

Antimicrobial Activity of Extracts of some Plants Collected from the Kingdom of Saudi Arabia

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Abstract. The antimicrobial activity of twenty wild plants growing in the western regions of the Kingdom of Saudi Arabia was tested. The methanolic extracts from the aerial parts was determined using the agar diffusion method. Eight microorganisms were used in this study, namely, *Escherichia coli*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, *Sarcina lutea*, *Bacillus subtilis*, *Mycobacterium phlei* and *Candida albicans*. The results revealed that thirteen extracts exhibited a significant broad-spectrum antibacterial activity against both Gram-positive and Gram-negative bacteria. On the other hand, seven extracts showed only a narrow spectrum activity against Gram-positive bacteria. All the tested extracts with the exception of one plant showed a significant anti-mycobacterium effect, while they have variable antifungal effects.

Keywords: Antimicrobial activity, Antibacterial, Antifungal, Wild plants.

Introduction

Over the past decade, herbal medicine has become a topic of augmented global importance, having impacts on both world health and international trade. In terms of world health, traditional medicinal plants continue to play a central role in the healthcare systems of large proportions of the

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world's population^[1]. This is particularly true in developing countries, where traditional systems of medicine have a long and uninterrupted history of use. Recognition and development of the medicinal and economic benefits of traditional medicinal plants is on the increase in both developing and industrialized countries, although it varies greatly from region to region^[2]. Since their discovery, antimicrobial drugs have proved remarkably effective for the control of bacterial infections. However, it was soon evident that bacterial pathogens were unlikely to surrender unconditionally, because some pathogens rapidly become resistant to many of the first discovered effective drugs^[3]. The antimicrobial compounds from plants may inhibit bacterial growth by different mechanisms than those presently used as antimicrobials and may have a significant clinical value in the treatment of resistant microbial strains^[4].

Materials and Methods

Plant Materials

Twenty plant species were collected from Jeddah-Al-Taif Road, Al-Hadda and Al-Taif district. The collected plants were identified by Dr. Abdulaziz Fayed, Professor of Plant Taxonomy, College of Science, Assuit University, Egypt. Plants were air-dried at room temperature, powdered and stored in dark colored bottles.

Preparation of Crude Extracts

Fifty grams of each plant material was extracted by reflux twice with methanol (150 ml) for 15 min followed by evaporation of combined extracts using rotary evaporator under vacuum. The residue of each extract was kept in refrigerator until use.

Test Organisms

Seven strains of bacteria and one yeast were used in all antimicrobial screening. The microorganisms were: *Escherichia coli* (ATCC10536), *Proteus vulgaris* (NCTC4175), *Pseudomonas aeruginosa* (CNCMA21), *Staphylococcus aureus* (ATCC4175), *Sarcina lutea* (laboratory collection strain), *Bacillus subtilis* (NCTC6633), *Mycobacterium phlei* (laboratory collection strain) and *Candida albicans* (ATCC60193).

Agar-Diffusion Method

The agar-diffusion method was used^[5] to assess the antimicrobial activities of the methanolic extracts of the plants against representatives of acid fast bacilli (*M. phlei*), gram-positive bacteria (*B. subtilis*, *S. aureus* and *Sarcina lutea*), Gram-negative bacteria (*E. coli*, *Proteus vulgaris* and *P. aeruginosa*) and yeast (*C. albicans*). Applying the agar diffusion method, cups (0.5 cm in diameter) were made using No. 3 cork borer using trypticase soy agar (Difco). Extracts were dissolved in dimethylsulfoxide (DMSO) at concentration of 100 mg/mL then 50 μ l (containing 5 mg of the extract under test) were aseptically added to the cups (5 mg/cup). Plates were incubated inverted at 37°C for 24-48 hr. After incubation, the inhibition zones were recorded in mm. Diameters less than 5 mm indicated no effect. Fifty microlitre of DMSO were used as a negative control, while ofoxacin (Ofx, Oxoid) and amphotericin B (Amp.B, Oxoid); 5 μ g/disc each were used as positive controls.

