



The Antibacterial Effects of Çiriş (*Asphodelus aestivus* Brot.) on Some Foodborne Pathogenic Bacteria

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Abstract

This study aimed to determine the antibacterial effects of Çiriş (*Asphodelus aestivus* Brot.) on eight important food-borne pathogenic bacteria by the disc diffusion method. The highest inhibition effect was determined on *Escherichia coli* with a 15.63 mm zone diameter, followed by, *Staphylococcus aureus* (14.78 mm), *Listeria monocytogenes* (10.72 mm), *Shigella flexneri* (10.06 mm), *Enterobacter aerogenes* (9.22 mm) and *Enterococcus faecalis* (8.15 mm) (P<0.05). On the other hand, Çiriş showed the lowest inhibition effect on *Salmonella Typhi* and *Salmonella Typhimurium*, with 7.28 and 6.42 mm zone diameters, respectively. Comparing the data obtained to the effects of the standard antibiotics on the same pathogens, it was found that *Escherichia coli* were susceptible while *Staphylococcus aureus* and *Shigella flexneri* were moderately susceptible to the ethanol extract of Çiriş. On the other hand, it was determined that *Salmonella Typhi*, *Listeria monocytogenes*, *Enterococcus faecalis*, *Salmonella Typhimurium*, and *Enterobacter aerogenes* exhibited resistance.

Anahtar Kelimeler: *Asphodelus aestivus* Brot., Pathogens, Antibacterial effect, Disc diffusion.

Çirişin (*Asphodelus aestivus* Brot.) Bazı Gıda Kaynaklı Patojen Bakteriler Üzerindeki Antibakteriyel Etkisi

Öz

Bu çalışmada çirişin (*Asphodelus aestivus* Brot.) sekiz önemli gıda kaynaklı patojen bakteri üzerindeki antibakteriyel etkisinin disk difüzyon metodu ile belirlenmesi amaçlanmıştır. Araştırma sonucunda en fazla inhibisyon etkinin; 15.63 mm zon çapı *Escherichia coli* üzerinde olduğu, bunu sırasıyla *Staphylococcus aureus* (14.78 mm), *Listeria monocytogenes* (10.72 mm), *Shigella flexneri* (10.06 mm), *Enterobacter aerogenes* (9.22 mm) ve *Enterococcus faecalis* (8.15 mm) takip ettiği belirlenmiştir (P<0.05). Buna karşın en düşük inhibisyon etkinin ise; sırasıyla 7.28 ve 6.42 mm zon çaplarıyla *Salmonella Typhi* ve *Salmonella Typhimurium* üzerinde olduğu gözlemlenmiştir. Elde edilen veriler aynı patojenler üzerinde standart antibiyotiklerin etkileri ile kıyaslandığında çirişin etanol ekstraktına karşı; *Escherichia coli*'nin duyarlı, *Staphylococcus aureus* ve *Shigella flexneri*'nin orta duyarlı olduğu belirlenmiştir. Buna karşın; *Salmonella Typhi*, *Listeria monocytogenes*, *Enterococcus faecalis*, *Salmonella Typhimurium* ve *Enterobacter aerogenes*'in ise, dirençli olduğu tespit edilmiştir.

Keywords: *Asphodelus aestivus* Brot., Patojen, Antibakteriyel etki, Disk difüzyon.

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1. Introduction

Today, foodborne diseases have reached a level that can pose a serious threat to public health. One of the most important causes of these diseases is the pathogenic bacteria that contaminated foods in various ways (Karatepe & Patir 2012). Most of the bacteria causing foodborne infections to have gained resistance to antimicrobial drugs, especially antibiotics due to reasons including misuse, unnecessary use, etc. As a result of this clinical problem, people started to prefer products with natural antimicrobial properties (Oskay et al, 2005). Natural products are increasingly accepted as they are better tolerated in the human body and, also, because of their unique advantages (Silva et al, 2017).

As is the case in all the world, plants that have important medical effects have been used for decades in the treatment of various diseases by the public in Turkey (Acibuda & Bostan Budak, 2018). Due to its location in the world, Turkey has a very rich flora and hosts about 12000 plant taxa (Akcicek & Vural, 2007).

