İstanbul Journal of Pharmacy

İstanbul Journal of Pharmacy,
IJP 2025, 55 (1): 134–148

https://doi.org/10.26650/IstanbulJPharm.2025.1441299
Submitted: 22.02.2024
Revision Requested: 26.07.2024
Last Revision Received: 14.10.2024
Accepted: 22.10.2024

Original Article

Open Access

Exploring the potential of *Lavandula stoechas* in smoking cessation: A molecular docking study of α4β2 nicotinic acetylcholine receptor interactions

Elif Barış [1] & Hüseyin Saygın Portakal [2]

[1] İzmir University of Economics, Faculty of Medicine, Medical Pharmacology, İzmir, Türkiye
[2] İzmir University of Economics, Faculty of Engineering, Genetics and Bioengineering, İzmir, Türkiye

Abstract

Background: *Lavandula stoechas*, commonly known as lavender, has traditionally been used in various therapeutic applications, including smoking cessation. The molecular interaction of Lavandula stoechas compounds with the α4β2 nicotinic acetylcholine receptors, which are crucial for smoking cessation, is not well understood. This study aims to analyze these interactions and compare them with the known smoking cessation drug varenicline tartrate.

Methods: Molecular docking analysis was performed on essential compounds of *Lavandula stoechas* to assess their binding affinities to the α4β2 nicotinic acetylcholine receptors. The study utilized the crystal structure of the receptor and conducted virtual drug screening using AutoDock Vina in the PyRx Virtual Screening Tool. ADME (Absorption, Distribution, Metabolism, and Excretion) and toxicity profiles were also predicted using in silico methods.

Results: The molecular docking revealed that several *Lavandula stoechas* compounds exhibited significant binding affinities to the α4β2 receptor. Compounds with the highest binding affinities were identified and compared with varenicline. The ADME and toxicity profiles indicated that these compounds had more favorable properties than varenicline, suggesting their potential as alternative smoking cessation agents.

Discussion: The findings demonstrate that *Lavandula stoechas* contains compounds with significant binding affinities to the α4β2 nicotinic acetylcholine receptors, similar to varenicline. This indicates a potential role for *Lavandula stoechas* in smoking cessation therapy. The favorable ADME and toxicity profiles of these compounds further support their potential as alternatives to current smoking cessation drugs. This study paves the way for further research into the therapeutic applications of Lavandula stoechas in smoking cessation.

Keywords Molecular docking · α4β2 Nicotinic Receptor · Varenicline · Lavandula Stoechas · Smoking cessation

Citation: Barış, E. & Portakal, H. S. (2025). Exploring the potential of *Lavandula stoechas* in smoking cessation: A molecular docking study of α4β2 nicotinic acetylcholine receptor interactions. *İstanbul Journal of Pharmacy*, *55*(1), 134-148. https://doi.org/10.26650/IstanbulJPharm.2025.1441299

This work is licensed under Creative Commons Attribution-NonCommercial 4.0 International License.
© 2025. Barış, E. & Portakal, H. S.

Corresponding author: Hüseyin Saygın Portakal saygin.portakal@izmirekonomi.edu.tr

INTRODUCTION

Lavenders (*Lavandula spp.*) are plant species belonging to the Lamiacae family and have been used for therapeutic and cosmetic purposes for centuries. Considering their traditional use, lavender oil is believed to have antibacterial, antifungal, sedative, and antidepressant effects. The first evidence of the use of lavender as a therapeutic agent dates back to the ancient Romans and Greeks. *Lavandula stoechas* is a plant that has been frequently used in Mediterranean countries since ancient times due to its analgesic, antiseptic, and wound-healing effects. This plant is popularly known as karabaş otu in Turkey (Oraloglu, 2018; Şahinler et al., 2022).

The chemical composition of the plant differs according to the plant subspecies. Lavenders are classified into four main categories: *Lavandula latifolia, Lavandula angustifolia, Lavandula stoechas*, and *Lavandula x intermedia*. The main components of lavender oil are linalool, linalyl acetate, 1,8-cineole, beta-ocimene, terpinen-4-ol, and camphol. Lavender essential oil is usually produced by steam distillation from flower heads and leaves. In a study, *L. angustifolia* essential oil significantly reduced anxiety, craving, systolic blood pressure, and heart rate in smokers (de Almeida Cunha et al., 2018). The linalool substance in it has a protective effect against smoking-induced lung inflammation (Ma et al., 2015).

Ocimene and Terpinolene compounds are not only responsible for the plant's refreshing aroma but also offer potential antimicrobial effects (Betlej et al., 2023). Camphene, present in two isomeric forms, (+)-Camphene and (-)-Camphene, exudes a cool, fir-like odor, adding another layer of complexity to Lavandula's scent profile. Geraniol and Linalool, with their sweet, floral notes, are pivotal in defining the quintessential lavender fragrance. These compounds are celebrated not only for their delightful scent but also for their potential calming and anti-anxiety effects (Dos Santos et al., 2022). Menthol, in its various forms including (-)-Menthol, offers a cooling sensation, often associated with Lavandula's use in soothing balms and ointments, which contribute to the plant's antioxidant properties (Chen et al., 2023). Lastly, compounds like Thymoquinone, trans-anethol, and 1,8-cineole contribute to the multifaceted profile of lavender, offering potential antioxidant, antimicrobial, and therapeutic benefits (Alam et al., 2022). The synergy of these compounds makes *Lavandula spp.* a plant of immense interest in aromatherapy, phytotherapy, and the fragrance industry, where its extracts are cherished for their aromatic allure and potential health benefits.

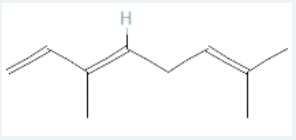
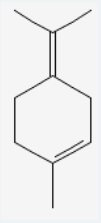
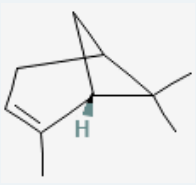
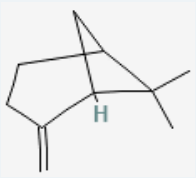
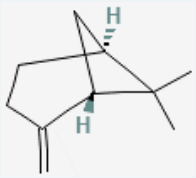
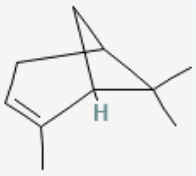
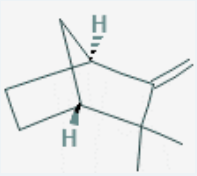
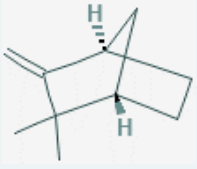
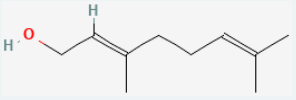
Regarding the cholinergic receptors, 17 subunits nACh receptor have been identified. The nAChR subunits most widely distributed in the central nervous system are the α4β2 (~90%) and α7 (~10%) subunits. The α4β2 subunit is the target structure of smoking cessation drugs. It has been suggested that borneol or camphor compounds of *Lavandula stoechas* can be used as nACh receptor antagonists as adjuvants in smoking addiction (Angioni et al., 2006). In an in silico study, the interaction of *Lavandula stoechas* and its camphol (camphor) and menthone structures with nACh receptors were shown to exhibit agonist properties. It has been reported that the hydroxyl group in menthol and carvacrol structures may have reduced plant activity (Hajiyeva, 2018).

