ÖZGÜN ARAŞTIRMA ORIGINAL RESEARCH

Med J SDU / SDÜ Tıp Fak Derg ▶ 2019:26(4):436-442 doi: 10.17343/sdutfd.534049

EVALUATION OF ANTIMICROBIAL ACTIVITIES AND INVITRO CYTOTOXIC ACTIVITIES OF GUNDELIA TOURNEFORTII L. PLANT EXTRACTS

GUNDELİA TOURNEFORTİİ L. BİTKİ EKSTRAKTLARININ ANTİMİKROBİYAL AKTİVİTELERİNİN VE İNVİTRO SİTOTOKSİK AKTİVİTELERİNİN DEĞERLENDİRİLMESİ

Betül ÖZALTUN[1], Taner DAŞTAN[2]

[1] Nigde Omer Halisdemir University, Faculty of Medicine, Cardiology, Nigde

[2] Cumhuriyet University, Yıldızeli Vocational School, Department of Medical Services and Techniques, Sivas

Cite this article as: Özaltun B, Daştan T. Evaluation of Antimicrobial Activities and In Vitro Cytotoxic Activities of Gundelia tournefortii L. Plant Extracts. Med J SDU 2019; 26(4): 436-442.

Öz

Giriş

Kenger, Asteraceae familyasından Gundelia tournefortii bitkisi, tıbbi bir bitkidir. Kenger'in çiçeklerinin, yapraklarının, tohumlarının ve köklerinin besin kaynağı olarak kullanıldığı bilinmektedir.

Gereç ve Yöntem

Bu çalışmada Sivas ilinde, taze filizleri soyularak çiğ ve pişirilerek yenilen Kenger'in gövde kısımlarının su fazı ve uçucu yağ ekstraksiyonu yapılarak, özütlerin antimikrobiyal ve in vitro antiproliferatif özellikleri bakımından değerlendirilmesi amaçlanmıştır. Sitotoksik aktiviteleri Meme Kanseri Hücre Hattı (MCF-7) ve insan endotelyal hücre hattı (HUVEC) kullanılarak araştırılmıştır. Bitki özütlerinin hücre kültürlerindeki etkileri, XTT yöntemiyle Eliza reader cihazında absorbans bulunarak elde edilmiştir. Kenger bitki özütlerinin MCF-7 hücreleri ve endotelyal hücreler üzerinde sitotoksik etkileri belirlenmiştir.

Bulgular

Kenger bitki özütlerinin zayıf antimikrobiyal etkilerinin olduğu MIC değerleriyle ortaya konulmuştur.

Sonuç

Sivas ilinde halk arasında şifalı olduğu bilinen, bahar ve yaz aylarında büyüyen bitki kısımları kesilip soyularak iç kısımlarının besin olarak tüketildiği, çeşitli hastalıkların tedavisinde kullanılan Kenger bitkisinin, denediğimiz hücreler üzerinde sitotoksik etkilerinin olduğu, zayıf antimikrobiyal etkisinin bulunduğu görülmektedir.

Anahtar Kelimeler: Kenger,antimikrobiyal,antikanserojen,ekstraksiyon

Abstract

Objective

Kenger, Gundelia tournefortii plant from Asteraceae family, is a medicinal plant. Flowers, leaves, seeds and roots of Kenger have been reported to be used as a source of food.

Material and Methods

In this study, it was aimed to evaluate the antimicrobial and in vitro antiproliferative properties of Kenger which fresh sprouts are peeled and eated cooked or uncooked in Sivas. The water phase and essential oil

İletişim kurulacak yazar/Corresponding author: betulozaltun@hotmail.com

Müracaat tarihi/Application Date: 01.03.2019 • Kabul tarihi/Accepted Date: 03.05.2019

Available online at http://dergipark.gov.tr/sdutfd

Makaleye http://dergipark.gov.tr/sdutfd web sayfasından ulaşılabilir.

extraction of the body parts of Kenger were aimed to be evaluated in terms of their antimicrobial and in vitro antiproliferative properties.

