

**TOTAL PHENOLIC CONTENT, ANTIOXIDANT, ANTIBACTERIAL AND
ANTIFUNGAL ACTIVITIES, FT-IR ANALYSES OF *Brassica
oleracea* L. var. *acephala* AND *Ornithogalum umbellatum* L.**

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Brassica oleracea var. *acephala* and *Ornithogalum umbellatum* are two plant species which belong Brassicaceae and Liliaceae, respectively. Both of them are used as food and medicinally. *O. umbellatum* is used as anticancer, diuretic anti anti emetic. *B. oleracea* var. *acephala* is used for gastritis, gastric and duodenal ulcers and preventing some cancer forms such as stomach cancer, breast cancer and lung cancer. The aim of this research was to search antioxidant, antibacterial and antifungal activities and FT-IR analyses of ethanol extracts of stem and flower parts of *B. oleracea* var. *acephala* and *O. umbellatum*. Extracts were obtained by Soxhlet extraction. Ethanol extracts of *B. oleracea* var. *acephala* showed higher antibacterial activity than ethanol extracts of *O. umbellatum*. The most effective antifungal extract was ethanol extract of flower part of *O. umbellatum* against the fungi *Candida. tropicalis* and *Candida. parapsilosis*. Antioxidant activity of the extracts were also studied. The highest total phenolic and flavonoid contents were found in flower part of *B. oleracea* var. *acephala* as 50.7 ± 0.007 $\mu\text{g GAE/mL}$ and stem part of *O. umbellatum* as 81.37 ± 0.006 $\mu\text{g CE/mL}$, respectively. BHT which used as standard antioxidant had higher total antioxidant activity than tested ethanol extracts. DPPH activity of the extracts increased in the following order: Flower part of *B. oleracea* var. *acephala* > Stem part of *O. umbellatum* > Flower part of *O. umbellatum* > Stem part of *B. oleracea* var. *acephala*. These results which obtained from this study demonstrate that *B. oleracea* var. *acephala* and *O. umbellatum* species might be utilized as an alternative to synthetic antioxidants and antimicrobials.

Keywords: Plant extract, antimicrobial activity, antioxidant activity.

INTRODUCTION

Natural products which is obtained from plants both pure compounds and extracts, ensure many opportunities for new drugs. Because of their different chemical structures, interest has gained in worldwide for producing therapeutic drugs from plants (SASIDHARAN *et al.*, 2011). The phytochemicals which exist in plants have been found to serve as antioxidants through

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scavenging free radicals and many possess features for the remedy of illnesses caused by oxidative stress (SHAHADI *et al.*, 1992). Antioxidants are substances that remarkably retard or inhibit oxidation of an oxidizable substrate when exist in even low concentrations (HALLIWELL and GUTTERIDGE, 1999). Antioxidants are capable of detoxify ROS and hinder their damage by preventative, repair, physical defenses mechanisms (YADAV *et al.*, 2018). It was seen from the previous studies which carried out by many researchers that plants were recorded to possess stronger antioxidant capacity (YADAV *et al.*, 2018; DARKWAH *et al.*, 2018; KASKA *et al.*, 2018; CESUR *et al.*, 2017).

Because of using excessive antibiotics in clinical, agriculture and veterinary applications, many microbial strains gained antibiotic resistance. This situation led to the investigate effective antimicrobial drugs which obtained from medicinal plants. Plant phytochemicals has variety of chemical structures which overcome the antibiotic resistance. Plants have important secondary metabolites that generally utilized in the pharmaceutical industry. For many centuries, plants have been utilized to heal infectious illnesses and regarded as considerable source of new antimicrobial drugs (BEREKSI *et al.*, 2018). Many studies have been performed to screen the antimicrobial capacities of medicinal plants (BEKTAŞ *et al.*, 2018; KAYA *et al.*, 2018; MOSTAFA *et al.*, 2018).

Liliaceae family is mostly distributed in temperate regions of the Northern Hemisphere. Many Liliaceae are significant decorative plants, widely grown for their charming flowers and involved in a major floriculture of cut flowers and dry bulbs. Many species of Liliaceae are popular cultivated plants in private and public areas (TRIPATHI *et al.*, 2016). *Ornithogalum umbellatum* L. belongs to the Liliaceae family. It possesses some medicinal properties such as healing acne and some kinds of cancer, using diuretic and anti emetic (GÜL and DINLER, 2016).

Brassicaceae is one of ten economically important plant family in the world. It is one of the largest Angiosperm family represented by 380 genera and 3700 species distributed worldwide. Most members of Brassicaceae family are located in the northern temperate zone. *Brassica oleracea* L. var. *acephala* is a member of Brassicaceae family. It contributes neurological protection. Moreover, it helps to decrease many cancer forms and heart diseases (ÇÖMLEKÇİOĞLU and KUTLU, 2018).

In this study, it was aimed to investigate antioxidant, antibacterial and antifungal activities, total phenolic and flavonoid contents and FT-IR analysis of ethanol extracts of stem and flower parts of *Brassica oleracea* var. *acephala* and *Ornithogalum umbellatum*.

MATERIALS AND METHODS

Plant samples

Ornithogalum umbellatum and *Brassica oleracea* var. *acephala* have collected from local market in Giresun, Turkey in 2018 and they were identified by Associate Professor Zafer TÜRKMEN.