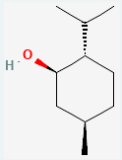
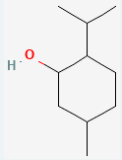
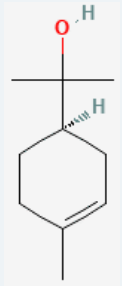
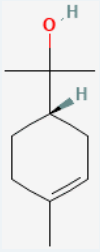
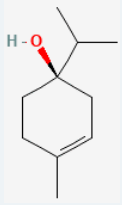
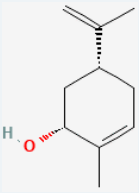
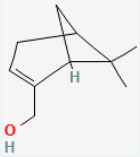
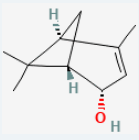
Three drugs are currently used as first-line pharmacotherapy for smoking cessation: nicotine replacement therapy, bupropion hydrochloride (sustained release), and varenicline tartrate. These drugs work by modulating the nicotinic system activity on the central nervous system. In smoking cessation therapy, as smoking behavior continues, the success rate of treatment declines. In patients using varenicline and bupropion as a smoking cessation treatment, the success rates at the end of 1 year were 20.5% and 18.6%, respectively. Although there is no significant difference between these drugs in terms of smoking cessation, the very low adherence to treatment (31.5%) supports the need for new agents in the treatment of smoking cessation (Benli et al., 2017). On the other hand, *Lavandula stoechas* has been traditionally used in smoking cessation therapy, although there is a lack of evidence about its pharmacological effects.

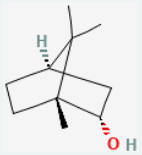
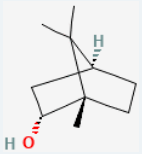
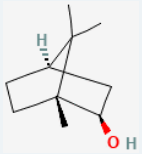
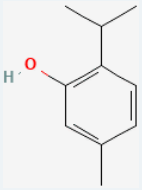
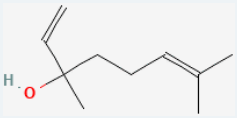
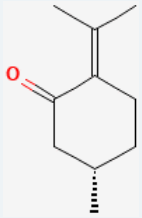
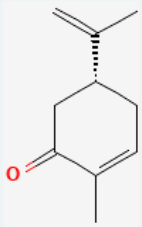
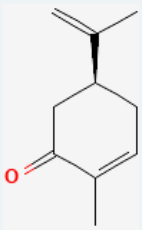
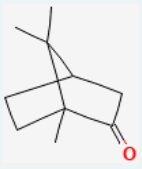
Varenicline, a medication used for smoking cessation, acts as a partial agonist at α4β2 neuronal nicotinic acetylcholine receptors (Mihalak et al., 2006). As a partial agonist, it has a dual mechanism of action: it stimulates these receptors to a moderate extent, mimicking the effects of nicotine but to a lesser degree, while also competitively binding to these receptors and blocking nicotine from attaching to them. This unique pharmacological profile not only helps to reduce the intensity of nicotine cravings but also diminishes the pleasurable sensations associated with smoking (Mihalak et al., 2006).

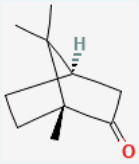
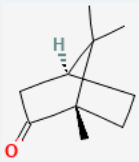
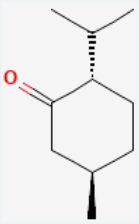
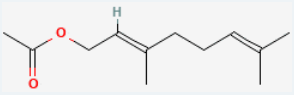
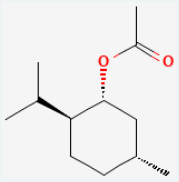
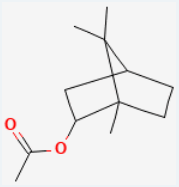
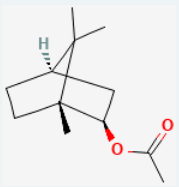
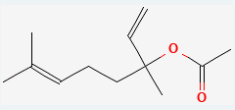
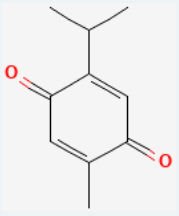
In this study, the molecular docking analysis of *Lavandula stoechas* essential compounds (Table 1) with α4β2 nicotinic acetylcholine receptors was investigated (Kirmizibekmez et al., 2009). Additionally, the binding and pharmacokinetic properties of these compounds were compared with those of varenicline. Once the *Lavandula stoechas* essential compounds having the highest binding affinity to the related protein were identified, ADME and toxicity analyses were predicted using in silico approaches.

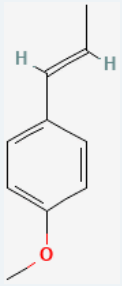
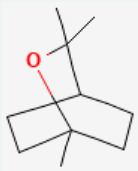
Table 1. Essential compounds of *Lavandula stoechas*

Group	Compound name	PubChem ID	Chemical Structure
Hydrocarbon	Ocimene	5281553	
	Terpinolene	11463	
	(1S)-(-)α-pinene	12223112	
	²(+)-β-Pinene	14896	
	²(-)-β-Pinene	440967	
	²α-pinene	6654	
	(+)-Camphene	92221	
	(-)-Camphene	440966	
Alcohol	Geraniol	637566	

Group	Compound name	PubChem ID	Chemical Structure
	(-)-Menthol	16666	
	Menthol	1254	
	(R)-(+)-α-Terpineol	442501	
	(S)-(-)-α-Terpineol	443162	
	²(-)-terpinen-4-ol	5325830	
	²(-)-Carveol	330573	
	(1R)-(-)-myrtenol	10582	
	(S)cis-verbenol	87839	

Group	Compound name	PubChem ID	Chemical Structure
Aldehydes and Ketones	(+)-Borneol	6552009	
	(-)-Borneol	1201518	
	Isoborneol	6321405	
	Thymol	6989	
	Linalool	6549	
	(S)-(-)-Pulegone	638012	
	(R)-(-)-Carvone	439570	
	(S)-(-)-Carvone	16724	
	(±)-Camphor, EP Ref,std (F)	2537	

Group	Compound name	PubChem ID	Chemical Structure
	(1R)-(+)-Camphor	159055	
	(1S)-(+)-Camphor	444294	
	(−)-Menthone	26447	
	Geranyl acetate	1549026	
	(1R)-(-)-menthyl acetate	220674	
	²(-)-Bornyl acetate	6448	
Esters	ısobornyl acetate	637531	
	Linalyl acetate	8294	
	Thymoquinone	10281	

Group	Compound name	PubChem ID	Chemical Structure
Others	trans-anethol	637563	
	1,8-cineole	2758	

MATERIALS AND METHODS

Protein Preparation

The 17nACh receptor subunit α4β2 crystal structure, including the varenicline drug with PDB ID: 6UR8 was retrieved in .pdf format from the Protein Data Bank (PDB). The electron microscopy analysis of the selected structure had a 3.17 Å resolution. The downloaded protein structure was imported in UCSF Chimaera software version 1.16 and prepared with the Dock Prep module of the software. The ligands, solvent (water) molecules, and heteroatoms of the protein were removed from the structure, hydrogen atoms and partial charges were added, and the side chains were replaced using the Dunbrack 2010 rotamer library. Once the preparation was completed, the prepared protein was exported in .pdf format and imported into the PyRx Virtual Screening Tool for use in molecular docking studies.

