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The Antibacterial Effects of the Different Extracts of *Oenothera biennis* and *Origanum minutiflorum* O. Schwarz et. P. H. Davis on Food-borne Pathogenic Bacteria

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ABSTRACT

This study aimed to determine the antibacterial effects of *Oenothera biennis* and *Origanum minutiflorum* O. Schwarz et. P. H. the leaves extrats on food-borne pathogenic bacteria. The highest antibacterial effect on bacterial strains, the lowest Minimum Inhibitory Concentration (MIC) and the lowest Bactericidal Concentration (MBC) values of both plants was determined in extracts obtained from the leaves using diethyl ether ($P < 0.05$). Diethyl ether extracts of both plants showed the highest antibacterial activity on *Listeria monocytogenes* (37.23 and 23.98-mm-zone diameters, respectively) ($P < 0.05$). The lowest MIC and MBC effect of the diethyl extract of *Oenothera biennis* on bacterial strains was determined to be 0.011mg/L and 7.81 mg/mL, respectively, on *Bacillus cereus*. However, the highest values were determined to be 0.750 mg/ L and >500 mg/mL, respectively, in acetone extract detected on *Pseudomonas aeroginosa*. The lowest MIC value of the diethyl extract of *Origanum minutiflorum* O. Schwarz et. P. H. Davis was determined to be 0.029 mg/L on *Listeria monocytogenes* and *Bacillus cereus* whereas the lowest MBC was determined to be 7.81 mg/mL on *Bacillus cereus*.

1. Introduction

Oenothera biennis is a biennial plant from the *Convolvulaceae* family. It is commonly known as evening primrose. The plant grows well in sandy, loamy and clayey soils in almost every place such as fields, roadsides and meadows (Morrison and Reekie, 1995). The oil obtained from the seeds of the plant was determined to have various pharmacological properties (Arimura, 2003a). The seed oil of the plant is very rich in γ-linolenic acid content, an important fatty acid found in the composition of prostaglandins and related hormones that are effective in regulating muscle and vascular contractions in human physiology. (Becker, 1983; Shukla et al., 1999). Also, recent studies have shown that ethanol extract obtained from the seeds of the plant shows specific anti-tumor activity (Arimura et al., 2003b; Arimura et al., 2004).

The *Origanum* genus belonging to the *Lamiaceae* family has 24 species in Turkey. Of these species, 16 are endemic (Aslım and Yücel, 2008; Albayrak and Aksoy, 2017). They are widely grown in the Mediterranean region (Azizi et al., 2009; Oke and Aslim, 2010). The species has high-level biological properties and reported to possess antimicrobial, antifungal, antioxidant, antimutagenic, anticarcinogenic, antifungal, antinematodal, antiparasitic and antiemetic activities (Bostancioglu et al., 2012; Chishti et al., 2013; Karaboduk et al., 2014; Sarikurkcu et al., 2015). *Origanum minutiflorum* O. Schwarz et. P. H. Davis (Turkish oregano) is an endemic species that grows in the Sütçüler region of Isparta in Turkey (Baydar, 2005). It is widely used as a spice and herbal tea (Ozen et al., 2014).

This study aimed to determine the antibacterial effects of the extracts obtained from the leaves of *Oenothera biennis* and *Origanum minutiflorum* O. Schwarz et. P. H. on foodborne pathogenic bacteria.

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2. Materials and Methods

2.1. Materials

Oenothera biennis used in the study was obtained from the villages in Antalya while *Origanum minutiflorum* O. Schwarz et. P. H. Davis was obtained from the Sütçüler region in Isparta, Turkey.

2.2. Bacterial Strains Used in This Study

In the study; *Staphylococcus aureus* (ATCC 6538), *Yersinia enterocolitica* (ATCC 9610), *Salmonella Typhimurium* (ATCC 14028), *Listeria monocytogenes* (ATCC 51774), *Escherichia coli* (ATCC 25922) *Enterococcus feacalis* (ATCC 29212), *Enterobacter aerogenes* (ATCC 13048) ve *Shigella flexneri* (ATCC 12022) *Bacillus cereus* (ATCC 14579) *Pseudomonas aeroginosa* (ATCC 15442), *Escherichia coli* (ATCC 25922) species of bacteria were used.

