VAL130  
TYR132  
TRP137  
PHE169  
ILE179  
ILE187  
ALA188  
ARG189



*Trans*-cinnamyl acetate

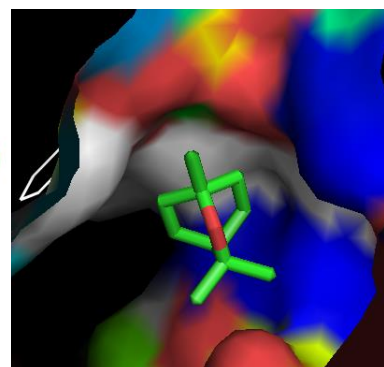
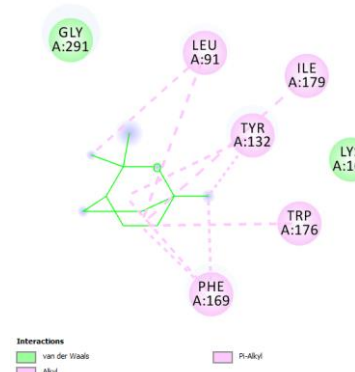
LEU091  
SER096  
SER097  
ASN098  
VAL130  
TYR132  
TRP137  
PHE169  
TRP176  
ILE179



BACE1

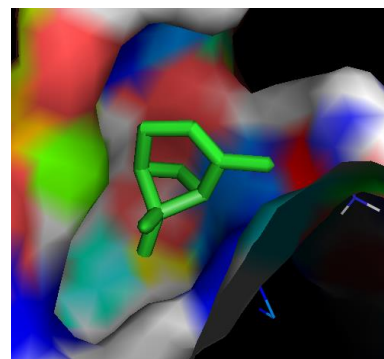
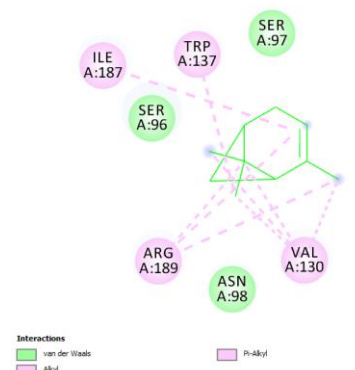
1,8-cineole