Asphodelus aestivus Brot. is a member of the Liliaceae, *Asphodellus* L. genus. In Turkey, it is locally called “Çiriş” or “Yalancı Çiriş”. It has sword-shaped leaves with 35-45 cm length and 3 cm width. The plant yields pea-shaped fruits with 6 seeds, 7 mm in size. It grows naturally in arid, uncultured soils. Along with Turkey, it naturally grows in Africa, Arab countries and some parts of Europe (Baytop, 1999).

Çiriş is conventionally consumed fresh as well as it can be consumed as a cooked meal. Also, it is used in the traditional treatment due to its antirheumatic, wound healing, antihemorrhoidal, alopecia, eczema and abscess-curing effects (Baytop, 1999; Ugurlu et al, 2009).

This study aimed to determine the antibacterial effect of Çiriş on some important foodborne pathogenic bacteria by the disc diffusion method.

2. Material and Methods

2.1. Materials

Çiriş samples used in the study were provided by the Çakıroğlu Baharat (Afyonkarahisar/Turkey) company by collecting from the natural habitat of the plant (Aegean Coast-Aydin / TURKEY) in April-May.

2.2. Bacterial Strains Used in this Study

In the study; *Staphylococcus aureus* (ATCC 6538), *Salmonella Typhi* (ATCC 19430), *Listeria monocytogenes* (ATCC 51774), *Escherichia coli* (ATCC 25922), *Enterococcus faecalis* (ATCC 29212), *Salmonella Typhimurium* (ATCC 14028), *Enterobacter aerogenes* (ATCC 13048) and *Shigella flexneri* (ATCC 12022) species of bacteria were used.

2.3. Method

2.3.1. Preparation of Ethanol Extracts of the Plant

Çiriş samples used in the study were washed and cleaned and then dried at room temperature. The dried plants divided into small pieces were ground in a laboratory mill (D300, I Sundem, Turkey) until obtaining a powder form. Then, a 300 g sample was weighed in a precision balance (Radwag PS 510 R, Poland) and 400 mL 1:3 (w/v) 85% ethanol (Merck, 100983, Germany) was added. The prepared samples were shaken at 120 rpm for 24 hours using a shaker (Wiseshake SHO-2D, Witeg, Germany). After the extraction was filtered with a sterile filter paper (Whatman No. 32), the solvent in the filtrate was removed on a rotary evaporator (Heidolph, Germany). The extract was taken to a sterile glass vial and covered with an aluminum foil and stored in a cold environment.

2.3.2. Determination of Antibacterial Activity

Homogeneous turbidity was formed by suspending individual colonies from young cultures (24-48 h) produced on selective medium specific for each bacterial culture with a sterile loop in 9 mL sterile ringer (Merck, 115525, Germany). The density of the inoculum suspension (BIOS that, 1B, Turkey) was adjusted to 0.5 McFarland standard with densitometry. The inocula were taken using transport swap (Firatmed, Turkey) and uniformly inoculated onto the surface of freshly prepared Muller Hinton Agar (Merck 1.05437, Germany) (MHA) at 22 °C (Bauer et al, 1966; Akarca, 2019).

After letting the media impregnate the inoculate for 10 minutes, 10 ml Çiriş essential oil was impregnated. Then, blank antibiogram disks (Bio-Disk 316.010.001, Turkey) impregnated with 10 µL laurel essential oil were placed onto the surface of the medium provided that the zones that will be formed will not be in contact with each other and the Petri dishes were incubated under conditions specified by Akarca et al. (2019) (Incucell, MMM, Germany). The zones formed at the end of the period were measured with the help of a digital caliper (Mitutoyo, 500-181-30, Japan) under daylight.

2.3.3. Statistical Evaluation

SPSS 23.0.0. (SPSS Inc, USA) statistical package program was used to statistically analyze the results. The data obtained from the analyses were evaluated by variance analysis technique in a randomized block experimental design. The Duncan test was used to determine the level of difference between groups.

