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Investigation of the chemical composition, antioxidant and anticholinesterase activities of *Consolida orientalis*

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ABSTRACT

Aims: It is known that a decrease in the amount of acetylcholine in the body, which is known to be responsible for learning and cholinergic activity in the nervous system, causes Alzheimer's disease. Acetylcholine is destroyed by acetylcholinesterase (AChE) and Butyrylcholinesterase (BChE) enzymes in the nervous system. *Consolida orientalis (C. orientalis)* is a species that belongs to the Ranunculaceae family and grows naturally in many parts of the world. It is known that it plays a role in many biological activities thanks to its content of important phytochemical components such as phenolics and alkaloids. In this study; It was aimed to investigate the antioxidant activity, AChE and BChE enzyme inhibition activities of *C. orientalis* flower extracts.

Methods: The chemical content of ethanol extracts obtained from the flowers of *C. orientalis* plant, which was collected and identified from Sivas İmaret village between June and July 2023, was examined with 6 different reference substances (gallic acid, rosmarinic acid, myrcetin, quercetin, apigenin and camphorol). Antioxidant activity was determined by 2,2-Diphenyl-1-picrylhydrazyl (DPPH) and 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) tests. Ascorbic acid and trolox were used as positive controls. The inhibition capacity of the samples on AChE and BChE enzymes was determined by the Ellman method.

Results: Chemical content analysis of the extract was performed by high pressure liquid chromatography (HPLC) and only gallic acid was detected among the standard compounds. When C. orientalis ethanol extracts were examined with DPPH and ABTS tests, they showed low-moderate antioxidant activity (IC50 (µg/ml) DPPH=4.8, IC50 (µg/ml) ABTS=4.4) compared to standard substances. *C. orientalis* ethanol extract was studied at a concentration of 20 µg/ml. The extract inhibited the AChE enzyme at 66.5% and the BChE enzyme at 53.2%. It was observed that the extract inhibited both enzymes at moderate to good levels, although not higher than galantamine used as positive control.

Conclusion: This study shows us that *C. orientalis* flowers have therapeutic potential for the effective management of neurological disorders due to their antioxidant and anticholinesterase activity. It is thought that our data will contribute to the literature as a preliminary study for the development of a new phytotherapeutic agent in the treatment of Alzheimer's disease.

Keywords: Acetylcholinesterase, Alzheimer's disease, antioxidant, butyrylcholinesterase, *Consolida orientalis*

INTRODUCTION

When the activity of free radicals in the body exceeds the body's own defense mechanism, a condition called oxidative stress occurs. Oxidative stress caused by the imbalance between the neutralization and production of free radicals by the antioxidant mechanism leads to irreversible cell damage, cardiovascular disease, cancer, accelerated aging, and many diseases, including neurodegenerative disorders such as Alzheimer's and Parkinson's.[1,2]

Efforts are being made to elucidate why and how the disease occurs with different hypotheses such as cholinergic, amyloid and oxidative stress. Among these, the cholinergic hypothesis; It is the only hypothesis that clarifies the cause of Alzheimer's disease and is currently accepted in the scientific world. According to this hypothesis, a decrease in the amount of acetylcholine, an important neurotransmitter that increases learning and cholinergic activity in the nervous system, causes Alzheimer's. Acetylcholine is destroyed by AChE and BChE enzymes in the nervous system.[3] For this reason, recently the phytochemicals contained in plants; Its inhibitory effects on acetyl and butyrylcholinesterase enzymes, which play a major role in the treatment of neurodegenerative diseases such as Alzheimer's and dementia, are being investigated.

Traditionally used herbal medicines are of great interest as pharmacological targets in the prevention and treatment of various diseases. Although compounds isolated from plants

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play a very important role in the pharmaceutical industry; it is known that many plants are still not sufficiently researched in terms of their phytochemical components and biological activities.