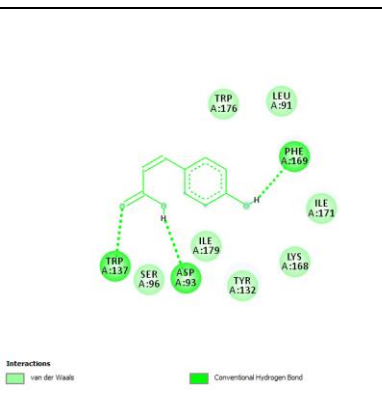
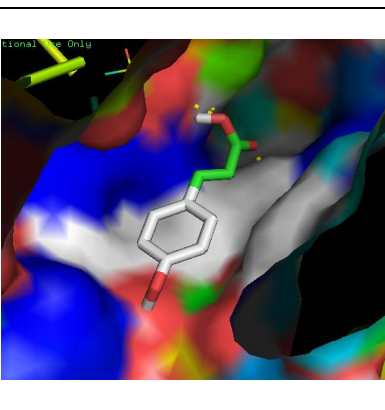
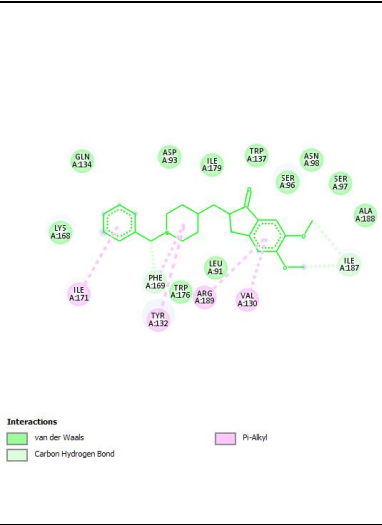
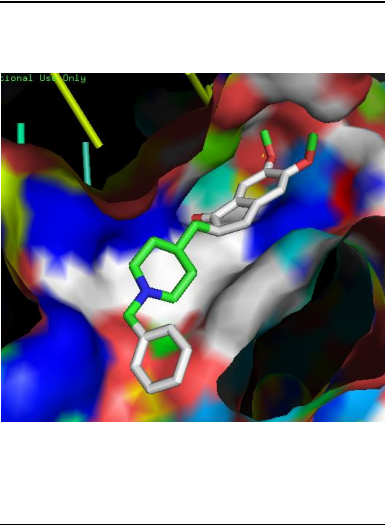
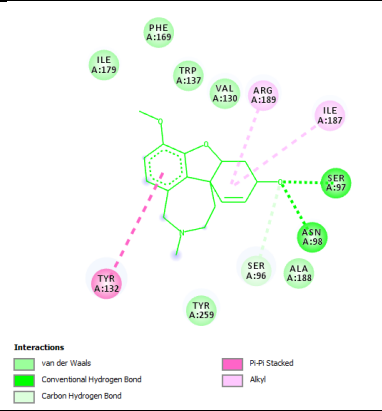
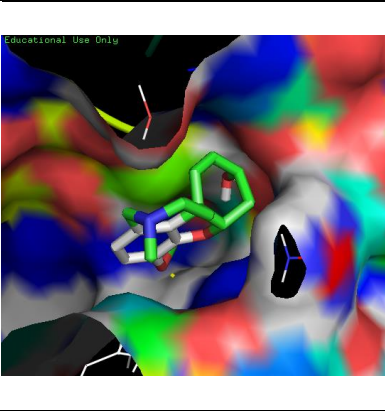
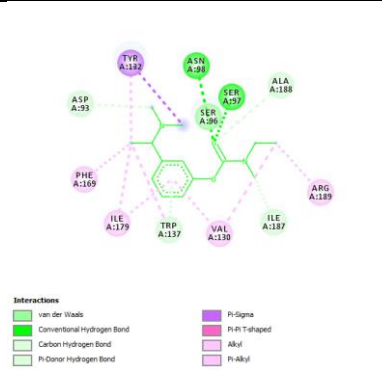
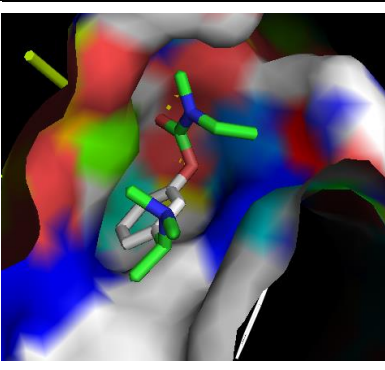
LEU091  
TYR132  
LYS168  
PHE169  
TRP176  
ILE179  
GLY291



$\alpha$ -pinene

SER096  
SER097  
ASN098  
VAL130  
TRP137  
ILE187  
ARG189



	Coumaric acid	LEU091 ASP093 SER096 TYR132 TRP137 LYS168 PHE169 ILE171 TRP176 ILE179		
BACE1	Donepezil	LEU091 ASP093 SER096 SER097 ASN098 VAL130 TYR132 GLN134 TRP137 LYS168 PHE169 ILE171 TRP176 ILE179 ILE187 ALA188 ARG189		
	Galantamine	ASP093 SER096 SER097 ASN098 VAL130 TYR132 TRP137 PHE169 ILE179 ILE187 ALA188 ARG189		
	Rivastigmine	ASP075 TYR124 SER293 PHE295 TRP286 VAL294 ARG296 PHE297 TYR337 PHE338 TYR341		