Microorganisms

While seven bacteria were used in antibacterial studies; four fungi were used in antifungal studies. Table 1 summarizes bacteria and fungi strains which used in the study.

Table 1. Bacteria and fungi which used in the study.

Microorganism	Bacteria/Fungi	Where obtained from		
<i>Salmonella enterica</i> ATCC 14028	Bacteria	Giresun	Province	Control
		Laboratory		
<i>Proteus vulgaris</i> FMC 1	Bacteria	Firat University		
<i>Enterobacter aerogenes</i> CCM 2531	Bacteria	Firat University		
<i>Gordonia rubripertincta</i> (lab isolate)	Bacteria	Yeditepe University		
<i>Bacillus subtilis</i> IMG 22	Bacteria	Firat University		
<i>Klebsiella pneumoniae</i> (laboratory isolate)	Bacteria	Yeditepe University		
<i>Enterococcus faecalis</i> ATCC 29212	Bacteria	Rize University		
<i>Candida albicans</i> FMC 17	Fungi	Firat University		
<i>Candida tropicalis</i> ATCC 13803	Fungi	Firat University		
<i>Saccharomyces cerevisiae</i> ATCC 9763	Fungi	Giresun	Province	Control
		Laboratory		
<i>Candida parapsilosis</i> ATCC 22019	Fungi	Giresun University		

Preparation of extracts

Flower and stem parts of *O. umbellatum* and *B. oleracea* var. *acephala* was separated, dried on laboratory and then they were grounded into powder, separately. Thirty gram flower and stem parts of *O. umbellatum* and *B. oleracea* var. *acephala* were extracted with 300 mL ethanol with a Soxhlet extractor, separately (KUMAR *et al.*, 2012).

Antibacterial and antifungal activities

The antibacterial and antifungal potentials of the ethanol extracts of *O. umbellatum* and *B. oleracea* var. *acephala* were identified through utilizing Minimum Inhibition Concentration (MIC) assay. Ethanol extracts were dissolved in dimethyl sulfoxide (DMSO) at 30 mg/mL concentration. Dissolved extracts were sterilized through 0.45 µm pore sized filter. The 96 well plates were prepared by dispensing into each well 95 µL of Müeller Hinton Broth and 5 µL of the inoculum. 100 µL ethanol extracts of initially prepared at the concentration of 1 mg/mL was added into the first wells. Then, 100 µL from their serial dilutions were added into consecutive wells. This 96 well plate was incubated at 37°C for bacteria overnight and 35°C for 48 h for fungi. The MIC was expressed as the lowest concentration of the test compounds to inhibit the growth of microorganisms (YIĞIT *et al.*, 2009).

Total phenolic content

Total phenolic content of the extracts was determined by the procedure of SLINKARD and SINGLETON (1977) using gallic acid standard. 0.1 mL extract and 4.5 mL distilled water were mixed. Then, 0.1 mL Folin–Ciocalteu reagent (previously diluted 3 fold with distilled water) was put into the mixture. After 3 minutes, 0.3 mL Na₂CO₃ (2%) was added. The absorbance was measured at 760 nm, after incubating the mixture for 90 min (SLINKARD and SINGLETON, 1977). The quantity of the total phenolic compounds was denoted as µg gallic acid equivalent (GAE)/mL. The tests were carried out three times.

*Antioxidant activity**Total flavonoid content*

Total flavonoid content of the extracts was determined by the method of ZHISHEN *et al.* (1999). 0.25 mL extract was added to 1.25 mL distilled water followed by 75 μL NaNO_2 (5%) and incubated for 5 min. Afterwards, 150 μL $\text{AlCl}_3 \cdot 6\text{H}_2\text{O}$ (10%) was added to the mixture and then incubated for 5 min, the reaction mixture was treated with 0.5 mL NaOH (1M) and 275 μL distilled water. Absorbance was read spectrometrically at 510 nm (ZHISHEN *et al.*, 1999). The amount of total flavonoid compounds was calculated as μg catechin equivalent (CE)/mL. The tests were carried out three times.

Total antioxidant capacity

Total antioxidant capacity of the extracts were defined by the method of PRIETO *et al.*, (1999). Absorbance was measured at 695 nm (PRIETO *et al.*, 1999). The results were calculated as μg ascorbic acid equivalent (AAE)/mL plant sample from ascorbic acid standard graphical equation. The tests were carried out three times.

1,1-diphenyl-2-picryl-hydrazyl (DPPH) radical scavenging activity

DPPH radical scavenging activity of the extracts was established by DPPH. Appropriate dilution series (50-200 $\mu\text{g}/\text{mL}$) were prepared for ethanolic extracts in DMSO. 0.75 mL of each solution was added to 1.5 mL of a 6×10^{-5} M methanolic solution of DPPH. The mixture was stirred vigorously and allowed to stand in the dark at the room temperature for 30 min. Decrease in absorbance of the solution against methanol was measured at 517 nm with a Shimadzu 1240 UV-Vis spectrophotometer (BLOIS, 1958). Rutin and Butylated hydroxytoluene (BHT) used as standard antioxidants. The DPPH radical scavenging activity was calculated using the following equation:

$$\left(\frac{\text{ABS of the control} - \text{ABS of the sample}}{\text{ABS of the control}} \right) \times 100$$

Copper reducing antioxidant capacity (CUPRAC)

CUPRAC test was employed by the method of ÖZYÜREK *et al.* (2009). 0.5 mL extract (prepared in 50-200 $\mu\text{g}/\text{mL}$), 1.0 mL CuCl_2 solution (1×10^{-2}), 1.0 mL neocuproine solution (7.5×10^{-3} M) and 1.0 mL ammonium acetate buffer (1.0 M, pH: 7.0) were mixed in a test tube. Then, the tube was vortexed and stored in a dark place for 30 min. absorbance was read at 450 nm (ÖZYÜREK *et al.*, 2009). BHT was used as standard antioxidant agent.