2.3. Preparation of Plant Extracts

Leaves of the plants were cut into small pieces and mixed with 400 mL ethanol (85%: Merck, 100983, Germany), methanol (Merck, 106009 Germany), diethyl ether (Merck, 100921, Germany), acetone (Merck, 100014, Germany) or chloroform (Merck, 102445, Germany) at 1:3 (w/v) ratio. The mixtures were then shaken at 22 °C in a shaker (Wiseshake SHO-2D, Witeg, Germany) at 120 rpm for 24 hours. After the extracts were filtered through sterilized filter paper (Whatman No. 32), the solvents were removed from the extract by rotary evaporator (Heidolph, Germany). The extracts were stored in colored glass bottles (100 ml, glass bottle, Turkey) at 4°C in refrigerator (Arçelik 554271, Turkey).

2.4. Preparation of Discs Containing Plant Extracts

For the preparation of discs containing the plant extracts, 10 µL samples of the extracts of *Oenothera biennis* and *Origanum minutiflorum* O. Schwarz et. P. H. Davis were taken into Petri dishes (Sterile, 90 x 15, Fıratmed, Turkey) using sterile tipped pipettes (Research Plus, Eppendorf) and dropped on 6-mm-diameter empty antibiogram discs (Bio-Disk 316010001). The Petri dishes were kept closed at 4 °C in refrigerator (Arçelik 554271, Turkey) for 60 minutes for the discs to absorb the extracts. The extract-impregnated discs were then dried in a laminar flow cabinet (Cryste, Puricube 1200) at room temperature for 8-10 hours.

2.5. Preparation of the Inocula

The young (24-hour) bacterial strains produced on non-selective media were taken from single growing colonies using a sterile loop and suspended in physiological saline (Merck, 115525, Germany) until homogeneous turbidity occurred. The density of the inoculum suspension was adjusted to 0.5 McFarland standard using a densitometer (Biosen, 1B, Turkey). The inocula were taken using a transport swap (Fıratmed, Turkey) and inoculated on the surface of Mueller Hinton Agar (1.05437, Merck, Germany) (MHA) and spread homogeneously (Bauer et al., 1966; Akarca, 2019).

After waiting for 10 minutes for the medium to absorb the inocula, the antibiogram discs containing plant extracts were placed in the Petri dishes and incubated in an incubator (Incucell, MMM, Germany) as described by Anonymous (2018) and Cruz-Gálvez et al. (2018). The zones formed at the end of the period were measured in mm using a digital caliper (Mitutoyo, 500-181-30, Japan) under sufficient daylight.

2.5. Minimum Inhibitory Concentration (MIC) and Minimum Bactericidal Concentration (MBC) Values

From the extracts of both plants obtained with five different solvents, 1 mL of Nutrient Broth (Merck, 1.05443, Germany) was added to the first tubes. Then, 1 mL of the mixtures formed in the first tubes was taken and transferred to the next tubes. This procedure was applied to all tubes in series. As a result, the mixtures were obtained in equal amounts in each tube but only half of the concentration of the previous tube. Also, positive and negative control tubes were formed.

Of the bacterial strains used in the study, 1 µl (10^6 cfu/mL) were inoculated into all other tubes except for the negative control tube and incubated at the appropriate temperature, time and conditions. At the end of the period, turbidity in the tubes, membrane formation on the surface and sediment at the bottom were regarded as positive. Also, no growth was determined in the negative control tube whereas growth was determined in the positive control tube. The MIC value was determined by taking half of the sum of the concentrations of the first tube evaluated as positive growth and the tubes previously evaluated as negative growth (By Aamer et al., 2015; Chikezie, 2017; Akarca, 2019).

The first tube, which was evaluated as negative growth in MIC analysis, was inoculated into Muller Hinton Agar (Merck, 195437, Germany) by taking 1 µl from all the tubes at the following concentrations and then incubated at the appropriate temperature, time and conditions for each bacterial species. The value of the first concentration with no growth at the end of the period was evaluated as MBC (Dhiman et al., 2011; By Aamer et al., 2015; Akarca, 2019).

2.6. Statistical Analysis

The results of the study were determined by SPSS (V 23.0.0) statistical software and the differences were determined by the Duncan test ($P < 0.05$).

3. Results and Discussion

The antibacterial effect (mm-zone-diameter) of the extracts of *Oenothera biennis* from five different solvents on ten foodborne pathogenic bacteria is shown in Table 1.