Protein

Ligand

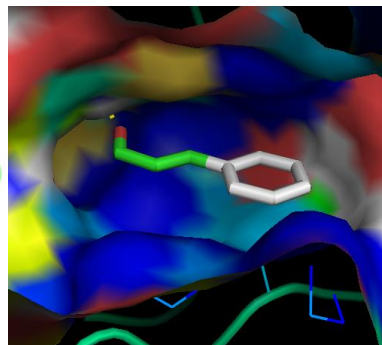
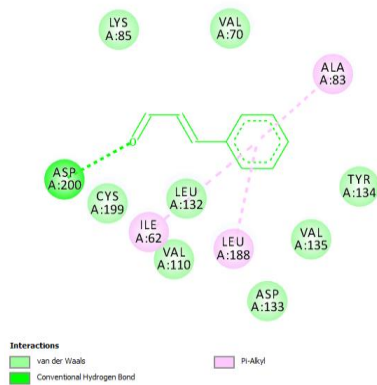
Amino  
Acid Bone

2D Visualization

3D Visualization

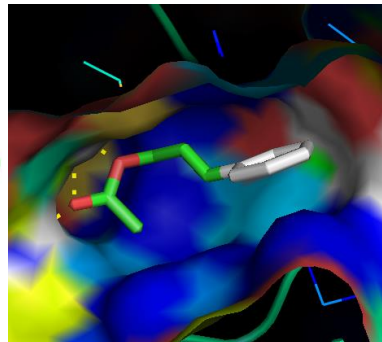
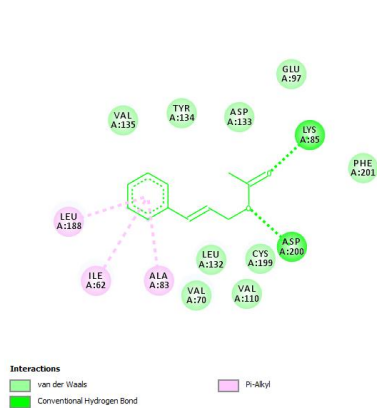
Cinnamaldehyde

ILE062  
VAL070  
ALA083  
LYS085  
VAL110  
ASP133  
LEU132  
TYR134  
VAL135  
LEU188  
CYS199  
ASP200



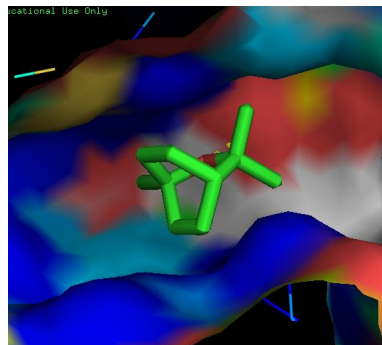
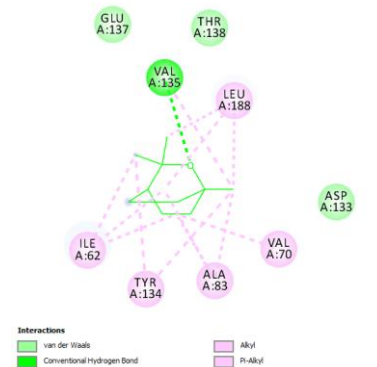
*Trans*-cinnamyl acetate

ILE062  
VAL070  
ALA083  
LYS085  
GLU097  
VAL110  
LEU132  
ASP133  
TYR134  
VAL135  
LEU188  
CYS199  
ASP200  
PHE201



GSK-3

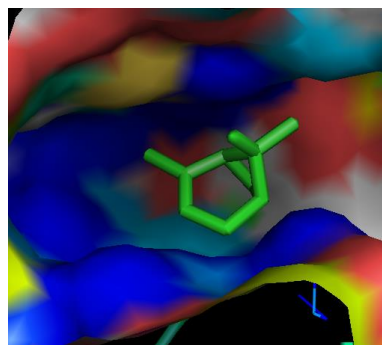
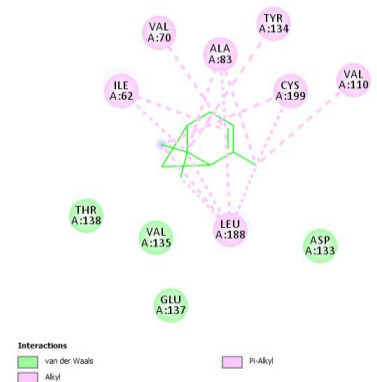
ILE062  
ALA083  
VAL070  
ASP133  
TYR134  
VAL135  
GLU137  
THR138  
LEU188