Consolida is a genus belonging to the *Ranunculaceae* family, consisting of approximately 52 species that grow naturally in many parts of the world, such as Western Europe, the Mediterranean and Asia. Many members of the genus *Consolida* contain important phytochemicals such as phenolics and alkaloids.[1,4] In 1960, the chemical structure of 11 alkaloids obtained from the *Consolida* plant was determined and it was reported that these alkaloids could affect the neuromuscular pathway.[5] Turkiye's flora is very rich in species belonging to the *Consolida* genus from the *Ranunculaceae* family. It is known that there are 29 different species, 14 of which are endemic. *Consolida*; it is a breed that can easily grow in dry climatic conditions and is frequently seen in steppes, deserts and dry stony slopes.[6,7] *C. orientalis* is a medium-tall, sticky-hairy annual plant with simple or branched stems 20-74 cm long and numerous ribbon-like sword leaves. The flowers are purple in color and in the form of dense clusters. It is frequently preferred in the cut flower trade and dry plant designs as dried and fresh cut flower plants.[8]

Phenolic acids, tyrosol derivatives, diterpene alkaloids, lignans and stilbenes, mainly flavonoids, are responsible for the biological activity of *C. orientalis*.[1,4] Previous *in vivo* and *in vitro* studies have shown that; It has antioxidant, antibacterial, anti-tyrosinase, anticancer effects as well as polyphenols and phytosterols (β-sitosterol).[1,9,10]

The purpose of this study; Determination of phytochemical content of *C. orientalis* flower extracts by HPLC, investigation of antioxidant activity, AChE and BChE enzyme inhibition activities.

METHODS

Ethics

No human or animal biological material was used in the study. This is a laboratory study with *Consolida orientalis* plants. Therefore, ethics committee decision is not required. All procedures were carried out in accordance with the ethical rules and the principles.

Plant Material

The *C. orientalis* plant used in the study was collected in June-July 2023 from the area with an altitude of 1400 m, at the coordinates of 39°41'40"N, 37°02'25"E, in Sivas İmaret village. The taxonomic description of the collected samples was made by Anadolu University Faculty of Pharmacy Faculty Member Professor Yavuz Bülent KÖSE. A sample of the collected plant was labeled with the herbarium number (16195) and was recorded and stored in the Herbarium of Anadolu University Faculty of Pharmacy.

Preparation of Plant Extract

Plant extracts prepared from the flower parts of *C. orientalis* were used in our study. After the samples were washed first with tap water and then with pure water, they were dried on blotting papers, ground in a grinder and 100 grams were taken and 1000 ml of ethanol was added. It was kept at room temperature in a shaker at 150 rpm for 24 hours. At the end of the extraction process, the extract was filtered through filter paper, and then the solvent was removed in a rotary evaporator at 40°C. The obtained extract was placed in a dark glass bottle and stored at -20 °C to be used in experimental procedures.[11]

High Pressure Liquid Chromatography (HPLC)

C. orientalis ethanol extract was studied at a concentration of 10 mg/ml and analyzed by filtering through a 0.22 µm membrane filter after dissolving in ethanol. Solutions of the standard substances used (gallic acid, rosmarinic acid, myrcetin, quercetin, apigenin and camphorol) were prepared with methanol. HPLC analyzes were carried out with a UV-DAD detector connected to the Agilent 1100 HPLC system. While C18 column (250 x 4.6mm, 5μm) was used as the stationary phase, A [acetonitrile: distilled water: formic acid (10:89:1, v/v)] and B (acetonitrile: distilled water: formic acid [89:10:1, v/v)] were used as the mobile phase. The flow rate was set to 1.0 ml/min and gradient flow was provided. For the gradient flow B mobile phase, a range of 15-100% was studied in the 40-minute method. Each sample was injected in triplicate. Peaks were analyzed at 330 nm. Injection volume was set as 20 µL, column temperature was set at 40°C.[12]

Antioxidant Capacity

2.2-Diphenyl-1-picrylhydrazyl (DPPH) antioxidant activity test: The total antioxidant capacity of *C. orientalis* ethanol extract was determined using the DPPH method described by Blois et al.[13] The reaction mixture contained 100 µM DPPH• and formulations in methanol. After 30 min, absorbance was read at 517 nm using a UV spectrophotometer (UV-1800, Shimadzu, Japan) at 25±2 °C.[14] Ascorbic acid was used as a positive control. Results are calculated as IC50 (µg/ml).

2.2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid) (ABTS) antioxidant activity test: The antioxidant capacity of the samples was determined by Re et al.[15] ABTS was measured using the radical cation decolorization protocol described by. ABTS and 2.45 mm potassium persulfate dissolved in 7 mm water; mixed to form ABTS. The mixture was kept in a dark room at 25°C for 16 hours before use. Ethanol was added to the mixture and absorbance was measured at 734 nm at 25°C. The process was carried out in three repetitions. Ethanol was used as a negative control and trolox as a positive control.[14] Results are calculated as IC50 (µg/ml).

