Afyon Kocatepe Üniversitesi Fen ve Mühendislik Bilimleri Dergisi
Afyon Kocatepe University – Journal of Science and Engineering
https://dergipark.org.tr/tr/pub/akufemubid

Araştırma Makalesi / Research Article
DOI: https://doi.org/10.35414/akufemubid.1359936
AKÜ FEMÜBİD **24** (2024) 035401 (620-627)
AKU J. Sci. Eng. **24** (2024) 035401 (620-627)

# Biological Activities of Extracts of Red and Yellow Hawthorn Fruits in Different Solvents

***Makale Bilgisi / Article Info***
Alındı/Received: 14.09.2023
Kabul/Accepted: 06.05.2024
Yayımlandı/Published: 27.06.2024

## Kırmızı ve Sarı Alıç Meyvelerinin Farklı Çözücülerdeki Ekstraktlarının Biyolojik Aktiviteleri

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#This manuscript was presented as an oral presentation at the 6th International Anatolian Agriculture, Food, Environment and Biology Congress (TARGID 2022) held in Kütahya/Turkey between 07-09/October/2022 and this manuscript is the improved version of the abstract of the paper published in the congress abstract book.



**Öz**

Bu çalışmada Afyonkarahisar İl'inde doğal ortamda yetişen alıç meyvelerinden etanol, metanol ve aseton çözgenleri kullanılarak farklı ekstraktlar elde edilmiştir. Bu ekstraktların önceden belirlenmiş olan hedef mikroorganizmalar üzerine antibakteriyel ve antifungal aktiviteleri incelenmiştir. Çalışma sonucunda elde edilen verilere göre en yüksek antibakteriyel aktivite kırmızı alıcın etanol ekstraktında 12.06 mm zon çapı ile *Listeria monocytogenes* üzerinde tespit edilmiştir. En yüksek antifungal aktivite ise sarı alıcın etanol ekstraktında 17.22 mm zon çapı ile *Aspergillus flavus* üzerinde tespit edilmiştir. Bunun yanında en düşük minimum inhibitör konsantrasyon (bakteriyel) değeri kırmızı alıcın etanol ekstraktında 46.87 µg/mL ile Listeria monocytogenes üzerinde belirlenmiştir. En düşük minimum inhibitör konsantrasyon (fungal) değeri ise sarı alıcın etanol ekstraktında 35.15 µg/mL ile *Aspergillus flavus* üzerinde gözlenmiştir. En düşük minimum bakterisidal konsantrasyon değeri 93.75 µg/mL ile kırmızı alıcın etanol ekstraktında *Staphylococcus aureus* üzerinde tespit edilirken, en düşük fungisidal konsantrasyon değeri 23.44 µg/mL ile sarı alıcın etanol ekstraktında hem *Aspergillus flavus*, hem de *Penicillium notatum* üzerinde tespit edilmiştir. Çalışma sonuçları sarı alıcın ve kırmızı alıcın özellikle etanol ekstraktlarının yüksek antibakteriyel ve antifungal etkilere sahip olduğunu göstermiştir.

**Anahtar Kelimeler:** Antibakteriyel; Antifungal; Ekstrakt; Listeria monocytogenes; Aspergillus flavus.

**Abstract**

In this study, different extracts were obtained from hawthorn fruits growing in the natural environment in Afyonkarahisar Province, using ethanol, methanol, and acetone solvents. The antibacterial and antifungal activities of these extracts on predetermined target microorganisms were examined. According to the data obtained as a result of the study, the highest antibacterial activity was detected on *Listeria monocytogenes* in the ethanol extract of red hawthorn with a zone diameter of 12.06 mm. The highest antifungal activity was detected in the ethanol extract of yellow hawthorn on *Aspergillus flavus* with a zone diameter of 17.22 mm. In addition, the lowest minimum inhibitory concentration (bacterial) value was determined on *Listeria monocytogenes* in the ethanol extract of red hawthorn with 46.87 µg/mL. The lowest minimum inhibitory concentration (fungal) value was observed on *Aspergillus flavus* in the ethanol extract of yellow hawthorn with 35.15 µg/mL. The lowest minimum bactericidal concentration value was detected on *Staphylococcus aureus* at 93.75 µg/mL in the ethanol extract of red hawthorn, while the lowest fungicidal concentration value was detected on both *Aspergillus flavus* and *Penicillium notatum* at 23.44 µg/mL in the ethanol extract of yellow hawthorn. The study results showed that especially ethanol extracts of yellow and red hawthorn have high antibacterial and antifungal effects.