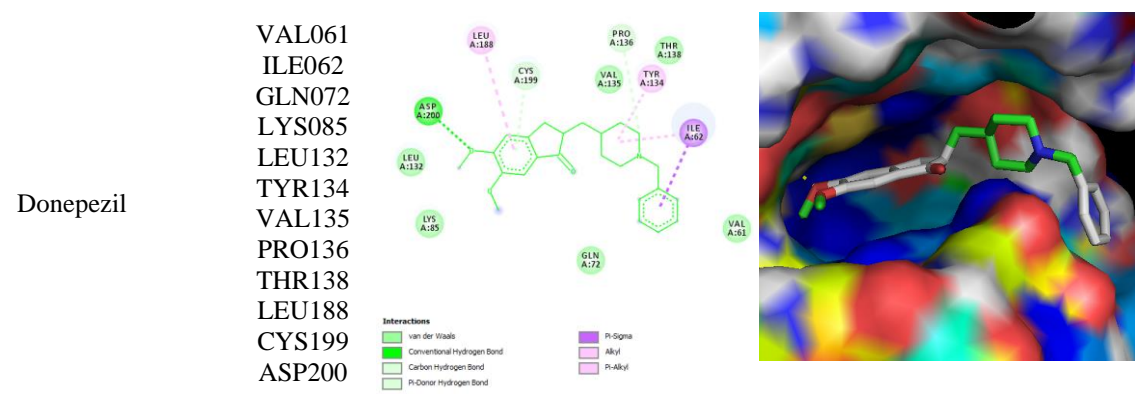
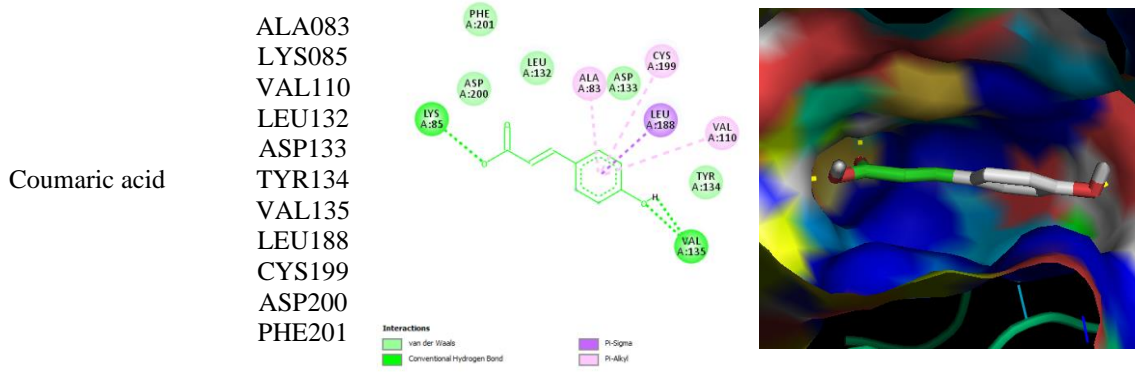
1,8-cineole

