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Antimicrobial Activities of Fruits of *Crataegus* and *Pyrus* Species

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Abstract

The antimicrobial activity of the ethyl acetate extracts from fruits of *Crataegus tanacetifolia*, *Crataegus x bornmülleri*, *Pyrus communis* subsp. *communis*, and *Pyrus serikensis* (Rosaceae) were investigated by the agar-well diffusion assay, and the extracts exhibited antimicrobial effect against most of the bacteria and all of the yeasts tested. Although the plants differed in their activities against the microorganisms tested, the extracts of *C. x bornmülleri* and *P. communis* subsp. *communis* displayed no antifungal activity against the fungi tested. *P. serikensis* extract inhibited the growth of all bacteria tested but not *Aspergillus niger*, *Aspergillus parasiticus*, *Aspergillus citri*, and *Aspergillus fumigatus*. In conclusion, *P. serikensis* was the most active antimicrobial plant. Particularly, *P. serikensis* extract showed significant antifungal activities and could be used as an antimicrobial agent in new drug therapy.

Keywords: Antimicrobial activity, *Crataegus tanacetifolia*, *Crataegus x bornmülleri*, endemic, *Pyrus communis* subsp. *communis*, *Pyrus serikensis*.

Introduction

Hawthorne (*Crataegus* spp.) (Rosaceae) consists of small trees and shrubs. Common names for hawthornes include May blossom, quick thorn, whitethorn, haw hazels, gazels, halves, hagthorn, and bread and cheese tree. They are usually multibranched 2–5 m shrubby trees that can reach a height of up to 10 m. The hawthorne tree prefers the forest margins of lower and warmer areas. The color of the ripe fruit ranges from yellow through green to red and on to dark purple. Most of the species ripen their fruit in early to mid-autumn (Özcan et al., 2005).

Crataegus tanacetifolia (Lam.) Pers. fruits are yellow, sometimes suffused with red, globose, and 2 cm in diameter. *C. x bornmuelleri* Zabel resembles *C. tanacetifolia* in its glandular-serrate leaves but differs in its larger (3–5 cm), villous leaves. Both *Crataegus* spp. are endemic to Turkey (Davis, 1972).

Pyrus communis subsp. *communis* (pear) tree is up to 20 m and is common in Europe and Turkey. Fruits are pyriform to subglobose, 2–4 cm long, and yellowish green. *Pyrus communis* subsp. *communis* is described from Europe and Turkey (Davis, 1972).

Pyrus serikensis A. Güner et H. Duman are trees or shrubs up to 10 m high. The trunk is up to 60 cm in diameter. It is endemic as an East Mediterranean element and is called a "zingit" by local people. Fruits are 0.7–1.5 cm in diameter, globose, reddish brown, with white spots (Güner & Duman, 1991).

The medicinal use of hawthorne has a long tradition with written records dating back to ancient Roman times (Veveris et al., 2004). Some *Crataegus* constituents are predicted to be good antioxidants. The flower and fruit constituents responsible for free radical scavenging activity are epicatechin and chlorogenic acid. They are among the best antilipoperoxidants (Bahorun & Trotin, 1994; Rakotoarison & Greisser, 1997; Škerget, 2005). Traditionally, the fruits or the berries are used for their astringent properties in heavy menstrual bleeding and in diarrhea. Both the flowers and berries act as diuretics and can be used to treat kidney problems and dropsy. Apart from their delicious flavor, hawthorne fruits have been shown to have a tonic effect on the heart. Fruits of our native species are often used in the treatment of weak heart conditions, especially if this is accompanied by high blood pressure (Baytop, 1984; Schussler & Holzl, 1995). Nowadays, *Crataegus* extracts are mainly utilized for the treatment of mild forms of congestive heart failure according to the New York Heart Association functional class II (Veveris et al., 2004).

