

The Antimicrobial Activity of Some Honey Bee Products and some Saudi Folkloric Plant Extracts

Nevine B. Ghanem

*Department of Food Nutrition, College of Agriculture and Nutrition,
King Faisal University, Al-Ahsa, Saudi Arabia
nevineghanem@yahoo.com*

Abstract. Alcoholic and aqueous extracts from 17 Saudi Arabia folkloric plants and some honey bee products were screened for antimicrobial activity against some tested pathogenic microorganisms by zone of inhibition assay and minimal inhibitory concentration (MIC). Compared to honey and bee wax, ethanolic extract of propolis showed the highest zone of inhibition (23mm) against *S. aureus* ATCC255923. Aqueous extract of Alum (Ming Fan) was highly effective against all tested strains with inhibition zones ranging from 25 to 29 mm followed by *Juglans regia* (28mm) with *S. epidermidis* ATCC12228, *Rhazya stricta* (24mm) with *Str. pyogenes* ATCC19615, and *Commiphora myrrha* (24mm) with *S. aureus* ATCC255923. The combined effect of equimixture of ethanolic extracts of propolis and Bee wax was more effective than a single extract showing 1.5 fold increase of inhibition zone against *S. aureus* ATCC255923 and *C. albicans* NCTC2708. Extracts with the most potent action against microorganisms were tested to determine their minimum inhibitory concentration (MIC). Alum (Ming Fan) was found to have the greatest activity with MIC mean value of 0.29% (w/v) followed by *Rhazya stricta*, *Juglans regia* and propolis with mean MIC values of 0.4, 2.66, and 3.75, respectively.

Keywords: Minimal inhibitory concentration (MIC), folkloric medicinal plants, propolis, honey, zone of inhibition assay, honey bee products.

Introduction

In developed countries, traditional, complementary and alternative medicine is becoming more popular. For example, the percentage of the

population that has used such medicine at least once is 48% in Australia, 31% in Belgium, 70% in Canada, 49% in France and 42% in the United States of America^[1]. In Saudi Arabia, different kinds of herbs are available; many species of these herbs are used directly in human food or as medicine, such as *Zingiber officinale*, *Thymus capitatus*, *Crocus stiva*, *Nigella sativa*, *Coriandrum sativum*, *Mentha piperita*, *Commiphora myrrha*, etc. In addition, honey products, natural gums and alum are known as folkloric medicines^[2, 3, 4, 5].

Honey is the natural sweet product produced by honey bees as they collect nectar or blossoms from the secretion of living parts of plants or excretions of plants, transform and combine with specific substances of their own to ripen and mature^[6]. One of the most important features of honey is that it can be kept for a long period of time without becoming spoiled^[7]. Honey is usually contaminated with numerous microorganisms^[8]. In honey, aerobic *Bacillus* as well anaerobic *Clostridium* spores and small fragments of moulds may appear^[9]. Osmophilic yeast such as *Saccharomyces*, *Schizosaccharomyces* and *Torula* predominate. This reservoir for microbes status however does not diminish the many important uses that honey is known for. In fact, the antimicrobial status of honey is giving it a continued place in the management of wounds and injuries^[10]. Honey was used to treat wounds as long ago as 2000 years before bacteria were discovered to be the cause of infection. More recently, honey has been reported to have an inhibitory effect to around 60 species of bacteria including aerobes and anaerobes, gram –positives and gram negative^[11].

Propolis is a resinous hive product collected by bees from tree buds and mixed with secreted bees wax. Bees use the propolis as a glue to seal the opening of the hives^[12]. Propolis known in folk medicine since ancient times, has attracted much attention in recent years as a useful ingredient applied in medicine and food products^[13]. It is known that the ethanolic extract of propolis exhibits various pharmacological activities such as antimicrobial, antiviral, antifungal and anti-inflammatory properties^[13, 14, 15].

Bee wax is also a product of bees, secreted from the wax gland of bee workers. It is a mixture of esters, fatty acids, higher alcohols and saturated hydrocarbons in addition to aromatic substances and

pigments^[16]. Zanoschi *et al.*, (1991) reported the use of bee wax together with other products for burns treatment.

Since ancient time herbs and oleo-gum resins such as gum myrrh were widely used in unprocessed form for fragrance and in folk medicines. They have been used in a number of medicinal contexts for a long time and still today in several countries across Europe, India, Africa, China and Middle East. Furthermore, they continued to find modern pharmacological applications most of them as claimed by traditional therapies^[18,19].