Acetylcholinesterase and Butyrylcholinesterase Inhibition

Cholinesterase inhibition was determined spectrophotometrically by the modified Ellman method. Standard galantamine was applied as a positive control, and the solvent ethanol was used as a negative control. Sodium phosphate buffer (pH:8.0), AChE enzyme stock solution and *C. orientalis* ethanol extract at a concentration of 20 µg/ml were mixed and incubated for 30 minutes. Following the incubation, the reaction was started after adding Ellman's reagent 5.5-dithio-bis-(2-nitrobenzoic acid) (DTNB) and acetylthiocholine. Butyrylthiocholine chloride was used as a

substrate to test the BChE enzyme under the same conditions. The reaction was observed by monitoring the formation of the yellow 5-thio-2-nitrobenzoate anion, which forms as a result of the reaction between DTNB and thiocholine, at a wavelength of 412 nm. All experiments were repeated three times and results are reported as % inhibition value.[16]

RESULTS

High Pressure Liquid Chromatography (HPLC)

The column leaving times (*t*R) of the standard compounds used for HPLC analyzes (gallic acid, rosmarinic acid, myrcetin, quercetin, apigenin and camphorol) and the presence of these standards in *C. orientalis* flower extract are given in Table 1.

Table 1. Retention times of standard compounds and their presence in the extract

Standard compound	t_R	Status on statement
Gallic acid	3.752	Detected
Rosmarinic acid	9.595	None
Myrcetin	10.437	None
Quercetin	14.334	None
Apigenin	17.060	None
Camphorol	18.354	None

t_R: Retention time

The HPLC chromatogram of *C. orientalis* flower extract is shown in Figure. When compared to the chromatogram of standard compounds, only gallic acid with the same *t*R value was detected in the extract.

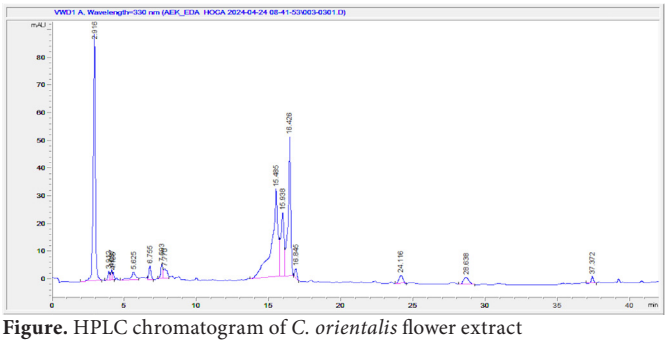

Figure. HPLC chromatogram of *C. orientalis* flower extract

Antioxidant Capacity

The antioxidant capacity of *C. orientalis* ethanol extract measured using DPPH and ABTS methods (ascorbic acid and trolox were used as positive control, respectively) is shown in Table 2. When the antioxidant capacity of the extract was evaluated according to the results of the DPPH test, the IC50 value was calculated as 4.8 µg/ml, and when evaluated according to the results of the ABTS test, it was calculated as 4.4 µg/ml.

Table 2. DPPH and ABTS activities of *C. orientalis* ethanol extract

	C. orientalis ethanol extract	Positive control
	IC50 (µg/ml)	
DPPH•	4.8	0.002 (Ascorbic acid)
ABTS•	4.4	0.01 (Trolox)

DPPH: 2,2-Diphenyl-1-picrylhydrazyl, ABTS: 2,2'-azino-bis (3-ethylbenzothiazoline-6-sulfonic acid)

In the study on the antioxidant activity of *C. orientalis* methanolic extract,[17] the antioxidant capacity was calculated in % and it was determined that different results were obtained at different concentrations. Considering the concentration closest to that used in this study (0.2 mg/ml), the antioxidant capacity of the methanol extract in the study was determined to be 8.97%. This result is compatible with the results obtained in our study.

Acetylcholinesterase and Butyrylcholinesterase Inhibition

C. orientalis ethanol extract was studied at a concentration of 20 µg/ml. The results of the AChE and BChE activity tests of the extract are given in Table 3. The extract inhibited the AChE enzyme at 66.5% and the BChE enzyme at 53.2%. It was observed that the extract inhibited both enzymes at moderate to good levels, although not higher than galantamine used as positive control.