***Keywords:*** *Hawthorn; Antibacterial; Antifungal; Extract; Listeria monocytogenes; Aspergillus flavus*

## 1. Introduction

Hawthorn (Crataegus spp.) is a fruit in the Roseceae family. It is a plant species that can grow spontaneously in many regions of Turkey, especially on mountainsides. It has a structure in the form of a bush or small tree, generally varying between 5 and 10 m in height (Ahmadipour *et al*. 2019). It is reported that hawthorn varieties, which can have different colors depending on the environment in which they grow, mostly have yellow, red, and dark purple colors (Polatcı and Taşova 2017). Hawthorn is a fruit with high nutritional value. It is rich in sugar, vitamin C, and mineral substances such as Ca, P, K, Mg, and Fe (Tüysüz *et al*. 2021). It can be used in various ways. It can be consumed directly, fresh or dried, or after being turned into processed products such as jam, marmalade, molasses, vinegar, and wine (Çalışkan *et al*. 2018, Türkhan *et al*. 2018). The hawthorn plant, especially its fruit, leaves, and flowers, have been used among the public for therapeutic purposes since ancient times (Çoklar and Akbulut 2016, Caliskan and Karaman 2018, Rüzgar and Yazıcı 2022). The variety and amount of

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biochemical substances found in the fruit are effective in having many positive effects on the human health. Bioactive compounds such as chlorogenic acid, epicatechin, and hyperoside in the structure of hawthorn make the fruit a powerful source of antioxidants and provide free radical scavenging activity (Altınbaşak and Çelik 2021).

Previous studies have shown that hawthorn and its products have antimicrobial, antioxidant, and anti-inflammatory effects. It has also been found to have positive effects on high blood pressure, high cholesterol, and cardiovascular diseases (Kaya *et al*. 2019). When the literature was examined, no study was found on the effect of extracts obtained from different hawthorn species with the help of different solvents. The aim of this study was to determine the effects of extracts obtained with the use of different solvents on predetermined target pathogenic bacteria and spoilage molds.

## 2. Materials and Methods

### *2.1. Material*

The yellow (*Crataegus tanacetifolia*) and red (*Crataegus monogyna*) hawthorn fruits used in the study were obtained from the rural areas of Büyükkalecik-Afyonkarahisar, Türkiye (38°40'50''N, 30°29'20''E, Altitude: 1501) in October 2022.

### *2.2. Solvent extraction*

Yellow and red hawthorns used in the study were dried in the laboratory at room temperature for two days. Then, the dried fruits were weighed as 150 g each, and 400 ml of 80% (v/v) each solvent (ethanol, methanol, acetone) was added. Then, it was shaken in a dark room at 120 rpm using a shaker (WiseShake® SHO-2D) for 24 hours. At the end of the period, the mixtures were filtered through sterilizing paper (Whatman, Grade 54, Diameter 55 mm) and taken into a rotary evaporator (Heidolph Hei-VAP value), and the ethanol and extract parts were separated from each other at the rate of 100 rpm and the temperature of 40 °C.

### *2.3. Microorganisms used in the study*

In this study, both antibacterial and antifungal activities of the extracts were determined. For this reason, *Escherichia coli* (ATCC 25922), *Staphylococcus aureus* (ATCC 6538), *Listeria monocytogenes* (ATCC 51774) and *Salmonella Enteritidis* (ATCC 13076) were used as test bacteria, while *Aspergillus flavus* (ATCC 204304), *Aspergillus niger* (ATCC 16888), *Penicillium notatum* (ATCC 9478), and *Penicillium chrysogenum* (ATCC 10106) were used as test molds.