Results

Table 1 lists plant extracts and their activities against the various organisms. In the present investigation, extracts of 20 plants belonging to different families were screened, all of which showed activity against at least four of the test organisms. *Sarcina lutea* was the most inhibited microorganism by 12 of the tested plant extracts and weakly inhibited by 8 plant extracts. *S. aureus* was the second most inhibited microorganism with 10 plant extracts, while 10 others showed a weak activity. *Bacillus subtilis* was partially inhibited by 15 plant extracts and weakly inhibited by the others.

The extracts of *Achillea biebersteinii* Afan., *Argemone ochroleuca* Sweet, *Rumex nervosus* and *Vernonia schimperii* strongly inhibited the growth of *Mycobacterium phlei*, while 5 plant extracts had a moderate effect and three showed a weak effect, while eight plant extracts were inactive. Gram-negative bacteria were weakly inhibited by 14 of the plant extracts. *Candida albicans* was inhibited by 9 tested plant extracts and 6 showed a weak activity.

In general, the antibacterial activity of plant extracts appears to be more inhibitory to Gram-positive bacteria than Gram-negative bacteria. It should be remembered that penicillin and some of the other prominent

Table 1. Inhibition zones obtained from the tested plant extracts.

Plant name	Family	SN	Place of collection	1	2	3	4	5*	6	7*	8
<i>Achillea biebersteinii</i> Afan.	Asteraceae	AB1064	BTR	11	10	17	18	22	18	21	20
<i>Argemone ochroleuca</i> Sweet	Papaveraceae	AO1128	ED	11	12	18	18	22	18	25	22
<i>Maerua crassifolia</i>	Capparaceae	MC1034	HR	10	15	13	12	10	12	-	-
<i>Commicarpus grandiflorus</i>	Nyctaginaceae	CG1126	T	9	15	16	14	11	10	12	12
<i>Commicarpus plumbagineus</i>	Nyctaginaceae	CP1127	HR	-	-	-	10	9	12	10	10
<i>Crotalaria emarginella</i>	Fabaceae	CE1119	HR	-	-	-	12	15	15	12	-
<i>Ochradenus baccatus</i> Delile	Resedaceae	OB1131	T	10	12	20	19	16	16	18	18
<i>Rumex nervosus</i>	Polygonaceae	RN1129	HR	10	10	19	14	22	17	22	18
<i>Stachys</i> Sp. Aff. <i>Schimperi</i> Vatke	Lamiaceae	SS1095	BTR	11	14	18	21	21	20	17	22
<i>Echolium viride</i> (Forsk.)	Acanthaceae	EV1006	S, T	-	-	-	12	9	14	17	-
<i>Dipterygium glaucum</i> Decne.	Cruciferae	DG1037	J	12	10	12	12	11	12	-	-
<i>Jasminum grandiflorum</i> L.Subsp. <i>floribundum</i>	Oleaceae	JG1125	B	14	12	22	20	20	15	-	22
<i>Cleome ramosissima</i> Webb ex Parl	Capparaceae	CR1040	HR	9	9	12	12	13	15	-	13
<i>Lavandula pubescens</i> Decne	Lamiaceae	LP1089	HR	9	12	14	10	10	9	10	-
<i>Chenopodium schraderianum</i> Schult.	Chenopodiaceae	CS1049	B	-	-	-	9	18	15	-	14
<i>Fioria dictyocarpa</i> (Webb)	Malvaceae	FD1124	T	-	-	-	10	19	14	-	10
<i>Echium arabicum</i>	Boraginaceae	EA1045	B	-	-	-	9	15	14	-	12
<i>Pulicaria guesatii</i> Rawi	Astreaeae	PG1063	MR	8	12	10	9	23	16	18	15
<i>Plectranthus asirensis</i> J.R.I	Lamiaceae	PA1093	B	12	11	18	16	20	16	14	19
<i>Vermonia schimperii</i>	Asteraceae	VS1054	HR	11	10	10	16	10	19	22	10

1, *E. coli*; 2 *Pseudomonas aeruginosa*; 3, *Proteus vulgaris*; 4, *Staphylococcus aureus*; 5, *Sarcina lutea*; 6, *Bacillus subtilis*; 7, *Mycobacterium phlei*; 8, *Candida albican*.

Al-Baha-Al Taif Road: BTR; Al-Hadda Road: HR; Al-Baha: B; Al-Sheffa: S; Al-Taif: T; ED: Ekrema dame; Jeddah: J; Jeddah-Al-Hadda Road: JHR;

Al-Maddina Road: MR; Specimen number: SN.