3. Results and Discussion

Table 1 shows the antibacterial effects of Çiriş ethanol extracts on eight different pathogenic bacteria according to the disc diffusion method.

Table 1. Antibacterial Effects of *Asphodelus aestivus* Brot. Ethanol Extract on Some Food Borne Pathogens (mm zone diameter)

Bacteria	Antibacterial Effect (mm Zone Diameter)
<i>Staphylococcus aureus</i>	14.78±1.75 ^{ab}
<i>Salmonella Typhi</i>	7.28±1.05 ^c
<i>Listeria monocytogenes</i>	10.72±1.90 ^{bc}
<i>Escherichia coli</i>	15.63±2.28 ^a
<i>Enterococcus faecalis</i>	8.15±2.79 ^c
<i>Salmonella Typhimurium</i>	6.42±1.15 ^c
<i>Shigella flexneri</i>	10.06±2.31 ^c
<i>Enterobacter aerogenes</i>	9.22±1.47 ^c

It was determined that, of the eight different food-origin pathogenic bacteria, Çiriş showed the highest inhibition effect on *Escherichia coli* with a 15.63±2.28 mm zone diameter, followed by *Staphylococcus aureus* (14.78±1.75 mm), *Listeria monocytogenes* (10.72±1.90 mm), *Shigella flexneri* (10.06±2.31 mm), *Enterobacter aerogenes* (9.22±1.47 mm) and *Enterococcus faecalis* (8.15±2.79 mm). Çiriş showed the lowest inhibition effect on *Salmonella Typhi* and *Salmonella Typhimurium*, with 7.28±1.05 ve 6.42±1.15 mm zone diameters, respectively.

Oskay et al. (2007) have reported the antimicrobial activity of Çiriş on various bacteria ranged from 2 to 15 mm zone diameter, with the highest activity being on *Staphylococcus aureus*, *Pseudomonas fluorescens*, and *Escherichia coli*. Fatal et al. (2016) have reported that the highest antibacterial effect of oil extracted from *Asphodelus aestivus* Brot. seeds were 9.3 mm and on *Klebsiella pneumonia*, whereas the lowest effect was on *Escherichia coli* with a 6.7 mm zone diameter.

Various studies have reported that *Asphodelus* species contained anthraquinone, naphthalene, polysaccharide (Li et al, 2000; Zhang et al, 2000), sesquiterpene, lactone, flavonoids, arylcoumarin and glycoside compounds (Peksel et al, 2012; Bayrak 2013). The antibacterial effect of Çiriş was associated with these components.

Comparing the zone diameters of the Çiriş ethanol extract on eight different foodborne pathogens to the zone diameters of standard antibiotics determined by Clinical and Laboratory Standards Institute (CLSI) (2015) and European Committee on Antimicrobial Susceptibility Testing (EUCAST) (2018) (Table 2), it was found that only *Escherichia coli* were susceptible (S), *Staphylococcus aureus* and *Shigella flexneri* were moderately susceptible (I), whereas *Salmonella Typhi*, *Listeria monocytogenes*, *Enterococcus faecalis*, *Salmonella Typhimurium* and *Enterobacter aerogenes* were found to be resistant (R) (Table 3).

4. Conclusion

The present study investigated the antibacterial effect of *Asphodelus aestivus* Brot. on eight different food-borne pathogenic bacteria and it was found that Çiriş had antibacterial effects particularly on *Escherichia coli*, *Staphylococcus aureus* and *Shigella flexneri*.

The resistance of bacteria to the antibiotics used today is increasing day by day. Therefore, studies on the search for alternative substances have gained momentum in recent years. Particularly, studies on the antimicrobial properties of the preparations obtained from different parts of many spices and plants yielded positive results.

The results obtained in the present study are of importance in terms of showing that Çiriş can be used for these purposes and constituting a source for future studies. As an advantage, this plant, unlike many other plants and spices, grows naturally in Turkey in abundance and can be purchased for very reasonable prices.