Results
Cytotoxic activities were investigated using the Breast Cancer Cell Line (MCF-7) and human endothelial cell line (HUVEC). The effects of plant extracts on cell cultures were determined by the absorbance of Eliza reader by XTT method. Kenger plant extracts have been shown to have weak antimicrobial effects with MIC values.

Conclusions
In the province of Sivas, growing parts of the plant in the spring and summer are cut and peeled into the inner parts of the nutrients. Kenger, which is used in the treatment of various diseases, has cytotoxic effects on the cells we tested and has a weak antimicrobial effect.

Keywords: Kenger, antimicrobial, anticarcinogen, extraction

Introduction

Due to their unique flavor and aroma, antimicrobial, antioxidant and anticarcinogenic properties, wild plants have a wide bioactivity profile (1). The geographic structure and climatic features of our country provide an extremely rich variety of plants (2). However, in our country, there are limited studies on the identification of wild plants consumed as food in different regions, their intended use and nutritional values (3-8).

As in all over the world plants and obtained products like extract, drug etc. are widely used in the treatment of various diseases in our country, too. However, scientific data on the biological effects and mechanisms of most of the extracts from medicinal plants are still insufficient. On the other hand, various plants are used to be developed as potential antibacterial, antitumoral and antiviral materials in recent years, synthetic or semi-synthetic production of the active ingredients obtained from these plants is accelerated. It has been reported that it has been used in our country and used for medical purposes among the people,

Kenger (Gundelia tournefortii) plant, which is well known in Sivas, has not been investigated in detail, but in the province, lohences fed to increase milk, crushed and mixed with honey for those who want to have children, people who have palpitations and anxiety problems are recommended to be consumed by boiling the root parts, used as stomach soothing by chewing gum in digestive disorders and seeds are crushed into coffee, used locally for many medical purposes. According to the literature, Gundelia tournefortii is a medical plant from the family Asteraceae (Compositae) and grown temperate regions of western Asia (Turkey, Cyprus, Egypt, Iran, Israel, Jordan, Azerbaijan and Turkmenistan). In Turkey, the local name known as “kenger”. Gundelia tournefortii ‘s flowers, leaves, seeds and roots have been reported to be used as a source of food (9). In the Middle East fresh and undeveloped flower buds are often sold in regional markets (10). The dry seeds of Gundelia tournefortii in Eastern Anatolia are used for the treatment of vitiligo. Fresh seeds are used in pickling and are also an effective diuretic. Gundelia tournefortii is used externally for latex cuts obtained from roots and also chewed as gum (11). Gundelia tournefortii ‘s antioxidant activity is investigated (12,13) and seeds have higher antioxidant potential than other parts of the plant. In Iran, the body part of the plant is considered as hepatoprotective and blood cleaner (14-15). Gundelia tournefortii L. extracts caffeic acid (CA) and three caffeic acid [neochlorogenic acid (3-COA), Cryptoclorogenic acid (4-CQA), chlorogenic acid (5-CQA)] derivatives by HPLC. Wagner et al. (1984) found seven saponin compounds in this plant and and the mixture of these compounds have a strong mollussicidal activity (16). Sekeroglu et al. (2012) found high amounts of essential unsaturated fatty acids linoleic and oleic acid in this plants’ seeds (17). In the literature Gundelia tournefortii extracts have not been published about anticancer activity. In Sivas province, it is considered to be beneficial, nowadays, plant culture studies are continuing. It is aimed to evaluate the antimicrobial and antiproliferative properties of the water phase extraction and essential oil extraction of the body parts of the Kenger plant in this study.

Material and Method

Plant Specimen

Kenger, Gundelia tournefortii L., plant was collected from pasture lands in the Sivas region during the May-October period (B7 Sivas, step area) . The plants was diagnosed according to the book “ Flora of Turkey” by Dr. Mustafa Sevindik (1502) (18). The collected plant samples were cut and dried in room conditions and in the shade then stored in colored jars for use in analysis.