### *2.4 Preparation of discs containing hawthorn extracts*

Extracts obtained from two different hawthorn varieties using different solvents were taken into separate 100 µL petri dishes (Sterile, 90 x 15, Fıratmed, Turkey) using a sterile-tipped automatic pipette (Research Plus, Eppendorf). Blank antibiogram discs (Bioanalyse 316010001) were placed on them. For the discs to impregnate the extracts, the petri dishes were kept in the refrigerator (Arçelik, 554271, Turkey) at 4℃ for 60 minutes with their lids closed. At the end of the specified period, the discs were dried in a laminar flow cabinet (Cryste, Puricube 1200).

### *2.5. Determination of antibacterial and antifungal activities in discs containing extracts*

The methods specified by Akarca (2019) were used to determine the antibacterial activities, while the methods specified by Alastruey-Izquierdo *et al*. (2015) were used to determine the antifungal activities with some minor modifications.

Young cultures grown on specific media were used to determine antimicrobial activities. For this purpose, the following media were used for each microorganism in determining the antibacterial activity, respectively. For E. coli: Chromocult TBX Agar (Merck 1.16122), for S. aureus: Baird Parker Agar (Merck 1.05406), for L. monocytogenes: Oxford Agar (Merck 1.07004), for S. Enteritidis: Brilliant Green Phenol Red Lactose Sucrose Agar (Merck 1.10747). Bacteria were incubated in an incubator (Incucell MMM, Germany) at 37±0.1℃ for 16 – 20 hours (*L. monocytogenes* in a 5 % $CO^2$ environment). To determine antifungal activity, mold strains were subjected to incubation periods of 5-7 days at 25℃ on Sabouraud 2 % Dextrose Agar (Merck 1.07315). At the end of the incubation period, the cultures were transferred with a sterile loop and suspended in Ringer's solution. In both analyses, the density of the suspended strains was adjusted to 0.5 McFarland Standard using a densitometer (Biosan, 1B, Turkey). The density-adjusted inoculums were taken with the help of transport swap (Fıratmed, Turkey) and homogeneously inoculated onto the freshly prepared Muller Hinton Agar (Merck 1.05437) surface at 25℃ for antibacterial activity analysis. On the other hand, for the determination of antifungal activity, again, the inoculums were homogeneously inoculated onto the surface of Muller Hinton Agar, this time modified with 2 % glucose and 0.5 mg/L methylene blue at 25℃. Then, 10 minutes were waited for the inoculations to be absorbed by the media. (Bauer *et al*. 1966, Akarca 2019, Akarca and Tomar 2019). Then, 100 µL of the extract was impregnated and dried into each blank antibiogram disk

(Bio-Disk 316010001, Turkey) and placed on the surface of the media at a distance so that the zones that would form would not touch each other. Petri dishes were then incubated (bacteria at 37±0.1℃ for 16 – 20 hours, molds at 25±0.1℃ for 72 – 96 hours). At the end of the period, The diameters of the zones formed in all petri dishes were measured with the help of a digital caliper (Mitutoyo, 500-181-30, Japan) under sufficient sunlight.

### *2.6. Statistical analysis*

The data obtained within the scope of the study were evaluated using the SPSS V 23.0.0 statistical package program. The study was conducted in double replication and double parallel, and the analysis of variance technique was used to evaluate the data obtained. The level of differences was determined by the Duncan test ($p<0.05$).