Ligand Preparation

To perform virtual drug screening, a library containing 37 *Lavandula stoechas* essential compounds was prepared. The ligands were retrieved from the PubChem database (Table 1). The library was imported into the PyRx software, and the ligands were prepared by following the energy minimization module of the PyRx Virtual Screening Tool.

Molecular Docking

Virtual drug screening was conducted on the AutoDock Vina package loaded in the PyRx Virtual Screening Tool by docking all ligands to the QMR binding site of the 17nACh receptor subunit α4β2. During the molecular docking strategy, the file format of the ligands was converted to .pdt, and the grid box coordinates were defined as x = 118.33, y = 196.96, z = 188.304. Grid box dimensions were set to x = 10, y = 10, z = 10. Once the molecular docking had been completed, the data demonstrating the binding affinity, rmsd/ ub, and rmsd/lb were exported in .cs format. The ligands with the highest binding affinity and modes with 0 values for each rmsd/ub and rmsd/lb parameters were selected, and the receptor-ligand interactions were analysed using both the Biovia Discovery Studio Visualiser and PyMol software.

Validation

In order to validate the molecular docking strategy, the structure of the QMR ligand found in the crystal structure of the protein had been exported as .pdf format and re-docked by following ligand preparation and molecular docking protocols. Furthermore, the known ligand varenicline was also retrieved from the PubChem Database (PubChem ID: 170361) and docked to the ligand-binding site using the same strategy.

Table 2. Molecular docking results of Lavandula essential compounds as well as QMR (re-docking) and Varenicline to 17nACh nicotinic receptor subunit α4β2.

Docking Results			
Compound	PubChem ID	Docking Score (kcal/mole)	Interacting Amino Acids
C1	1254	−6,3	TRP 57, TYR 100, TRP 156, TYR 197, CYS 199
C2	2537	−7,3	TYR 100, TRP 156, TYR 197, TYR 204
C3	2758	−6,7	TRP 57, TRP 156, TYR 197, CYS 199, CYS 200, TYR 204
C4	6448	−7,6	TRP 57, TRP 156, TYR 197, CYS 199
C5	6549	−6,5	LEU 121, TRP 156, TYR 204
C6	6654	−7,6	TRP 57, TYR 100, TRP 156, TYR 197, CYS 199, TYR 204
C7	6989	−6,1	LEU 121, TRP 156, TYR 197, CYS 199, TYR 204
C8	8294	−6,6	LEU 121, TRP 156
C9	10281	−6,7	TRP 57, LEU 121, TRP 156, CYS 199
C10	10582	−6,7	TRP 57, TYR 100, LEU 121, TRP 156, TYR 197, TYR 204
C11	11463	−6,5	TYR 100, PHE 119, LEU 121, TYR 197, CYS 199, CYS 200, TYR 204
C12	14896	−7,8	TRP 57, TYR 100, LEU 121, TRP 156, TYR 197, TYR 204
C13	16666	−6,3	TRP 57, TYR 100, TRP 156, TYR 197
C14	16724	−6,8	TRP 57, TYR 100, TYR 197, TYR 204
C15	26447	−6,7	TRP 156, TYR 197
C16	87839	−7,9	TRP 57, TYR 100, TRP 156, TYR 197, CYS 199, TYR 204
C17	92221	−7,1	TRP 57, TYR 100, TRP 156, TYR 197, TYR 204
C18	159055	−7,3	TYR 100, TRP 156, TYR 197, TYR 204
C19	220674	−6,4	ARG 81, LEU 121, TRP 156, THR 157, TYR 197, CYS 199, TYR 204
C20	330573	−6,7	TRP 57, TYR 100, PHE 119, LEU 121, TRP 156, TYR 197, CYS 199, CYS 200
C21	439570	−6,7	TYR 100, TYR 197, TYR 204
C22	440966	−7	TRP 57, TYR 100, TRP 156, TYR 197
C23	440967	−7,8	TRP 57, TYR 100, LEU 121, TRP 156, TYR 197, TYR 204
C24	442501	−7	TRP 57, TYR 100, TRP 156, TYR 197, CYS 199, TYR 204
C25	443162	−7	TRP 57, TYR 100, TRP 156, TYR 197, CYS 199, TYR 204
C26	444294	−7,2	TRP 57, TRP 156, TYR 197, CYS 199
C27	637531	−7,6	TYR 100, TRP 156, TYR 197, TYR 204
C28	637563	−6,1	TRP 57, ASN 109, VAL 111, LEU 121, TRP 156, CYS 199, CYS 200
C29	637566	−6,4	TRP 57, LEU 121, TYR 197, CYS 199, CYS 200
C30	638012	−6,7	TRP 157, TYR 100, TYR 197
C31	1201518	−6,9	TRP 57, LEU 121, TRP 156, TYR 197, CYS 199
C32	1549026	−6,9	TRP 57, TYR 100, LEU 121, TRP 156, TYR 197, CYS 199, TYR 204
C33	5281553	−6,3	TRP 57, TYR 100, PHE 119, LEU 121, TRP 156, CYS 199
C34	5325830	−6,8	TRP 57, TYR 100, TRP 156, TYR 197, CYS 199, TYR 204
C35	6321405	−7,1	TYR 100, TRP 156, TYR 197, CYS 199, TYR 204
C36	6552009	−7,2	TYR 100, TRP 156, TYR 197, TYR 204
C37	12223112	−7,6	TRP 57, TYR 100, TRP 156, TYR 197, CYS 199, TYR 204
QMR	CTRL	−9,1	TRP 57, TYR 100, LEU 121, TRP 156, CYS 199
Varenicline	170361	−9,1	TRP 57, PHE 119, LEU 121, TRP 156, TYR 197, CYS 199, TYR 204

ADME Study and Toxicity Profile

The absorption, distribution, metabolism, and Excretion (ADME) and toxicity properties of the three ligands with the highest binding affinities, as well as varenicline, were analysed using both the OSIRIS Property Explorer tool and the SwissADME server. The analysis included physico-chemical properties (formula, molecular weight, molar refractivity, and topological polar surface area (TPSA)), lipophilicity parameters (iLOGP, XLOGP3, WLOGP, MLOGP, SILICOS-IT, and Consensus Log Po/w), solubility properties (Log S, SILICOS-IT solubility in both mg/ml and mol/l units, and solubility class), drug-likeness properties (druglikeness and drug-score), pharmacokinetics parameters gastrointestinal (GI) absorption, blood brain barrier (BBB) permeant, P-gp substrate, CYP1A2 inhibitor, CYP2C19 inhibitor, CYP2C9 inhibitor, CYP2D6 inhibitor, and CYP3A4 inhibitor), and toxicity profiles (mutagenicity, tumorigenicity, irritant effects, and reproductive effects) of related ligands were revealed with swissADME server and possible toxicity properties including mutagenicity, tumorigenicity, reproductive effect, and irritant effect as well as drug likeness and drug scores have been predicted with OSIRIS property explorer tool.