*Laboratory collection strains

antibiotic agents of fungal origin are also rather selective in their inhibitory action, most of them being inhibitory to Gram-positive bacteria. Unlike Gram-positive bacteria, the lipopolysaccharide layer along with proteins and phospholipids are the major components in the outer surface of Gram-negative bacteria^[6]. The outer lipopolysaccharide layer hinders access of most compounds to the peptidoglycan layer of the cell wall. This explains the resistance of Gram-negative strains to the lytic action of most extracts exhibiting activity. The negative results obtained against Gram-negative bacteria were not unexpected since this class of bacteria is usually more resistant than Gram-positive bacteria^[7]. The antimicrobial extracts of tested plants can be assumed to be useful to the producing plant in warding off infectious diseases and there is therefore a compelling reason to suppose that anti-infective agents could be active against human pathogens as was suggested by folkloric and historical accounts^[8].

Infections caused by *Mycobacterium phlei* are among the most difficult to treat with conventional antibiotics. The growth of *Mycobacterium phlei* was completely inhibited by four plants extracts and partially by five extracts. *Bacillus* species are common microbes found in most natural environments including soil, water, plant and animal tissues. While most *Bacillus* species are regarded as having a little pathogenic potential, both *Bacillus cereus* and *Bacillus subtilis* have been known to act as primary invaders or secondary infectious agents in a number of diseases and have been implicated in some cases of food poisoning^[9]. Figures 1-4 illustrated the results of some selected plant extracts, which have a significant antimicrobial activity.

Conclusion

Our results allow us to conclude that the crude extracts of *Achillea biebersteinii* Afan. *Ecbolium viride* (Forssk) and *Rumex nervosus* exhibited a significant antimicrobial activity and properties that support folkloric use in the treatment of some diseases as broad-spectrum antimicrobial agents^[10]. This probably explains the use of these plants by the indigenous people against a number of infections since generations^[11]. Researches have some reports on the chemical nature of the antimicrobial active ingredients of different species belonging to the same genera listed above, which could be helpful in future investigations targeting the identification of the chemical nature of the antimicrobial

active ingredients present in the most efficacious indigenous species used in this study.

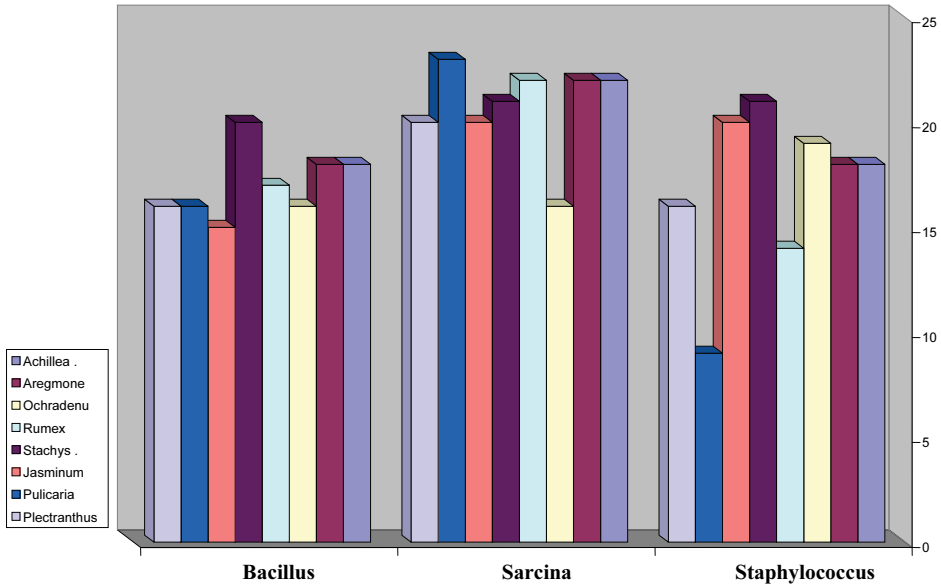


Fig. 1. Antimicrobial activity of plant extracts showing the highest activity against Gram-positive bacteria.