**Table 1.** Antibacterial activity of yellow and red hawthorn extracts on selected pathogen bacteria (zone diameter, mm)

| | Yellow Hawthorn | | | | | |
|---|---|---|---|---|---|---|
| | **Methanol** | **ABE** | **Ethanol** | **ABE** | **Acetone** | **ABE** |
| ***E. coli*** | 6.02±0.01$^{D}$ | - | 9.15±0.91$^{A}$ | ++ | 7.32±0.29$^{BC}$ | - |
| ***S. aureus*** | 7.02±0.01$^{D}$ | - | 10.28±0.30$^{B}$ | ++ | 9.21±0.23$^{C}$ | ++ |
| ***L. monocytogenes*** | 6.02±0.01$^{D}$ | - | 18.5±0.52$^{A}$ | +++ | 6.45±0.07$^{CD}$ | - |
| ***S. Enteritidis*** | 9,06±0.19$^{A}$ | ++ | 7.82±0.71$^{B}$ | - | 6.81±0.37$^{BC}$ | - |
| | **Red Hawthorn** | | | | | |
| | **Methanol** | **ABE** | **Ethanol** | **ABE** | **Acetone** | **ABE** |
| ***E. coli*** | 8.48±0.68$^{AB}$ | + | 7.77±0.33$^{BC}$ | - | 6.89±0.22$^{CD}$ | - |
| ***S. aureus*** | 8.39±0.37$^{C}$ | + | 11.36±0.57$^{A}$ | +++ | 8.74±0.22$^{C}$ | + |
| ***L. monocytogenes*** | 7.795±0.87$^{C}$ | - | 12.06±0.87$^{B}$ | +++ | 7.78±0.35$^{C}$ | - |
| ***S.* Enteritidis** | 6.505±0.45$^{C}$ | - | 7.85±0.32$^{B}$ | - | 6.78±0.48$^{BC}$ | - |

ABE: Anti Bacterial Effect, A - D (→): Values with the different capital letters in the same line for each analysis differ significantly, ($p < 0.05$). 6-8(-): Resistant, 8-9(+): Moderately Sensitive, 9-11(++): Sensitive, 11≥ (+++): Ultrasensitive.

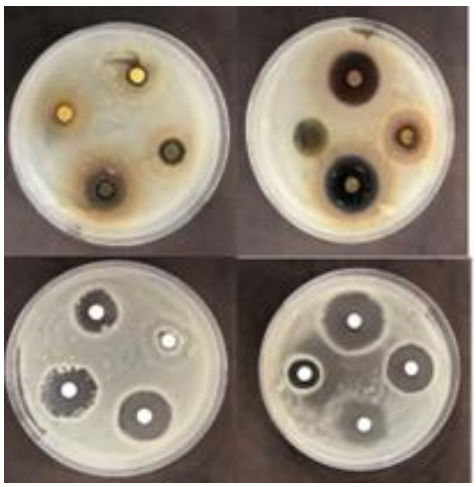

**Figure 1.** Antibacterial inhibiton zones of yellow and red hawthorn extracts

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## 3. Results and Discussions

### *3.1. Antibacterial activities of yellow and red hawthorn extracts*

The antibacterial activities of yellow and red hawthorn extracts obtained using methanol, ethanol and acetone on selected pathogenic bacteria have been shown in Table 1 and Picture 1.

### *3.2. MIC (Bacterial) values of yellow and red hawthorn extracts*

MIC (bacterial) values of yellow and red hawthorn extracts obtained using methanol, ethanol and acetone on selected pathogenic bacteria have been shown in Table 2.

### *3.3. MBC values of yellow and red hawthorn extracts*

MBC values of yellow and red hawthorn extracts obtained using methanol, ethanol and acetone on selected pathogenic bacteria have been shown in Table 3.

### *3.4. Antifungal activities of yellow and red hawthorn extracts*

The antifungal activities of yellow and red hawthorn extracts obtained using methanol, ethanol and acetone have been shown in Table 4.