Preparation of plant extracts
For the water phase extract obtained from the Kenger plant; 100 g plant was used. The amounts used for extraction are dry plant weight. The plant parts were milled in house rondo. Distilled water (solid: liquid ratio 1:10) at 1000 mL 80 0C was added to 100 g of plant sample and the plant sample was allowed to infuse for 30 minutes. It was then filtered hot through filter paper and lyophilized to remove the solvent. Samples were weighed for quantification of the extractable compounds. Extracts were obtained in 2.92% and 2.38% (w/w) yields in leaf and root extractions, respectively. Etracts were kept at -20 ºC in color bottles until analysis. In order to investigate the biological activities of plant extracts, each extract was dissolved in distilled water at 1 mg / mL to form stock material. Subsequent steps included dimethyl sulfoxide (DMSO) for microbial activities and dilutions in Dulbecco's modified Eagle's medium (DMEM) for cell cultures.

Extraction of the essential oil
For extracting essential oil from the body parts of the Kenger plant, dried plant parts were hydrodistilled for 3 hours using Clevenger and the yield of essential oil was found to be 0.2% (v/w). After drying with sodium sulfate in desiccator, the oil obtained was stored at -20 ° C until used.

In vitro cytotoxic activity measurements
In experiments Human breast adenocarcinoma cell (MCF-7) and normal human umbilical vein endothelial cell line (HUVECs) were used.

Amplification and reproduction of cells
MCF-7 and HUVEC cells were transplanted into 96 well plates during incubation of cells at 25 cm2 flasks in a 37 ° C 5% CO2 incubator, DMEM (high glucose, 2mM L-glutamine and sodium pyruvate) and 10% Cultured on FBS-containing media. The flask was selected and the sowing process was started. 5 ml of Trypsin-EDTA solution (0.25%) and phosphate buffer (PBS) were added to remove the flask adhering cells. The flask was then transferred to the falcon so that a flask would be a falcon. The upper phase was poured, the cells were slowly struck by the falcon and the cells were added with 20 ml of DMEM and 5 ml of FBS. A mixture of 200µl was placed in 96 wells (5 x 10 3 cells in 100 µl// plate cavity) and 96 well plates were placed to incubator.

Application of Herbal Extract to Cells
Plant extracts were dissolved in water. 5 different doses were applied to the cells. The extracts of different concentrations were added to the cells on 96-well plates, respectively, with 3 replicates, and 96 well plates were placed in the incubator for 24 and 48 hours. Plant extracts prepared at a stock concentration of 1 mg/ mL were applied to cell cultures for 24 and 48 hours, with varying doses of 50 µg/mL to 1000 µg/mL.

Administration to cells XTT and Cell Vitality
XTT (2, 3-bis- (2-methoxy - 4 - nitro-5 - sulfophenyl) - 2Htetrazolium-5 - carboxanilide) in which the tetrazolium salt is present. This salt can only be converted into formazan by cells that have metabolic activity to detect living cells. XTT is a formazan salt with water solubility. When used in conjunction with Medium, which does not contain phenol red, it creates a color close to orange color. It is based on the principle that XTT, the yellow tetrazolium salt of metabolically active cells, converts into orange formazan paint. This transformation occurs only in living cells. Formazan is soluble in solutions and can be directly measured by microplate reader. When starting trials, XTT solution should be prepared just before use. 50 µl of XTT solution was added to each well and 96 plates were placed to the incubator. Plates were incubated at 37 ºC for 5 hours in the incubator. After the incubation period, the plate was shaken slightly and the paint was re-mixed in the wells and were placed on Elisa device and absorbance reading was made at 450-500 nm.