$\alpha$ -pinene

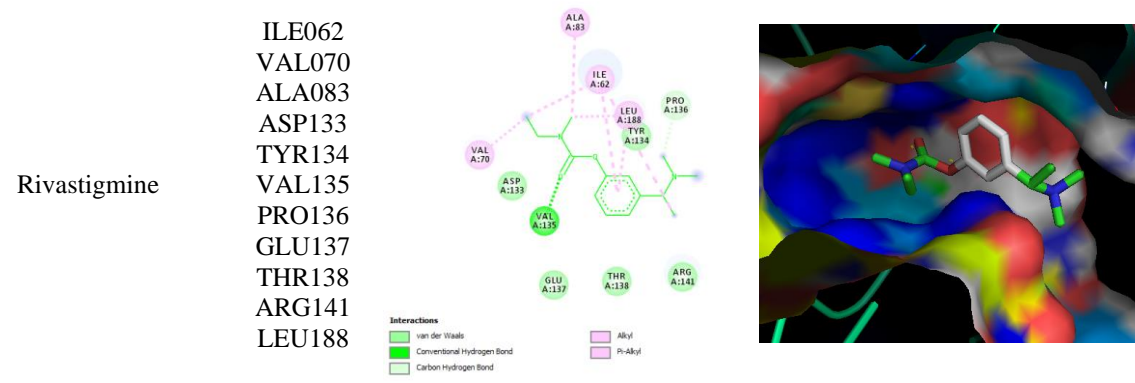
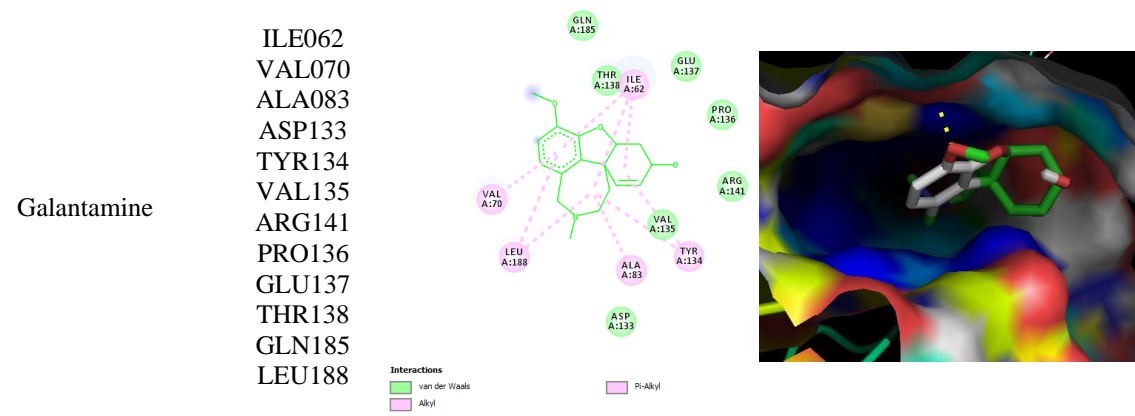
ILE062  
VAL070  
ALA083  
VAL110  
ASP133  
TYR134  
VAL135  
THR138  
GLU137  
LEU188  
CYS199







GSK-3



**CONCLUSION**

Molecular Docking of main phytochemical compounds of *C. zeylanicum* Blume essential oil has potency as Alzheimer's disease drug. These findings implicated that compounds could actively block the

acetylcholinesterase, glycogen synthase kinase-3, and  $\beta$  site APP cleaving enzyme 1 activity. The best interaction model of the compound was shown by *trans*-cinnamyl acetate and coumaric acid. The binding energy of the compounds is lower than AD drugs (donepezil,

rivastigmine, galantamine), but it is not significantly different from AD drugs to inhibit the receptors. Our study can be used as the basis for conducting further research. In vitro studies, in vivo studies, pharmacokinetics and bioavailability of compounds, compound structures, and structure-activity relationships are needed to ensure the potency of *C. zeylanicum* Blume essential oil as an AD drug.

#### ACKNOWLEDGMENTS

All authors contributed to this study. This Study was supported by the Departemen of Physiology and Medical Biochemistry and Department of Neurology, Faculty of Medicine, Universitas Airlangga, Indonesia.

#### CONFLICT OF INTEREST

There are no potential conflicts of interest to declare in this study.