Table 2. CLSI and EUCAST Clinical Microbiological Zone Diameter Standards (mm) (CLSI, 2015; EUCAST, 2018)

Antibiotics	<i>Esherichia coli</i>			<i>Staphylococcus aureus</i>			<i>Salmonella Typhimurium</i>			<i>Enterobacter aerogenes</i>			<i>Listeria monocytogenes</i>			<i>Shigella flexneri</i>			<i>Salmonella Typhi</i>			<i>Enterococcus faecalis</i>		
	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R	S	I	R
Ampicilin	≥17	14-16	≤13	NT	NT	NT	≥17	14-16	≤13	≥17	14-16	≤13	NT	NT	NT	≥17	14-16	≤13	≥17	14-16	≤13	≥17	14-16	≤13
Benzylpenicillin	≥14	-	≤14	≥26	-	≤26	≥14	-	≤14	≥14	-	≤14	≥13	-	≤13	≥14	-	≤14	≥14	-	≤14	≥14	-	≤14
Amoxicillin-clavulanic acid	≥18	14-17	≤13	NT	NT	NT	≥18	14-17	≤13	≥18	14-17	≤13	NT	NT	NT	≥18	14-17	≤13	≥18	14-17	≤13	≥18	14-17	≤13
Gentamicin GN10	≥15	13-14	≤12	≥15	13-14	≤12	≥15	13-14	≤12	≥15	13-14	≤12	NT	NT	NT	≥15	13-14	≤12	≥15	13-14	≤12	≥15	13-14	≤12
Penicilin	NT	NT	NT	≥29	-	≤28	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT
Netilmicin	≥15	13-14	≤12	≥18	-	≤18	≥15	13-14	≤12	≥15	13-14	≤12	NT	NT	NT	≥15	13-14	≤12	≥15	13-14	≤12	≥15	13-14	≤12
Erythromycin	NT	NT	NT	≥23	14-22	≤13	NT	NT	NT	NT	NT	NT	≥25	-	≤25	NT	NT	NT	NT	NT	NT	NT	NT	NT
Streptomycin	≥15	12-14	≤11	NT	NT	NT	≥15	12-14	≤11	≥15	12-14	≤11	NT	NT	NT	≥15	12-14	≤11	≥15	12-14	≤11	≥15	12-14	≤11
Chloramphenicol	≥18	13-17	≤12	≥18	13-17	≤12	≥18	13-17	≤12	≥18	13-17	≤12	NT	NT	NT	≥18	13-17	≤12	≥18	13-17	≤12	≥18	13-17	≤12
Sulfonamides	≥17	13-16	≤12	≥17	13-16	≤12	≥17	13-16	≤12	≥17	13-16	≤12	NT	NT	NT	≥17	13-16	≤12	≥17	13-16	≤12	≥17	13-16	≤12
Kanamycin	≥18	14-17	≤13	≥18	14-17	≤12	≥18	14-17	≤13	≥18	14-17	≤13	NT	NT	NT	≥18	14-17	≤13	≥18	14-17	≤13	≥18	14-17	≤13
Tetracycline	NT	NT	NT	≥19	15-18	≤14	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT
Clindamycin	NT	NT	NT	≥21	15-20	≤14	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT
Fusidic Acid	NT	NT	NT	≥24	-	≤44	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT
Penicilin	NT	NT	NT	≥29	-	≤28	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT	NT

NT: No Test Record, S: Sensitive, R: Resistant, I: Medium Sensitive.

Table 3 Determination of the antibacterial effect of the extract of *Asphodelus aestivus* Brot. on eight different foodborne pathogenic bacteria according to the standard values determined by EUCAST (2018) and CLSI (2015)

Bacteria	Antibacterial Effect
<i>Staphylococcus aureus</i>	I
<i>Salmonella Typhi</i>	R
<i>Listeria monocytogenes</i>	R
<i>Escherichia coli</i>	S
<i>Enterococcus faecalis</i>	R
<i>Salmonella Typhimurium</i>	R
<i>Shigella flexneri</i>	R
<i>Enterobacter aerogenes</i>	R