Antimicrobial Activity Measurements
Microdilution Broth Method
The microdilution broth method was used to determine the minimum inhibition concentration (MIC) of microorganisms of Kenger plant extracts against microorganisms (19). In this study, Staphylococcus aureus (ATCC 29213), Enterococcus faecalis (ATCC 29212), Pseudomonas aeruginosa (ATCC 27853), Escherichia coli (ATCC 25922), Bacillus cereus (ATCC11778), Klebsiella pneumonia (ATCC 13883), Candida albicans (ATCC 10231) and Candida tropicalis (DSM11953) microorganisms were used. Mueller Hinton Broth (Accumix® AM1072) were used for bacteria. Sabouraud Dextrose Broth (Himedia ME033) was used for Candida albicans and Candida tropicalis. 90 µl was added to the wells first line of the microtiter plates and 50 µl was added to the other wells. The 11th wells were used as sterility control and 100 µl was added to the broth (20,21). The twelfth wells were used as reproductive control. 10 µl of extract was added to the first row of wells and serial dilution was made. The microorganisms were extracted from the microorganisms grown on the blood agar broth and the suspension was prepared from the microorganisms in the McFarland 0.5 turbidity. 50 µl of a microorganism suspension was added to each well to 5 x 10 5 CFU / mL for bacteria and 0.5-2.5

x 103 CFU / mL for Candida albicans. Plaques with added bacteria were incubated for 16-24 hours at 37 °C, with Candida albicans incubated at 35 °C. 50 µl 2 mg/ml 2,3,5-triphenyltetrazolium chloride (TTC) (Merck, Germany) was added to each well to make the growth visible at the end of the incubation period and incubated at 37 °C for 2 hours. The first wells without color change in the wells were accepted as the MIC value. The test was repeated 3 times.

Statistical Analysis

SPSS 22.0 (IBM Corporation, Armonk, New York, United States) program was used to analyze the data. Normal distribution of univariate data Kolmogorov-Smirnov test with Lilliefors correction, Mardia for multivariate normal distribution with Shapiro-Wilk test and Variability coefficients; (Dornik and Hansen omnibus) test, Levene test was used for homogeneity of variance. The Independent-Samples T test was used with the Bootstrap results in comparison of the two independent groups. One-Way Anova (Robust Test: Brown-Forsythe) test was used in conjunction with the Bootstrap results, and LSD, Dunnett and Games Howell tests were used for post hoc analysis. Quantitative data are expressed as mean ± std. (Standard deviation) values in the tables. Categorical data are expressed by n (number) and percentages (%) Data were examined at 95% confidence level and p value was considered to be significant if less than 0.05.

Results

Antimicrobial activity results

The antimicrobial activity values of the studied samples were significant when they were 0.1 mg/ml or lower, they were reported to be moderately effective in the range of 0.1 < MIC ≤ 0.625 mg/ml and weakly effective when the MIC value was more than 0.625 mg/ml (22,23). Kenger plant parts were found to be weak on all microorganisms we tested (Table 1).

Cytotoxic activity results

Plant extracts were applied to MCF-7 and HUVEC cell lines for 24 and 48 hours at 50, 100, 250, 500 and 1000 µg/mL concentrations and XTT test was performed. All plant extracts were found to have cytotoxic activity on MCF-7 cells and HUVEC cells both at 24 hours and 48 hours and in general at all administration doses (*P< 0.05,**P< 0.01; Table 2, Figure 1 and 2). Kenger plant essential oils have been found to be the most effective in cell lines according to both doses and time. On the other hand, root and leaf extracts were generally found to have similar levels of cytotoxic activity. When the effects of extracts were compared according to application doses, differences between all doses were found to be significant (Table 2). In addition, as a result of the applications during 24 h and 48 h, cytotoxic changes in cell lines were found to be significant (p <0.005; Table 2). Although the cytotoxic effects of the different extracts of the kenger plant on both cell lines were statistically significant, plant extracts on MCF-7 cell lines were much more cytotoxic (Figure 1,2).