FT-IR analyses

Infra red (IR) analysis was done with the aid of infra red spectrophotometer (JASCO OFTIR 6600 with ATR attachment) at the Central Research Laboratory in Giresun University. A drop of purified extract was placed carefully on crystal surface. It was fixed on the infra red beam. The infra red data was compared to the table of IR frequencies.

RESULTS AND DISCUSSION

Antibacterial and antifungal activities

Table 2 represents MIC values of the tested extracts. In this survey, all of the tested extracts exhibited varying degree of antibacterial and antifungal action against test bacteria and fungi. The extracts were more effective against fungi than bacteria. MIC values of the extracts range from 0.625 mg/mL to 5 mg/mL against bacteria and range from 0.0078 mg/mL to 5 mg/mL against fungi.

Table 2. MIC values of ethanol extracts of stem and flower parts of *B. oleracea* var. *acephala* and *O.umbellatum* (mg/mL).

Microorganisms	Stem part of <i>B. oleracea</i> var. <i>acephala</i>	Flower part of <i>B. oleracea</i> var. <i>acephala</i>	Stem part of <i>O. umbellatum</i>	Flower part of <i>O. umbellatum</i>
<i>B. subtilis</i>	5	0.625	2.5	2.5
<i>K. pneumoniae</i>	5	2.5	10	2.5
<i>G. rubripertincta</i>	5	2.5	2.5	5
<i>E. faecalis</i>	5	2.5	2.5	1.25
<i>P. vulgaris</i>	5	2.5	2.5	2.5
<i>E. aerogenes</i>	5	2.5	2.5	2.5
<i>S. enterica</i>	5	2.5	5	5
<i>C.albicans</i>	1.25	0.625	5	1.25
<i>C. tropicalis</i>	0.625	0.625	0.15625	0.0078
<i>S. cerevisiae</i>	1.25	0.625	1.25	1.25
<i>C. parapsilosis</i>	1.25	0.625	0.15625	0.0078

The antibacterial and antifungal activity of *O. umbellatum* and *B. oleracea* var. *acephala* was confirmed with other studies in literatures. For example, ERTÜRK *et al.* (2018) searched ethanol extract of leaves of *B. oleracea* var. *acephala* and it was found activity against *Pseudomonas aeruginosa*, *Proteus vulgaris*, *Escherichia coli*, *Listeria monocytogenes*, *Candida albicans*, *Clostridium perfringens*, *Salmonella enterica*, *Bacillus subtilis*, *Micrococcus luteus*, *Staphylococcus aureus* and *Aspergillus niger* (ERTÜRK *et al.*, 2018). In agreement with this study, we found activity in ethanol extracts of stem and flower parts of *B. oleracea* var. *acephala* against *P. vulgaris*, *S. enterica*, *B. subtilis* and *C. albicans*. Antibiotic properties of glucosinolates of *B. oleracea* var. *acephala* was also studied by CARBONE *et al.* (2016). AYAZ *et al.* (2016) found antimicrobial activity against *S. aureus*, *E. faecalis*, *B. subtilis* and *M. catarrhalis* in nine phenolic acids found in the leaves of *B. oleracea* var. *acephala* (AYAZ *et al.*, 2008). Similarly, we found activity in the ethanol extracts of stem and flower parts of *B. oleracea* var. *acephala* against *E. faecalis* and *B. subtilis*. DEMIRKOL *et al.* (2017) screened *O. umbellatum* species collected from different locations from Ordu. It was concluded from this study, methanol/water extracts of *O. umbellatum* had activity against *S. aureus* and *E. coli* but no activity against *Listeria monocytogenes* (DEMIRKOL *et al.*, 2017). YIĞIT *et al.* (2013) found no activity in chloroform and methanol extracts of the leaves of *O. umbellatum* against *P.*

aeruginosa, *E. coli*, *E. aerogenes*, *S. aureus* (YIĞIT *et al.*, 2003). On the other hand, we found activity in ethanol extracts of stem and flower parts of *O. umbellatum* against *E. aerogenes*.

Different results could be explained with collected plant materials from different locations, using different solvents, using different parts of the same plant and using different concentrations.

Total phenolic content

Phenolic compounds possess redox characteristics, which provide them to role as antioxidants. Since their free radical scavenging property is attributed by their hydroxyl groups, the total phenolic concentration might be utilized for quick screening of antioxidant capacity (BABAA and MALIKB, 2015).