Antimicrobial activity against Gram negative bacteria

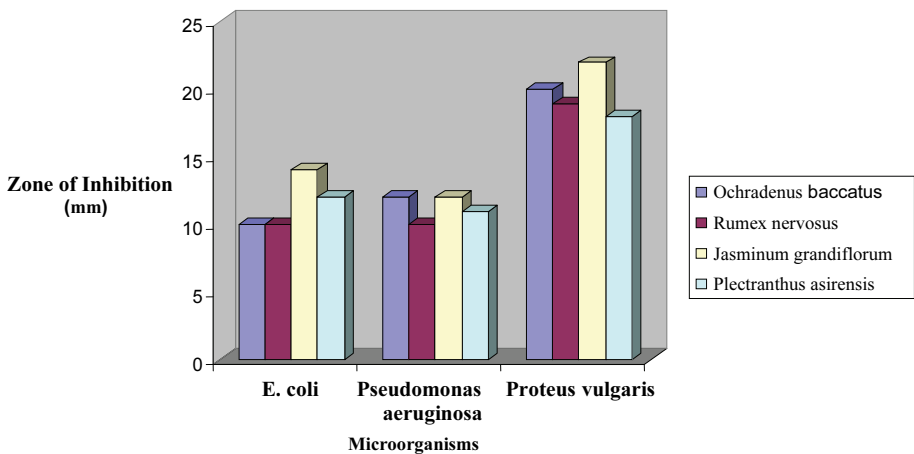


Fig. 2. Antimicrobial activity of plant extracts showing the highest activity against Gram-negative bacteria.

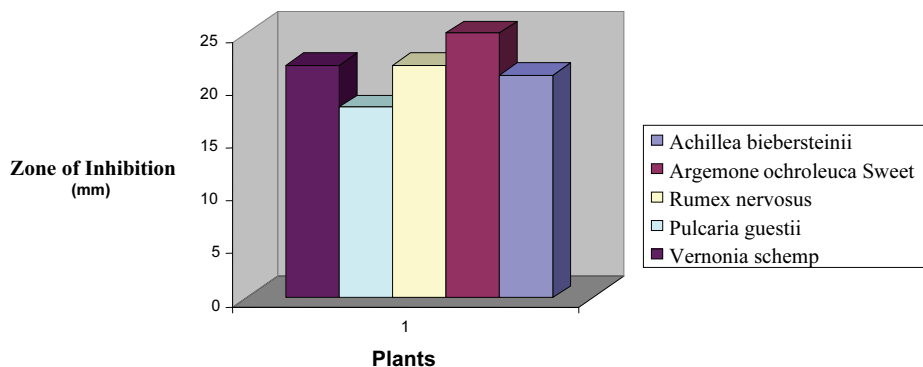


Fig. 3. Antimicrobial activity of plant extracts showing the highest activity against *Mycobacterium pheli*.

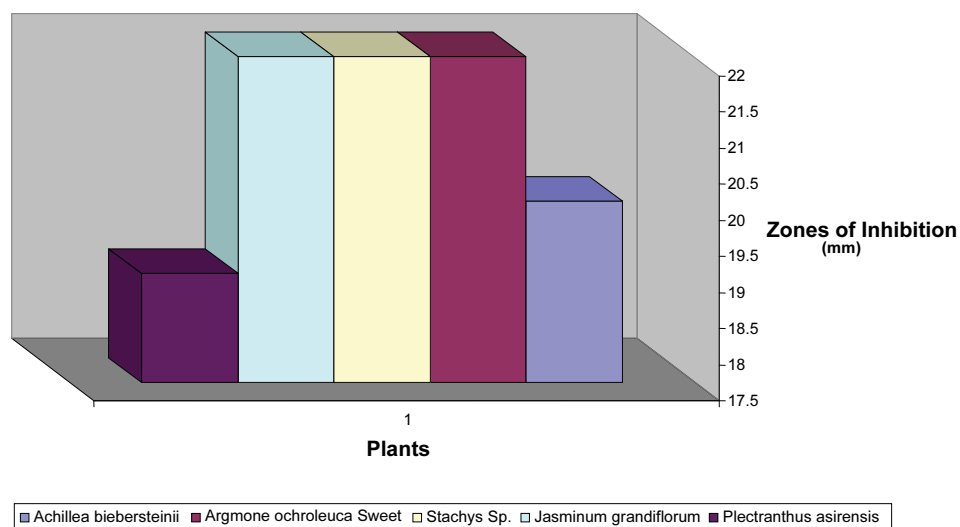


Fig. 4. Antifungal activity of plant extracts showing the highest activity against *Candida albicans*.