Discussion

Gundelia tournefortii, is called Kenger in Sivas locally, is known to be used in the treatment of various diseases among people, however, their effects with scientific data have not been studied extensively. Nowadays, it is important to reveal its biological activities due to the fact that it is a plant which has been started to be cultured in agriculture. In this study, the antimicrobial activity of this plant, its cytotoxic activities in MCF-7 and HUVEC cell cultures were investigated and it was aimed to reveal the potential effects in alternative medicine applications. According to our results, we can say that different parts of this plant have no effective antimicrobial activity on the microorganisms we tested. On the other hand, serious cytotoxic effects of the extracts in MCF-7 cancer cell line and HUVEC cells were determined within 24 and 48 hours (P> 0.05). Decreases in cell viability were observed due to dose and duration and the results were statistically significant (Table 2, figure 1-2).

Konak et al. (2017) According to their study, the bioavailability of antioxidants of Kenger plant was determined as approximately 95% by ABTS method. In conclusion, this study shows that Gundelia tournefortii is a potential source of antioxidants and can be consumed on a daily basis as a natural source of antioxidants (24). In another study by Karaaslan et al. (2014), Gundelia tournefortii was found to be rich in vitamin C and reduced glutathione (GSH). The low MDA content of the plant is considered to be an indication that the plant is not under oxidative stress. In addition, Gundelia tournefortii plant is soluble in water-soluble antioxidants and vitamins such as C, E is stated to be a rich plant (25). In our study, the cytotoxic effect of kenger plant extracts in cell lines can be related to the high antioxidant and oxidative damage of the plant highlighted in both studies. Abu Lafi et al. (2019) in a study of the methanol, hexane and water phase of Gundellia tournefortii extract Human Colon cancer (HCT-116) cell line, anticancer activities were looked at and other extracts of the plant outside the water phase, cytotoxic effect was found (26). In our study, extracts obtained from different parts of Gun-

dellia tournefortii plant have been found to be cytotoxic in human breast cancer (MCF-7) cell line.

The fact that no synthetic extract or any natural substance derived from plants or plants or cytotoxic effects of a single active substance on cancerous cells, while not damaging the intact cells, may have the potential for developing drug substances for the future. Considering that the toxic effects of synthetic cancer drugs and treatment methods on normal cells and their use in the clinic are quite costly, the demands for drugs developed from natural plant sources are increasing day by day.

In order to find the molecules responsible for the anti-proliferation activity observed in our study, we need to conduct further research to separate the kenger extract into its components and to purify the bioactive compounds individually, and to plan projects in the field of pharmacognosy. On the other hand, kenger (Gundelia tournefortii) plant should be studied in many different cell lines except MCF-7 cells and its

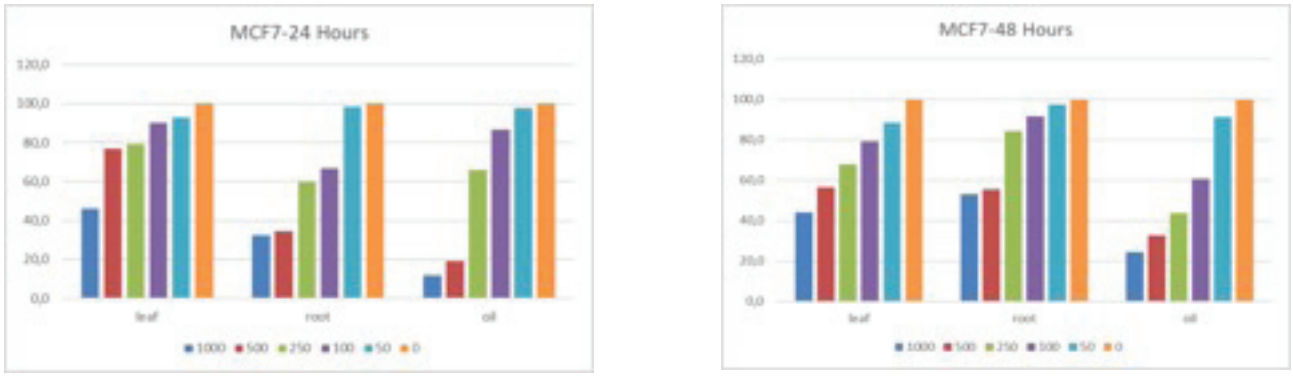

Figure 1. Cytotoxic activities of MCF-7 cell line depending on doses of plant extracts.
A. 24 hours administration B. 48 hours administration

