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Molecular docking of various bioactive compounds from essential oil of *Trachyaspermum ammi* against the fungal enzyme Candidapepsin-1

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ABSTRACT

The bioactive compounds from essential oil of *Trachyaspermum ammi* using gas chromatography–mass spectrometry and their inhibition potential against the enzyme Candidapepsin-1 were studied. The research work focuses on the molecular simulation of bioactive compounds against the enzyme that acts as a potential drug target and support the drug discovery process. Candidapepsin-1 has been reported to be the cause for biofilm formation, superficial skin infections, and oral infections. Fifteen active compounds and their interactions with Candidapepsin-1 were studied in this research work. The compounds satisfied Lipinski's rule of five in order to be used as an oral drug. ADMET properties of the compounds used to determine pharmacodynamic and pharmacokinetic properties which were reported in the study. The compounds were docked against the enzyme with the help of AutoDock 4.2.6 software. Ligustilide has the lowest free binding energy of −5.75 kcal/mol against the Candidapepsin-1 with three hydrogen bond interactions at Ile 223, Tyr 225, and Thr 222 at the active site of the enzyme followed by cedrane with −5.20 kcal/mol. The hydrogen bond interactions, Vander Waals interactions, and two-dimensional and three-dimensional interactions were studied.

INTRODUCTION

Trachyaspermum ammi often known as Ajwain or carom seeds or bishop's weed is a plant seed that are widespread across diverse areas of India and are cultivated predominantly in north-western states of India like Gujarat and Rajasthan, Madhya Pradesh and in other countries like Afghanistan, Bangladesh, Egypt Iran, and Iraq. *Trachyaspermum ammi* belongs to the family of Apiaceae, dicot and possess high medicinal properties. *Trachyaspermum ammi* is used by the people traditionally as it possesses several properties to cure diverse liver, lungs, and stomach disorders. *Trachyaspermum ammi* is commonly used as an herbal medicine by the people. The seed also possesses carminative and antispasmodic properties which make it an effective remedy for various disorders, such as indigestion, abdominal pain, piles, amenorrhea, bronchial inflammation, asthma, hepatoprotective agent, and chronic diarrhea (Bairwa *et al*., 2012). Research has shown the significant antibacterial properties of *T. ammi* against a wide variety of bacteria, such as *Staphylococcus aureus, Escherichia coli, Enterococcus faecalis, Shigella flexneri*, and *Salmonella typhi* (Kaur and Arora, 2009). Multiple drug resistant fungal strains, such as *Candida albicans, Candida krusei, Candida tropicalis*, and *Candida glabrata*, were also inhibited by *T. ammi* (Khan *et al*., 2010). Volatile oils of *T. ammi* has the potential to for antifungal properties against *C. albicans* and *Aspergillus* species (Ishwar and Singh, 2000). The plant and its seeds contain several phytochemicals and essential oils, such as thymol, g-Terpinene, isobornylisobutyrate, o-Cymene, p-Cymene, a-Pinene, silphine, verbenene, ionone myrcene, thymyl acetate, etc. (Dhaiwal *et al*., 2017). Thymol is the major constituent of *T. ammi* seeds. *Trachyaspermum ammi* contains 50% of thymol. Thymol is an essential oil obtained from the seeds of *T. ammi* and finds applications in toothpaste and perfume industries (Joshi, 2000).

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Phytochemical analysis

Trachyaspermum ammi contains diverse phytochemicals. Carbohydrates make the predominant principal composition (38.6%), fats (18.1%), proteins (15.4%), flavone and saponins (7%). Ajwain also contains trace elements of calcium, nicotinic acid, and iron (Pruthi and Jiwan Singh, 1998). Thymol (36%–60%) is the major proportion of ajwain fruits essential oils (Ishikawa *et al*., 2001). Other principal oils in *T. ammi* are carvone (46%), dillapiole, and limonene. Non-thymol (thymene) fractions are γ-terpenine, dipentene, α-terpenine, α-pinene, β-pinene, para-cymene, α-3-carene, and carvacrol. Ethanolic extracts of ajwain fruits yields hydroscopic saponins. Characteristic yellow colored flavone isolated from ajwain fruits possesses 6-O-β-glucopyranosyloxythymol, oleoresin, and volatile oils (Garg and Kumar, 1998).

Pharmacological and medicinal activities of Trachyaspermum ammi (ajwain)

Trachyaspermum ammi (Ajwain) has a characteristic aromatic and pungent smell. Often, the seeds are used as spices in recipes, perfume industry, and food preservatives. In Ayurveda, ajwain is used to heal stomach- and liver-related disorders. A fraction of crushed fruits of ajwain are used to cure colonic pains. Dry and hot fomentations of fruit extracts are applied externally onto the chest to relieve asthma- and lung-related disorders. Ajwain seeds have shown potential to have anti-inflammatory property, antifungal (Saeidnia *et al*., 2005), antibacterial, antifilarial (Mathew *et al*., 2008), digestive stimulant, antiplasmodic, broncho-diliating, galactogogic (Kaur, 1998), detoxification of toxins G1 (AFG 1) (Velazhahan *et al*., 2010), diuretic (Ahsan *et al*., 1990), antihelminthic, hypolipidemic (Kumari and Prameela, 1992), and gastroprotective properties. *Trachyaspermum ammi* has shown potential to cure carbon tetrachloride- and paracetamol-induced liver damage in male albino rats (Gilani *et al*., 2005). *Trachyaspermum ammi* also has proven records that to cure hypertension and colonic diarrhea (Gilani *et al*., 2005).