RESULTS

In this study, the binding affinities of *Lavandula* essential compounds to the 17nACh nicotinic receptor subunit α4β2 were investigated using in silico techniques. A total of 37 compounds of *Lavandula* species were molecularly docked to the ligand-binding site of the receptor. Data demonstrating the PubChem IDs, binding affinities of the compounds and the amino acid residues that interact with them are listed in Table 2. The results demonstrated that the binding affinities of the compounds ranged from 6.1 to 7.9 kcal/mole interval. In particular, compounds C4 (2(-)-Bornyl acetate), C6 (2α-pinene), C12 (2(+)-β-Pinene), C16 ((S)cis-verbenol), C23 (2(-)-β-Pinene), C27 (ısobornyl acetate), and C37 ((1S)-(-)α-pinene) exhibited the highest binding affinities within the created library. As such, these compounds, along with varenicline, were filtered for further analysis based on Lipinski's rule of five and the ADME properties (Table 3).

Table 3. Filtering of the compounds having higher binding affinity and Varenicline with Lipinski's rule of five ADME properties.

Compounds	Lipinski's Rule of Five			
	<10 H-bond acceptors	<5 H-bond donors	<500 kDa Mw	<5 ClogP
C4	Yes	Yes	Yes	Yes
C6	Yes	Yes	Yes	No
C12	Yes	Yes	Yes	No
C16	Yes	Yes	Yes	Yes
C23	Yes	Yes	Yes	No
C27	Yes	Yes	Yes	Yes
C37	Yes	Yes	Yes	No
Varenicline	Yes	Yes	Yes	Yes

In addition, varenicline and its derivative QMR, which is found within the crystal structure of the protein, were docked to the ligand-binding region by repeating the molecular docking strategy. Both ligands exhibited a binding affinity of −9.1 kcal/mol to the protein. The re-docking of the QMR and the interactions between the ligands and the protein are demonstrated in Figure 1. While the re-docking result demonstrated the sufficiency of the developed docking strategy, it was revealed that both QMR may interact with TRP 57, TYR 100, LEU 121, TRP 156, and CYS 199 amino acids via carbon-hydrogen bond, pi-sigma, pi-sulfur, pi-pi T shaped, alkyl, and pi-alkyl interactions. Similarly, varenicline may interact with TRP 57, PHE 119, LEU 121, TRP 156, TYR 197, CYS 199, and TYR 204 amino acids via conventional hydrogen bond, pi-pi T-shaped, alkyl, and pi-alkyl interactions. In addition, the RMDS value demonstrating the sufficiency of the docking study through the analysis of the conformational convenience of the re-docked ligand with the compound in the crystal structure has been analysed with the DockRMSD online server. According to the computation of the server RMSD value between the QMR compounds in both the re-docked form and the crystal structure was defined as 0.880. Considering the binding affinities, RMSD value, which is less than 2.0, and the similarities between interacting amino acids and interaction types, the sufficiency of the study has been evaluated as sufficiently qualified for further studies. Furthermore, it was revealed that TRP 57, LEU 121, TRP 156, and CYS 199 are common amino acids with significant roles in the interaction with chemical compounds.

Among the *Lavandula* essential compounds, it was discovered from molecular docking and Lipinski's rule of five filtering that C4 (2α-pinene; Hydrocarbon), C16 ((S)cis-verbenol, alcohol), and C27(ısobornyl acetate, ester) are the most promising candidates, as they have binding affinities of 7.6, 7.9, and 7.6 kcal/mol, respectively, without any Lipinski violation. The interactions between these compounds and the related protein were visualized, demonstrating that C4 may interact with TRP 57, TRP 156, TYR 197, and CYS 199 amino acids; C16 may interact with TRP 57, TYR 100, TRP 156, TYR 197, CYS 199, and TYR 204 amino acids; and C27 may interact with TYR 100, TRP 156, TYR

197, and TYR 204 amino acids through conventional hydrogen bonds, pi-sigma, alkyl, and pi-alkyl interactions (Figure 2). Considering these results, it was demonstrated that these compounds have significant binding affinities to the ligand-binding site of the 17nACh nicotinic receptor subunit α4β2, interacting with common amino acids that play a role in binding with chemical compounds. Commonly TRP 57, TYR 100, TRP 156, TYR 197, and CYS 199 amino acids of the protein have been recognized by most of the docked ligands. In addition, these amino acids and those in the same location are primarily recognized by known inhibitors of the protein. As such, the essential amino acids that might be recognized to achieve inhibitory activity were revealed with this in silico study.

Furthermore, the ADME and toxicity properties of the three selected ligands and varenicline were predicted using in silico techniques (Table 4). The results demonstrated that the lipophilicity and solubility properties of the selected ligands were much more potent than those of varenicline. Although varenicline has the highest score among these compounds and shows no toxicity, the drug-likeness properties of the selected ligands suggest that they might only have an irritant effect, yet they are still assessed as possessing ideal drug characteristics with proper ADME features. In addition, while C4 and C27 have only a CYP2C9 inhibitory effect, C16 has no CYP isoform inhibitory activity and varenicline has a CYP2D6 inhibitory activity. Considering these results, it was evaluated that the selected ligands are promising candidates for targeting the 17nACh nicotinic receptor subunit α4β2 ligand binding region comparable to varenicline, as they have strong binding affinities, may recognize common interacting amino acids, and possess sufficient ADME and toxicity profiles. As such, these compounds that are isolated from *Lavandula* species could be repurposed for use in smoking cessation treatment.