Bilia et al. (1994) isolated flavanol glycoside from the aerial parts of *Pyrus bourgaeana*. Jin and Sato (2003)

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determined that aqueous axtracts from succulent young shoots of the pear *Pyrus* spp. exhibited strong antibacterial activity against the bacterium *Erwinia amylovora* bv. 4.

As there are vast, renewable resources containing diverse natural products, the search for novel defense compounds from plants continues. Development of such plant-based compounds for microbial control is of interest to countries where there is an extensive indigenous flora. There is not much work on the antimicrobial activity of hawthorne and *Pyrus* fruits as indicated above. Therefore, attempts were made in this study to determine the antibacterial and antifungal activity of matured endemic hawthorne and *Pyrus* fruit collected from Turkey.

Materials and Methods

Plant materials and extraction procedure

Fresh wild hawthorne and *Pyrus* fruits (Davis, 1972; Güner & Duman, 1991) were collected from typical localities in Turkey. *Crataegus tanacetifolia* fruits were collected from Civan harmani, Kalabak-Eskişehir (October, 2003; 1510 m), *Crataegus x bornmülleri* fruits were collected from Dalli yayla, Kalabak-Eskişehir (October, 2003; 1520 m); *Pyrus communis* subsp. *communis* and *Pyrus serikensis* fruits were collected from Bati, Serik-Antalya (November 2003; 20 m). The specimens collected were identified with the help of *Flora of Turkey and the East Aegean Islands* (Davis, 1972) and *The Karaca Arboretum Magazine* (Güner & Duman, 1991). The voucher specimens were deposited in the Herbarium of the Faculty of Science, Anadolu University (ANES). The fruits were transported in polypropylene bags and held at room temperature. Fruits were cleaned manually and rinsed with water; 20 g of fresh fruit sample was ground in a mortar and then extracted with 150 ml of ethyl acetate (Fluka, Italy) for 24 h by using Soxhlet equipment. The extracts were filtered using Whatman filter paper (no. 1) and then concentrated *in vacuo* at 70°C. The residues were stored in a refrigerator until subsequent use. For the bioassay, the extracts were suspended in ethyl acetate at a concentration of 10 mg/ml.

Microorganisms

A total of 40 microbial genera including 28 bacteria, 3 yeasts, and 9 mold species were used in this study. Table 1 shows the isolates used in the study and their source of origin.

Antimicrobial activity

The antimicrobial activities of the ethyl acetate extract of the samples were evaluated by means of the agar-well diffusion assay (Rojas et al., 2003; Şahin et al., 2003) with some modifications. Twenty milliliters of the specified molten agar (45°C) were poured into 9-cm, sterile Petri dishes. A suspension (100 µl) containing 10^8 cfu/ml bacteria, 10^6 cfu/ml yeasts, or 10^4 spore/ml of fungi was spread on the plates of Nutrient agar (Merck, USA), Mueller-Hinton agar (Oxoid), or Sabouraud dextrose agar (Oxoid, UK) medium, respectively. Once the plates were dried aseptically, 6-mm wells were bored using a sterile cork borer. Extracts (50 µl) were placed into the wells and the plates were incubated for 37°C for 24 h for bacterial strains, 48 h for yeast, and at room temperature for 72 h for fungi. Chloramphenicol (10 mg/ml) for bacteria and ketoconazole (10 mg/ml) for yeast and fungi were used as standard antibiotics. The tests were carried out in triplicate. Antimicrobial activity was evaluated by measuring zone of inhibition against the test organism.

Results and Discussion

The microorganisms tested in this study comprised a panel of human pathogens, plant pathogens, food toxicants, and saprophytes (Table 1). The antimicrobial activities of *Crataegus tanocetifolia*, *Crataegus x bornmüllerii*, *Pyrus serikensis*, and *P. communis* subsp. *communis* ethyl acetate extracts against microorganisms were examined in the current study and their potency was qualitatively assessed by the presence or absence of inhibition zones and zone diameter (Table 1).