Candidapepsin-1 enzyme structure

The Candidapepsin-1 (EC: 3.4.23.24) is an apoenzyme and belongs to the class of Secreted aspartic proteinase (Sap) family (Sanglard *et al*., 1997; Smolenski *et al*., 1997). The gene responsible for this enzyme is SAP1 and is located on the chromosome 6 of the organism. The Candidapepsin-1 is a proteolytic virulent enzyme from the endophytic polymorphic fungal species *C albicans* and is responsible for superficial Candida infections like oral and skin infections in immunocompromised individuals (Correia *et al*., 2010; Meenambiga *et al*., 2018; Staib *et al*., 2000). The enzyme invades and adheres to the host tissue by digesting the host cell membrane and small molecules to acquire nutrition. The enzyme digests the cell host cell membrane to inhibit the attack from the host immune system (Naglik *et al*., 2003). The enzyme attacks the oxygen carrier molecule hemoglobin through its proteolytic activity and releases various antimicrobial hemocidins to combat against various microbes of the same niche (Schaller *et al*., 2001). The enzyme is very stable and active at pH 5.0 (Aoki *et al*., 2011). This enzyme is similar in structure and function with its isoenzyme Candidapepsin-5. There are 10 subclasses of acidic hydrolases in the Sap family from Candidapepsin-1 to 10. The structural investigation discloses the highly conserved overall secondary structure of Candidapepsin-1 enzyme (Borelli *et al*., 2008). However, Candidapepsin-1, Candidapepsin-2, and Candidapepsin-3 differ from Candidapepsin-5 in two aspects, namely, the net overall electrostatic charge and structural conformation of its entrance toward the active site of the enzyme (Borelli *et al*., 2008). The active site of the enzyme holds a net negative electrostatic charge due to the presence of basic amino acids. The enzyme contains two chains A and B with 391 amino acid residues and molecular weight of 41.6 KDa. The active site is located between the positions at Asp82 and Asp267 of the enzyme. The entry toward the active site of the enzyme is wider. Figure 1 denotes the three-dimensional structure of the Candidapepsin-1 enzyme. The most predominant inhibitor of the enzyme is Pepstatin A. Hence, molecular docking of the enzyme with bioactive plant secondary metabolites supports the existing drug development process as molecular chemical interactions can be comprehensively studied.

MATERIALS AND METHODS

Bioactive compounds obtained from gas chromatography–mass spectrometry (GC-MS) analysis of Trachyaspermum ammi essential oils

The bioactive compounds from essential oils of *T. ammi* were obtained from the conventional hydrodistillation process using Dean–Stark apparatus (Dhaiwal *et al*., 2017; Javed *et al*., 2012). The essential oils were analyzed using GC–MS (Dhaiwal *et al*., 2017).

The information about the bioactive compounds, such as IUPAC name, structure, and chemical formula, were retrieved from PubChem database (Table 1). The bioactive compounds mentioned in Table 1 were used for molecular docking against the enzyme Candidapepsin-1. Lipinski properties and ADMET properties of the compounds were studied.

Enzyme target preparation

The enzyme Candidapepsin-1 (PDB Id: 2qzw) was used as the drug target in this research work with resolution 2.05 Å and the method of incorporation was done using X-ray diffraction. The

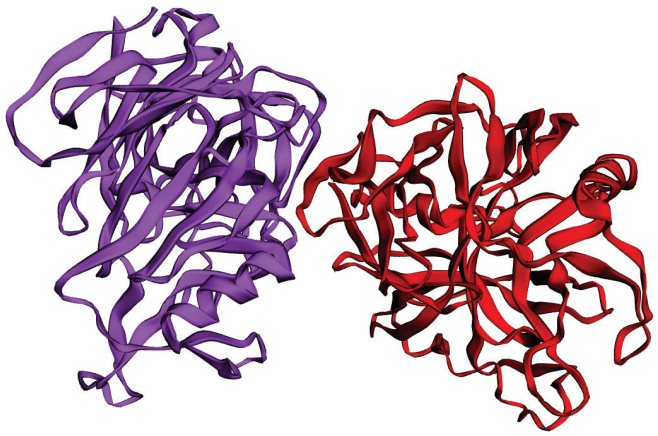
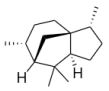
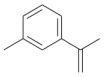
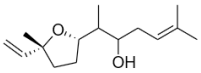
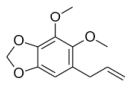
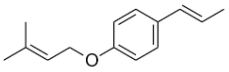
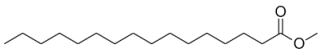
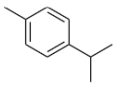
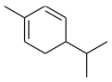
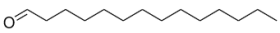
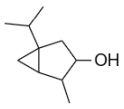
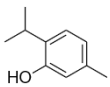
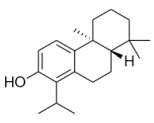


Figure 1. Three-dimensional structure of Candidapepsin-1 (PDB Id: 2qzw) with chain A and B.

Table 1. Bioactive compounds from essential oil of *T. ammi* analyzed through GC-MS analysis reported by Dhaiwal *et al*. (2017).

S. no	Compound	IUPAC name	Structure	Chemical formula
1	Cedrane	(1S,2R,5S,7S,8R)-2,6,6,8-tetramethyltricyclo [5.3.1.0(1,5)] undecane		$C_{15}H_{26}$
2	Cineole	1,8-Epoxy-p-menthane		$C_{10}H_{18}O$
3	m-Cymene	1-methyl-3-prop-1-en-2-yl benzene		$C_{10}H_{14}$
4	Davanol	2-[(2S,5R)-5-ethenyl-5-methyloxolan-2-yl]-6-methylhept-5-en-3-ol		$C_{15}H_{26}O_2$
5	Dillapiole	4,5-dimethoxy-6-prop-2-enyl-1, 3-benzodioxole		$C_{12}H_{14}O_4$
6	Foeniculin	1-(3-methylbut-2-enoxy)-4-[(E)-prop-1-enyl] benzene		$C_{14}H_{18}O$
7	Ligustilide	(3Z)-3-Butylidene-4,5-dihydro-2-benzofuran-1-one		$C_{12}H_{14}O_2$
8	Methyl palmitate	Methyl hexadecanoate		$C_{17}H_{34}O_2$
9	o-Cymene	1-methyl-2-propan-2-ylbenzene		$C_{10}H_{14}$
10	p-Cymene	1-methyl-4-propan-2-ylbenzene		$C_{10}H_{14}$
11	Phellandrene	2-methyl-5-propan-2-ylcyclohexa-1, 3-diene		$C_{10}H_{16}$
12	Tetradecanal	Tetradecanal		$C_{14s}H_{28}O$
13	Thujanol	4-methyl-1-propan-2-ylbicyclo [3.1.0] hexan-3-ol		$C_{10}H_{18}O$
14	Thymol	5-methyl-2-propan-2-ylphenol		$C_{10}H_{14}O$
15	Totarol	(4bS,8aS)-4b,8,8-trimethyl-1-propan-2-yl-5,6,7,8a,9,10-hexahydrophenanthren-2-ol		$C_{20}H_{30}O$

protein was retrieved from the Protein Data Bank database. The protein consists of two chains, namely, A and B. One of the protein chains (chain A) was used this study. This was done to improve the accuracy of ligand binding (Sasikala and Meena, 2016). The interfering crystallographic water molecules were also removed from the protein for effective ligand binding (Meenambiga *et al*., 2018).