Total phenolic contents of the tested extracts were summarized in Table 3. The total phenolic contents of the ethanolic stem part of *B. oleracea* var. *acephala* extract, the ethanolic flower part of *B. oleracea* var. *acephala*, ethanolic stem part of *O. umbellatum* var. *acephala* extract and the ethanolic flower part of extract *O. umbellatum* var. *acephala* were detected as 21.63 ± 0.001 $\mu\text{g GAE/mL}$, 50.7 ± 0.007 $\mu\text{g GAE/mL}$, 19.26 ± 0.001 $\mu\text{g GAE/mL}$ and 18.96 ± 0.001 $\mu\text{g GAE/mL}$, respectively. *O. umbellatum* var. *acephala* extracts had lower phenolic content than *B. oleracea* var. *acephala* extracts.

Table 3. Total phenolic content of the ethanol extracts of *B. oleracea* var. *acephala* and *O. umbellatum*.

Samples	Total Phenolic Content ($\mu\text{g GAE/mL}$)
Stem part of <i>B. oleracea</i> var. <i>acephala</i>	21.63 ± 0.001
Flower part of <i>B. oleracea</i> var. <i>acephala</i>	50.7 ± 0.007
Stem part of <i>O. umbellatum</i>	19.26 ± 0.001
Flower part of <i>O. umbellatum</i>	18.96 ± 0.001

FERREIRA *et al.* (2015) determined that total phenolic content of methanol extract of *B. oleracea* var. *acephala* as 0.606 mg GAE/mL (FERREIRA *et al.*, 2015). FRATIANNI *et al.* (2014) found total phenolic content of ethanol extracts of *B. oleracea* var. *acephala* which was collected from Italy as 6.37 mM GAE/ml (FRATIANNI *et al.*, 2014). On the other hand, different results were obtained in our study. Using different extraction methods and collecting plant from different locations might be caused different results. ÇÖMLEKÇİOĞLU and KUTLU (2018) searched seasonal variation of total phenolic content of *B. oleracea* var. *acephala* and it was detected that total phenolic content of *B. oleracea* var. *acephala* were ranged from 7.32 ± 0.025 mg GAE/g and 11.63 ± 0.057 mg GAE/g between November and February (ÇÖMLEKÇİOĞLU and KUTLU, 2018). Apaydın and Yolcu (2017) studied total phenolic contents of methanol extracts of fresh and cooked *O. umbellatum* which was collected from Giresun and it was found that total phenolic contents of fresh and cooked *O. umbellatum* as 5.821 ± 0.008 mg/g dry plant and 5.056 ± 0.004 mg/g dry plant , respectively. Total phenolic content was decreased when the heat process was applied (APAYDIN and YOLCU, 2017). In our study, we found total phenolic content of ethanol extracts of stem and flower parts of fresh *O. umbellatum* as 19.26 ± 0.001 $\mu\text{g GAE/mL}$ and 18.96 ± 0.001 $\mu\text{g GAE/mL}$, respectively. The difference between our study and Apaydın's study might be arised from using different plant parts, using different solvents and using different

methods. ADAWIA *et al.* (2016) found total phenolic contents of methanol extracts of bulbs and aerial parts of *O. umbellatum* as 5.50 mg GAE/g and 23.04 mg GAE/g, respectively (ADAWIA *et al.*, 2016).

Antioxidant activity

Total flavonoid content

Flavonoids are secondary plant metabolites which have important chelating and antioxidant properties. Antioxidant property of flavonoids depends on the structure and substitution pattern of hydroxyl groups (STANKOVIC, 2011).

Total flavonoid content of studied extracts was determined using spectrophotometric method with aluminum chloride. The content of flavonoids was expressed in Table 4. While total flavonoid content of *B. oleracea* var. *acephala* extracts ranged from 30.59±0.002 to 56.18±0.014 µg CE/mL, total flavonoid content of *O. umbellatum* extracts ranged from 23.55±0.002 to 81.37±0.006 µg CE/mL.

Table 4. Total flavonoid content and total antioxidant capacity of the ethanol extracts of *B. oleracea* var. *acephala* and *O. umbellatum*

Samples	Total Flavonoid Content (µg CE/mL)	Total Antioxidant Capacity (µg AAE/mL)
Stem part of <i>B. oleracea</i> var. <i>acephala</i>	30.59±0.002	80.35±0.011
Flower part of <i>B. oleracea</i> var. <i>acephala</i>	56.18±0.014	146.56±0.010
Stem part of <i>O. umbellatum</i>	81.37±0.006	105.50±0.025
Flower part of <i>O. umbellatum</i>	23.55±0.002	63.19±0.007

ÇÖMLEKÇİOĞLU and KUTLU (2018) also searched seasonal variation of total flavonoid content of *B. oleracea* var. *acephala* and it was detected that total flavonoid content of *B. oleracea* var. *acephala* were ranged from 2.01±0.07 mg CE/g and 3.96±0.09 µg CE/g between November and February (ÇÖMLEKÇİOĞLU and KUTLU, 2018)

APAYDIN and YOLCU (2017) studied total flavonoid contents of methanol extracts of fresh and cooked *O. umbellatum* which was collected from Giresun and it was found that total flavonoid contents of fresh and cooked *O. umbellatum* as 3.258±0.028 mg/g dry plant and 2.546±0.052 mg/g dry plant, respectively (APAYDIN and YOLCU, 2017). In contrast to Apaydin's study, we found total flavonoid content of ethanol extracts of stem and flower parts of fresh *O. umbellatum* as 81.37±0.006 µg CE/mL and 23.55±0.002 µg CE/mL, respectively. The difference could be arised from using different solvents and using different methods. ADAWIA *et al.* (2016) found total flavonoid contents of methanol extracts of bulbs and aerial parts of *O. umbellatum* as 1.82 mg rutin equivalent/g and 52.30 mg rutin equivalent/g, respectively (ADAWIA *et al.*, 2016).