The results showed that the extract of *Crataegus tanacetifolia* mediated some degree of activity against bacteria and yeasts (Table 1). *Penicillium notatum* was the only fungus that was inhibited by the. *C. tanacetifolia* extract. *Salmonella typhimurium*, *P. fluorescens*, and *Pseudomonas gingeri* isolates were not inhibited by the ethyl acetate extract of *C. tanacetifolia*. The most significant result was that yeast isolate *Rhodotorola rubra* was inhibited by all of the plant extracts, and the inhibition zone diameter was higher than the standard antibiotic ketoconazole.

The extract of *Crataegus x bornmülleri* showed some degree of activity against some bacteria and all yeasts (Table 1) but not fungi. *C. x bornmülleri* did not inhibit the growth of *Yersinia enterocolitica* and *P. fluorescens*. None of the fungi showed any inhibition zone against *C. bornmülleri* extracts. Inhibiton zone diameters of *C. bornmülleri* were generally higher than *C. tanacetifolia* extract.

The extract of *Pyrus serikensis* exhibited some degree of activity against all the bacteria tested and some yeasts and fungi. Although inhibition zone diameters were lower than the standard antibiotics, *P. serikensis* extract had higher activity degree than *P. communis* subsp. *communis* extract (Table 1). The most significant results with *P. serikensis* extract were that *P. serikensis* extract was the only test material that inhibited the growth of *P. fluorescens*. However, *Aspergillus niger*, *A. parasiticus*,

Table 1. Strains used in the study and their source.

Microorganism	Source
Bacillus cereus NRRL B-3711	USDA, Agricultural Research Service, Peoria, IL, USA
Bacillus subtilis NRRL 744	
Salmonella typhimurium NRRL B-4420	
Microccoccus luteus NRRL B-4375	
Proteus vulgaris NRRL B-123	
Xanthamonas campestris pv. *campestris* NRRL-B1459	
Enterobacter aerogenes NRRL B-3567	
Streptococcocus feacalis NRRL B-14617	
Rhodotorula rubra NRRL Y-2505	
Fusarium oxysporum NRRL 5836	Ege University, Faculty of Science, Department of Biology, İzmir, Turkey
Fusarium moniliforme NRRL 1866	
Penicillium notatum NRRL 807	
Aspergillus fumigatus NRRL 163	
Aspergillus parasiticus NRRL 2999	
Aspergillus niger NRRL 321	
Escherichia coli ATCC 25922	Ankara University, Faculty of Veterinary, Ankara, Turkey
Pseudomonas aeroginosa ATCC 27853	
Staphylococcus aureus ATTC 6538 T	
Aeromonas hydrophila	J.B. Jones, University of Florida, IFAS, Plant Pathology Department
Listeria monocytogenes	
Klebsiella pneumoniae	
Yersinia enterocolitica	
Pseudomonas fluorescens B 130	
Bacillus pumilus B122	
Pseudomonas syringae pv. tomato 32	
Pseudomonas tobacco 8	
Xanthamonas campestris pv. vesicatoria 75-3	
Pseudomonas lachrymans	
Pseudomonas syringae -1	
Pseudomonas syringae pv. glycine PG1-T	
Pseudomonas syringae pv. phaseolicola NCPPB 52	Vidaver, A. K. University of Nebraska Lincoln, Institute of Agricultural and Natural Resources
Pseudomonas gingeri 3146	
Pseudomonas gladioli pv. agricola RR3	J.G. Turner, University of East Anglia, UK
Brucella spp.	Osmangazi University Medical Faculty
Pseudomonas maltophila	
Candida albicans	
Candida globrata	
Geotrichum graminis	
Cladosporum herbarum	Anadolu University Faculty of Science
Aspergillus citri	

A. citri, and *A. fumigatus* were not inhibited by the extract, although the other fungi were susceptible.

The *Pyrus communis* subsp. *communis* extract inhibited the growth of many bacteria and all the yeasts but not the fungi (Table 1). *S. typhimurium*, *Brucella* spp., *Streptococcus feacalis*, *P. fluorescens*, *Pseudomonas tobacco*, *Pseudomonas lachrymans*, *P. s.* pv. *phaseolicola*, *Pseudomonas gingeri*, and *R. rubra* were not inhibited by the *P. communis* subsp. *communis* extract.