ADMET properties of bioactive compounds from essential oil

Prediction of absorption, distribution, metabolism, and excretion properties of the compounds were done using the freely available SwissADME software package (Kramer *et al*., 2017). This was performed to enhance the success of drug discovery and development process.

Molecular docking studies using the AutoDock 4.2.6 software

Molecular docking was performed using the 15 bioactive compounds depicted in the Table 1. The bioactive compounds were docked against the Candidapepsin-1 enzyme using the comprehensive bioinformatics tool AutoDock 4.2.6 software. The AutoDock 4.2.6 is relied on the principle of Lamarckian genetic algorithm and is the most reliable automated tool used by the researchers to understand the protein-ligand interactions and protein–protein interactions (Meenambiga *et al*., 2015). The ligand-protein structure-based drug designing was performed using this software. AutoDock 4.2.6 is dependent on two techniques, namely, the Rapid-grid based estimation and systematic search of torsional freedom for the ligand-protein molecular docking (Meenambiga *et al*., 2015).

Grid parameters

Default grid size of 20 Å was set. Total grid points per map were 64,000. Grid spacing was 0.375 Å (default). The center grid box sizes were *x* center: −16.302, *y* center: −23.34, and −16.245, respectively.

Discovery studio 3.1- visualizer

Discovery studio 3.1 is a visualizer programmed and developed by the Accelrys. This software is free of cost, provides comprehensive information, and is the most often used by the scientific community to view the ligand-receptor interactions. The software provides us with the necessary information about the interactions of small and large molecules taking part in the interaction. The software deals with various aspects of molecular docking, such as macromolecule engineering, ligand-receptor interactions, pharmacophore modeling, antibody modeling & optimization simulations, macromolecule design, and protein–protein interactions (Almagro *et al*., 2011; Luu *et al*., 2011; Sutter *et al*., 2011). The two-dimensional and three-dimensional interactions images displayed in this study were developed through this software.

RESULTS AND DISCUSSION

The following are the bioactive compounds obtained from the GC–MS analysis of essential oil from the seeds of *Trachyaspermum ammi*: (1) cedrane, (2) cineole, (3) m-cymene, (4) davanol, (5) dillapiole, (6) foeniculin, (7) ligustilide, (8) methyl palmitate, (9) o-cymene, (10) p-cymene, (11) phellandrene, (12) tetradecanal, (13) thujanol, (14) thymol, and (15) totarol. These bioactive compounds were reported by Dhaiwal *et al*. (2017). Although the bioactive compounds were found to be an effective antioxidant, there were no reports available for the potential of *T. ammi* against the virulence of fungal enzyme, namely, Candidapepsin-1.

Molecular docking of ligands against the active site of the enzyme will elucidate the interactions between them. This will pave way for discovery of novel phytomedicines in the field of drug discovery and development.

The compounds from the GC–MS analysis satisfied the Lipinski's rule of five. This rule comprises of five sub rules, namely, (1) molecular weight (<500), (2) log P (<+5.6), (3) Number of hydrogen donors (<5), (4) Number of hydrogen

Table 2. Lipinski properties of bioactive compounds from essential oil of *T. ammi*.

S. no	Compound name	Molecular weight (<500 Da)	Log P (<5.6)	H-bond donor (<5)	H-bond acceptor (<10)	Molar refractivity (40–130)
1	Cedrane	206	4.49	0	0	64.60
2	Cineole	154	2.74	0	1	45.52
3	m-Cymene	132	3.02	0	0	45.88
4	Davanol	238	3.46	1	2	71.92
5	Dillapiole	222	2.16	0	4	59.56
6	Foeniculin	202	4.06	0	1	66.07
7	Ligustillide	190	2.87	0	2	54.48
8	Methyl palmitate	270	5.60	0	2	82.32
9	o-Cymene	134	3.11	0	0	45.26
10	p-Cymene	134	3.11	0	0	45.26
11	Phellandrene	136	3.16	0	0	45.84
12	Tetradecanal	212	4.88	0	1	67.14
13	Thujanol	154	2.04	1	1	45.16
14	Thymol	150	2.82	1	1	46.93
15	Totarol	286	5.54	1	1	88.99

acceptors (<10), and (5) Molar refractivity (40–130). The Lipinski's rule of five is utilized to appraise the drug-likeliness of a compound; in other words, it is very essential criteria for a compound to satisfy this rule in order to be administered orally. The Lipinski's rule of five is also used to assess the durability of a drug molecule (Benet *et al*., 2016). Hence, this is rule is essential for a bioactive compound to be considered an oral drug (Lipinski *et al*., 2012). Table 2 represents the detailed information about the bioactive compounds with their respective properties.

The *in silico* analysis through molecular docking revealed the importance of structure-based drug designing strategy toward the development for novel drugs against the inhibition of potential drug target. The virulent enzyme responsible for superficial skin infections like candidiasis and biofilm formation is Candidapepsin-1 (Korting *et al*., 1998). The enzyme's active site was docked with several bioactive compounds from *T. ammi*. The binding energy for each bioactive compound against the Candidapepsin-1 enzyme, interaction of hydrogen bonds, Vander Waals interactions, and essential details were listed in Table 3. Ligustilide has the lowest binding energy of −5.75 kcal/mol and has three hydrogen bond interactions with amino acids Ile 223, Tyr 225, and Thr 222 at the active site. Lower the binding energy, greater is the binding efficiency. Greater the hydrogen bonds between the enzyme and ligand determines the strength of binding (Kortemme *et al*., 2003). The two-dimensional and three-

Table 3. Molecular docking result analysis of bioactive compounds from essential oil of *T. ammi* against Candidapersin-1 (PDB Id: 2qzw) enzyme.