The objective of this study was to examine the antimicrobial activity of honey and some of bees products such as propolis and bee wax and some folkloric herbs, natural gum and alum against some pathogenic gram positive and gram negative bacteria as well as *Candida albicans*.

Materials and Methods

Test Microorganisms

The pathogenic standard strains used as test organisms were *Staphylococcus aureus* (ATCC25923), *Staphylococcus epidermidis* (ATCC12228), *Streptococcus pyogenes* (ATCC19615), *Escherichia coli* (ATCC25922), *Proteus mirabilis* (ATCC14153), *Salmonella typhimurium* (ATCC14028), *Pseudomonas aeruginosa* (ATCC27853), *Bacillus subtilis* (ATCC27853) and *Candida albicans* (NCTC2708). They were obtained from the Department of Microbiology, Faculty of Science, Alexandria University, Egypt. Bacterial strains were maintained on nutrient broth and nutrient agar media (Oxiod Ltd.), while *Candida albicans* was cultivated on Sabouraud agar medium provided with 2% (ml /v) glucose under aerobic conditions for 48 h at 37°C.

Sources of Tested Plants Products

Medicinal folk plants *Rhazya stricta*, *Nigella sativa*, *Sambucus nigra*, *Commiphora myrrh*, *Zingiber officinale*, *Zizyphus spina Christi*, *Ferula asafoetida*, *Eugenia caryophyllus*, *Cuminum cyminum*, *Juglans regia*, Arabic lubban, *Ricinus communis* *Nasturtium officinale* and Alum (Ming Fan) are listed in Table (1).

Table 1. List of Folkloric medicinal plants used.

Sample No.	Scientific name	Common name	Plant part	Folkloric utilization in traditional medicine
1	<i>Rhazya stricta</i>	Harmal	Fruit	as a reputed tonic and curative for rheumatic pain, sore throat, syphilis ,diabetes, helminthiasis, inflammatory conditions, fever and other diseases ^[20] .
2	<i>Nigella sativa</i>	Black Cumin	Seed	For the treatment of gastrointestinal and respiratory diseases ^[21] .
3	<i>Sambucus nigra</i>	Elder	Flower	as a remedy for inflammation caused by colds and fever ^[22] .
4	<i>Commiphora myrrha</i>	Myrrha	Gum resin from the bark of trees	to treat wounds, intestinal disorders, diarrhea, cough and chest ailments ^[23,24] .
5	<i>Zingiber officinale</i>	Ginger	Rhizome	as spice and to treat nausea ^[25] .
6	<i>Ziziphus spina-christi</i>	Sidr	Leaf	for the treatment of ulcers, wounds, eye diseases , bronchitis wounds, skin diseases and as an anti-inflammatory ^[26] .
7	<i>Ferula asafoetida</i>	Angedan gum	Gum	fighting flu ,natural antiviral bronchitis and even hysteria ^[27] .
8	<i>Eugenia caryophyllus</i>	cloves	Floral bud	in cooking , for dental pain, used to treat diarrhea, intestinal worms and other digestive ailments, had antimicrobial properties against fungi and bacteria ^[28] .
9	<i>Cuminum cyminum</i>	Cumin	fruit	seeds help with digestion ^[29] .
10	<i>Juglans regia</i>	walnut	Bark	as a tooth cleaner ^[30] .
11	<i>Boswellia sacra</i> (Frankincense)	Arabic lubbān	Gum	treat a variety of ailments including nausea, indigestion, chest coughs, hypertension, and post-childbirth recovery ^[31] .
12	Alum)	Ming Fan	-----	natural deodorant by inhibiting the growth of the bacteria ^[32] .
13	<i>Ricinus communis</i>	castor bean	Seed	treatment of warts, cold tumors, indurations of the abdominal organs, whitlows, lacteal tumors, indurations of the mammary gland ^[33] .
14	<i>Nasturtium officinale</i>	Water cress	Seed	acts as a stimulant, a source of phytochemicals ,antioxidants, a diuretic, an expectorant, , digestive aid., antiangiogenic cancer-suppressing properties ^[34] .