We have evaluated plants that are naturally grown in Turkey and preferably endemic and are potential useful resources. Therefore, these studies will be beneficial from medicinal and economic standpoints.

It is well-known that an extract of succulent young shoot tissue of pear exhibits strong antibacterial activity, and this activity was found in 12 cultivars of Chinese pears (*P. ussuriensis*), Japanase pears (*P. pyrifolia*), and European pears (*P. communis*) (Jin & Sato, 2003). However, this is the first study that shows the antimicrobial activities of ethyl acetate extract of fruits of *Pyrus communis* subsp. *communis* and endemic *Pyrus serikensis*, *Crataegus tanacetifolia*, and *Crataegus x bornmulleri*.

Table 2. Antimicrobial activity of *Crataegus bornmülleri*, *C. tanacetifolia*, *Pyrus serikensis*, and *P. C.* Subsp. *communis* ethyl acetate extracts against the bacterial, yeast, and fungal strains tested based on agar well-diffusion method.

	Inhibiton zone in diameter (mm/sensitive strains)				
Microorganism	*C. tanacetifolia*	*C. x bornmülleri*	*Pyrus serikensis*	*P. c.* subsp. *communis*	Standard antibiotic
B. cereus	12	12	13	10	30
B. subtilis	7	9	9	8	40
S. typhimurium	—	10	15	—	38
M. luteus	12	12	10	10	50
P. vulgaris	9	12	13	8	31
E. coli	9	10	12	8	37
P. aeroginosa	12	11	14	8	23
P. maltophila	9	9	12	8	38
S. aureus	9	9	11	9	35
E. aeroginosa	12	10	12	12	36
Y. enterocolitica	12	—	12	12	33
Brucella spp.	10	10	15	—	25
S. feacalis	10	12	10	—	35
A. hydrophila	8	9	10	12	30
L. monocytogenes	14	18	19	10	30
K. pneumoniae	12	12	17	7	30
P. fluorescens	—	—	8	—	32
B. pumilus	10	12	13	9	25
P. s. pv. tomato	14	17	20	14	35
P. tobacco	11	13	17	—	38
X. campestris pv. campestris	12	14	15	8	43
X. campestris pv. vesicatoria	10	12	15	8	42
P. lachrymans	12	14	14	—	31
P. syringae	13	14	18	1	36
P. s. pv. glycine	14	15	19	8	33
P. s. pv. phaseolicola	12	14	17	—	38
P. gingeri	—	9	9	—	35
P. gladioli pv. agricola	10	16	16	8	28
C. albicans	13	14	14	10	34
C. globrata	12	14	10	10	34
R. rubra	13	13	15	11	8
G. graminis	—	—	10	—	25
F. oxysporum	—	—	15	—	20
F. moniliforme	—	—	15	—	25
C. herbarum	—	—	15	—	30
P. notatum	10	—	10	—	24
A. parasiticus	—	—	—	—	40
A. niger	—	—	—	—	20
A. citri	—	—	—	—	15
A. fumigatus	—	—	—	—	13

Vierling et al. (2003) investigated the pharmaceutical and pharmacological equivalence of different Hawthorne extracts. They concluded that all extracts had a relaxant effect on aorta, although aqueous-alcoholic extracts were characterized by similar procyanidin, flavonoid, total vitexin, and total phenols content, and aqueous extract showed lower concentration of procyanidins, flavonoids, and total phenols but a similar total vitexin content.