S. no	Compound name	Binding Energy Kcal/mol	No of Vander Waal's interaction	No. of hydrogen bonds	Hydrogen bond interaction	Total polar and non-polar bonding
1.	Cedrane	−5.20	GLY 220, ASP 86, THR 221, GLY 85, ASP 218, SER 35, GLY 34, LEU 216, ILE 223, ASP 32, ILU 123, ILE 30, TYR 84.	0	0	GLY 220, ASP 86, THR 221, GLY 85, ASP 218, SER 35, GLY 34, LEU 216, ILE 223, ASP 32, ILU 123, ILE 30, TYR 84.
2.	Cineole	−4.20	ILE 123, ILE 30, ASP 86, SER 35, TYR 84, GLY 85, GLY 34, ASP 218, ASP 32, GLY 220.	0	0	ILE 123, ILE 30, ASP 86, SER 35, TYR 84, GLY 85, GLY 34, ASP 218, ASP 32, GLY 220.
3.	m-Cymene	−3.58	ASP 32, GLY 34, SER 35, ILE 305, LEU 216, ASP 218, GLY 220. ILE 123, ASP 86, TYR 84.	0	0	ASP 32, GLY 34, SER 35, ILE 305, LEU 216, ASP 218, GLY 220. ILE 123, ASP 86, TYR 84.
4.	Davanol	−2.94	THR 78, GLY 135, PHE 80, LEU 94, ALA 134, ALA 133, TYR 81, PRO 79.	0	0	THR 78, GLY 135, PHE 80, LEU 94, ALA 134, ALA 133, TYR 81, PRO 79.
5.	Dillapiole	−3.91	GLY 220, ASP 86, TYR 225, THR 221, VAL 12, SER 13, ILE 30, ASP 32, ILE 123	1	THR 222	GLY 220, ASP 86, TYR 225, THR 221, VAL 12, SER 13, ILE 30, ASP 32, ILE 123
6.	Foeniculin	−4.36	VAL 12, GLY 220, THR 222, THR 221, TYR 225, GLY 85, ILE 305, ASP 86, ILE 30, SER 13	0	0	VAL 12, GLY 220, THR 222, THR 221, TYR 225, GLY 85, ILE 305, ASP 86, ILE 30, SER 13
7.	Ligustillide	−5.75	ILE 305, THR 221, VAL 12, ILE 30, SER 13, GLY 220, ASP 86	3	ILE 223, TYR 225, THR 222	ILE 305, THR 221, VAL 12, ILE 30, SER 13, GLY 220, ASP 86
8.	Methyl Hexadecanoate	+0.92	LEU 76, THR 78, PRO 79, PHE 80, TYR 81, TYR 81, ALA 133, GLY 135	0	0	LEU 76, THR 78, PRO 79, PHE 80, TYR 81, TYR 81, ALA 133, GLY 135
9.	o-Cymene	−3.62	GLY 220, ASP 86, THR 221, ASP 218, GLY 34, SER 35, ILE 123, TYR 84, ASP 32, ILE 30	0	0	GLY 220, ASP 86, THR 221, ASP 218, GLY 34, SER 35, ILE 123, TYR 84, ASP 32, ILE 30
10.	p-Cymene	−3.65	ASP 86, THR 221, ILE 305, ASP 218, TYR 84, GLY 34, GLY 220, ASP 32, ILE 30, ILE 123	0	0	ASP 86, THR 221, ILE 305, ASP 218, TYR 84, GLY 34, GLY 220, ASP 32, ILE 30, ILE 123
11.	Phellandrene	−3.97	ILE 123, GLY 220, ASP 32, TYR 84, THR 221, ASP 218, ASP 86	0	0	ILE 123, GLY 220, ASP 32, TYR 84, THR 221, ASP 218, ASP 86
12.	Tetradecanal	−3.17	THR 221, TYR 225, ASP 86, TYR 84, ASP 32, ILE 123, ILE 30, GLY 220, SER 13, VAL 12	1	THR 222	THR 221, TYR 225, ASP 86, TYR 84, ASP 32, ILE 123, ILE 30, GLY 220, SER 13, VAL 12
13.	Thujanol	−4.14	ASP 86, TYR 84, SER 35, GLY 85, GLY 34, THR 221, ASP 218, ILE 305, ILE 30, ILE 123, ASP 32, GLY 220	1	ASP 218	ASP 86, TYR 84, SER 35, GLY 85, GLY 34, THR 221, ASP 218, ILE 305, ILE 30, ILE 123, ASP 32, GLY 220
14.	Thymol	−4.05	GLY 220, ASP 218, THR 221, GLY 85, ASP 86, ILE 305, GLY 34, SER 35, TYR 84, ILE 30, ILE 123	1	ASP 32	GLY 220, ASP 218, THR 221, GLY 85, ASP 86, ILE 305, GLY 34, SER 35, TYR 84, ILE 30, ILE 123
15.	Totarol	−4.87	VAL 12, SER 13, TYR 84, ILE 123, ILE 30, SER 35, ASP 86, GLY 220, THR 221, THR 222	1	ASP 32	VAL 12, SER 13, TYR 84, ILE 123, ILE 30, SER 35, ASP 86, GLY 220, THR 221, THR 222

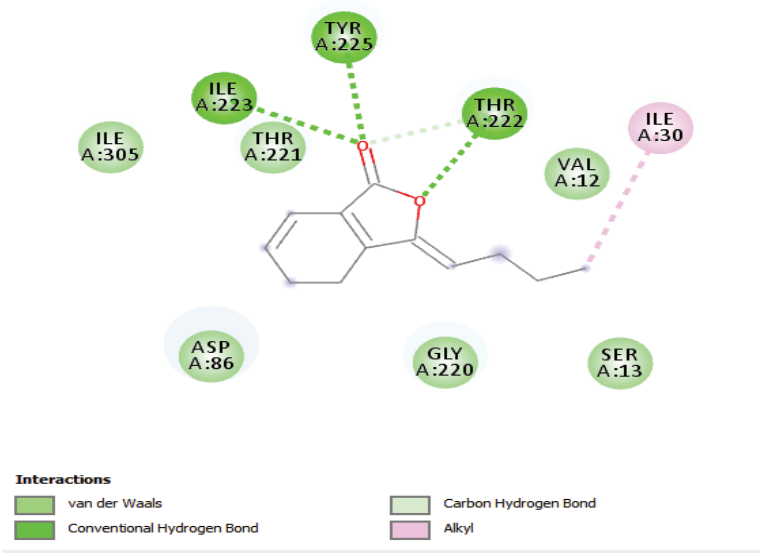


Figure 2. Two-dimensional and three-dimensional residual interactions map of ligustilide against the active site of Candidapepsin-1.