It was determined that the diethyl ether extract of *Oenothera biennis* showed the highest antibacterial

effect on *Listeria monocytogenes* with a 37.23-mm-zone-diameter, followed by *Bacillus cereus* with a 32.46-mm-zone diameter ($P < 0.05$). In contrast, the acetone extract exhibited the lowest antibacterial effect ($P < 0.05$). *Pseudomonas aeroginosa* was the bacteria species on which this extract showed the lowest antibacterial effect with a 7.33-mm-zone-diameter ($P < 0.05$).

It was determined that the highest antibacterial effect was determined in the *Origanum minutiflorum* O. Schwarz et. P. H. extract obtained using diethyl ether, followed by extracts obtained using ethanol and chloroform (Table 2; $P < 0.05$).

Table 1
Antibacterial Effects of Different Extracts of *Oenothera biennis* (mm Zone Diameter)

Species of Bacteria	Solvent				
	Ethanol	Methanol	Diethyl ether	Acetone	Chloroform
Staphylococcus aureus	16.02 ± 1.39^{Bde}	15.08 ± 0.65^{Bbc}	20.55 ± 0.93^{Ad}	9.93 ± 0.25^{Cbc}	14.28 ± 1.02^{Bcd}
Yersinia enterecolitica	18.00 ± 1.57^{Ad}	10.29 ± 0.96^{Bef}	18.18 ± 0.54^{Aef}	12.29 ± 1.14^{Bab}	13.79 ± 1.66^{ABcd}
Salmonella Typhimurium	21.85 ± 1.24^{Ac}	14.61 ± 1.12^{Bbc}	23.18 ± 0.34^{Ac}	13.80 ± 2.67^{Ba}	15.87 ± 0.51^{Bbc}
Listeria monocytogenes	35.81 ± 0.62^{Aa}	16.76 ± 1.39^{Bb}	37.23 ± 0.90^{Aa}	15.15 ± 1.03^{Ba}	17.30 ± 0.88^{Bb}
Enterococcus feacalis	17.86 ± 0.38^{Ad}	12.01 ± 0.14^{Bde}	18.43 ± 0.66^{Aef}	9.35 ± 0.67^{Cbc}	10.22 ± 0.30^{Cf}
Enterobacter aerogenes	14.77 ± 0.40^{Ade}	10.02 ± 0.51^{Bef}	16.25 ± 0.11^{Af}	9.16 ± 0.44^{Bbc}	10.71 ± 0.53^{Bf}
Shigella flexneri	16.96 ± 1.28^{ABde}	13.77 ± 0.58^{Bcd}	17.24 ± 0.91^{Aef}	10.08 ± 0.07^{Cbc}	13.94 ± 1.07^{ABcd}
Bacillus cereus	30.65 ± 1.70^{Ab}	19.28 ± 0.80^{Ba}	32.46 ± 0.71^{Ab}	15.28 ± 1.11^{Ca}	20.36 ± 0.78^{Ba}
Pseudomonas aeroginosa	13.36 ± 0.82^{Be}	8.86 ± 0.22^{Cf}	19.23 ± 1.12^{Ade}	7.33 ± 0.21^{Cc}	9.04 ± 0.23^{Cf}
Escherichia coli	17.05 ± 1.34^{ABde}	9.32 ± 0.16^{Cf}	20.12 ± 1.37^{Ad}	15.19 ± 0.96^{Ba}	11.27 ± 1.10^{Cde}

a-f (↓): Values with the same capital letters in the same column for each analysis differ significantly ($P < 0.05$).
A-C (→): Values with the same capital letters in the same rows for each analysis differ significantly ($P < 0.05$).

Table 2
Antibacterial Effects of Different Extracts of *Origanum minutiflorum* O. Schwarz et. P. H. (mm Zone Diameter)