Total antioxidant capacity

Total antioxidant capacity test a quantitative method, is based on the reduction of Phosphate-Mo (VI) to Phosphate Mo (V) by the sample and subsequent formation of a bluish green colored phosphate/Mo (V) complex at acid pH (ARCEAMPONG *et al.*, 2018).

The result of total antioxidant activity (TAC) is shown in Table 4. The highest total antioxidant capacity was found in ethanol extract of flower part of *B. oleracea* var. *acephala* and the lowest total antioxidant capacity was found in ethanol extract of flower part of *O. umbellatum*.

Table 5. CUPRAC and DPPH radical scavenging activities of the ethanol extracts of *B. oleracea* var. *acephala* and *O. umbellatum*

Samples	Concentration ($\mu\text{g/mL}$)	CUPRAC activity (nm)	DPPH scavenging activity (% inhibition)
Stem part of <i>B. oleracea</i> var. <i>acephala</i>	50	0.066 \pm 0.051	NA
	100	0.077 \pm 0.058	2.84 \pm 0.004
	150	0.108 \pm 0.001	4.85 \pm 0.012
	200	0.121 \pm 0.004	13.25 \pm 0.008
Flower part of <i>B. oleracea</i> var. <i>acephala</i>	50	0.117 \pm 0.018	18.77 \pm 0.004
	100	0.135 \pm 0.002	33.78 \pm 0.010
	150	0.157 \pm 0.001	64.14 \pm 0.008
	200	0.170 \pm 0.003	80.80 \pm 0.023
Stem part of <i>O. umbellatum</i>	50	0.063 \pm 0.003	5.08 \pm 0.004
	100	0.169 \pm 0.009	9.24 \pm 0.002
	150	0.275 \pm 0.067	16.75 \pm 0.007
	200	0.311 \pm 0.006	24.92 \pm 0.012
Flower part of <i>O. umbellatum</i>	50	0.120 \pm 0.020	1.48 \pm 0.013
	100	0.136 \pm 0.002	5.62 \pm 0.006
	150	0.224 \pm 0.018	9.99 \pm 0.008
	200	0.259 \pm 0.007	15.31 \pm 0.002
BHT	50	0.526 \pm 0.022	80.22 \pm 0.014
	100	0.554 \pm 0.040	84.49 \pm 0.013
	150	0.714 \pm 0.010	88.09 \pm 0.002
	200	0.862 \pm 0.015	94.06 \pm 0.002
Rutin	50		58.31 \pm 0.025
	100		61.08 \pm 0.003
	150	NT	64.28 \pm 0.001
	200		75.57 \pm 0.013

NT: Not Tested

Reagent of CUPRAC is CuCl_2 which was combined with neocuproine in ammonium acetate buffer pH 7. Cu (II) will be reduced to Cu (I). Complex Cu (I) – neocuproine gives yellow color and show characteristic absorption at wavelength 450 nm²⁰. Intensity of yellow color depends on amount of Cu (II) that is reduced to Cu (I). If a sample reduces Cu (II) to Cu (I), at the same time it will be oxidized, so that sample can act as antioxidant (FIDRIANNY *et al.*, 2016).

CUPRAC activity of the extracts were presented in Table 5. CUPRAC activity of the extracts increased with the increasing concentration. *O. umbellatum* extracts exhibited higher

activity than *Brassica oleracea* var. *acephala* extracts. BHT which used as standard antioxidant had higher activity than tested extracts.

DPPH radical scavenging activity of the extracts were summarized in Table 5. DPPH activity of the extracts increased with the increasing concentration. DPPH activity of the extracts increased in the following order: Flower part of *B. oleracea* var. *acephala*> Stem part of *O. umbellatum*> Flower part of *O. umbellatum*> Stem part of *B. oleracea* var. *acephala*. Standard antioxidants exhibited higher activity than extracts except for flower part of *B. oleracea* var. *acephala*.