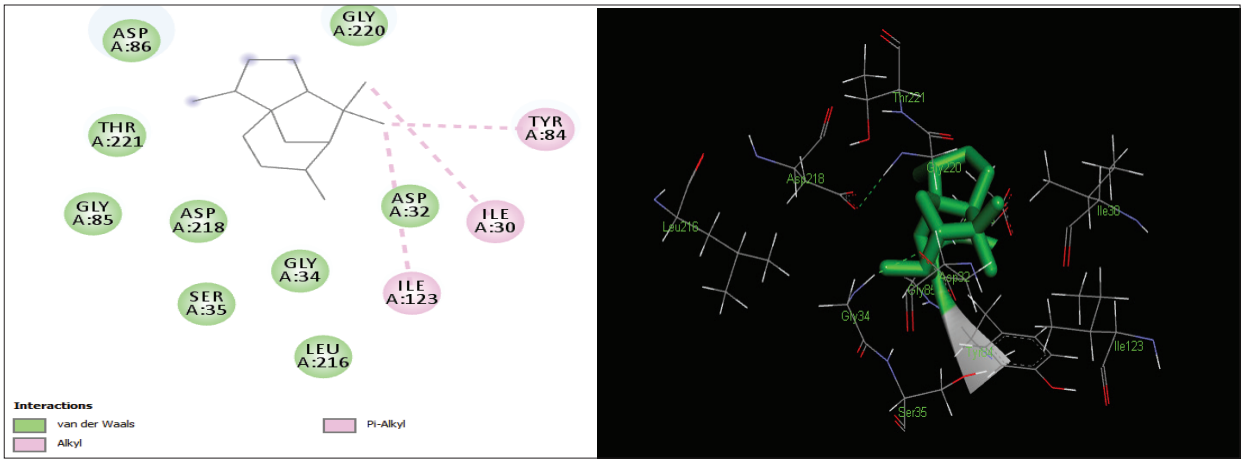


Figure 3. Two-dimensional and three-dimensional residual interactions map of cedrane against the active site of Candidapepsin-1.

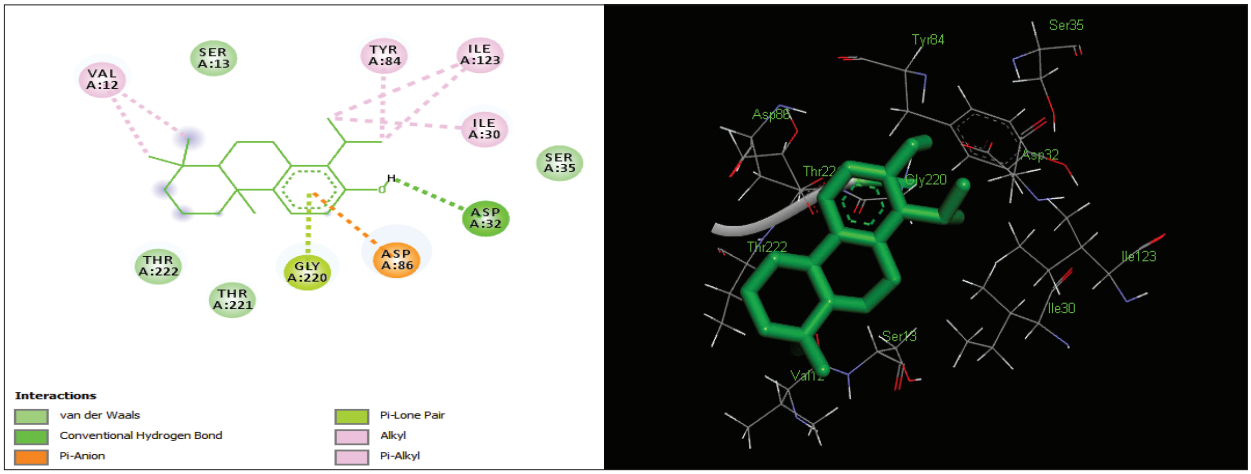


Figure 4. Two-dimensional and three-dimensional residual interactions map of totarol against the active site of Candidapepsin-1.

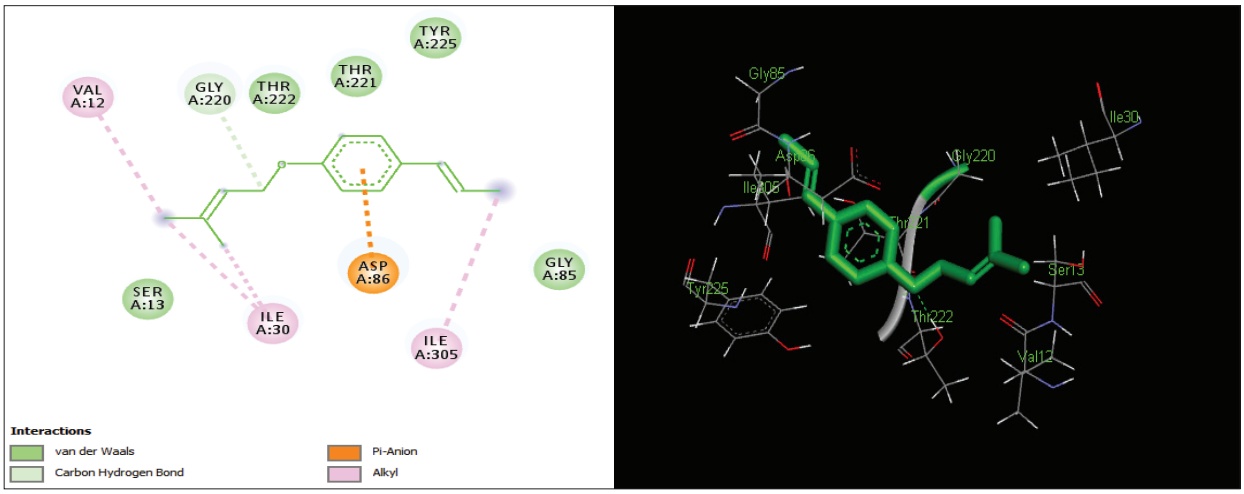


Figure 5. Two-dimensional and three-dimensional residual interactions map of foeniculin against the active site of Candidapepsin-1.