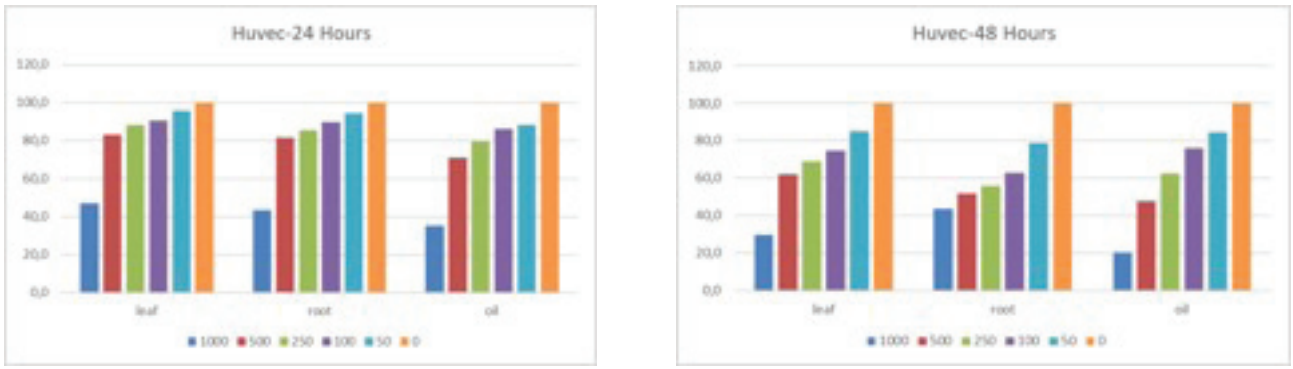

Figure 2. Cytotoxic activities of plant extracts in HUVEC cell line depending on doses
A: 24 hours administration B: 48 hours administration

Table 1 Antimicrobial activities of Gundellia tournefortii plant parts

	E.coli	S.aureus	P.aeruginosa	K.pneumoniae	B.cereus	E.faecalis	C.albicans	C.tropicalis
	ATCC 25922	ATCC 29213	ATCC 27853	ATCC 13883	ATCC11778	ATCC29212	ATCC10231	DSM11953
root	>5	0.5	0.625	>2.5	2.5	0.625	0.625	>5
leaf	>5	0.5	0.625	>2.5	>2.5	0.625	0.625	>5
Ess.oil	>5	0.5	0.625	>2.5	2.5	0.625	0.625	>5

Table 2 Comparison of the effects of various parts of the Gundelia tournefortii plant on cell lines in terms of time, dose and cell lines.