FERREIRA *et al.* (2015) found DPPH scavenging activity of methanol extract of *B. oleracea* var. *acephala* as 63.20% (FERREIRA *et al.*, 2015). In an another study, it was reported that the methanolic extract of *B. oleracea* var. *acephala* showed inhibition percentages higher than 70% (MELO *et al.*, 2006). In our study, we found DPPH scavenging activities of ethanol extracts of stem and flower parts of *B. oleracea* var. *acephala* as 13.25% and 80.80%, respectively. The different results between our study and the studies above might be arised from collecting plant samples from different countries and using different solvents. FRATIANNI *et al.* (2014) found EC values of DPPH activity of *B. oleracea* var. *acephala* which was collected from Italy as 1.53 ± 0.009 mg (FRATIANNI *et al.*, 2014). Çömlekçioğlu and Kutlu (2018) searched seasonal variaiton of antioxidant activity of *B. oleracea* var. *acephala* and it was detected IC₅₀ values of DPPH activity of *B. oleracea* var. *acephala* ranged from 1.91 ± 0.002 mg/ml and 1.41 ± 0.002 mg/ml between November and February (ÇÖMLEKÇIOĞLU and KUTLU, 2018). Ayas *et al.* (2017) investigated DPPH activity of methanol extract of *O. umbellatum* and it was concluded that IC₅₀ value of DPPH activity was 7.1 mg/g. (AYAS *et al.*, 2017). ADAWIA *et al.* (2016) found IC₅₀ value of DPPH activity of methanol extracts of bulbs and aerial parts of *O. umbellatum* as 0.96 mg/ml and 0.29 mg/ml, respectively (ADAWIA *et al.*, 2016).

FT-IR Analyses

From the result in Figure 1, the ethanolic flower extracts of *O. umbellatum* has absorption bands, the wave number (cm⁻¹) of the peaks obtained were summarized in Table 6. The IR spectrum of different extracts reveals structural information about major and minor constituents. The peak at 3282.25 cm⁻¹ assigned to the O-H stretching. In addition, the peak at 1608.34 cm⁻¹ assigned to the C=C stretching means that some aromatic compounds existed in the extract. The peaks at 1245.79 cm⁻¹ belong to O-H bending and peak at 2919.7 cm⁻¹ assigned to aliphatic C-H bond. The peak at 2352.73 cm⁻¹ was assigned to C≡N stretching or C≡C stretching. Presence of C≡C, C≡N, C=C, O-H and C-O bonding structures are responsible for the formation of alkyne, nitrile, alkene and phenol.

From the result in Figure 2, the ethanolic stem extracts of *O. umbellatum* contained absorption bands as were describe in Table 7. The band at 2919.7 cm⁻¹ and 2854.13 cm⁻¹ assigned to aliphatic C-H stretching. Meanwhile, the peak at 1608.34 cm⁻¹ and 1511.92 cm⁻¹ aromatic compounds. The presence of peak at 3305.39 cm⁻¹ belongs to an phenolic compound. Another characteristics peak is at 1052.94 cm⁻¹ was assigned to C-O stretching.

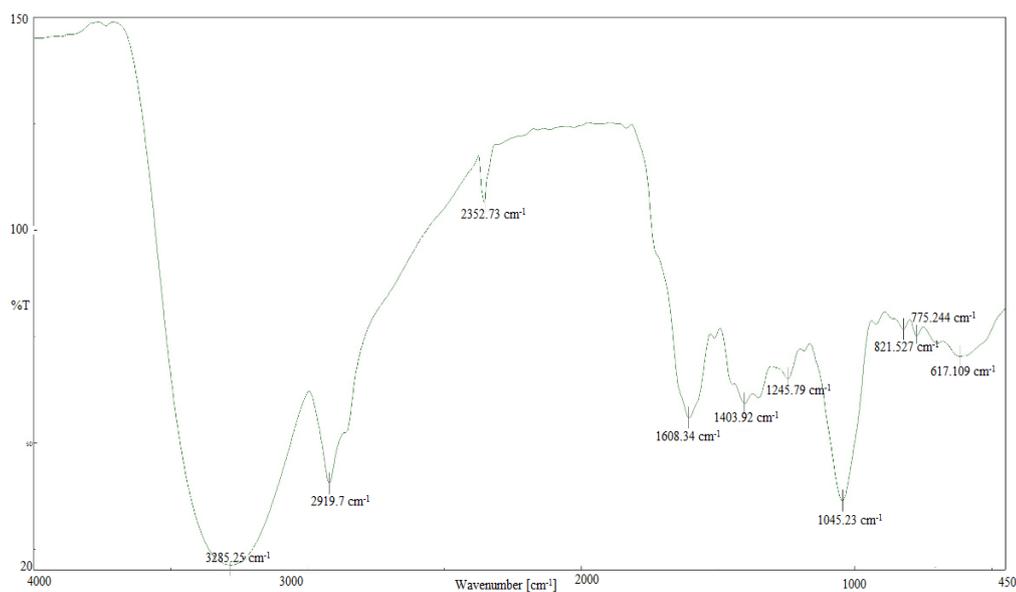


Figure 1. FT-IR analysis of ethanolic extract of flower part of *O. umbellatum*

Table 6. Results of FT-IR analysis of ethanol extract of flower part of *O. umbellatum*

Wave numbers (cm ⁻¹)	Bond
3282.25	O-H stretching
2919.7	Aliphatic C-H bond
2352.73	C≡N stretching or C≡C stretching
1608.34	Aromatic C=C stretching
1403.92	C-H in-plane bending
1245.79	O-H bending
1045.23	C-O stretching

Figure 3 and Table 8 shows ethanolic flower extract of *B. oleracea* var. *acephala*. The peak at 3367.1 cm⁻¹ revealed the presence the alcohols, phenols (O–H stretch). The peak at 2919.7 cm⁻¹ and 2854.13 cm⁻¹ refers to the presence of aliphatic compounds (C–H bond). The peak at 1731.76 cm⁻¹ corresponds the carboxylic acid group (C=O stretch). A peak at 3066.26 cm⁻¹ denotes aromatic compounds (C-H bond). The peak of 2360.44 cm⁻¹ indicate the alkyne or nitrile (C≡N bond or C≡C bond).