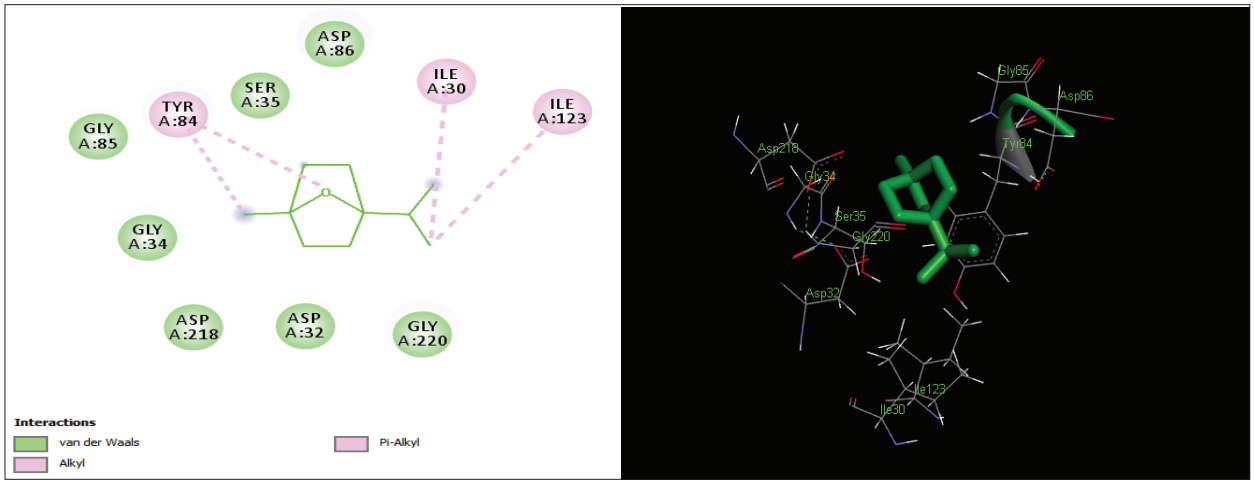


Figure 6. Two-dimensional and three-dimensional residual interactions map of cineole against the active site of Candidapepsin-1.

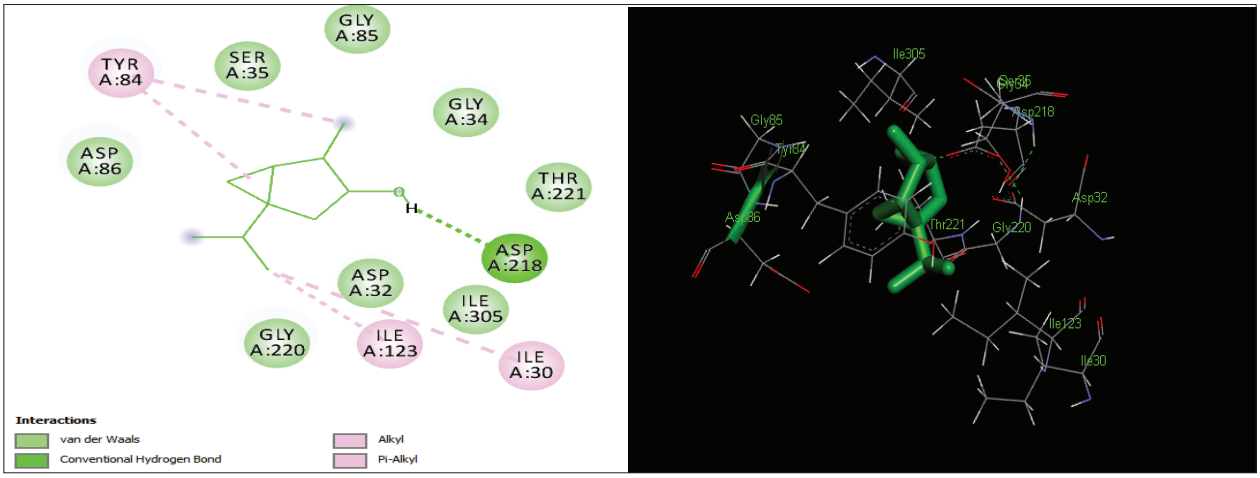


Figure 7. Two-dimensional and three-dimensional residual interactions map of thujanol against the active site of Candidapepsin-1.

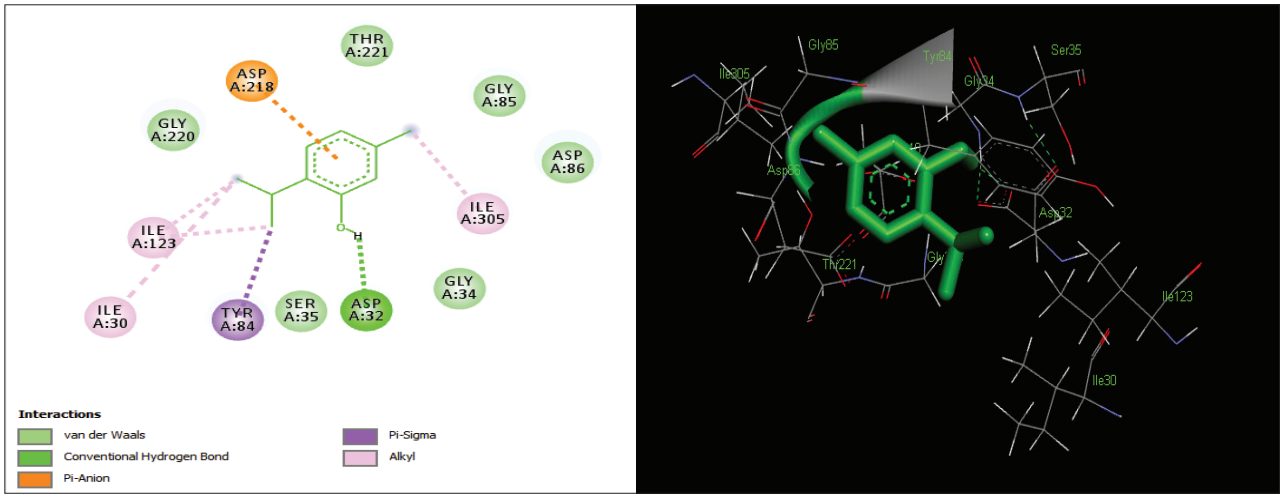


Figure 8. Two-dimensional and three-dimensional residual interactions map of thymol against the active site of Candidapepsin-1.