1- *Rhazya stricta*, 2- *Nigella sativa*, 3- *Sambucus nigra*, 4- *Commiphora myrrha*, 5- *Zingiber officinale*, 6- *Ziziphus spina –Christi*, 7- *Ferula asafoetida*, 8- *Eugenia caryophyllus*, 9- *Cuminum cyminum*, 10- *Juglans regia*, 11- Arabic lubbān, 12- Alum (Ming Fan), 13- *Ricinus communis*, 14- *Nasturtium officinale*.

They were purchased and collected from a well known traditional and folk herbal store in Al Ahsa city (Saudi Arabia). Folk plants were identified by a plant taxonomist at Life Science Department, College of Science, King Faisal University.

Preparation of Tested Extracts

Tested Folkloric materials were air-dried and ground into fine powder by a Braun Multi-Mill and passed through a sieve (24- mesh)^[35]. 5 g of finely ground-dried samples were extracted with adequate amount of water to a concentration of 12.5% (w/v) then mixed in a blender. The extracts were filtered through Whatman Filter paper No. 1 (Whatman Limited, England) to remove large particles, and the extracts were passed through a 0.2 µm filter at room temperature and stored at 4°C until used in microbial assay^[36].

Honey Samples

Honey samples collected from Al AHSA local market were stored in tightly closed glass containers wrapped in aluminum foil and kept at room temperature. The honey dilutions were prepared just before use to ensure that there was no loss of hydrogen peroxide. Sample of 10 g of honey was added to 10 ml distilled water and mixed to achieve 50% (w/v) solution^[37].

Propolis Samples

The propolis samples were kindly provided from a local apiary in Al Ahsa region, Saudi Arabia. Propolis specimens were further dehydrated with a low vacuum pump, and the extracts of dried propolis samples were prepared as described by Koo and Park (1997). The dried propolis samples were ground into a fine powder and 2g of the propolis powder were mixed with 25 ml of 80% ethanol in plastic centrifuge tube (LXG-50-C) and shaken at 70°C for 30 min. After extraction, the mixtures were centrifuged to obtain the supernatants, which were designated as ethanolic extracts of propolis (EEP).

Bee Wax

A sample of crude bee wax was thoroughly washed using distilled water, dried in open air, then broken down, and extracted with 70% ethanol for 48 h at 37°C using a shaker (150 rpm). The ethanol sample

was filtered through Whatman no.1 filter paper. The solvent was removed under reduced pressure at 40°C using rotary evaporator^[39, 40].

Microbiological Analysis of Honey, Propolis and Bee Wax

A volume of Seder honey sample was diluted with distilled water and stirred to achieve 50% (w/v) concentration. Serial decimal dilutions were prepared in duplicate, then 1 or 0.1 ml samples of appropriate dilutions were poured on agar plates. Mesophilic aerobic bacteria were counted on plate count agar (Merck) incubated for 48h at 35-37°C. Coli forms were enumerated after plating on Violet Red Bile (Merck) agar plates with a cover layer of the same medium and incubated for 20-24 h at 37°C. Yeast colonies were inoculated on Sabouraud agar medium (Merck) and incubated for 24-48 h at 30°C.

Determination of Antimicrobial Activity

Antimicrobial activity of honey products and herbs extracts was evaluated by agar diffusion method^[41]. 100 µl of diluted bacterial suspension (5×10^6 CFU/ ml) were spread onto the surface of plate count agar medium (PCA). Wells (0.6 mm in diameter) were cut from the agar with a sterile cork borer. Then 100 µl of honey products and/ or herbs extract were added to each well. Ethyl alcohol and water were used as negative control in all experiments. Plates were then incubated at 37°C for 24 h. Antimicrobial activity was evaluated by measuring the diameter of the clear inhibition zone [expressed in millimeters (mm)] formed around each tested substance. All tests were performed in triplicate and the mean of three readings was calculated and used in the analysis. Measurement of the antimicrobial activity of honey products mixture were tested, the measured diameter of inhibition zones developed were compared with the expected inhibition zones according to the following equation^[39].

(Measured inhibition zone of honey products mixture - Expected inhibition zone of honey products mixture).

The interaction between propolis and Bee wax mixture was then calculated by subtracting the expected value of inhibition zone from the measured one.