**Table 2.** MIC (bacterial) values (µg/mL) of yellow and red hawthorn extracts on selected pathogen bacteria

| | Yellow Hawthorn | | |
|---|---|---|---|
| | **Methanol** | **Ethanol** | **Acetone** |
| ***E. coli*** | 750.00±0.00$^{A}$ | 562.50±265.16$^{A}$ | 750.00±0.00$^{A}$ |
| ***S. aureus*** | 750.00±0.00$^{A}$ | 140.63±66.29$^{C}$ | 375.00±0.00$^{B}$ |
| ***L. monocytogenes*** | 750.00±0.00$^{A}$ | 93.75±0.00$^{B}$ | 750.00±0.00$^{A}$ |
| ***S. Enteritidis*** | 750.00±0.00$^{A}$ | 750.00±0.00$^{A}$ | 750.00±0.00$^{A}$ |
| | **Red Hawthorn** | | |
| | **Methanol** | **Ethanol** | **Acetone** |
| ***E. coli*** | 562.50±265.16$^{A}$ | 562.50±265.16$^{A}$ | 750.00±0.00$^{A}$ |
| ***S. aureus*** | 375.00±0.00$^{B}$ | 70.31±33.15$^{C}$ | 375.00±0.00$^{B}$ |
| ***L. monocytogenes*** | 562.50±265.16$^{A}$ | 46.87±0.00$^{B}$ | 750.00±0.00$^{A}$ |
| ***S. Enteritidis*** | 750.00±0.00$^{A}$ | 750.00±0.00$^{A}$ | 750.00±0.00$^{A}$ |

A - C (→): Values with the different capital letters in the same line for each analysis differ significantly ($p < 0.05$).

**Table 3.** MBC values (µg/mL) of yellow and red hawthorn extracts on selected pathogen bacteria

| | Yellow Hawthorn | | |
|---|---|---|---|
| | **Methanol** | **Ethanol** | **Acetone** |
| ***E. coli*** | 1000.00±0.00$^{A}$ | 500.00±0.00$^{AB}$ | 750.00±0.00$^{AB}$ |
| ***S. aureus*** | 750.00±353.55$^{A}$ | 187.50±88.38$^{B}$ | 375.00±0.00$^{AB}$ |
| ***L. monocytogenes*** | 1000.00±0.00$^{A}$ | 125.00±0.00$^{D}$ | 750.00±0.00$^{B}$ |
| ***S.* Enteritidis** | 1000.00±0.00$^{A}$ | 750.00±353.55$^{AB}$ | 750.0±0.00$^{AB}$ |
| | **Methanol** | **Ethanol** | **Acetone** |
| ***E. coli*** | 750.00±353.55$^{AB}$ | 375.00±176.78$^{B}$ | 750.00±353.55$^{AB}$ |
| ***S. aureus*** | 375.00±176.77$^{AB}$ | 93.75±44.19$^{B}$ | 375.00±176.78$^{AB}$ |
| ***L .monocytogenes*** | 500.00±0.00$^{C}$ | 125.00±0.00$^{D}$ | 375.00±176.77$^{C}$ |
| ***S. Enteritidis*** | 1000.00±0.00$^{A}$ | 500.00±0.00$^{B}$ | 1000.00±0.00$^{A}$ |

A - D (→): Values with the different capital letters in the same line for each analysis differ significantly ($p < 0.05$).

**Table 4.** Antifungal activities of yellow and red hawthorn extracts on selected molds (zone diameter, mm)

| | Yellow Hawthorn | | | | | |
|---|---|---|---|---|---|---|
| | **Methanol** | **AFE** | **Ethanol** | **AFE** | **Acetone** | **AFE** |
| ***A. flavus*** | 10.15±0.68$^{BC}$ | ++ | 17.22±0.43$^{A}$ | +++ | 10.18±0.31$^{BC}$ | ++ |
| ***A. niger*** | 8.74±0.05$^{C}$ | + | 15.21±0.43$^{A}$ | +++ | 8.69±0.11$^{C}$ | + |
| ***P. notatum*** | 9.60±0.46$^{BC}$ | ++ | 16.76±0.44$^{A}$ | +++ | 9.88±0.53$^{BC}$ | ++ |
| ***P. chrysogenum*** | 8.41±0.51$^{C}$ | + | 11.46±0.44$^{A}$ | +++ | 12.58±0.63$^{A}$ | +++ |
| | **Red Hawthorn** | | | | | |
| | **Methanol** | **AFE** | **Ethanol** | **AFE** | **Acetone** | **AFE** |
| ***A. flavus*** | 9.27±0.72$^{C}$ | ++ | 10.68±0.48$^{B}$ | ++ | 11.18±0.23$^{B}$ | +++ |
| ***A. niger*** | 7.69±0.43$^{C}$ | - | 11.07±0.57$^{B}$ | +++ | 7.97±0.55$^{C}$ | - |
| ***P. notatum*** | 9.13±0.46$^{C}$ | ++ | 10.28±0.04$^{B}$ | ++ | 9.10±0.15$^{C}$ | ++ |
| ***P. chrysogenum*** | 8.87±0.02$^{BC}$ | + | 9.36±0.22$^{BC}$ | ++ | 9.87±0.61$^{B}$ | ++ |