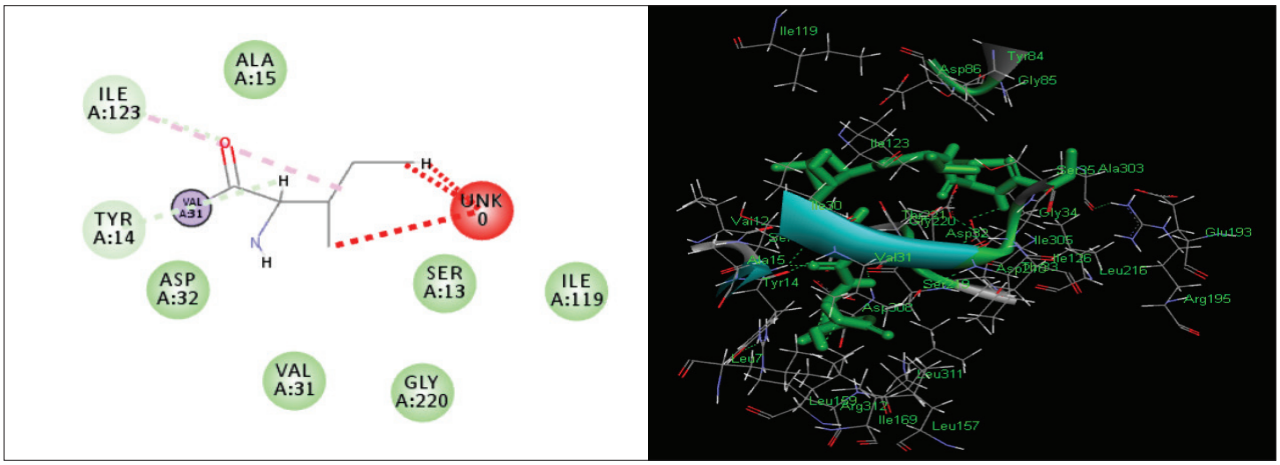


Figure 9. Two-dimensional and three-dimensional residual interactions map of Pepstatin A against the active site of Candidapepsin-1.

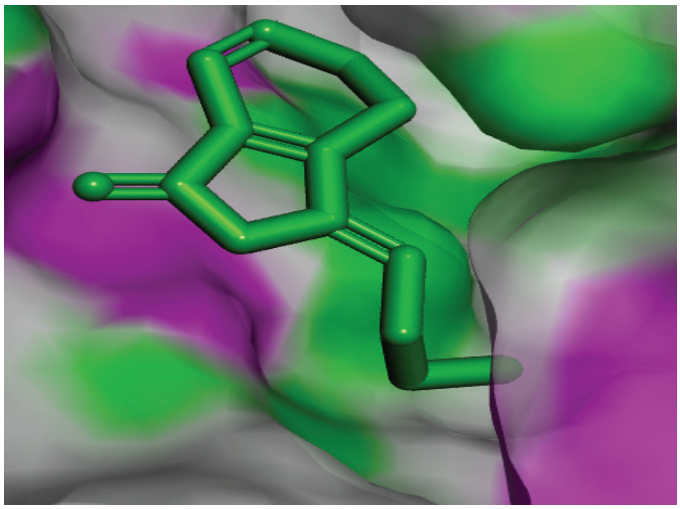


Figure 10. Docked conformation of ligustilide against the active site of Candidapepsin-1.

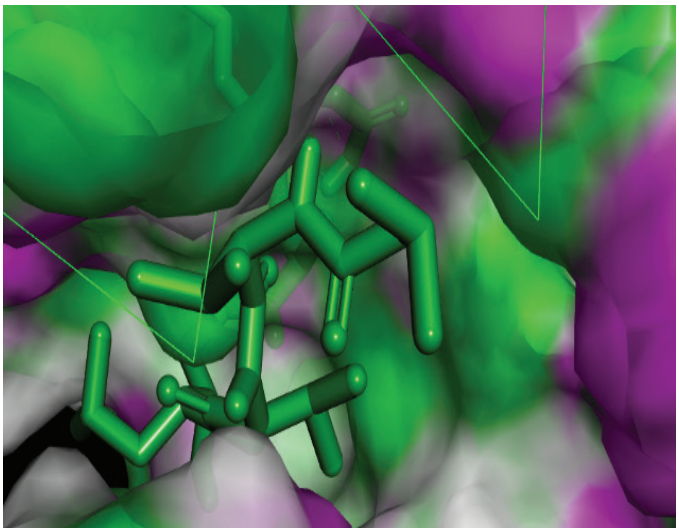


Figure 11. Docked conformation of Pepstatin A against the active site of Candidapepsin -1.

Table 4. Absorption properties of various bioactive compounds.

S. No	Compound	Water solubility (log mol/l)	$CACO_2$ permeability (Log Pabb in 10^{-6} cm/Sec)	GI absorption (%)	Skin permeability (Log Kp)	P-glycoprotein substrate	P-glycoprotein I inhibitor	P-glycoprotein II inhibitor
1	Cedrane	−6.06	1.38	94.42	−2.12	No	No	No
2	Cineole	−2.77	1.51	96.26	−2.13	Yes	No	No
3	m-Cymene	−4.10	1.53	93.65	−1.21	No	No	No
4	Davanol	−3.24	1.64	93.41	−2.29	No	No	No
5	Dillapiole	−2.44	1.83	95.10	−2.45	No	No	No
6	Foeniculin	−4.65	1.80	95.04	−1.30	No	No	No
7	Ligustillide	−3.09	1.62	96.30	−2.22	No	No	No
8	Methyl palmitate	−6.93	1.60	92.33	−2.60	No	No	No
9	o-Cymene	−4.11	1.53	93.88	−1.18	No	No	No
10	p-Cymene	−4.08	1.53	93.54	−1.19	No	No	No
11	Phellandrene	−3.85	1.41	96.55	−1.51	No	No	No
12	Tetradecanal	−6.49	1.48	93.02	−2.07	No	No	No
13	Thujanol	−2.51	1.50	94.78	−2.13	No	No	No
14	Thymol	−2.79	1.61	90.84	−1.62	No	No	No
15	Totarol	−5.91	1.56	92.77	−2.65	No	No	Yes

Table 5. Distribution properties of various bioactive compounds.