#### REFERENCES

- Alzheimer's Disease International and Alzheimer's Australia (2014) *Dementia in the Asia Pacific Region*, Alzheimer's Disease International. London: Alzheimer's Disease International. Available at: <https://www.alz.co.uk/dementia-in-the-asia-pacific>.
- Bhat, R. *et al.* (2003) 'Structural Insights and Biological Effects of Glycogen Synthase Kinase 3-specific Inhibitor AR-A014418', *Journal of Biological Chemistry*, 278(46), pp. 45937–45945. doi: 10.1074/jbc.M306268200.
- Cai, Z. M. *et al.* (2020) '1,8-Cineole: a review of source, biological activities, and application', *Journal of Asian Natural Products Research*. Taylor & Francis. doi: 10.1080/10286020.2020.1839432.
- Cassani, R. *et al.* (2018) 'Systematic review on resting-state EEG for Alzheimer's disease diagnosis and progression assessment', *Disease Markers*, 2018, pp. 1–26. doi: 10.1155/2018/5174815.
- Cortes-Canteli, M. and Iadecola, C. (2020) 'Alzheimer's Disease and Vascular Aging', *Journal of the American College of Cardiology*, 75(8), pp. 942–951. doi: 10.1016/j.jacc.2019.10.062.
- Cumming, J. N. *et al.* (2012) 'Structure based design of iminohydantoin BACE1 inhibitors: Identification of an orally available, centrally active BACE1 inhibitor', *Bioorganic and Medicinal Chemistry Letters*. Elsevier Ltd, 22, pp. 2444–2449. doi: 10.1016/j.bmcl.2012.02.013.
- Dá Mesquita, S. *et al.* (2016) 'Insights on the pathophysiology of Alzheimer's disease: The crosstalk between amyloid pathology, neuroinflammation and the peripheral immune system', *Neuroscience and Biobehavioral Reviews*. Elsevier Ltd. doi: 10.1016/j.neubiorev.2016.06.014.
- Dhage, P. A. *et al.* (2021) 'Leveraging hallmark Alzheimer's molecular targets using phytoconstituents: Current perspective and emerging trends', *Biomedicine and Pharmacotherapy*. Elsevier Masson SAS, 139, pp. 1–16. doi: 10.1016/j.biopha.2021.111634.
- Fahad, M. *et al.* (2018) 'Medicinal and pharmacological role of traditional Asian Food condiment: *Cinnamomum zeylanicum* Blume', *International Journal of Advances in Pharmacy Medicine and Bioallied Sciences*, 6(1), pp. 22–30. Available at: <http://biomedjournal.com/medicinal-and-pharmacological-role-of-traditional-asian-food-condiment-cinnamomum-zeylanicum-blume/>.
- Fan, L. *et al.* (2020) 'New Insights into the Pathogenesis of Alzheimer's Disease', *Frontiers in Neurology*, 10, pp. 1–12. doi: 10.3389/fneur.2019.01312.
- Ferreira, P. S. *et al.* (2018) 'A Review of Analytical Methods for p-Coumaric Acid in Plant-Based Products, Beverages, and Biological Matrices', *Critical Reviews in Analytical Chemistry*. Taylor & Francis. doi: 10.1080/10408347.2018.1459173.
- Frydman-Marom, A. *et al.* (2011) 'Orally administered cinnamon extract reduces  $\beta$ -amyloid oligomerization and corrects cognitive impairment in Alzheimer's disease animal models', *PLoS ONE*, 6(1), pp. 1–11. doi: 10.1371/journal.pone.0016564.
- Gnanaraj, C. *et al.* (2022) 'In Silico Molecular Docking Analysis of Karanjin against Alzheimer's and Parkinson's Diseases as a Potential Natural Lead Molecule for New Drug Design, Development and Therapy', *Molecules*, 27. doi: 10.3390/molecules27092834.
- Hajinejad, M. *et al.* (2020) 'Natural Cinnamaldehyde and Its Derivatives Ameliorate Neuroinflammatory Pathways in Neurodegenerative Diseases', *BioMed Research International*. doi: 10.1155/2020/1034325.
- Hamidpour, R. *et al.* (2015) 'Cinnamon from the selection of traditional applications to its novel effects on the inhibition of angiogenesis in cancer cells and prevention of Alzheimer's disease, and a series of functions such as antioxidant, anticholesterol, antidiabetes, antibacteri', *Journal of Traditional and Complementary Medicine*, 5, pp. 66–70. doi: 10.1016/j.jtcme.2014.11.008.
- Jagust, W. (2018) 'Imaging the evolution and pathophysiology of Alzheimer disease', *Nature Reviews Neuroscience*. Springer US, 19, pp. 687–700. doi: 10.1038/s41583-018-0067-3.
- Kallel, I. *et al.* (2019) 'Optimization of Cinnamon (*Cinnamomum zeylanicum* Blume) Essential Oil Extraction: Evaluation of Antioxidant and Antiproliferative Effects', *Evidence-based Complementary and Alternative Medicine*. Hindawi, 2019. doi: 10.1155/2019/6498347.
- Kareti, S. R. and Pharm, S. M. (2020) 'In Silico Molecular Docking Analysis of Potential Anti-Alzheimer's Compounds Present in Chloroform Extract of *Carissa carandas* Leaf Using Gas Chromatography MS/MS', *Current Therapeutic*