Škerget et al. (2005) determined phenols, proanthocyanidins, quercetin, and apigenin in extracts of hawthorne (*C. laevigata*) and concluded that these groups were responsible for the antioxidative activity.

Bilia et al. (1994) isolated a new flavanol glycoside, isorhamnetin 5-*O*-β-D galactopyranoside from the aerial parts of *Pyrus bourgaeana*, but there is no record on the effect of this substance in pharmaceutical and pharmacological use.

Jin and Sato (2003) determined that aqueous extracts from succulent young shoots of the pear *Pyrus* spp. exhibited strong antibacterial activity against the bacterium *Erwinia amylovora* bv. 4. The substance was identified as benzoquinone (2,5-cyclohexadiene-1,4-dione) by NMR spectra, mass spectra, and HPLC analysis. However, there is no record on the chemical composition of fruits of *Pyrus* spp.

In conclusion, *C. tanacetifolia*, *C. bornmülleri*, *P. serikensis*, and *P. communis* subsp. *communis* collected from Turkey exhibited a broad range of antimicrobial activity to varying degrees. Particularly, *P. serikensis* extract showed significant antifungal activities and could be used as antimicrobial agent in new drug therapy. Investigations on the chemical composition of the ethyl acetate extracts of fruits of *Pyrus* spp. and *Crataegus* spp. should be carried out in further studies to reveal their antimicrobial effect.

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References

Baytop T (1984): *Treatment with Plants in Turkey*. Publication No. 3255. İstanbul, İstanbul University.

Bahorun T, Trotin F (1994): Antioxidant activities of *Crataegus monogyna* extracts. *Planta Med 60*: 323–326.

Bilia AR, Gonzalez HM, Morelli I, Nieri E, Rubio ME (1994): Flavonol glycosides from *Pyrus bourgaeana*. *Phytochemistry 35*: 1378–1380.

Davis PH (1972): *Flora of Turkey and the East Aegean Islands*, Vol. 4. Edinburgh, Edinburgh University Press, pp. 135–136, 162–163.

Güner A, Duman H (1991): Critics on *Pyrus boissieriana* Buhse subsp. *crenulata* Browicz. *The Karaca Arboretum Magazine 2(4)*: 165–170.

Jin S, Sato N (2003): Benzoquinone, the substance essential for antibacterial activity in aqueous extracts from succulent young shoots of the pear *Pyrus* spp. *Phytochemistry 62*: 101–107.

Özcan M, Haciseferoğullari H, Marakoğlu T, Arslan D (2005): Hawthorne (*Crataegus* spp.) fruit: Some physical and chemical properties. *J Food Eng 69*: 409–413.

Rakotoarison DA, Greisser B (1997): Antioxidant activities of phenolic extracts from flowers, in vitro callus and cell suspension cultures of *Crataegus monogyna*. *Pharmazie 52*: 60–64.

Rojas R, Bustamante B, Bauer J, Fernández I, Albán J, Lock O (2003): Antimicrobial activity of selected Peruvian medicinal plants. *J Ethnopharmacol 88*: 199–204.

Schussler M, Holzl J (1995): Myocardial effects of flavanoids from *Crataegus* species. *Arzneimittel-Forsch 45*: 842–845.

Škerget M, Kotnik P, Hadolin M, Hraš AR, Simonič M, Knez Ž (2005): Phenols, proanthocyanidins, flavones and flavonols in some plant materials and their antioxidant activities. *Food Chem 89*: 191–198.

Veveris M, Koch E, Chatterjee SS (2004): *Crataegus* special extract WS® 1442 improves cardiac function and reduces infarct size in a rat model of prolonged coronary ischemia and reperfusion. *Life Sci 74*: 1945–1955.

Vierling W, Brand N, Gaedcke F, Sensh KH, Schneider E, Scholz M (2003): Investigation of the pharmaceutical and pharmacological equivalence of different Hawthorn extracts. *Phytomedicine 10*: 8–16.