Species of Bacteria	Solvent				
	Ethanol	Methanol	Diethyl Ether	Acetone	Chloroform
Staphylococcus aureus	13.88 ± 0.64^{Bb}	10.99 ± 0.25^{Cb}	16.30 ± 0.83^{Ac}	10.72 ± 0.36^{Cc}	11.33 ± 0.21^{Cbc}
Yersinia enterecolitica	11.09 ± 0.23^{B}	10.01 ± 0.36^{Bb}	12.84 ± 0.32^{Aef}	9.96 ± 0.09^{Bc}d	10.63 ± 0.50^{Bcd}
Salmonella Typhimurium	12.84 ± 0.39^{ABbcd}	10.65 ± 0.97^{BCb}	13.35 ± 0.86^{Ad}	10.25 ± 0.47^{Ccd}	12.07 ± 0.19^{ABCb}
Listeria monocytogenes	20.86 ± 0.32^{Ba}	15.55 ± 0.93^{Ca}	23.98 ± 0.27^{Aa}	13.14 ± 0.52^{Db}	16.77 ± 0.46^{Ca}
Enterococcus feacalis	10.61 ± 0.75^{Ae}	9.45 ± 0.71^{ABb}	10.90 ± 0.53^{Af}	8.13 ± 0.44^{Bf}	10.13 ± 0.21^{ABcd}
Enterobacter aerogenes	11.00 ± 0.33^{Ade}	9.77 ± 0.50^{Ab}	11.56 ± 1.16^{Aef}	9.33 ± 0.30^{Ade}	10.45 ± 0.12^{Acd}
Shigella flexneri	10.97 ± 0.35^{Bde}	9.92 ± 0.37^{Bb}	13.23 ± 0.41^{Aef}	9.84 ± 0.29^{Bcd}	9.78 ± 0.33^{Bd}
Bacillus cereus	19.42 ± 1.04^{ABa}	16.10 ± 0.73^{Ca}	21.56 ± 1.07^{Ab}	14.42 ± 0.24^{Ca}	17.02 ± 0.64^{BCa}
Pseudomonas aeroginosa	13.52 ± 0.82^{Abc}	9.13 ± 0.30^{BCb}	13.80 ± 0.38^{Ad}	8.49 ± 0.04^{Cf}	10.28 ± 0.37^{Bcd}
Escherichia coli	11.67 ± 0.83^{Acde}	11.04 ± 0.35^{ABb}	11.86 ± 0.28^{Aef}	9.16 ± 0.63^{Bde}	10.33 ± 0.60^{ABcd}

a-f (↓): Values with the same capital letters in the same column for each analysis differ significantly ($P < 0.05$).
A-D (→): Values with the same capital letters in the same rows for each analysis differ significantly ($P < 0.05$).

Table 3
Antibacterial Effects of Different Extracts of *Oenothera biennis* and *Origanum minutiflorum* O. Schwarz et. P.

Species of Bacteria	Solvent									
	Ethanol		Methanol		Diethyl Ether		Acetone		Chloroform	
	Ob	*Om*	*Ob*	*Om*	*Ob*	*Om*	*Ob*	*Om*	*Ob*	*Om*
Staphylococcus aureus	++	++	++	+	+++	++	+	+	++	+
Yersinia enterecolitica	++	+	+	+	+++	++	++	+	++	+
Salmonella Typhimurium	+++	++	++	+	+++	++	++	+	++	++
Listeria monocytogenes	+++	+++	++	++	+++	+++	++	++	++	++
Enterococcus feacalis	++	+	++	+	+++	+	+	-	+	+
Enterobacter aerogenes	++	+	+	+	++	+	+	+	+	+
Shigella flexneri	++	+	++	+	+	++	+	+	++	+
Bacillus cereus	+++	+++	+++	++	+++	+++	++	+	+++	++
Pseudomonas aeroginosa	++	++	-	+	+++	++	-	-	+	+
Escherichia coli	++	+	+	+	+++	+	++	+	+	+

7-9 mm zone diameter: -, 9-12 mm zone diameter: +,12-18 mm zone diameter :++, >18 mm zone diameter:+++, *Ob*: *Oenothera biennis*, *Om: Origanum minutiflorum* O. Schwarz et. P. H. Davis

Table 4
MIC (mg/L) and MBC (mg/mL) Values of Different Extracts of *Oenothera biennis*.