Table 3. Cholinesterase enzyme inhibition of *C. orientalis* ethanol extract (20 µg/ml)

Tested samples	AChE inhibition	BChE inhibition
C. orientalis ethanol extract	%66.5	%53.2
Galantamine (Positive control)	%99.2	%86.9

Studies show that medicinal plants have a wide range of therapeutic effects and have therapeutic potential for the effective management of neurological disorders associated with AChE dysregulation.[18] For this reason, this study was designed about AChE and BChE inhibition of *C. orientalis* plant.

DISCUSSION

There are not enough studies in the literature regarding the medical effects of *C. orientalis*. In particular, no anticholinesterase activity study has been found for this species. Our study is the first in this sense. When the studies were examined, it was determined by researchers that it has antioxidant, antibacterial, anti-tyrosinase and anticancer effects.

The *Consolida* genus contains important bioactive components such as phenolic compounds and alkaloids. In the study conducted with methanol extracts of 6 different species (*C. glandulosa, C. hellospontica, C. raveyi, C. regalis, C. staminosa* and *C. stenocarpa*); 236 flavonoids, 93 phenolic acids, 78 tyrosol derivatives, 49 diterpene alkaloids, 29 lignans and 7 stilbenes were found in the content of the plants. It is known that other phytochemicals, especially polyphenolic compounds (gallic acid, quercetin) and diterpene alkaloids, are responsible for many biological activities such as antimicrobial, antiparasitic, antioxidant and antitumor in the plant.[4,19,20]

In the studies conducted, norditerpenoid, diterpenoid and norditerpene (18-demethylpubeskenine) alkaloids were found in the content of *C. orientalis*.[19,21,22]

DPPH and ABTS radical scavenging tests of ethanol extracts of different organs of the *Consolida regalis* plant belonging to the *Consolida* genus were examined in terms of antioxidant activity. It was concluded that all extracts showed cleaning

activity depending on concentration, thus the plant has antioxidant activity.[23]

The antioxidant activity of methanol, ethyl acetate and water extracts of *C. orientalis* was investigated, the extract with the best antioxidant capacity was found to be methanol, and the extract with the lowest antioxidant capacity was ethyl acetate.[17]

In the study where the antibacterial and antioxidant activities of different extracts of *C. orientalis* were evaluated against selected bacteria; The extracts have high antimicrobial activity against *Proteus mirabilis*, *Enterobacter cloacae*, *Klebsiella pneumonia* and *Staphylococcus aureus*; It has also been reported to display weak nitric oxide scavenging activity and Fe^{2} chelating ability.[9]

In a study conducted to determine the cytotoxic effect of *C. orientalis* flower extract; It was determined that plant extracts did not show any cytotoxic effect on the WI-38 human fibroblast cell line even at a concentration of 5 mg/100 µL.[24]

In another study; It was aimed to evaluate the *in vitro* cytotoxic activity of the ethanol extract of *C. orientalis* collected from Mazandaran in the north of Iran using the human cervical carcinoma cell line HeLa. In the results of working; The anticancer potential of the ethanolic extract of the plant in the HeLa cell line has been proven. And this result showed us the presence of cytotoxic compounds in ethanolic extracts of *C. orientalis.*[10]

In the study conducted with different species of the *Consolida* genus (*C. glandulosa, C. hellospontica, C. raveyi*); As a result of cholinesterase inhibition analyses, it was observed that extracts of *C. hellospontica* and *C. glandulosa* exhibited high inhibitory effects.[4]

Ghanbarpour et al.[25] reported that *Consolida* extract could be used as a strategy to control tick resistance to synthetic acaricides.[26]

Limitqations

The most important limitation of the study is that only 6 of the phytochemical components in the plant extract (gallic acid, rosmarinic acid, myrcetin, quercetin, apigenin and camphorol) could be investigated. In addition, the absence of similar studies in the literature limits our study in terms of references.

CONCLUSION

Acetylcholinesterase inhibitors used in current treatment reduce the symptoms of Alzheimer's disease and slow the progression of the disease, but do not provide a complete cure. Therefore, recently in the prevention and treatment of Alzheimer's disease; studies on the detection of acetylcholinesterase and butyrylcholinesterase inhibitors obtained from new and natural sources and without toxicity have gained great importance.