Determination of the Minimum Inhibitory Concentration (MIC)

Extracts which exhibited high activities against one or several pathogenic organisms were further assayed for their minimum inhibitory concentration (MIC). This was carried out by the two fold serial dilution of the tested extracts in nutrient broth or Sabouraud broth for *Candida albicans* (2 ml volumes), then inoculated with 100 µl inoculum size with the test organisms. The alcoholic and aqueous crude extracts were prepared at concentrations of 5, 2.5; 1.3; 0.6; 0.3 and 0.2% (w/v). The MIC was determined by the broth dilution method^[42]. Nutrient broth samples (10 ml) were inoculated with different concentrations of the crude extracts and with 100 µl of active inocula of microorganisms (approximately 10⁸ CFU/ml) for 24h at 37°C for bacteria and at 30°C for yeast. The MIC was determined as the lowest concentration of the extract which inhibited the organism^[36]. The results of antimicrobial activity were expressed as the mean obtained upon three independent analyses.

Results & Discussion

Freshly produced honey must lack pathogenic microorganisms. The aerobic mesophilic bacteria counts of Seder honey sample examined in this study was 220 ± 14.14 CFU/g (Data not shown). The absence of other microbial groups may be attributed to diluting honey with water that supports the growth of nonpathogenic bacterial strains and kills pathogenic strains. According to published data total aerobic viable count values for honeys can range from zero to several thousand per gram. This variation in bacterial counts may be due to the type of sample (raw, finished or retailed), the freshness of the honey, the time of harvest and the analytical techniques used^[43]. The presence of a certain type of bacteria indicates the contamination from secondary sources during manipulations and previous processes. The contamination with fungi and bacteria indicate inadequate hygiene conditions during collections, manipulation, processing and storage^[44]. On the other hand, samples of propolis and Bee wax were free from microbial growth, this may be due to the chemical composition of propolis which inhibited the presence of microbes^[45, 46].

Honey has been shown to be bactericidal to many pathogenic microorganisms. This is due to its osmotic properties, acidity mainly

from hydrogen peroxide and photochemical factors^[10, 47]. Unexpected data were recorded in this study using Seder honey which showed slight inhibitory antibacterial effect on *B. subtilis* and *P. mirabilis* (Table 2).

Table 2. Antimicrobial activity of Honey and Bees products extracts against some pathogens expressed as zone of inhibition (mm).

Microorganism	Bee products		
	Zone of Inhibition in (mm)		
	Honey Seder	Propolis	Bee Wax
<i>S. aureus</i> ATCC25923	0	23	7
<i>S. epidermis</i> ATCC12228	0	20	6.5
<i>B. subtilis</i> ATCC27853	1	14	7
<i>P. auruginosa</i> ATCC27853	0	6	4
<i>E. coli</i> ATCC25922	0	2	3
<i>Strept. pyogenes</i> ATCC19615	0	8	6.5
<i>S.typhimurium</i> ATCC14028	0	0	0
<i>P. mirabilis</i> ATCC14153	1	0	0
<i>C. albicans</i> NCTC2708	0	10	20

These results are in contrast with those recorded by numerous reports^[11, 48, 49, 50] dealt with the antimicrobial activities of honey. This negative effect may be attributed to heat processing of honey. Other workers have however shown a reduction in antibacterial activity of honey on dilution to four times^[10]. It is also important that honeys used as an antimicrobial agent must be stored at low temperature and not exposed to light, so that none of the glucose oxidase activity is lost although all honey will stop the growth of bacteria because of its high sugar content^[10].

The inhibitory effect of ethanolic extract of propolis (EEP) sample was pronounced on most tested microorganisms, it is worth noting that gram positive bacteria were generally more sensitive to EEP extract than gram negative. *Proteus mirabilis* ATCC14153 and *Salmonella typhimurium* ATCC14028 seemed to be the least inhibited by EEP extract compared to the other tested organisms (Table 2). The maximum inhibition zone (23 mm) was recorded against *Staphylococcus aureus*, followed by *Staphylococcus epidermidis* (20mm) and *Bacillus subtilis* (14 mm). It is concluded that EEP could be used as antibacterial agent^[31, 51, 52, 53].

Crude bee wax, unlike propolis is more known and is easier to obtain. It is well known in revealing therapy for gastric ulcer^[30]. Zanoschi *et al.* (1991) reported on the use of bee wax for the treatment of

burns. Bee wax sample used in this study was found effective against the studied gram positive and gram negative bacteria and showed pronounced inhibitory effect with *Candida albicans* NCTC2708 (20mm) as well (Table 2). This is not in agreement with data obtained by Hasanain (1997) who reported no in vitro inhibitory effect of bee wax against some pathogenic bacteria including *B. subtilis*. This may be due to the immiscibility of the wax with the agar medium.