	Doze (µg/mL)	MCF7		HUVEC		P Values			
		24 h	48 h	24 h	48 h	MCF7 (24-48)	Huvec (24-48)	24 s (Huvec-MCF7)	48 s (Huvec-MCF7)
leaf	1000	45,9±3,35	44,0±3,24	47,1±0,65	29,9±0,04	0,266	0,001	0,612	0,017
	500	76,9±3,42	56,6±4,56	82,8±2,07	62,2±5,74	0,001	0,244	0,062	0,256
	250	79,5±0,56	67,8±1,73	88,0±0,76	69,1±0,47	0,001	0,001	<0,001	0,260
	100	90,0±0,11	79,2±0,56	90,5±0,29	74,6±0,54	0,001	0,001	0,072	0,031
	50	92,8±0,16	88,7±0,82	95,7±2,44	85,0±3,00	0,001	0,001	0,176	0,112
	0	100,0±1,15	100,0±0,22	100,0±2,45	100,0±3,13	1	1	1	1
root	1000	32,3±1,38	53,1±0,65	43,5±0,92	43,5±2,02	0,001	0,768	<0,001	0,001
	500	34,4±2,14	55,4±0,73	81,4±3,24	51,7±7,98	0,001	0,001	<0,001	0,501
	250	59,7±2,88	84,5±0,22	85,4±0,76	55,8±0,71	0,001	0,001	<0,001	<0,001
	100	66,6±2,85	91,6±0,53	89,7±1,59	62,9±0,54	0,001	0,001	<0,001	<0,001
	50	98,5±0,67	97,6±0,96	94,3±0,88	78,8±0,88	0,245	0,001	0,003	<0,001
	0	100,0±0,17	100,0±2,43	100,0±1,24	100,0±4,39	1	1	1	1
ess.oil	1000	11,6±0,67	24,2±1,81	35,0±10,43	20,3±0,75	0,241	0,113	0,018	0,025
	500	19,1±0,78	32,7±2,08	70,7±5,72	47,7±4,84	0,001	0,001	0,004	0,008
	250	66,1±6,54	43,6±8,26	79,5±0,34	62,5±0,42	0,241	0,001	0,070	0,058
	100	86,6±3,23	60,7±2,34	86,0±0,44	75,9±0,75	0,001	0,001	0,757	<0,001
	50	97,5±1,64	91,3±0,72	88,3±0,56	84,5±1,26	0,001	0,001	0,001	0,001
	0	100,0±1,51	100,0±0,42	100,0±0,39	100,0±2,66	1	0,731	1	1

Independent T test (Bootstrap) - Paired T Test (Bootstrap) All data were shown mean±standard deviation.

effects on cytotoxic activities and cell growth should be studied in detail.

Also different studies should be planned using the experimental animal to test the cardiovascular effect of this plant. We believe that this study, which we have tried to reveal the bioactive pharmacological properties of Gundelia tournefortii plant, will contribute to the current literature knowledge. We believe that the selection and design of model molecules can be used for future studies.

Acknowledgement
This study was supported by Sivas Cumhuriyet University Scientific Research Projects Commission (Project code: YMYO-007). In our study, we see the support of the microorganism strains we use in the study we would like to thank to Lecturer Mehmet Ataş for their support. We would like to thank Birgül Canikligil and Dr. Mustafa Sevindik for the helps of providing and diagnosing the plant samples.

References

1. Çoban ÖE, Patır B. Use of some spices and herbs antioxidant affected in foods. Electronic Journal of Food Technologies 2010; 5(2): 7-19.
2. Önde S, Vurdu H. Bitki çeşitliliği ve unutulan gen kaynakları. Tabiat ve İnsan 1988; 22(2): 27-31.
3. Yücel E, Güney F, Şengün İY. The wild plants consumed as a food in Mihalıççık district (Eskişehir/Turkey) and consumption forms of these plants. Biological Diversity and Conservation 2010; 3(3): 158-175.
4. Yapıcı Ü, Hoşgören H, Saya Ö. Ethnobotanic features of Kurtalan (Siirt) province. Dicle University Journal of Ziya Gökalp Faculty of Education 2009; 12: 191-196.
5. Kırbağ S, Zengin F. Antimicrobial activies of some medical plants in Elazığ. Journal of Agricultural Science 2006; 16(2): 77-80.
6. Demir H. Chemical composition of some wild (Polygonum cgnatum, Tragopoxgom reticulatus and Berberis vulgaris) plants collected from Erzurum. Bahçe 2006; 35(1-2): 55-60.
7. Kaya İ, İncekara N, Nemli Y. Ingredients of some weeds consumed as foof in aegean region. Journal of Agricultural Science 2004; 14(1): 1-6.
8. Yücel E, Tülükoğlu A. Gediz (Kütahya) çevresinde halk ilacı olarak kullanılan bitkiler. Ekoloji (Çevre Dergisi) 2000; 9(36): 12-14.
9. Ertuğ F. An ethnobotanical study in central Anatolia (Turkey).