Species of Bacteria	Solvent									
	Ethanol		Methanol		Diethyl Ether		Acetone		Chloroform	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
Staphylococcus aureus	0.070±0.023^{Bab}	11.72±3.91^{Aa}	0.093±0.000^{Bab}	15.63±0.00^{Ac}	0.035±0.012^{Ba}	11.72±3.91^{Ab}	0.562±0.188^{Aab}	156.25±93.75^{Ab}	0.139±0.045^{Bab}	39.07±23.44^{Acd}
Yersinia enterecolitica	0.046±0.000^{Bb}	15.63±0.00^{Ba}	0.281±0.094^{Aab}	62.50±0.00^{Ac}	0.070±0.024^{ABa}	11.72±9.91^{Bb}	0.281±0.094^{Abc}	39.07±23.44^{ABb}	0.093±0.000^{ABab}	23.44±7.81^{ABcd}
Salmonella Typhimurium	0.035±0.012^{Aab}	11.72±3.91^{Aa}	0.234±0.141^{Aab}	23.44±7.81^{Ac}	0.023±0.000^{Aa}	19.53±11.72^{Ab}	0.187±0.000^{Ac}	31.25±0.00^{Ab}	0.281±0.094^{Aab}	23.44±7.81^{Acd}
Listeria monocytogenes	0.017±0.06^{Ab}	15.63±0.00^{Aa}	0.070±0.024^{Ab}	19.53±11.72^{Ac}	0.017±0.06^{Aa}	7.81±0.00^{Ab}	0.117±0.071^{Ac}	23.44±7.81^{Ab}	0.035±0.012^{Ab}	15.63±0.00^{Ad}
Enterococcus feacalis	0.035±0.012^{Bab}	11.72±3.91^{Aa}	0.140±0.047^{Bab}	62.50±0.00^{Ac}	0.029±0.018^{Ba}	39.07±23.44^{Ab}	0.562±0.188^{Aab}	312.50±187.5^{Aa}	0.140±0.047^{Bab}	187.50±62.5^{Ab}
Enterobacter aerogenes	0.070±0.024^{Bab}	15.63±0.00^{Aa}	0.281±0.094^{ABab}	93.75±32.25^{Abc}	0.070±0.024^{Ba}	62.50±0.00^{Aab}	0.374±0.000^{Abc}	312.50±187.5^{Aa}	0.105±0.082^{Bab}	156.25±93.75^{Abc}
Shigella flexneri	0.046±0.000^{Ab}	93.75±31.25^{Aa}	0.234±0.141^{Aab}	125.00±0.00^{Abc}	0.035±0.012^{Aa}	46.88±15.63^{Aab}	0.140±0.047^{Ac}	187.50±62.5^{A}b	0.070±0.024^{Ab}	78.13±46.87^{Abcd}
Bacillus cereus	0.017±0.006^{Ab}	11.72±0.00^{Aa}	0.035±0.012^{Ab}	20.03±12.22^{Ac}	0.011±0.000^{Aa}	15.63±0.00^{Ab}	0.093±0.000^{Ac}	15.63±0.00^{Ab}	0.138±0.092^{Aab}	11.72±3.91^{Ad}
Pseudomonas aeroginosa	0.140±0.047^{Ba}	132.82±117.18^{Ba}	0.374±0.000^{ABa}	375.00±125.00^{ABa}	0.035±0.012^{Ba}	93.75±31.25^{Ba}	0.750±0.000^{Aa}	>500.00	0.469±0.282^{ABa}	500.00±0.00^{Aa}
Escherichia coli	0.117±0.071^{Aab}	93.75±31.25^{Aa}	0.281±0.094^{Aab}	312.50±187.50^{Aab}	0.058±0.035^{Aa}	39.07±23.44^{Ab}	0.058±0.035^{Ac}	46.88±15.62^{Ab}	0.105±0.082^{Aab}	125.00±0.00^{Abcd}

a-d (↓): Values with the same capital letters in the same column for each analysis differ significantly (P < 0.05).
A-C (→): Values with the same capital letters in the same rows for each analysis differ significantly (P < 0.05).

Table 5
MIC (mg/L) and MBC (mg/mL) Values of Different Extracts *Origanum minutiflorum* O. Schwarz et. P. H.