- Research. Elsevier Inc., 93. doi: 10.1016/j.curtheres.2020.100615.
- Khaerunnisa, S., Suhartati and Awaluddin, R. (2020) *Penelitian in silico untuk pemula*. Surabaya: Airlangga University Press.
- Khoshnazar, M., Parvardeh, S. and Bigdeli, M. R. (2020) 'Alpha-pinene exerts neuroprotective effects via anti-inflammatory and anti-apoptotic mechanisms in a rat model of focal cerebral ischemia-reperfusion', *Journal of Stroke and Cerebrovascular Diseases*. Elsevier Inc., 29(8). doi: 10.1016/j.jstrokecerebrovasdis.2020.104977.
- Kumar, A. et al. (2022) 'Alzheimer Disease', in *In: StatPearls [Internet]*. Treasure Island (FL): StatPearls Publishing. Available at: <https://www.ncbi.nlm.nih.gov/books/NBK499922/>.
- Kumar, Shivani, Chowdhury, S. and Kumar, Suresh (2017) 'In silico repurposing of antipsychotic drugs for Alzheimer's disease', *BMC Neuroscience*. BioMed Central, 18(76), pp. 1–16. doi: 10.1186/s12868-017-0394-8.
- Lipinski, C. A. et al. (2001) 'Experimental and computational approaches to estimate solubility and permeability in drug discovery and development settings', 46, pp. 3–26. Available at: <https://pubmed.ncbi.nlm.nih.gov/11259830/>.
- Litke, R. et al. (2021) 'Modifiable Risk Factors in Alzheimer Disease and Related Dementias: A Review', *Clinical Therapeutics*. Elsevier Inc., 43(6), pp. 953–965. doi: 10.1016/j.clinthera.2021.05.006.
- Long, J. M. and Holtzman, D. M. (2019) 'Alzheimer Disease: An Update on Pathobiology and Treatment Strategies', *Cell*. Elsevier Inc., 179(3), pp. 312–339. doi: 10.1016/j.cell.2019.09.001.
- Lucey, B. P. (2020) 'It's complicated: The relationship between sleep and Alzheimer's disease in humans', *Neurobiology of Disease*. Elsevier, 144, pp. 1–8. doi: 10.1016/j.nbd.2020.105031.
- Momtaz, S. et al. (2017) 'Cinnamon, a promising prospect towards Alzheimer's disease', *Pharmacological Research*. Elsevier Ltd. doi: 10.1016/j.phrs.2017.12.011.
- National Center for Biotechnology Information (2022a) 'PubChem Compound Summary for CID 2758, Eucalyptol'. Available at: <https://pubchem.ncbi.nlm.nih.gov/compound/Eucalyptol>.
- National Center for Biotechnology Information (2022b) 'PubChem Compound Summary for CID 5282110, Cinnamyl acetate'. Available at: <https://pubchem.ncbi.nlm.nih.gov/compound/Cinnamyl-acetate>.
- National Center for Biotechnology Information (2022c) 'PubChem Compound Summary for CID 637511, Cinnamaldehyde'. Available at: <https://pubchem.ncbi.nlm.nih.gov/compound/Cinnamaldehyde>.
- National Center for Biotechnology Information (2022d) 'PubChem Compound Summary for CID 637542, 4-Hydroxycinnamic acid'. Available at: <https://pubchem.ncbi.nlm.nih.gov/compound/4-Hydroxycinnamic-acid>.