Economic Botany 2000; 54(2): 155-182.
10. Kunkel G. Plants for Human Consumption. 1th ed. Königstein: Koeltz Scientific Books; 1984.
11. Sezik E, Yeşilada E, Honda G, Takaishi Y, Takeda Y, Tanaka T. Traditional medicine in Turkey. Folk medicine in central Anatolia. Journal of Ethnopharmacology 2001; 75(2): 95-115.
12. Coruh N, Celep A S, Özgökçe F, İşcan M. Antioxidant capacities of Gundelia tournefortii L. extracts and inhibition on glutathione-S-transferase activity. Food Chemistry 2007; 100(3): 1249-1253.
13. Tawaha K, Alali F Q, Gharaibeh M, Mohammad M, El-Elimat T. Antioxidant activity and total phenolic content of selected Jordanian plant species. Food Chemistry 2007; 104(4): 1372-1378.
14. Jamshidzadeh A, Fereidooni F, Salehi Z, Niknahad H. Hepatoprotective activity of Gundelia tourenfortii. Journal of Ethnopharmacology 2005; 101(1): 233-237.
15. Haghi G, Hatami A, Arshi R. Distribution of caffeic acid derivatives in Gundelia tournefortii L. Food Chemistry 2011; 124(3): 1029-1035.
16. Wagner H, Nickl H, Aynehchi Y. Molluscicidal saponins from Gundelia tournefortii. Phytochemistry 1984; 23(11): 2505-2508.
17. Sekeroglu N, Senol F S, Orhan I E, Gulpinar A R, Kartal M, Sener B. In vitro prospective effects of various traditional herbal coffees consumed in Anatolia linked to neurodegeneration. Food Research International 2012; 45(1): 197-203.
18. Davis, P.H. Flora of Turkey. Edinburg at the University Press. 1975; 5: 325.
19. Eloff JN. A sensitive and quick microplate method to determine the minimal inhibitory concentration of plant extracts for bacteria. Planta Med 1998; 64: 711–713.
20. Franklin R, Matthew A, Alder J, Micheal N, George M, Ferraro MJ et al. Clinical and Laboratory Standards Institute, Methods for Dilution Antimicrobial Susceptibility Tests for Bacteria that Grow Aerobically, Approved Standard (9th ed.), CLSI document M07-A9. Clinical and Laboratory Standards Institute, 950 West Valley Road, Suite 2500, Wayne, Pennsylvania 19087, USA, 2012.
21. Pfaller MA, Chaturvedi V, Espinel-Ingroff A, Ghannoum MA, Gosey LL, Odds FC et al. The National Committee for Clinical Laboratory Standards. Reference Method for Broth Dilution Antifungal Susceptibility Testing of Yeasts, Approved Standard, (2nd ed)., NCCLS document M27- A2. CLSI, 940 West Valley Road, Suite 1400, Wayne, Pennsylvania 19087- 1898, USA, 2002.
22. Kuete V. Potential of Cameroonian plants and derived products against microbial infections: a review. Planta Med.2010; 76: 1479–1491.
23. Awouafack MD, McGaw LJ, Gottfried S, Mbouangouere R, Tane P, Spiteller M. and Eloff JN. Antimicrobial activity and cytotoxicity of the ethanol extract, fractions and eight compounds isolated from Eriosema robustum (Faba-ceae). BMC Complementary and Alternative Medicine 2013; 13: 289.
24. Konak M., Ateş M., Şahan Y. Evaluation of Antioxidant Properties of Gundelia tournefortii: A Wild Edible Plant. Journal of Agricultural Faculty of Uludag University, 2017; 31, 2, 101-108
25. Karaaslan Ö., Çötelli, E., Karataş, F., 2014. Investıgatıon of amounts of a, e, c vıtamıns wıth malondıaldehyde and glutathıone ın plant Gundelıa tournefortii. EÜFBED - Fen Bilimleri Enstitüsü Dergisi 2014; 7-2, 159-168
26. Abu-Lafi, S., Rayan B., Kadan, S., Abu-Lafi, M. Anticancer activity and phytochemical composition of wild Gundelia tournefortii. Oncology letters 2019; 17: 713-717.