Species of Bacteria	Solvent									
	Ethanol		Methanol		Diethyl Ether		Acetone		Chloroform	
	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC	MIC	MBC
Staphylococcus aureus	0.140±0.047^{ABa}	93.75±31.25^{Aab}	0.187±0.000^{Aba}	187.50±62.50^{Aab}	0.093±0.000^{Bab}	78.13±46.87^{Aa}	0.281±0.094^{Abc}	140.63±109.38^{Aab}	0.187±0.000^{Aba}	62.50±0.00^{Abc}
Yersinia enterecolitica	0.187±0.00^{Aa}	78.13±46.87^{ABab}	0.234±0.140^{Aa}	125.00±0.00A^{Bab}	0.140±0.047^{Aab}	46.88±15.63^{Ba}	0.374±0.000^{Abc}	187.50±62.50^{Aab}	0.281±0.094^{Aa}	125.00±0.00^{ABbc}
Salmonella Typhimurium	0.117±0.071^{Aa}	93.75±31.25^{Aab}	0.281±0.094^{Aa}	156.25±93.75^{Aab}	0.093±0.000^{Aab}	78.13±46.87^{Aa}	0.187±0.000^{Abc}	125.00±0.00^{Aab}	0.140±0.047^{Aa}	93.75±31.25^{Abc}
Listeria monocytogenes	0.046±0.00^{ABa}	19.53±11.72^{Ab}	0.070±0.024^{Aba}	46.88±15.63^{Ab}	0.029±0.018^{Bb}	11.72±3.91^{Aa}	0.093±0.000^{Ac}	39.07±23.44^{Ab}	0.070±0.018^{Aba}	31.25±0.00^{Ac}
Enterococcus feacalis	0.234±0.141^{Aa}	125.00±0.00^{Aab}	0.422±0.328^{Aa}	312.50±187.5^{Aa}	0.140±0.047^{Aab}	93.75±31.25^{Aa}	0.469±0.282^{Aab}	>500.00	0.187±0.000^{Aa}	187.50±62.5^{Abc}
Enterobacter aerogenes	0.140±0.047^{Aa}	187.50±62.5^{ABa}	0.187±0.000^{Aa}	250.00±0.00^{ABab}	0.234±0.147^{Aa}	125.00±0.00^{Ba}	0.140±0.047^{Abc}	375.00±125.0^{Aab}	0.281±0.094^{Aa}	250.00±0.00^{ABab}
Shigella flexneri	0.187±0.00^{Aa}	156.25±93.75^{Bab}	0.281±0.094^{Aa}	125.00±0.00^{Bab}	0.117±0.071^{Aab}	156.25±93.75^{Ba}	0.234±0.141^{Abc}	500.00±0.00^{Aab}	0.187±0.000^{Aa}	375.00±125.0^{ABa}
Bacillus cereus	0.078±0.32^{Aa}	39.07±23.44^{Aab}	0.117±0.071^{Aa}	46.88±15.63^{Ab}	0.029±0.018^{Ab}	7.81±0.00^{Aa}	0.058±0.035^{Ac}	78.13±46.88^{Ab}	0.117±.0.071^{Aa}	19.53±11.72^{Ac}
Pseudomonas aeroginosa	0.070±0.024^{Ba}	93.75±31.25^{Bab}	0.234±0.141^{Ba}	250.00±0.00^{Bab}	0.070±0.024^{Bab}	156.25±93.75^{Ba}	0.750±0.000^{Aa}	500.00±0.00^{Aa}	0.281±0.094^{Ba}	156.25±93.75^{Bbc}
Escherichia coli	0.140±0.047^{Aa}	62.50±0.00^{Aab}	0.187±0.000^{Aa}	93.75±31.25^{Aab}	0.140±0.047^{Aab}	125.00±0.00^{Aa}	0.187±0.000^{Abc}	312.50±187.5^{Aab}	0.140±0.047^{Aa}	93.75±31.25^{Abc}

a-d (↓): Values with the same capital letters in the same column for each analysis differ significantly (P < 0.05).
A-C (→): Values with the same capital letters in the same rows for each analysis differ significantly (P < 0.05)

The highest antibacterial effect was observed in the diethyl ether extract on *Listeria monocytogenes* with a zone diameter of 23.98 mm whereas the lowest antibacterial effect was on *Enterococcus faecalis* with a zone diameter of 8.13 mm in the acetone extract ($P < 0.05$).

Among the five different extracts, the highest antibacterial effect was determined in the extracts prepared with diethyl ether (Table 3), followed by ethanol and chloroform extracts ($P < 0.05$). Among the extracts, the lowest antibacterial effect on ten different pathogenic bacterial strains was determined in the acetone extract ($P < 0.05$). As a result of similar studies on the subject, it has been stated that the high antibacterial effects of extracts were caused by carvacrol and thymol, which are abundant in the structure of plants (Aslım and Yucel, 2008; Bostancioglu et al., 2012).

Of the five different extracts obtained from *Oenothera biennis*, the lowest MIC and MBC values were determined in diethyl ether, ethanol and methanol extracts, respectively, whereas the highest MIC and MBC values were determined in the acetone extract (Table 4; $P < 0.05$). The lowest MIC values of the extracts were on *Bacillus cereus* and *Listeria monocytogenes*, respectively, whereas the highest values were on *Pseudomonas aeroginosa*, *Yersinia enterecolitica* and *Enterobacter aerogenes*, respectively ($P < 0.05$).