S. no	Compound	VDss (human) (Log L/kg)	Fraction unbound (human) (Fu)	BBB permeability (Log BB)	CNS permeability (Log PS)
1	Cedrane	0.67	0.10	0.84	−1.44
2	Cineole	0.36	0.51	0.67	−2.55
3	m-Cymene	0.72	0.15	0.47	−1.39
4	Davanol	0.23	0.40	0.55	−2.84
5	Dillapiole	0.03	0.20	−0.22	−2.24
6	Foeniculin	0.68	0.11	0.62	−1.70
7	Ligustillide	0.30	0.42	0.56	−2.45
8	Methyl palmitate	0.23	0.07	0.75	−1.68
9	o-Cymene	0.74	0.16	0.48	−1.40
10	p-Cymene	0.70	0.16	0.48	−1.39
11	Phellandrene	0.41	0.43	0.76	−2.05
12	Tetradecanal	0.48	0.15	0.78	−1.67
13	Thujanol	0.35	0.47	0.66	−2.24
14	Thymol	0.51	0.20	0.41	−1.66
15	Totarol	1.16	0	0.55	−1.35

dimensional interactions of the ligustilide with the enzyme are depicted in Figure 2. Cedrane with the binding energy of −5.20 kcal/mol occupies the next spot in the ranking Figure 3. Totarol (−4.87 kcal/mol), foeniculin (−4.36 kcal/mol), cineole (−4.20 kcal/mol), thujanol (−4.14 kcal/mol), and thymol (−4.05 kcal/mol) follow up the order. The binding energies of bioactive compounds docked were compared with the binding energy of the reference ligand (Pepstatin A) (Fig. 9) against the enzyme with the same grid parameters. The binding energy of reference inhibitor ligand was found to be +2.77 kcal/mol, which is lower when compared to the binding energies of bioactive compounds from *T. ammi* (Fig. 11). The binding of bioactive compounds inhibits the activity of enzyme resulting in the neutralization the enzymes virulence (Hube *et al*., 1997). Ligustilide isolated from essential oil of Umbelliferae family has immense antifungal properties against several species of fungi, thus restricting the fungal growth and proliferations (Lee *et al*., 2008). Essential oils from *T. ammi* at different temperatures and pressures through minimum inhibitory concentration (MIC) studies against a variety of *Candida* species were studied by Rath and Mohapatra (2015). The MIC values for essential oil from *T. ammi* was 31.25 µl/ml for *C. albicans*, and *C. tropicalis,* 62.50 µl/ml for *Candida glabratra*, and 125 µl/ml for *Candida parapsilosis*, respectively (Rath and Mohapatra, 2015). This proves that the essential bioactive compounds from the oil has antifungal potential and can be used in therapeutic applications. Also, upon increasing the temperature and pressure had an augmented therapeutic inhibition potential of the essential oil against *Candida* infection (Pattnaik *et al*., 1995). Fungal hyphae are an important measure for the fungal growth and virulence. The development of the hyphae is inhibited by thymol, hence disturbing the cell membrane

Table 6. Metabolism properties of various bioactive compounds.

S. no	Compound	CYP2D6 substrate	CYP3A4 substrate	CYP1A2 inhibitor	CYP2C19 inhibitor	CYP2C9 inhibitor	CYP2D6 inhibitor	CYP3A4 inhibitor
1	Cedrane	No	Yes	Yes	No	No	No	No
2	Cineole	No	No	No	No	No	No	No
3	m-Cymene	No	No	Yes	No	No	No	No
4	Davanol	No	No	No	No	No	No	No
5	Dillapiole	No	Yes	Yes	No	No	No	No
6	Foeniculin	No	No	Yes	No	No	No	No
7	Ligustillide	No	No	No	No	No	No	No
8	Methyl palmitate	No	Yes	Yes	No	No	No	No
9	o-Cymene	No	No	Yes	No	No	No	No
10	p-Cymene	No	No	Yes	No	No	No	No
11	Phellandrene	No	No	No	No	No	No	No
12	Tetradecanal	No	Yes	No	No	No	No	No
13	Thujanol	No	No	No	No	No	No	No
14	Thymol	No	No	Yes	No	No	No	No
15	Totarol	No	Yes	Yes	No	No	No	No

Table 7. Excretion and toxicity properties of various bioactive compounds.

S. no	Compound	AMES toxicity	Max. tolerated dose (human) (Log mg/kg/day)	hERG I inhibitor	hERG II inhibitor	Oral Rat Acute Toxicity (LD50) (mol/kg)	Oral Rat Chronic Toxicity (LOAEL) (Log mg/kg)	Hepatotoxicity	Skin Sensitisation
1	Cedrane	No	−0.43	No	No	1.62	1.33	No	No
2	Cineole	No	0.59	No	No	1.85	2.07	No	Yes
3	m-Cymene	No	0.89	No	No	1.83	2.34	No	Yes
4	Davanol	No	0.39	No	No	1.85	2.02	No	Yes
5	Dillapiole	No	0.73	No	No	0.25	1.85	No	No
6	Foeniculin	No	0.86	No	No	1.78	1.31	No	Yes
7	Ligustillide	No	0.41	No	No	1.86	2.14	No	Yes
8	Methyl palmitate	No	0.18	No	No	1.64	2.99	No	Yes
9	o-Cymene	No	0.88	No	No	1.79	2.32	No	Yes
10	p-Cymene	No	0.90	No	No	1.83	2.33	No	Yes
11	Phellandrene	No	0.75	No	No	1.74	2.33	No	No
12	Tetradecanal	No	0.16	No	No	1.50	1.22	No	Yes
13	Thujanol	No	0.64	No	No	1.70	1.93	No	Yes
14	Thymol	No	1.00	No	No	2.07	2.21	Yes	Yes
15	Totarol	No	−0.21	No	Yes	2.60	1.16	No	Yes

thereby affecting the enzymes responsible for cell wall synthesis (Braga *et al*., 2007). Figures 2–10 list the two-dimensional and three-dimensional interactions of bioactive compounds with low binding energies against Candidapepsin-1. ADMET profiles of the compounds were depicted in Tables 4–7. All the compounds have high rates of gastrointestinal absorption (Table 4). From Table 5, it is clear that none of the drug penetrates the blood-brain barrier since the logBB value of all the compounds is less than three. For a drug to cross the blood-brain barrier, the logBB value must be greater than three (Muehlbacher *et al*., 2011).

CONCLUSION

Trachyaspermum ammi is traditionally used in ayurvedic medicine due to its anti-inflammatory, antifungal, antibacterial, anticancer, and antiarthritic potential. The current study revealed the inhibition potential of bioactive compounds from essential oil of *T. ammi* against the virulent enzyme Candidapepsin-1 of *C. albicans*. The bioactive compound ligustilide has the lowest binding energy of −5.75 kcal/mol. This proves the antifungal activity of the *T. ammi* against the *Candida* biofilm formation, there by inhibiting the virulence of the enzyme. The inhibition of the enzyme leads to novel discovery of plant-based therapeutic products. Computational molecular docking could be used as an effective supporting tool for the drug development process. Computational simulations also provide us with comprehensive results with high accuracy. Hence, their presence is necessary toward the development of drug discovery and development sector.

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CONFLICTS OF INTEREST

Authors declare that there are no conflicts of interest.

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