The possible synergistic interactions existing between propolis and Bee wax provided useful antimicrobial activity. Zone diameters were then compared to those developed around control wells receiving single samples, one at a time. The combined effect of equimixtures of propolis and Bee wax revealed highest positive interaction (7) on *Candida albicans*, followed by *Staphylococcus aureus* (5), (Table 3). Moreover, the zone of inhibition recorded showed 1.5 fold increases than that found with propolis alone. This experiment confirms the possibility of synergistic as well as antagonistic interactions between natural mixtures of Bee products^[55].

Table 3. Antimicrobial activity of equimixture of propolis and Bees wax against some pathogens expressed as zone of inhibition (mm).

Tested organism	Propolis and Bee wax mixture		
	Zone of Inhibition in (mm)		
	Measured	Expected	Interaction
<i>S. aureus</i> ATCC25923	20	15	5
<i>S. epidermis</i> ATCC12228	14	13.5	0.5
<i>B. subtilis</i> ATCC27853	12	10.5	1.5
<i>C. albicans</i> NCTC270	22	15	7

Interaction :(measured inhibition zone of propolis and Bee wax - Expected inhibition zone of propolis and Bee wax)

The antimicrobial activities of some herbs and gum extracts were evaluated; the results indicated various degrees of growth inhibition on the test microorganisms. Crude extract of *Juglans regia*, and Alum exhibited inhibitory effects against almost all tested strains. In Table 4 extract of *Commiphora myrrha* showed a high degree of antibacterial activity against *Staphylococcus aureus* (24mm). *Commiphora* species have a considerable antimicrobial activity against some gram positive and gram negative bacteria as recently, reported^[24]. Moreover, the extracts of *Rhazya stricta* could inhibit *Streptococcus. pyogenes* ATCC19615 with zone of inhibition of 24 mm. Whereas, *Nigella sativa*, *Ziziphus spina*, *Ferula asafoetida*, and *Cuminum cyminum* inhibited the

growth of *Salmonella typhimurium* ATCC14028. On the other hand *Zingiber officinale* and *Nasturtium officinale* showed no inhibitory effect on all the tested microorganisms^[56].

Table 4. Antimicrobial activity of Folkloric Herbs extracts against some pathogens expressed as zone of inhibition (mm).

Microorganism	Folkloric Herbs													
	1	2	3	4	5	6	7	8	9	10	11	12	13	14
<i>S. aureus</i> ATCC25923	-	-	-	24	-	10	-	19	-	20	-	25	-	-
<i>S. epidermidis</i> ATCC12228	17	-	-	-	-	20	--	20	-	28	-	29	-	-
<i>B. subtilis</i> ATCC27853	-	-	19	15	-	-	-	-	-	-	-	28	-	-
<i>P. auruginosa</i> ATCC27853	-	-	-	-	-	-	-	-	-	17	-	26	-	-
<i>E. coli</i> ATCC25922	-	-	-	-	-	-	-	-	-	23	-	28	-	-
<i>Strept.pyogenes</i> ATCC19615	24	14	-	-	-	-	-	-	-	19	17	25	-	-
<i>S.typhimurium</i> ATCC14028	-	20	-	-	-	20	20	-	23	-	-	27	14	-
<i>P. mirabilis</i> ATCC14153	-	-	-	-	-	-	-	17	-	20	-	25	-	-
<i>C. albicans</i> NCTC2708	-	-	-	-	-	-	-	-	-	20	-	25	-	-

1- *Rhazya stricta*, 2- *Nigella sativa*, 3- *Sambucus nigra*, 4- *Commiphora myrrha*, 5- *Zingiber officinale*, 6- *Ziziphus spina –Christi*, 7- *Ferula asafoetida*, 8- *Eugenia caryophyllus*, 9- *Cuminum cuminum*, 10- *Juglans regia*, 11- *Arabic lubbān*, 12- *Alum (Ming Fan)*, 13- *Ricinus communis*, 14- *Nasturtium officinale*.