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References

- [1] **Akerele O.** Medicinal plants and primary health care: an agenda for action. *Fitoterapia* 1988; **59**: 355–363.
- [2] **Zhang X.** *Regulatory Situation of Herbal Medicines*. A Worldwide Review. WHO: Geneva, Switzerland, 1998. 1–5.
- [3] **Cowan MM.** Plant products as antimicrobial agents. *Clin Microbiol Rev* 1999; **12**(4): 564–582.
- [4] **Harborne JB.** *Phytochemical Methods*, 3rd ed. London: Chapman Hall, 1998. 1–302.
- [5] **Tomas-Barberan FA, Msonthi JD, Hostettmann K.** Antifungal epicuticular methylated flavonoids from three Spanish *Helichrysum* species. *Phytochemistry* 1988; **27**: 753–755.
- [6] **Kirtikar KR, Basu BD.** *Indian Medicinal Plants*, vols. I and II. Lalit Mohan Basu, Allahabad, India, 1968.
- [7] **Turnbull PCB, Kramer JM.** Bacillus. In: Barlows A, Hausler JrWJ, Herrmann HD, Isenberg H, Shadomy HJ (eds.). *Manuals of Clinical Microbiology*. 5th Ed. American Society for Microbiology, Washington DC, 1991.
- [8] **Alzoreky NS, Nakahara K.** Antibacterial activity of extracts from some edible plants commonly consumed in Asia. *Int J Food Microbiol* 2003; **80**(3): 223-230.
- [9] **Desta B.** Ethiopian traditional herbal drugs. Part II: Antimicrobial activity of 63 medicinal plants. *J Ethnopharmacol* 1993; **39**(2): 129-139.
- [10] **Stamatis G, Kyriazopoulos P, Golegou S, Basayiannis A, Skaltsas S, Skaltsa H.** *In vitro* anti-Helicobacter pylori activity of Greek herbal medicines. *J Ethnopharmacol* 2003; **88**(2-3): 175-179.
- [11] **Getie M, Gebre-Mariam T, Rietz R, Höhne C, Huschka C, Schmidtke M, Abate A, Neubert RH.** Evaluation of the anti-microbial and anti-inflammatory activities of the medicinal plants *Dodonaea viscosa*, *Rumex Nervosus* and *Rumex Abyssinicus*. *Fitoterapia* 2003; **74**(1-2): 139-143.

دراسة التأثير المضاد للميكروبات لخلاصات بعض النباتات التي تنمو بالمملكة العربية السعودية

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المستخلص . فى هذه الدراسة تم اختبار التأثير المضاد للبكتيريا للخلاصات الكحولية المحضرة من الأعشاب البرية، التي تنمو فى المنطقة الغربية من المملكة العربية السعودية. تم قياس هذا التأثير باستخدام طريقة الأجار المنتشر. تم استخدام ثمانية ميكروبات مختلفة فى هذه الدراسة، هى: ايشير شياكولاي، والبروتيس فالجارس، والسودومونس، والميكروب العنقودي المكور الذهبى، والميكروب الأحمر، والسرشيا لوتيا، والسبسكيز العسوية، والميكوبكتيريا فيلاى، وخميرة الكنديدا البيكانس. تم اختبار عشرين خلاصة نباتية، وقيست مناطق تثبيط النمو الميكروبي بالمليمتر. وقد أسفرت النتائج أن ثلاثة عشر من النباتات المختبرة، لها تأثير مضاد واسع المدى للبكتيريا الموجبة والسالبة الجرام. ومن ناحية أخرى أظهرت النتائج، أن الخلاصات النباتية السبعة المتبقية، لها تأثير مضاد ضيق المدى على البكتيريا الموجبة الجرام فقط. ولقد لوحظ أن تسعة عشر من النباتات المختبرة، لها تأثيرا إيجابيا على ميكروب الميكوبكتيريا، ولوحظ أيضا تأثيرات مختلفة لخميرة الكنديدا.