The lowest MIC and MBC values of the plant extracts on ten different pathogenic bacteria strains were 0.011 mg/L and 7.81 mg/mL in the diethyl ether extract against *Bacillus cereus*, whereas the highest values were 0.750 mg/L and >500 mg/mL in the acetone extract against *Pseudomonas aeroginosa* (Table 4).

It was determined that the lowest MIC and MBC values, as in *Oenothera biennis*, were determined in the *Origanum minutiflorum* O. Schwarz et. P. H. extract obtained from the diethyl ether extract whereas the highest MIC and MBC values were determined in the acetone extract (Table 5; $P < 0.05$). The lowest MIC value of the diethyl extract was 0.029 mg/L on *Listeria monocytogenes* and *Bacillus cereus*, while the lowest MBC value of the diethyl extract was 7.81 mg/mL on *Bacillus cereus* ($P < 0.05$). The acetone extract of the *Origanum minutiflorum* O. Schwarz et. P. H. Davis had the highest MIC and MBC values against ten different food-borne pathogenic bacteria among the extracts obtained with five different solvents ($P < 0.05$). The highest MIC and MBC values of this extract were found to be 0.750 mg/L and >500 mg/mL on *Pseudomonas aeroginosa* ($P < 0.05$).

According to variance analysis results, the antibacterial effect of extracts obtained from two different plants using five different solvents on ten different foodborne pathogens, it was found that plant species, bacterial species, solvent species, solvent species x bacteria species interaction, solvent species x plant species interaction, bacterial species x plant species interaction and solvent species x bacterial species x plant species interaction were found to be significant ($P < 0.0001$; Table 6).

Similarly, bacterial species, solvent type, solvent type x bacterial species interactions had a significant effect on the MIC value ($P < 0.0001$). In terms of the effect on the MBC value, it was determined that plant species, bacterial species, solvent type interactions were significant at $P < 0.0001$, while solvent type x bacterial interaction was significant at $P < 0.05$ and bacterial species x plant species interaction was significant at $P < 0.01$ (Table 6).

Albayrak and Aksoy (2017), similar to our study, have stated that the antibacterial effect of *Origanum minutiflorum* was high. As a result of the research, the highest antibacterial effect of ethanol extract has been reported to be on *Aeromonas hydrophil*ic with a 24-mm-zone diameter and *Streptococcus pneumonia* with an 18-mm-zone diameter, respectively. The researchers have reported that the lowest MIC and MBC values of ethanol extract of the plant were 0.78 mg/mL and 0.78 mg/mL, respectively, on *Klebsiella pneumonia*.

Table 6
Analysis Results of Variance Analysis of Oenothera biennis and Origanum minutiflorum O. Schwarz et. P. H. Davis on Solvent, Antibacterial Effect, MIC and MBC Values (P value)

Factors	Antibacterial Effect	MIC	MBC
Plant species	<0.0001	0.057	<0.0001
Bacteria species	<0.0001	<0.0001	<0.0001
Solvent type	<0.0001	<0.0001	<0.0001
Solvent type x Bacteria species	<0.0001	<0.0001	0.05
Solvent type x Plant species	<0.0001	0.207	0.204
Bacteria species x Plant species	<0.0001	0.360	0.01
Solvent type x Bacteria species x Plant species	<0.0001	0.809	0.136

$P < 0.05$: Statistically significant, $P < 0.01$: Statistically very significant, $P < 0.0001$: Statistically too much significant, $P > 0.05$: Not statistically significant.

4. Conclusion

According to the results of this study, both plant species, especially *Oenothera biennis*, were found to have high antibacterial effects. The fact that the highest activity was found in diethyl ether extract among different solvents used showed that the components found in the composition of the leaves of the plants which exhibit antibacterial effect were best decomposed in this solvent.

In recent years, the trend towards the use of natural products as an alternative to artificial food additives and pharmaceuticals has led manufacturers to research this subject. Successful results obtained from many studies show the usability of such products that are obtained from a large number of plants.

The cultivation of these two plants, which have been consumed for different purposes for many years, in larger areas and the products such as extracts, essential oils, and essences to be obtained from these plants can be used as natural preservatives, shelf-life extenders and antibacterials in food industry, and medicine industry including pharmacology, medical and veterinary.

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