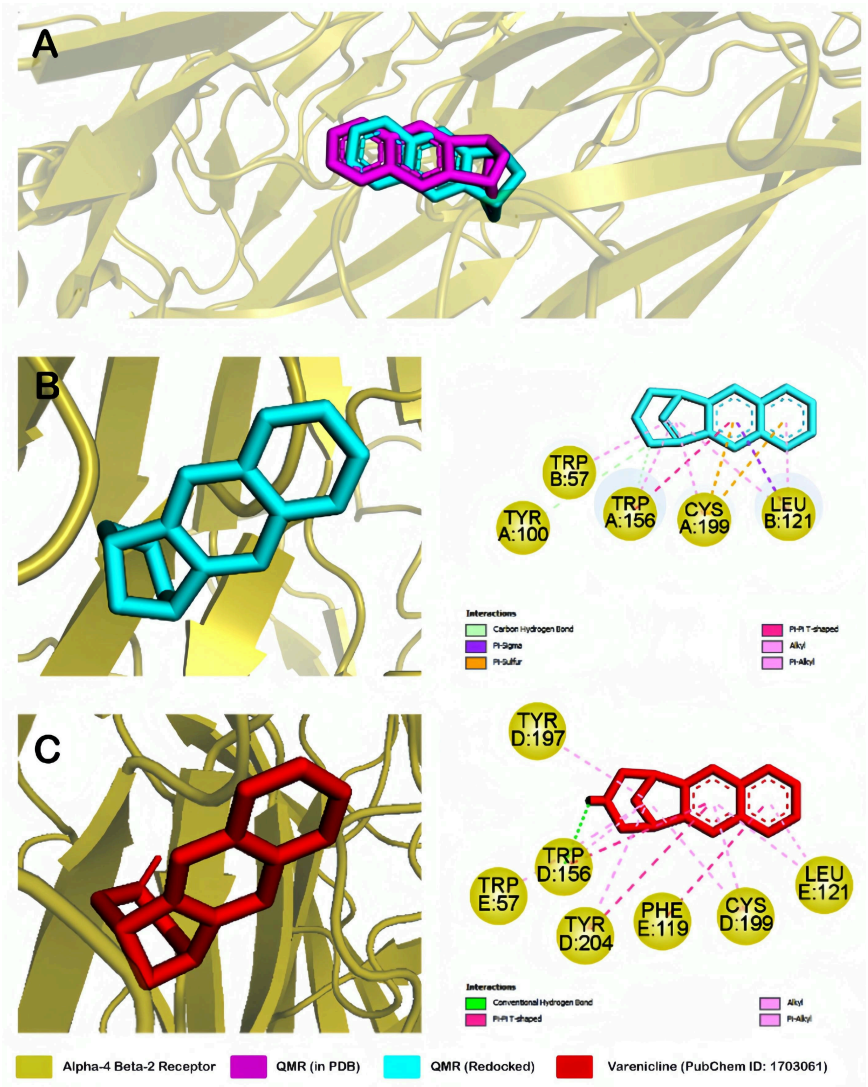


Figure 1. Validation results including poses of re-docked QMR and QMR in crystal structure of the protein; A) 3D demonstration of superimposition of re-docked QMR to protein icluding the inhibitor, B) 3D and 2D demonstrations of re-docked QMR interactions with the protein, and C) 3D and 2D demonstrations of varenicline interactions with the protein.

DISCUSSION

Lavandula, commonly known as lavender, is renowned for its captivating fragrance and a plethora of therapeutic benefits, largely attributed to its rich and complex composition of essential oils. Among the variety of compounds that contribute to its unique aromatic profile and medicinal properties, several stand out for their significant roles (Guo & Wang, 2020). *Lavandula spp.* and its essential oils have been shown to produce antispasmodic, vasodilator, antiviral, anti-inflammatory, antioxidant, antiallergic, antifungal and anticancer effects (Boukhatem et al., 2020; Lis-Balchin & Hart, 1999; Zuzarte et al., 2013). Among them, *L. stoechas* is traditionally used for headaches, *L. latifolia* as an abortifacient and *L. angustifolia* as a diuretic. However, the underlying mechanisms of the activities attributed to lavender oils have not yet been proven by extensive scientific studies (Cavanagh & Wilkinson, 2002).

In this study, the effects of the ingredients of *Lavandula* were compared with those of varenicline, a pharmacological agent used for smoking cessation therapy via alpha 4 beta 2 receptors. Varenicline has been associated with various side effects, particularly neuropsychiatric symptoms. These include symptoms commonly associated with quitting smoking and nicotine withdrawal, such as irritability, depression, and anxiety (McClure et al., 2009). Early reports suggested that varenicline might have psychiatric side-effects, including suicidal thoughts or psychosis (Smith et al., 2016). Additionally, varenicline has been linked to varenicline-induced psychotic depressive episodes in patients with bipolar disorder (Annagur & Bez, 2012). Varenicline is more effective at helping people to quit smoking than bupropion, or a single form of nicotine replacement therapy (NRT) and may be as or more effective than dual-form NRT. However, people taking varenicline are probably more likely to experience adverse events than those not taking it (Cahill et al., 2016), which suggests

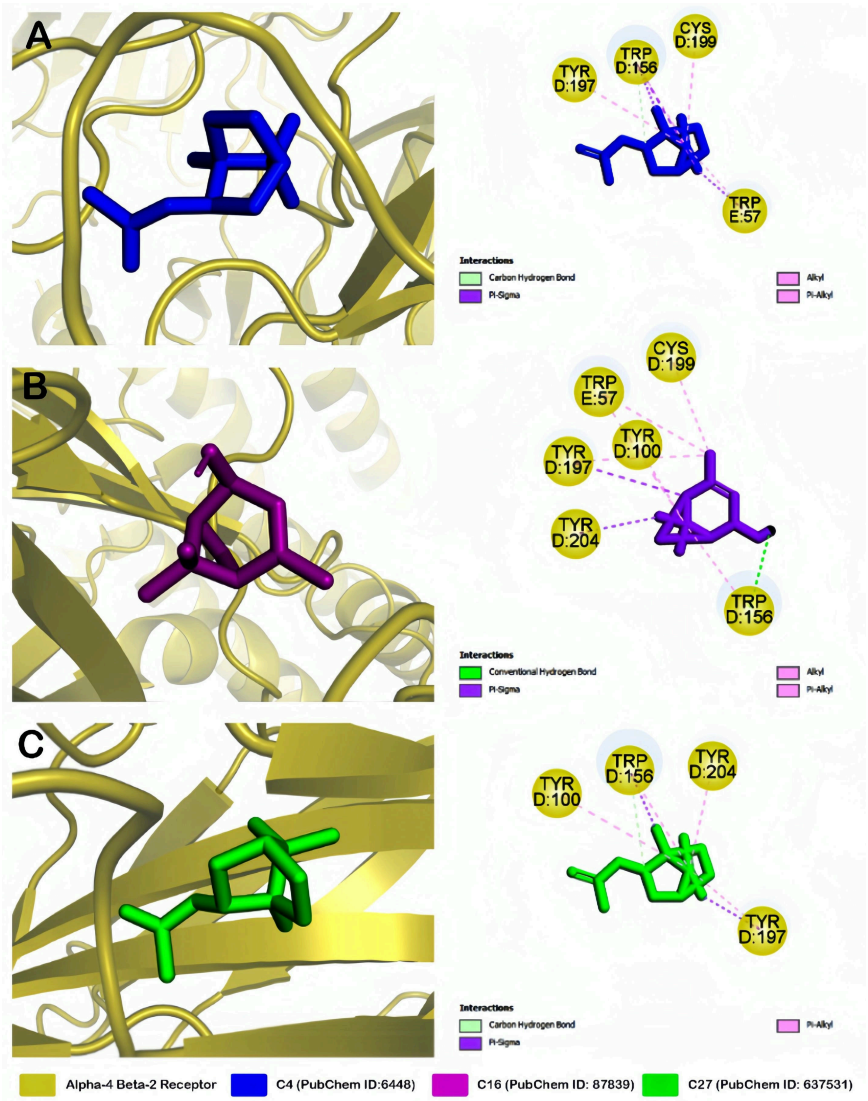


Figure 2. 3D and 2D demonstration of interaction types of selected Lavandula essential compounds with the protein's interacting amino acids; A) C4 (PubChem ID: 6448), B) C16 (PubChem ID: 87839), and C) C27 (PubChem ID: 637537).