AFE: Anti Fungal Effect, A - C (→): Values with the different capital letters in the same line for each analysis differ significantly, ($p < 0.05$). 6-8(-): Resistant, 8-9(+): Moderately Sensitive, 9-11(++): Sensitive, 11≥ (+++): Ultrasensitive.

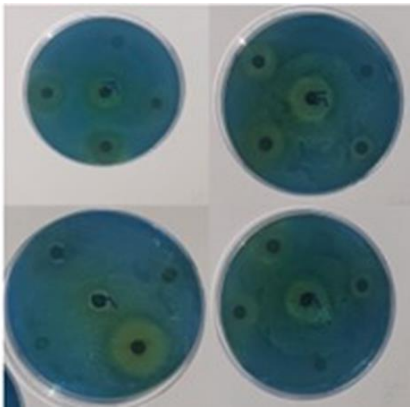

**Figure 2.** Antifungal inhibiton zones of yellow and red hawthorn extracts

### *3.5. MIC (Fungal) values of yellow and red hawthorn extracts*

MIC (fungal) values of yellow and red hawthorn extracts obtained using methanol, ethanol and acetone on selected molds have been shown in Table 5.

### *3.6. MFC values of yellow and red hawthorn extracts*

MFC values of yellow and red hawthorn extracts obtained using methanol, ethanol and acetone on selected molds have been shown in Table 6.

## 4. Discussion

### *4.1. Antibacterial activities of yellow and red hawthorn extracts*

The highest antibacterial activity on *E. coli*, one of the bacteria used in the study, was detected in the ethanol extract of yellow hawthorn with a zone diameter of 9.15 mm. The highest antibacterial activity on *S. aureus* was detected in the ethanol extract of red hawthorn with a zone diameter of 11.36 mm. The highest antibacterial activity on *L. monocytogenes* was detected in the ethanol extract of yellow hawthorn with a zone diameter of 18.5 mm, while the highest antibacterial activity on *S. Enteritidis* was detected in the methanol extract of yellow hawthorn with a zone diameter of 9.06 mm ($p<0.05$).

**Table 5.** MIC (fungal) values (µg/mL) of yellow and red hawthorn extracts on selected molds

| | Yellow Hawthorn | | |
|---|---|---|---|
| | **Methanol** | **Ethanol** | **Acetone** |
| ***A. flavus*** | 562.50±265.16$^{A}$ | 35.15±16.57$^{C}$ | 375.00±0.00$^{AB}$ |
| ***A. niger*** | 750.00±0.00$^{A}$ | 70.31±33.15$^{C}$ | 281.25±132.58$^{B}$ |
| ***P. notatum*** | 281.25±132.58$^{AB}$ | 58.59±49.72$^{B}$ | 140.63±66.29$^{AB}$ |
| ***P. chrysogenum*** | 187.50±0.00$^{A}$ | 93.75±0.00$^{A}$ | 70.31±33.15$^{A}$ |
| | **Red Hawthorn** | | |
| | **Methanol** | **Ethanol** | **Acetone** |
| ***A. flavus*** | 375.00±0.00$^{AB}$ | 117.19±99.44$^{BC}$ | 70.31±33.15$^{C}$ |
| ***A. niger*** | 750.00±0.00$^{A}$ | 46.87±0.00$^{C}$ | 375.00±0.00$^{B}$ |
| ***P. notatum*** | 562.50±265.16$^{A}$ | 70.31±33.15$^{B}$ | 562.50±265.16$^{A}$ |
| ***P. chrysogenum*** | 281.25±132.58$^{A}$ | 281.25±132.58$^{A}$ | 281.25±132.58$^{A}$ |

A - C (→): Values with the different capital letters in the same line for each analysis differ significantly, ($p < 0.05$).