- National Center for Biotechnology Information (2022e) 'PubChem Compound Summary for CID 6654, alpha-Pinene'. Available at: <https://pubchem.ncbi.nlm.nih.gov/compound/alpha-Pinene>.
- Ong, P. A. et al. (2021) 'Dementia Prevalence, Comorbidities, and Lifestyle Among Jatinangor Elders', *Frontiers in Neurology*, 12. doi: 10.3389/fneur.2021.643480.
- Pardridge, W. M. (2020) 'Treatment of alzheimer's disease and blood-brain barrier drug delivery', *Pharmaceuticals*, 13, pp. 1–25. doi: 10.3390/ph13110394.
- Rao, P. V. and Gan, S. H. (2014) 'Cinnamon: A multifaceted medicinal plant', *Evidence-based Complementary and Alternative Medicine*, pp. 1–12. doi: 10.1155/2014/642942.
- Samanta, S., Ramesh, M. and Govindaraju, H. (2022) 'Chapter 1: Alzheimer's is a Multifactorial Disease', in *Alzheimer's Disease: Recent Findings in Pathophysiology, Diagnostic and Therapeutic Modalities*, pp. 1–34. doi: 10.1039/9781839162732-00001.
- Tepe, A. S. and Ozaslan, M. (2020) 'Anti-Alzheimer, anti-diabetic, skin-whitening, and antioxidant activities of the essential oil of Cinnamomum zeylanicum', *Industrial Crops and Products*. Elsevier, 145, pp. 1–8. doi: 10.1016/j.indcrop.2019.112069.
- Tiwari, S. et al. (2019) 'Alzheimer's disease: pathogenesis, diagnostics and therapeutics', *International Journal of Nanomedicine*, Jul 2019(14), pp. 5541–5554. doi: 10.2147/IJN.S200490.
- Trang, A. and Khandhar, P. B. (2022) 'Physiology, Acetylcholinesterase', in *StatPearls [Internet]*. Treasure Island (FL): StatPearls Publishing. Available at: <https://www.ncbi.nlm.nih.gov/books/NBK539735/>.
- Vinicius, M. et al. (2019) 'Alzheimer's disease: risk factors and potentially protective measures', *Journal of Biomedical Science*. Journal of Biomedical Science, 26(33), pp. 1–11. Available at: <https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6507104/>.
- Wang, J. et al. (2020) 'Traditional uses, phytochemistry and pharmacological activities of the genus Cinnamomum (Lauraceae): A review', *Fitoterapia*. Elsevier, 146. doi: 10.1016/j.fitote.2020.104675.
- Yiannopoulou, K. G. and Papageorgiou, S. G. (2020) 'Current and Future Treatments in Alzheimer Disease: An Update', *Journal of Central Nervous System Disease*, 12, pp. 1–12. doi:

10.1177/1179573520907397.

Zhang, L. *et al.* (2015) 'Research progress of trans-cinnamaldehyde pharmacological effects', *China Journal of Chinese Materia Medica*, 40(23), pp. 4568–4572. Available at: <https://pubmed.ncbi.nlm.nih.gov/27141665/>.