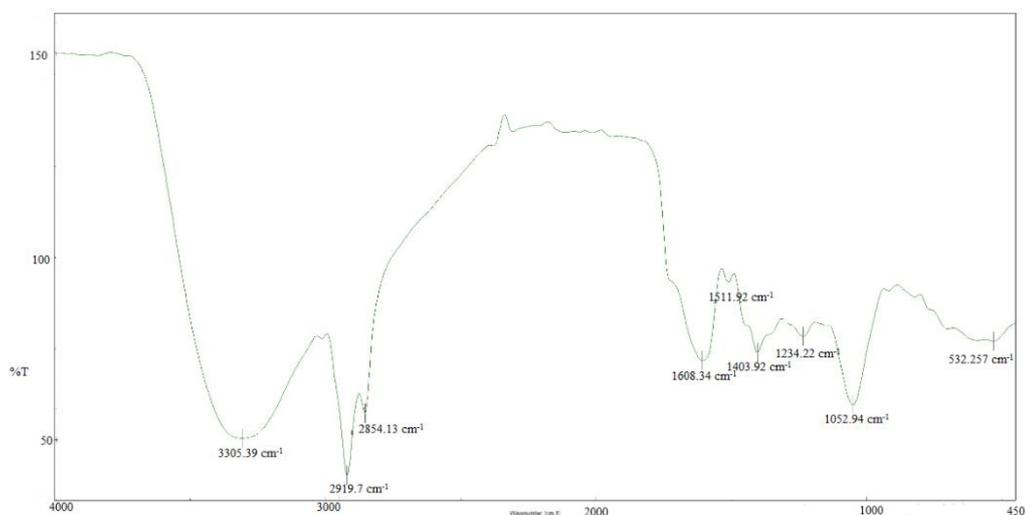


Figure 2. FT-IR analysis of ethanolic extract of stem part of *O. umbellatum*

Table 7. Results of FT-IR analysis of ethanol extract of stem part of *O. umbellatum*

Wave numbers (cm ⁻¹)	Bond
3305.39	O-H stretching
2919.7	Aliphatic C-H stretching
2854.13	Aliphatic C-H stretching
1608.34	Aromatic C=C stretching
1511.92	Aromatic C=C bond
1403.92	C-H in-plane bending
1234.22	O-H bending
1052.94	C-O stretching

Figure 4 and Table 9 shows ethanolic stem extract of *B. oleracea* var. *acephala*. The peak at 3332.99 cm⁻¹ revealed the presence the alcohols, phenols (O-H stretch). The peak at 2915.84 cm⁻¹ and 2850.27 cm⁻¹ refers to the presence of aliphatics (C-H stretch). The peak at 1662.34 cm⁻¹ corresponds the carboxylic acid group (C=O stretch). A peak at 1056.8 cm⁻¹ denotes C-O stretching.

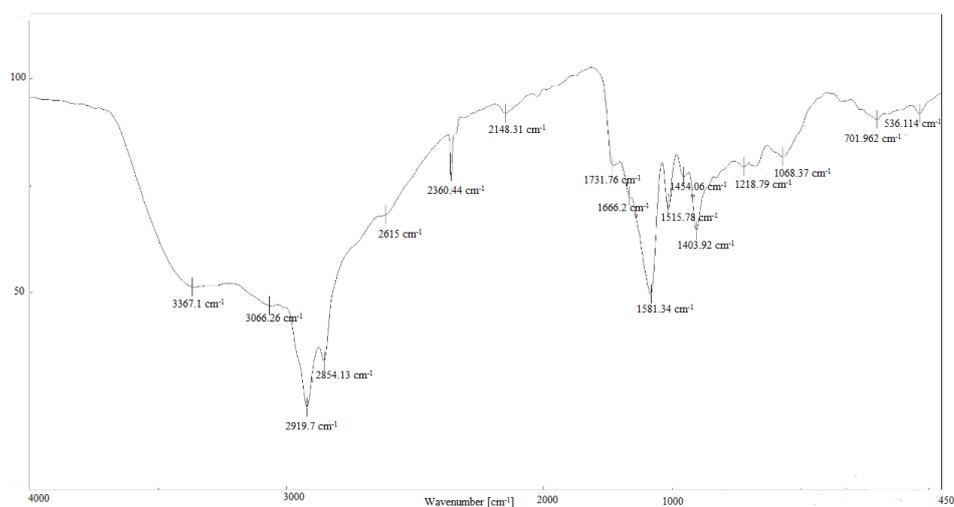


Figure 3. FT-IR analyses of ethanolic extract of flower part of *B. oleracea var. acephala*

Table 8. Results of FT-IR analysis of ethanol extract of flower part of *B. oleracea var. acephala*

Wave numbers (cm ⁻¹)	Bond
3367.1	OH stretching
3066.26	Aromatic C-H bond
2919.7	Alifatic C-H bond
2854.13	Alifatic C-H bond
2360.44	C≡N bond or C≡C bond
1731.76	C=O bond
1581.34	Aromatic C=C bond
1454.06	CH ₃ bending
1403.92	O-H bending
1218.79	O-H bending
1068.37	C-O stretching