**Table 6.** MFC values (µg/mL) of yellow and red hawthorn extracts on selected molds

| | Yellow Hawthorn | | |
|---|---|---|---|
| | **Methanol** | **Ethanol** | **Acetone** |
| ***A. flavus*** | 187.50±88.38$^{AB}$ | 23.44±11.05$^{B}$ | 375.00±176.77$^{A}$ |
| ***A. niger*** | 750.00±353.55$^{AB}$ | 46.875±22.10$^{C}$ | 187.50±88.39$^{C}$ |
| ***P. notatum*** | 562.50±437.50$^{A}$ | 23.44±11.05$^{A}$ | 93.75±44.20$^{A}$ |
| ***P. chrysogenum*** | 125.00±0.00$^{A}$ | 62.50±0.00$^{A}$ | 46.88±22.10$^{A}$ |
| | **Red Hawthorn** | | |
| | **Methanol** | **Ethanol** | **Acetone** |
| ***A. flavus*** | 375.00±176.77$^{A}$ | 78.13±66.29$^{B}$ | 46.88±22.10$^{B}$ |
| ***A. niger*** | 1000.00±0.00$^{A}$ | 31.25±0.00$^{C}$ | 375.00±176.78$^{BC}$ |
| ***P. notatum*** | 750.00±353.55$^{A}$ | 62.50±0.00$^{A}$ | 625.00±530.33$^{A}$ |
| ***P. chrysogenum*** | 187.50±88.39$^{A}$ | 156.25±132.58$^{A}$ | 187.50±88.39$^{A}$ |

A - C (→): Values with the diffrent capital letters in the same line for each analysis differ significantly, ($p < 0.05$).

In addition, it was determined that *E. coli* was sensitive to the ethanol extract of yellow hawthorn. In contrast, it was resistant to methanol and acetone extracts of yellow hawthorn. Furthermore, *E. coli* was moderately sensitive to the red hawthorn extract, while it was resistant to ethanol and acetone extracts of red hawthorn. It was determined that *S. aureus* was sensitive to both ethanol and acetone extracts of yellow hawthorn and resistant to methanol extract. At the same time, it was extremely sensitive to ethanol extract of red hawthorn and moderately sensitive to methanol and acetone extracts. It was observed that *L. monocytogenes* was extremely sensitive to ethanol extracts of both yellow and red hawthorn and resistant to other extracts. It was observed

that *S. Enteritidis* was sensitive only to the methanol extract of yellow hawthorn and was resistant to the other extracts. In a similar study, it was determined that the ethanol extract of hawthorn fruit had a bactericidal effect, especially on Gram-positive bacteria. Antibacterial activity was detected on *L. monoctogenes* with a zone diameter of 17 mm (Tadić *et al*. 2008). In a different study, it was reported that gold nanoparticles derived from the aqueous extract of hawthorn fruit showed a strong antibacterial effect (Baran *et al*. 2021). In a study examining the antibacterial properties of aqueous extracts of different plum varieties in the *Rosaceae* family, it was stated that all plum extracts except the Cancur plum showed antibacterial activity against the tested bacteria (Murathan *et al*. 2020). Bioactive compounds such as polyphenols found in the extracts are effective on antibacterial activity. (Ghendov-Moşanu et al. 2018). Antibacterial activity against Gram-negative bacilli is demonstrated by apigenin, vitexin, and saponarin flavones, while flavonoid compounds with two or three hydroxyl groups in the A or B rings show antibacterial effects against Gram-positive bacteria (Kostic *et al.* 2012).