In our study, the chemical content of the ethanol extracts of the flowers of the *C. orientalis* plant was examined by HPLC; Flavonoid-derived compounds (gallic acid, rosmarinic acid, myrcetin, quercetin, apigenin and camphorol), which are common in plants, were investigated and only gallic acid was detected in the extract. The reason for this is; It is known that the phytochemical profiles of plants vary depending on the time the plant was collected, the characteristics of the soil in which it grows, and other characteristics of the geographical region where it is located. Its antioxidant capacity and effects on acetylcholinesterase and butyrylcholinesterase enzymes were investigated, and a moderate to good effect of the plant was determined, especially on AChE and BChE enzymes. It is thought that our data will contribute to the literature as a preliminary study for the development of a new phytotherapeutic agent in the treatment of Alzheimer's disease. For further research, it is recommended to be supported by *in vitro* and *in vivo* studies.

ETHICAL DECLARATIONS

Ethics Committee Approval

No human or animal biological material was used in the study. This is a laboratory study with *Consolida orientalis* plants. Therefore, ethics committee decision is not required.

Informed Consent

No human or animal biological material was used in the study. This is a laboratory study with *Consolida orientalis* plants. Therefore, informed consent is not required.

Referee Evaluation Process

Externally peer-reviewed.

Conflict of Interest Statement

The authors have no conflicts of interest to declare.

Financial Disclosure

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Author Contributions

All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

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REFERENCES

1. Dehpour AA. Antioxidant activity and ısolation of β-sitosterol from ethyl acetate extract of aerial parts of *Consolida orientalis*. *Preprints (Basel).* 2016. doi:10.20944/preprints201609.0037.v1
2. Rayess YE, Nehme L, Ghanem C. Phenolic content, antioxidant and antimicrobial activities evaluation and relationship of commercial spices in the lebanese market. *BMC Chem.* 2023;17(1):157. doi:10.1186/s13065-023-01074-2
3. Demir Z, Türkan F. Asetilkolinesteraz ve bütirilkolinesteraz enzimlerinin alzheimer hastalığı ile ilişkisi. *Iğdır Üni Fen Bil Enstit Derg.* 2022;12(4):2386-2395. doi:10.21597/jist.1161271