Table 9. Results of FT-IR analysis of ethanol extract of stem part of *B. oleracea var. acephala*

Wave numbers (cm ⁻¹)	Bond
3332.39	O-H bond or N-H bond
2915.84	Alifatic C-H stretching
2850.27	Alifatic C-H stretching
1662.34	C=O stretching
1442.49	Aliphatic CH ₃ bending
1403.92	O-H bending
1056.8	C-O stretching

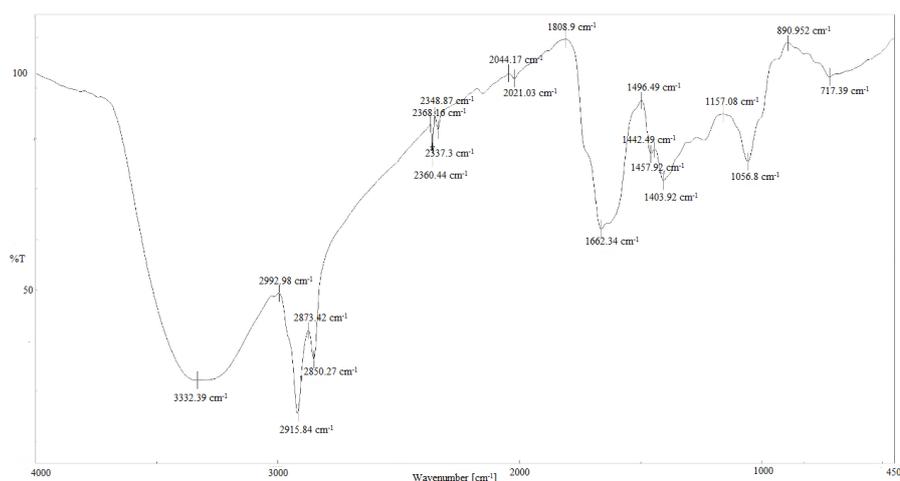


Figure 4. FT-IR analyses of ethanolic extract of stem part of *B. oleracea* var. *acephala*

CONCLUSION

Phenolic and flavonoid contents, antioxidant, antibacterial and antifungal activities, FT-IR analysis of ethanol extracts of stem and flower parts of *Brassica oleracea* var. *acephala* and *Ornithogalum umbellatum* were assessed. The ethanol extracts of *O. umbellatum* and *B. oleracea* var. *acephala* contain compounds which possess antioxidant, antibacterial and antifungal activities. Therefore, these plants might be an alternative to synthetic antimicrobial and antioxidant agents. Further study, however, needs to be carried out to determine and characterize the active compounds responsible for these biological activities.

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**UKUPNI SADRŽAJ FENOLA, ANTIOKSIDATIVNA, ANTIBAKTERIJSKA I
ANTIFUNGALNA AKTIVNOST, FTIR ANALIZA *Brassica oleracea* L. var. *acephala*
I *Ornithogalum umbellatum* L.**

Sinem AYDIN

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Izvod

Brassica oleracea var. *acephala* i *Ornithogalum umbellatum* su dve biljne vrste koje pripadaju familijama Brassicaceae i Liliaceae. Obe se koriste kao hrana i kao lekovi. *O. umbellatum* koristi se antikancerogeno, diuretski antiemetički. *B. oleracea* var. *acephala* koristi se za gastritis, čir na želucu i dvanaestopalačnom crevu i sprečava neke oblike raka poput raka želuca, raka dojke i pluća. Cilj ovog istraživanja bio je pretraživanje antioksidativnih, antibakterijskih i antifungalnih aktivnosti i FT-IR analiza etanolnih ekstrakata delova stabljike i cvetova *B. oleracea* var. *acephala* i *O. umbellatum*. Ekstrakti su dobijeni Sokhlet ekstrakcijom. Etanolni ekstrakti *B. oleracea* var. *acephala* su pokazali veću antibakterijsku aktivnost u odnosu na etanolne ekstrakte *O. umbellatum*. Najefikasniji antifungalni ekstrakt bio je etanolni ekstrakt cvetnog dela *O. umbellatum* protiv gljiva *Candida tropicalis* i *Candida parapsilosis*. Takođe je proučavana antioksidativna aktivnost ekstrakata.

Najveći ukupni fenolni i flavonoidni sadržaji pronađeni su u delu cveća *B. oleracea* var. *acephala* od $50,7 \pm 0,007 \mu\text{g GAE} / \text{mL}$, odnosno delova stabljike *O. umbellatum* od $81,37 \pm 0,006 \mu\text{g CE/mL}$. BHT koji se koristio kao standardni antioksidans imao je veću ukupnu antioksidativnu aktivnost u odnosu na testirane etanolne ekstrakte. DPPH aktivnost ekstrakata se povećala sledećim redosledom: cvetni delovi *B. oleracea* var. *acephala* > delovi stabljike *O. umbellatum* > cvetni delovi *O. umbellatum* > delovi stabljike *B. oleracea* var. *acephala*. Dobijeni rezultati pokazuju da se vrste *B. oleracea* var. *acephala* i *O. umbellatum* mogu koristiti kao alternativa sintetičkim antioksidansima i antimikrobnim sredstvima.

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