### *4.2. MIC (Bacterial) and MBC values of yellow and red hawthorn extracts*

Among the extracts of yellow hawthorn obtained using different solvents, the lowest MIC (bacterial) and MBC values were detected against *L. monocytogenes* in the ethanol extract as 93.75 µg/mL and 125.00 µg/mL ($p<0.05$). Among the red hawthorn extracts, the lowest MIC (bacterial) value was 46.87 µg/mL against *L. monocytogenes* in the ethanol extract, while the lowest MBC value was 93.75 µg/mL against *S. aureus* again in the ethanol extract. ($p<0.05$). In their study, Tadić *et al.* (2008) determined the MIC (bacterial) and MBC values of hawthorn fruit ethanol extract as 151 µg/mL and 187 µg/mL for both *L. monocytogenes* and *S. aureus*. It has been thought that location is effective in the difference between studies.

### *4.3. Antifungal activities of yellow and red hawthorn extracts*

In this study, the highest antifungal activity on *A. flavus*, *A. niger* and *P. notatum* was determined in the ethanol extract of yellow hawthorn with zone diameters of 17.22, 15.21 and 16.76 mm, respectively ($p<0.05$). Unlike the others, the highest antifungal activity on P. chrysogenum was detected in the acetone extract of yellow hawthorn with a zone diameter of 12.58 mm ($p<0.05$). However, it has been determined that *A. flavus* was ultrasensitive to the ethanol extract of yellow hawthorn and the acetone extract of red hawthorn, while it was sensitive to the other extracts of yellow and red hawthorns. *A. niger* was ultrasensitive to the ethanol extracts of both yellow and red hawthorn, moderately sensitive to the methanol and acetone extracts of yellow hawthorn, resistant to the methanol and acetone extracts of red hawthorn. It was observed that *P. notatum* was ultrasensitive only to the ethanol extract of yellow hawthorn and sensitive to all other extracts. It has been determined that *P. chrysogenum* was ultrasensitive to the both ethanol and acetone extracts of yellow hawthorn, sensitive to the ethanol and acetone extracts of red hawthorn, moderately sensitive to the methanol extracts of both yellow and red hawthorns.

In a study examining the antimicrobial properties of ethyl acetate extracts of various species of Hawthorne (*Crataegus* spp.) *(Rosaceae*), the strongest antifungal effect was detected on P. notatum (Güven *et al.* 2006).

### *4.4. MIC (Fungal) and MFC values of yellow and red hawthorn extracts*

Among the extracts of yellow hawthorn obtained using different solvents, the lowest MIC (fungal) value of 35.15 µg/mL was detected against *A. flavus* in the ethanol extract ($p<0.05$). The lowest MIC (fungal) value among red hawthorn extracts was detected against *A. niger* in the ethanol extract, with 46.87 µg/mL ($p<0.05$). The lowest MFC values were determined against *A. flavus* and *P. notatum* with a value of 23.44 µg/mL in the ethanol extract of the yellow hawthorn, while it was determined against *A. niger* with a value of 31.25 µg/mL in the ethanol extract of the red hawthorn ($p<0.05$).

The hawthorn fruit itself and the products obtained from this fruit have been used in the treatment of various diseases among the public for many years. In this study, it was tried to determine the antimicrobial activities of extracts obtained from two different hawthorn varieties with the help of different solvents. The results obtained showed that the hawthorn extracts have antimicrobial activity. Especially in recent years, the increasing demand of consumers for natural foods has led to greater emphasis on using natural additives in industrially produced foods. The results of this study will lead to new studies aimed at extending the shelf life of foods by adding these kinds of products to industrially processed foodstuffs.

**Declaration of Ethical Standards**

The authors declare that they comply with all ethical standards.

**Credit Authorship Contribution Statement**

Author-1: Conceptualization, investigation, supervision and writing – review and editing.
Author-2: Formal analysis, Data curation
Author-3: Methodology, conceptualization, investigation, supervision
Author-4: Data curation, Writing – original draft,

**Declaration of Competing Interest**

The authors have no conflicts of interest to declare regarding the content of this article.

**Data Availability Statement**

All data generated or analyzed during this study are included in this published article.

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