Table 5 shows the MIC of selected samples [(propolis, *Rhazya stricta*, *Juglans regia*, *Commiphora myrrha*, *Eugenia caryophyllus*, and *Alum (Ming Fan)*] extracts on the inhibition of eight test strains. A wide range of MIC values were recorded depending on the microbial strain. The ethanolic extract of propolis (EEP) showed MIC value of 2.5% (w/v) against *S. aureus* ATCC25923, and MIC of 5% (w/v) for *C. albicans* NCTC2708. So it can be useful for preventing candidal infections^[52]. *Rhazya stricta* is effective against both strains of *Staphylococcus epidermidis* ATCC12228 and *Streptococcus pyogenes* ATCC19615, with MIC (0.4%) (w/v). Similar result was obtained by Salamah *et al.* (1989). *Alum (Ming Fan)* extract showed a pronounced inhibitory activity against all tested strains with lower minimum inhibitory concentration of 0.3% (w/v) this may be due to the chemical composition which it composed of hydrated potassium aluminum sulfate {KAl(SO₄)₂·12(H₂O)}.

Table 5. Minimum inhibitory concentration (MIC) of EEP extract and some aqueous herb extracts.

Microorganisms	EEP and herb extracts MIC					
	1	2	3	4	5	6
<i>S. aureus</i> ATCC25923	2.5	--	5	2.5	2.5	0.3
<i>S. epidermis</i> ATCC12228	--	0.4	0.2	--	--	0.2
<i>P. auruginosa</i> ATCC27853	--	--	0.6	--	--	0.3
<i>E. coli</i> ATCC25922	--	--	--	--	--	0.3
<i>Strept. Pyogenes</i> ATCC19615	--	0.4	2.5	--	--	0.3
<i>S.typhimurium</i> ATCC14028	--	--	--	--	--	0.3
<i>P. mirabilis</i> ATCC14153	--	--	--	--	--	0.3
<i>C. albicans</i> NCTC2708	5	--	5	--	--	0.3
Overall mean MIC	3.75	0.4	2.66	2.5	2.5	0.29

1- Propolis, 2- *Rhazya stricta*, 3- *Juglans regia*, 4- *Commiphora myrrha*, 5- *Eugenia caryophyllus*, 6- Alum (Ming Fan).

Potassium alum is an astringent and antiseptic^[57]. For this reason, it can be used as a natural deodorant by inhibiting the growth of the bacteria responsible for body odor.

The samples extracts studied had different compounds and formulation. It is hoped that this study would lead to the establishment of some new and more potent antimicrobial drugs from natural origin. Therefore further studies are recommended in order to develop more effective treatments of combination of two or more antimicrobial drugs.

Acknowledgment

This research (2464) was funded by the Deanship of Scientific Research, College of Agriculture and Food Science, King Faisal University, Al- Ahsa. Saudi Arabia.

References

- [1] WHO, (2002) Traditional medicine. Report, EB111/9, World Health Organization, Geneva.
- [2] Abed, K.F. (2007) Antimicrobial activity of essential oils of some medicinal plants from Saudi Arabia. *Saudi J. Bio. Sci.*, **14**(1): 53- 60.
- [3] Adzu, B., Amos, S., Wambebe, C. and Gamaniel, K. (2001) Antinociceptive activity of *Zizyphus spina-christi* root bark extract. *Fitoterapia.*, **4**: 72, 344-350.
- [4] Aqel, M.B. (1993) Effects of *Nigella sativa* seeds on intestinal smooth muscle. *Int. J. Pharmacogn.*, **31**: 55-60.
- [5] Brown, D. (1995) Encyclopedia of Herbs and their uses. Dorling Kindersley London, ISBN0-7513-020-31.
- [6] Codex, A. (1989) Limentarius Cimmission Codex Standards for sugar (honey), Supplement 2 to codex A Limentarius volume 111, Food and Agriculture Organization of the United Nations and World Health Organization, Rome.