Table 4. ADME and Toxicity properties of selected Lavandula essential compounds and known α4β2 subunit inhibitor varenicline

ADME and Toxicity Properties					
Properties		Selected Compounds			Inhibitor
Physico-chemical properties	Ligand Name	C4	C16	C27	Varenicline
	Formula	C12H20O2	C10H16O	C12H20O2	C13H13N3
	Molecular Weight (g/mol)	196.29	152.23	196.29	211.26
	Molar Refractivity	56.33	46.38	56.33	66.88
	TPSA (topological polar surface area)	26.30 Å²	20.23 Å²	26.30 Å²	37.81 Å²
Lipophilicity	Log $P_{o/w}$ (iLOGP)	2.50	2.26	2.50	2.01
	Log $P_{o/w}$ (XLOGP3)	4.30	3.16	4.30	0.79
	Log $P_{o/w}$ (WLOGP)	2.76	1.97	2.76	1.42
	Log $P_{o/w}$ (MLOGP)	2.76	2.30	2.76	1.45
	Log $P_{o/w}$ (SILICOS-IT)	2.66	1.86	2.66	2.43
	Consensus Log Po/w	3.00	2.31	3.00	1.62
Solubility	Log S (SILICOS-IT)	−2.58	−1.44	−2.58	−4.31
	SILICOS-IT Solubility (mg/ml)	2.20e-01	5.57e-00	5.20e-01	1.04e-02
	SILICOS-IT Solubility (mol/l)	2.65e-03	3.665e-02	2.65e-03	4.94e-05
	Solubility Class	Soluble	Soluble	Soluble	Moderately Soluble
Druglikeness	Druglikeness	−3.69	−1.42	−3.69	0.86
	Drug-score	0.28	0.34	0.28	0.79
Pharmacokinetics	GI absorption	High	High	High	High
	BBB permeant	Yes	Yes	Yes	Yes
	P-gp substrate	No	No	No	Yes
	CYP1A2 inhibitor	No	No	No	No
	CYP2C19 inhibitor	No	No	No	No
	CYP2C9 inhibitor	Yes	No	Yes	No
	CYP2D6 inhibitor	No	No	No	Yes
	CYP3A4 inhibitor	No	No	No	No
Toxicity	Mutagenicity	No	No	No	No
	Tumorigenicity	No	No	No	No
	Irritant Effects	Yes	Yes	Yes	No
	Reproductive Effects	No	No	No	No

the need for alternative treatments, especially for people with intolerance. The *in silico* results of this study highlight the potential use of *L. stoechas* and its active compounds, providing insight beyond the traditional use of this plant (Kirmizibekmez et al., 2009). The molecular docking results of the study indicate that 2α-pinene (C4; Hydrocarbon), (S)cis-verbenol (C16, alcohol), and isobornyl acetate (C27, ester) are the most promising compounds of *L. stoechas*.

Bornyl acetate is a significant compound found in various plant extracts and essential oils (Wang et al., 2018). Structurally, isobornyl acetate is an ester derived from isoborneol, characterized by its pleasant woody and pine-like aroma, making it commonly used in fragrances and flavors. From a therapeutic standpoint, isobornyl acetate exhibits autonomic relaxation and antimicrobial properties. It also regulates myeloperoxidase activity, which correlates with the inhibition of lung inflammatory responses (Chen et al., 2014). The presence of an acetate group may enhance its lipid solubility, facilitating better interaction with lipid membranes and thereby increasing its bioavailability and efficacy in biological systems. Bornyl acetate is identified as a major constituent of *L. stoechas*, which exhibits various bioactivities, including

antimicrobial, antioxidant, and potential antiviral properties. Studies emphasized that the essential oil's chemical composition, highlighting bornyl acetate's role in contributing to these effects. Specifically, the essential oils from *L. stoechas*, characterized by the presence of bornyl acetate among other compounds, have been studied for their efficacy against bacterial and fungal pathogens, showing significant inhibitory activity (Benali et al., 2023). Moreover, the antioxidant capacity of these oils, presumably influenced by bornyl acetate and other components, suggests a protective role against oxidative stress (Kim et al., 2013; Kusumoto et al., 2014; Matsubara et al., 2011; Yang et al., 2014).Additionally, this compound inhibits inflammatory responses in the lungs via the modulation of myeloperoxidase activity (Chen et al., 2014). This study indicates that bornyl acetate may also play a crucial role in the smoking cessation effects of *L. stoechas* via alpha 4 beta 2 receptors. Along with its antioxidant and anti-inflammatory properties in the lungs, the compound may have promising effects on smoking-induced damage in the lungs in addition to its possible effects on smoking cessation.

α-Pinene has shown potential effects in prolonging thrombin time and preventing platelet aggregation, pointing out its potential for prevention of thrombosis and cardiovascular events. The hydrophobic nature of α-pinene enhances its ability to interact with cell membranes, potentially affecting membrane-bound enzymes and receptors. α-Pinene offers protection against H2O2-induced oxidative stress in neural cells. By reducing the production of inflammatory mediators such as IL-6 and TNF-α, α-pinene has demonstrated significant anti-inflammatory and analgesic effects (Salehi et al., 2019). Its structure, with two fused rings and a double bond, may contribute to its stability and reactivity, enhancing its biological activity. This study indicates that α-Pinene component *of L. stoechas* may also interact with alpha 4 beta 2 receptors. Together with its antioxidant and anti-inflammatory properties, the compound may have promising effects on smoking-induced cardiovascular effects with its possible effects on smoking cessation.

(S)-cis-verbenol compound of *L. stoechas* (Iscan & Oraloglu, 2019), derived from (-)-alpha-pinene in pine trees, demonstrates significant anti-ischemic and anti-inflammatory activities. It protects against cerebral ischemic injury by reducing neuronal cell death in oxygen-glucose deprivation models by reducing intracellular reactive oxygen species (ROS) levels and potently eliminates peroxyl radicals, indicating a strong antioxidant action. Additionally, it decreases the expression of pro-inflammatory cytokines in ischemic brain and immune-stimulated glial cells, highlighting its potential as a therapeutic agent due to its anti-oxidative and anti-inflammatory properties (Choi et al., 2010). The hydroxyl group in its structure enhances its polarity and solubility in water, which may improve its interaction with polar biomolecules and cellular components. Although its therapeutic properties have not been extensively studied in the past few years, the molecular docking analysis of this study indicates that the verbenol compound is one of the most promising components of *L. stoechas* without any possible pharmacokinetic interactions and might contribute to the smoking cessation effects of this plant. The unique chemical structures and physicochemical properties of 2α-pinene, (S)-cis-verbenol, and isobornyl acetate contribute to their outstanding performance in molecular docking studies. These properties may enhance their bioavailability, stability, and efficacy, leading to better inhibition of targeted pathways and providing therapeutic benefits, particularly in the context of smoking cessation and related health issues.