4. Rocchetti G, Zengin G, Cakmak YS. A UHPLC-QTOF-MS screening provides new insights into the phytochemical composition and biological properties of six Consolida species from Turkey. *Ind Crops Prod*. 2020;158:112966. doi:10.1016/j.indcrop.2020.112966
5. Olsen JD, Sisson DV. Toxicity of extracts of tall larkspur (*Delphinium barbeyi*) in mice, hamsters, rats and sheep. *Toxicol Lett*. 1991;56(1-2):33-41. doi:10.1016/0378-4274(91)90087-M
6. Ertuğrul K, Arslan E, Tugay O. Characterization of *Consolida* SF Gray (Ranunculaceae) taxa in Turkey by seed storage protein electrophoresis. *Turk J Biochemist*. 2010;35:99-104.
7. Liu Y, Tikunov Y, Schouten RE, Marcelis LFM, Visser RGF, Bovy A. Anthocyanin biosynthesis and degradation mechanisms in solanaceous vegetables: a review. *Front Chem*. 2018:6. doi:10.3389/fchem.2018.00052
8. Mansuroglu S, Karaguzel O, Ortacesme V. Effect of paclobutrazol on flowering, leaf and flower colour of *Consolida orientalis*. *Pak J Bot*. 2009;41(5): 2323-2332.
9. Dehpour AA, Ebrahimzadeh M, Roudgar A, Nabavi SF, Nabavi SM. Antioxidant and antibacterial activity of *Consolida orientalis*. *World Acad Sci Eng Technol*. 2011;73:162-166.
10. Nemati F, Dehpouri AA, Eslami B, Mahdavi V, Mirzanejad S. Cytotoxic properties of some medicinal plant extracts from Mazandaran, Iran. *Iran Red Crescent Med J*. 2013;15(11). doi:10.5812/ircmj.8871
11. Mata AT, Proença C, Ferreira AR, Serralheiro MLM, Nogueira JMF, Araújo MEM. Antioxidant and antiacetylcholinesterase activities of five plants used as Portuguese food spices. *Food Chem*. 2007;103(3):778-786. doi:10.1016/j.foodchem.2006.09.017
12. Okur ME, Ayla Ş, Karadağ AE, Çiçek Polat D, Demirci S, Seçkin İ. Opuntia ficus indica fruits ameliorate cisplatin-ınduced nephrotoxicity in mice. *Biol Pharm Bull*. 2020;43(5):831-838. doi:10.1248/bpb.b19-01044
13. Blois MS. Antioxidant determinations by the use of a stable free radical. *Nature*. 1958;181(4617):1199-1200. doi:10.1038/1811199a0
14. Karadağ AE, Demirci B, Çaşkurlu A. In vitro antibacterial, antioxidant, anti-inflammatory and analgesic evaluation of *Rosmarinus officinalis* L. flower extract fractions. *South African J Botany*. 2019;125:214-220. doi:10.1016/j.sajb.2019.07.039
15. Re R, Pellegrini N, Proteggente A, Pannala A, Yang M, Rice-Evans C. Antioxidant activity applying an improved ABTS radical cation decolorization assay. *Free Radic Biol Med*. 1999;26(9-10):1231-1237. doi:10.1016/S0891-5849(98)00315-3
16. Karadağ AE, Demirci B, Kültür Ş, Demirci F, Başer KHC. Antimicrobial, anticholinesterase evaluation and chemical characterization of essential oil *Phlomis kurdica* Rech. fil. growing in Turkey. *J Essent Oil Res*. 2020;32(3):242-246. doi:10.1080/10412905.2020.1743786
17. Zengin G, Mahomoodally MF, Picot-Allain CMN, Cakmak YS, Uysal S, Aktumsek A. In vitro tyrosinase inhibitory and antioxidant potential of *Consolida orientalis*, *Onosma isauricum* and *Spartium junceum* from Turkey. *South African J Botany*. 2019;120:119-123. doi:10.1016/j.sajb.2018.01.010
18. Patel SS, Raghuwanshi R, Masood M, Acharya A, Jain SK. Medicinal plants with acetylcholinesterase inhibitory activity. *Rev Neurosci*. 2018;29(5):491-529. doi:10.1515/revneuro- 2017-0054
19. Şener B, Orhan I, Özçelik B. Diterpenoid alkaloids from some Turkish *Consolida* species and their antiviral activities. *Arkivoc*. 2006;2007(7):265-272. doi:10.3998/ark.5550190.0008.722
20. Zengin G, Mahomoodally MF, Picot-Allain CMN, Cakmak YS, Uysal S, Aktumsek A. In vitro tyrosinase inhibitory and antioxidant potential of *Consolida orientalis*, *Onosma isauricum* and *Spartium junceum* from Turkey. *South African J Botany*. 2019;120:119-123. doi:10.1016/j.sajb.2018.01.010
21. Hohmann J, Forgo P, Hajdú Z, Varga E, Máthé I. norditerpenoid alkaloids from *Consolida orientalis* and complete 1H and 13C NMR signal assignments of some lycoctonine-type alkaloids. *J Nat Prod*. 2002;65(7):1069-1072. doi:10.1021/np020026z
22. Meriçli F, Meriçli AH, Ulubelen A, Desai HK, Pelletier SW. Norditerpenoid and diterpenoid alkaloids from Turkish *Consolida orientalis*. *J Nat Prod*. 2001;64(6):787-789. doi:10.1021/np0005558
23. Ucar E. *In-vitro* antioxidant and antimicrobial activities and various enzyme inhibitory activities of ethanol extracts from different organs of *Consolida regalis*. *Fresenius Environ Bull*. 2018;27(9):5950-5957.
24. Özpınar N. *Consolida orientalis* (Gay.) Schröd. ekstraktlarının *Acanthamoeba castellanii* trofozoit ve kist formları üzerine ameboisidal etkisi ve sitotoksik potansiyeli. *Inter J Academ Med Pharm*. 2020;EK-1(EK-1):34-39. doi:10.29228/jamp.42842
25. Ghanbarpour K, Tavassoli M, Shamsi L. Pesticide effects of *Consolida orientalis* extract on larval stage of *Argas persicus* (Acari: Argasidae). *Persian J Acarol*. 2019;8(3):265-270. doi:10.22073/pja.v8i3.45290
26. Demir Z, Türkan F. Asetilkolinesteraz ve bütirilkolinesteraz enzimlerinin alzheimer hastalığı ile ilişkisi. *J Instit Sci Technol*. 2022;12(4):2386-2395.