S: Sensitive, R: Resistant, I: Moderate Sensitive

References

- Acibuda, V. & Bostan-Budak, D. (2018) The place and importance of medicinal and aromatic plants in the World and Turkey. *Cukurova Journal of Agricultural Food Sciences*, 33(1), 37-44.
- Akarca, G. (2019). Composition and antibacterial effect on food borne pathogens of *Hibiscus sarrattensis* L. calyces essential oil. *Industrial Crops & Products*, 137, 285-289. <https://doi.org/10.1016/j.indcrop.2019.05.043>
- Akarca, G., Tomar, O., Guney, I., Erdur, S. & Gok, V. (2019) Determination of sensitivity of some food pathogens to spice extract. *Journal of Food Science and Technology*, 56(12), 5253-5261. <https://doi.org/10.1007/s13197-019-03994-1>
- Akcicek, E. & Vural, M. (2007). The endemic and rare non-endemic plants of Kumalar Mountain (Afyonkarahisar). *J. BAUN Inst. Sci. Technol*, 9(2),78-86.
- Bauer, A. W., Kirby, W. M. M., Sherris, J. C. & Turck, M. (1966). Antibiotic susceptibility testing by a standardized single disk method. *Am. J. Clin. Pathol*, 36, 493-496.
- Bayrak, B. B. (2013). Antioxidant activities of Çiriş (*Eremurus Spectabilis* Bieb.) and some sulfur compounds, Ph.D. Thesis, İstanbul University, Graduate school of natural and applied sciences, İstanbul, Turkey.
- Baytop, T. (1999). Treatment with plants in Turkey, Nobel Medical Bookstores, Istanbul, Turkey.
- CLSI. (2015). Clinical and Laboratory Standards Institute, Zone diameter and minimal inhibitory concentration (MIC) Standards.
- EUCAST. (2018). European Committee on Antimicrobial Susceptibility Testing, http://www.Eucast.org/fileadmin/src/media/PDFs/eucast_files/Breakpoint_tables/v_8.0_Breakpoint_Tables.pdf
- Fatal, T., Yilmaz, F. F., Birincioglu, S. S., Hosgor-Limoncu, M. & Kivrak, B. (2016). Fatty acid composition and antimicrobial activity of *Asphodelus aestivus* seeds. *Human & Veterinary Medicine, International Journal of the Bioflux Society*, 8(2), 103-107.
- Karatepe, P. & Patir, B. (2012). The effect of eugenol and thymol on chemical, microbiological and sensory quality of pasteurized butter. *Firat University Medical Journal of Health Sciences*, 26, 35-46.
- Li, C., Shi, J. G., Zhang, Y. P. & Zhang, C. Z. (2000). Constituents of *Eremurus chinensis*, *Journal of Natural Products*, 63, 653-656.
- Oskay, M., Tamer, A. U., Ay, G., Sari, D. & Akta, K. (2005) Antimicrobial activity of the leaves of *Lippia triphylla* (L'Her) O. Kuntze (Verbenaceae) against on bacteria and yeasts. *Journal of Biological Sciences*, 5, 620-622.
- Oskay, M., Aktas, K., Sari, D. & Azeri, C. A. (2007). Comparative study of antimicrobial activity using well and disk diffusion method on *Asphodelus aestivus* (Liliaceae). *Ekoloji*, 16, 62-65.
- Peksel, A., Altas-Kiyamaz, N. & Imamoglu, S. (2012). Evaluation of antioxidant and antifungal potential of *Asphodelus aestivus* Brot. growing in Turkey. *Journal of Medicinal Plants Research*, 62, 253-265.
- Silva, F. & Domingues, F. C. (2017). Antimicrobial activity of coriander oil and its effectiveness as food preservative. *Crit. Rev. Food Sci. Nutr*, 57, 35-47.
- Ugurly, I., Baslar, S., Yorek, N. & Dogan, Y. (2009). The investigation and quantitative ethnobotanical evaluation of medicinal plants used around Izmir province, Turkey. *J. Med. Plant Res*, 3(5), 345-367.
- Zhang, Y. P., Zhang, C. Z., Tao, B. Q. & Li, C. (2000). Chemical constituents from *Eremurus chinensis* Fedtsch. *China Journal of Chinese Materia Medica*, 25, 355-356.