In addition, a previously published study by Mohamed and his colleagues investigating the binding profiles of essential compounds of *Lupinus subcarnosus* as well as varenicline to alpha 4 beta 2 receptors have demonstrated that essential compounds may interact with TRP 156, THR 157, TYR 197, CYS 199, CYS 200 amino acids of the protein (Mohamed et al., 2022). In particular, that same amino acids have been recognized with both varenicline and the selected compounds of *Lavandula stoechas* demonstrate the sufficiency of the developed in silico strategy of the study. Furthermore, comparing the published data, the binding affinities of the discovered compounds in this study are quite enough to recognize alpha 4 beta 2 receptor.

CONCLUSION

In conclusion, this study underscores the significant potential of *Lavandula stoechas* and its key compounds—2α-pinene, (S)-cis-verbenol, and isobornyl acetate—in offering alternative therapeutic approaches for smoking cessation. By comparing the effects of these compounds with the pharmacological agent varenicline, known for its adverse neuropsychiatric effects, our research highlights a promising, natural avenue for addressing the challenges associated with smoking and nicotine withdrawal. Molecular docking results reveal that these compounds not only exhibit promising interactions with alpha 4 beta 2 receptors but also bring forth their intrinsic antioxidant, anti-inflammatory, and antimicrobial properties. These findings extend beyond the traditional applications of *L. stoechas*, proposing its compounds as potential multi-functional agents for smoking cessation therapies. Such natural alternatives could mitigate the side effects associated with current pharmacological treatments, offering a safer and po-

tentially more effective solution. Future studies should focus on clinical trials to fully elucidate the efficacy and safety of these compounds, paving the way for novel, plant-based interventions in addiction medicine.

Peer Review: Externally peer-reviewed.
Author Contributions: Conception/Design of Study: E.B.; Data Acquisition: H.S.P. ; Data Analysis/Interpretation: E.B., H.S.P.; Drafting Manuscript: E.B., H.S.P.; Critical Revision of Manuscript: E.B., H.S.P.; Final Approval and Accountability: E.B., H.S.P.
Conflict of Interest: The authors have no conflict of interest to declare.
Grant Support: The authors declared that this study has received no financial support.

Author Details

Elif Barış
[1] İzmir University of Economics, Faculty of Medicine, Medical Pharmacology, İzmir, Türkiye
0000-0001-6838-7932

Hüseyin Saygın Portakal
[2] İzmir University of Economics, Faculty of Engineering, Genetics and Bioengineering, İzmir, Türkiye
0000-0002-3582-4152 saygin.portakal@izmirekonomi.edu.tr

REFERENCES

Alam, M., Hasan, G. M., Ansari, M. M., Sharma, R., Yadav, D. K., & Hassan, M. I. (2022). Therapeutic implications and clinical manifestations of thymoquinone. *Phytochemistry*, *200*, 113213. https://doi.org/10.1016/j.phytochem.2022.113213

Angioni, A., Barra, A., Coroneo, V., Dessi, S., & Cabras, P. (2006). Chemical composition, seasonal variability, and antifungal activity of Lavandula stoechas L. ssp. stoechas essential oils from stem/leaves and flowers. *Journal of Agricultural and Food Chemistry*, *54*(12), 4364–4370. https://doi.org/10.1021/jf0603329

Annagur, B. B., & Bez, Y. (2012). Varenicline-induced psychotic depressive episode in a patient with bipolar disorder. *Therapeutic Advances in Psychopharmacology*, *2*(1), 35–37. https://doi.org/10.1177/2045125311430111

Benali, T., Lemhadri, A., Harboul, K., Chtibi, H., Khabbach, A., Jadouali, S. M., Quesada-Romero, L., Louahlia, S., Hammani, K., Ghaleb, A., Lee, L.-H., Bouyahya, A., Rusu, M. E., & Akhazzane, M. (2023). Chemical Profiling and Biological Properties of Essential Oils of Lavandula stoechas L. Collected from Three Moroccan Sites: In Vitro and In Silico Investigations. *Plants (Basel, Switzerland)*, *12*(6). https://doi.org/10.3390/plants12061413

Benli, A. R., Erturhan, S., Oruc, M. A., Kalpakci, P., Sunay, D., & Demirel, Y. (2017). A comparison of the efficacy of varenicline and bupropion and an evaluation of the effect of the medications in the context of the smoking cessation programme. *Tobacco Induced Diseases*, *15*, 10. https://doi.org/10.1186/s12971-017-0116-0

Betlej, I., Andres, B., Cebulak, T., Kapusta, I., Balawejder, M., Jaworski, S., Lange, A., Kutwin, M., Pisulewska, E., Kidacka, A., Krochmal-Marczak, B., & Borysiuk, P. (2023). Antimicrobial Properties and Assessment of the Content of Bioactive Compounds Lavandula angustifolia Mill. Cultivated in Southern Poland. *Molecules (Basel, Switzerland)*, *28*(17). https://doi.org/10.3390/molecules28176416

Boukhatem, M. N., Sudha, T., Darwish, N. H. E., Chader, H., Belkadi, A., Rajabi, M., Houche, A., Benkebailli, F., Oudjida, F., & Mousa, S. A. (2020). A New Eucalyptol-Rich Lavender (Lavandula stoechas L .) Essential Oil: Emerging. *Molecules*, *25*(3671), 1–17.

Cahill, K., Lindson-Hawley, N., Thomas, K. H., Fanshawe, T. R., & Lancaster, T. (2016). Nicotine receptor partial agonists for smoking cessation. *The Cochrane Database of Systematic Reviews*, *2016*(5), CD006103. https://doi.org/10.1002/14651858.CD006103.pub7

Cavanagh, H. M. A., & Wilkinson, J. M. (2002). Biological activities of lavender essential oil. *Phytotherapy Research*, *16*(4), 301–308. https://doi.org/10.1002/ptr.1103

Chen, N., Sun, G., Yuan, X., Hou, J., Wu, Q., Soromou, L. W., & Feng, H. (2014). Inhibition of lung inflammatory responses by bornyl acetate is correlated with regulation of myeloperoxidase activity. *Journal of Surgical Research*, *186*(1), 436–445. https://doi.org/10.1016/j.jss.2013.09.003

Chen, X., Shang, S., Yan, F., Jiang, H., Zhao, G., Tian, S., Chen, R., Chen, D., & Dang, Y. (2023). Antioxidant Activities of Essential Oils and Their Major Components in Scavenging Free Radicals, Inhibiting Lipid Oxidation and Reducing Cellular Oxidative Stress. *Molecules (Basel, Switzerland)*, *28*(11). https://doi.org/10.3390/molecules28114559

Choi, I.-Y., Lim, J. H., Hwang, S., Lee, J.-C., Cho, G.-S., & Kim, W.-K. (2010). Anti-ischemic and anti-inflammatory activity of (S)-cis-verbenol. *Free Radical Research*, *44*(5), 541–551. https://doi.org/10.3109/10715761003667562

de Almeida Cunha NB, Orozco CM, de Morais Pordeus LC, & Fernandes Braga JE. (2018). Effects of essential oil of Lavandula angustifolia in patients with cigarette craving. *Journal of Medicine and Therapeutics*, *2*(3), 1–6. https://doi.org/10.15761/JMT.1000133

Dos Santos, É. R. Q., Maia, J. G. S., Fontes-Júnior, E. A., & do Socorro Ferraz Maia, C. (2022). Linalool as a Therapeutic and Medicinal Tool in Depression Treatment: A Review. *Current Neuropharmacology*, *20*(6), 1073–1092. https://doi.org/10.2174/1570159X19666210920094504

Guo, X., & Wang, P. (2020). Aroma Characteristics of Lavender Extract and Essential Oil from Lavandula angustifolia Mill. *Molecules (Basel, Switzerland)*, *25*(23). https://doi.org/10.3390/molecules25235541

Hajiyeva K (2018) Design of nicotinic acetylcholine antagonists with virtual ligand scanning method. Unpublished Master Thesis, Hacettepe University, Department of Chemistry.

Iscan, G., & Oraloglu, Z. (2019). Karabaşotu Çaylarinda KâfurMiktaTayini. *Gıda*, *44*(5), 881–888. https://doi.org/10.15237/gida.gd19091

Kim, S. H., Lee, S. Y., Hong, C. Y., Gwak, K. S., Park, M. J., Smith, D., & Choi, I. G. (2013). Whitening and antioxidant activities of bornyl acetate and nezukol fractionated from Cryptomeria japonica essential oil. *International Journal of Cosmetic Science*, *35*(5), 484–490. https://doi.org/10.1111/ics.12069

Kirmizibekmez, H., Demirci, B., Yeşilada, E., Başer, K. H. C., & Demirci, F. (2009). Chemical composition and antimicrobial activity of the essential oils of Lavandula stoechas L. ssp. stoechas growing wild in Turkey. *Natural Product Communications*, *4*(7), 1001–1006.

Kusumoto, N., Zhao, T., Swedjemark, G., Ashitani, T., Takahashi, K., & Borg-Karlson, A.-K. (2014). Antifungal properties of terpenoids in Picea abies against Heterobasidion parviporum. *Forest Pathology*, *44*(5), 353–361. https://doi.org/https://doi.org/10.1111/efp.12106

Lis-Balchin, M., & Hart, S. (1999). Studies on the mode of action of the essential oil of lavender (Lavandula angustifolia P. Miller). *Phytotherapy Research: PTR*, *13*(6), 540–542. https://doi.org/10.1002/(sici)1099-1573(199909)13:6<540::aid-ptr523>3.0.co;2-i

Ma, J., Xu, H., Wu, J., Qu, C., Sun, F., & Xu, S. (2015). Linalool inhibits cigarette smoke-induced lung inflammation by inhibiting NF-κB activation. *International Immunopharmacology*, *29*(2), 708–713. https://doi.org/10.1016/j.intimp.2015.09.005

Matsubara, E., Fukagawa, M., Okamoto, T., Ohnuki, K., Shimizu, K., & Kondo, R. (2011). (-)-Bornyl acetate induces autonomic relaxation and reduces arousal level after visual display terminal work without any influences of task performance in low-dose condition. *Biomedical Research*, *32*(2), 151–157. https://doi.org/10.2220/biomedres.32.151

McClure, J. B., Swan, G. E., Jack, L., Catz, S. L., Zbikowski, S. M., McAfee, T. A., Deprey, M., Richards, J., & Javitz, H. (2009). Mood, side-effects and smoking outcomes among persons with and without probable lifetime depression taking varenicline. *Journal of General Internal Medicine*, *24*(5), 563–569. https://doi.org/10.1007/s11606-009-0926-8

Mihalak, K. B., Carroll, F. I., & Luetje, C. W. (2006). Varenicline is a partial agonist at alpha4beta2 and a full agonist at alpha7 neuronal nicotinic receptors. *Molecular Pharmacology*, *70*(3), 801–805. https://doi.org/10.1124/mol.106.025130

Mohamed, M., Alghamdi, S., Alquraini, A., Alghamdi, A., Shatla, I., El-Shorbagi, A.-N., Chaudhary, S., Aly, O., & Mustafa, M. (2022). Phytochemical and in silico study on Lupinus subcarnosus Hook, its effect on neuronal α4β2 nicotinic acetylcholine receptors (nAChRs) and the major alkaloids. *Octahedron Drug Research*. https://doi.org/10.21608/odr.2022.155742.1007

Oraloglu, Z. (2018). *Bitkisel Drog Monografı Hazırlanması: Lavandula stoechas L . Yüksek Lisans Tezi Zeynep ORALOĞLU Eskişehir 2018*. Anadolu University.

Şahinler, S. Ş., Sever Yilmaz, B., Sarıkurkcu, C., & Tepe, B. (2022). The importance of Lavandula stoechas L. in pharmacognosy and phytotherapy. *International Journal of Secondary Metabolite*, *9*(3), 360–375. https://doi.org/10.21448/ijsm.1098975

Salehi, B., Upadhyay, S., Erdogan Orhan, I., Kumar Jugran, A., L D Jayaweera, S., A Dias, D., Sharopov, F., Taheri, Y., Martins, N., Baghalpour, N., Cho, W. C., & Sharifi-Rad, J. (2019). Therapeutic Potential of α- and β-Pinene: A Miracle Gift of Nature. *Biomolecules*, *9*(11). https://doi.org/10.3390/biom9110738

Smith, R. C., Amiaz, R., Si, T.-M., Maayan, L., Jin, H., Boules, S., Sershen, H., Li, C., Ren, J., Liu, Y., Youseff, M., Lajtha, A., Guidotti, A., Weiser, M., & Davis, J. M. (2016). Varenicline Effects on Smoking, Cognition, and Psychiatric Symptoms in Schizophrenia: A Double-Blind Randomized Trial. *PloS One*, *11*(1), e0143490. https://doi.org/10.1371/journal.pone.0143490

Wang, H., Ma, D., Yang, J., Deng, K., Li, M., Ji, X., Zhong, L., & Zhao, H. (2018). An Integrative Volatile Terpenoid Profiling and Transcriptomics Analysis for Gene Mining and Functional Characterization of AvBPPS and AvPS Involved in the Monoterpenoid Biosynthesis in Amomum villosum. *Frontiers in Plant Science*, *9*, 846. https://doi.org/10.3389/fpls.2018.00846

Yang, H., Zhao, R., Chen, H., Jia, P., Bao, L., & Tang, H. (2014). Bornyl acetate has an anti-inflammatory effect in human chondrocytes via induction of IL-11. *IUBMB Life*, *66*(12), 854–859. https://doi.org/10.1002/iub.1338

Zuzarte, M., Gonçalves, M. J., Cavaleiro, C., Cruz, M. T., Benzarti, A., Marongiu, B., Maxia, A., Piras, A., & Salgueiro, L. (2013). Antifungal and anti-inflammatory potential of Lavandula stoechas and Thymus herba-barona essential oils. *Industrial Crops and Products*, 44, 97–103. https://doi.org/10.1016/j.in