- [7] **White, J.A.** (1975) Composition of honey. In: E Crane (ed.) Honey – a Comprehensive Survey. Heinemann, London. pp: 157-206.
- [8] **Migdal, W., Owczarczyk, H.B., Kedzia, B., Holderna-Kedzia, E. and Madajczk, D.** (2000) Microbiological decontamination of natural honey by irradiation. *Radiat. Phys. Chem.*, **57**: 285-288.
- [9] **Cheorum, J.A., Kim, J.K., Kang, J.K., Lee, E.Y. and Byun, M.W.** (2005) Irradiation effects on the Decontamination of Microorganisms in Honey. *International Symposium "New Frontier of Irradiated food and Non-Food Products, Bangkok, Thailand.*
- [10] **Olaitan, H.B., Adeleke, O.E. and Ola, I.O.** (2007) Honey: a reservoir for microorganisms and an inhibitory agent for microbes. *Afri. Health Sci.*, **7**(3): 159-165.
- [11] **Molan, P.C.** (1992) The antibacterial activity of honey 1. The nature of the antibacterial activity. *Bee World*, **73**(1): 5-28.
- [12] **Lu, L.C., Chen, Y.W. and Chou, C.C.** (2005) Antibacterial activity of propolis against *Staphylococcus aureus*. *Inter. J. Food Micro.* **102**: 213-220.
- [13] **Burdock, G.A.** (1998) Review of the biological properties and toxicity of bee propolis (propolis). *Food Chem. Toxicol.* **36**: 347-363.
- [14] **Park, Y.K., Koo, M.H., Abreu, J.A., Ikegaki, M., Cury, J.A. and Rosalen, P.N.** (1998) Antimicrobial activity of propolis on oral microorganisms. *Current Microbiology*, **36**: 24-28.
- [15] **Serkedjieva, J., Manolova, N. and Bankova, V.** (1992) Anti- influenza virus effect of some propolis constituents and their analogues (esters of substituted cinnamic acids). *J. Nat. Prod.*, **55**: 294-297.
- [16] **Carbajal, D., Molina V., Valdes, S., Arruzazabala, L., Rodeiro, I. and Mas, R.** (1995) Anti-ulcer activity of higher primary alcohols of beeswax. *J. Pharm. Pharmacol.*, **47**: 731-733.
- [17] **Zanoschi, C., Ciobanu, C., Verbuta, A. and Frincu, D.** (1991) The efficiency of some natural drugs in the treatment of burns. *Revista Medico – Chirurgicala a Societatii de Medici Si Naturalisti Din Lasi.*, **95**: 63-65.
- [18] **Massoud, A., Sisi, S.E., Salama, O. and Massoud, A.** (2001) Preliminary study of therapeutic efficacy of a new fasciolocidal drug derived from *Commiphora molmol (myrrh)*. *Am. J. Trop. Med. Hyg.*, **65**(2): 96-99.
- [19] **Muniruzzanan, M. and Chowdruy, M.B.R.** (2004) Sensitivity of fish pathogenic bacteria to various medicinal herbs. *Bangl. J. Vet. Med.*, **2**(1): 75-82.
- [20] **Baeshen, N.A., Lari, S.A., Aldoghaither, H.A. and Elkady A.I.** (2010) Biochemical evaluation of the effect of *Rhazya stricta* aqueous Leaves extract in liver and kidney functions in Rats. *Nature and science.*, **8**(4): 136-142.
- [21] **El-Tahir, K.E.H., Ashour, M.M.S. and Al-Harbi, M.M.** (1993) The respiratory effects of the volatile oil of the black seed (*Nigella sativa*) in guinea pigs: elucidation of the mechanism(s) of action. *Gen Pharmacol.*, **24**: 1115-1122.
- [22] **Zichria, Z.R., Noemi V., Moshe, Z., Orly, M., Liora R., Miriam, S. and Madeleine, M.** (2007) Inhibition of Several Strains of Influenza Virus in Vitro and Reduction of Symptoms by an Elderberry Extract (*Sambucus nigra* L.) during an Outbreak of Influenza B Panama, *The Journal of Alternative and Complementary Medicine*, **1** (4): 27.
- [23] **Ghazanfar S.A.** (1994) *Handbook of Arabian Medicinal Plants*. CRC Press Inc., Florida, USA.
- [24] **Rahman, M.M., Garvey, M., Piddock, L.J. and Gibbons S.** (2008) Antibacterial terpenes from the oleo-resin of *Commiphora molmol* (Engl.). *Phytother Res.* **10**: 1356-60.
- [25] **Arfeen Z., Owen H., Plummer J.L., Ilsley A.H., Sorby-Adams R.A. and Doecke C.J.** (1995) A double-blind randomized controlled trial of ginger for the prevention of postoperative nausea and vomiting. *Anaesth Intensive Care.*, **23**(4): 449-452.
- [26] **Abalaka M.E., Daniyan S.Y. and Mann A.** (2010) Evaluation of the antimicrobial activities of two *Ziziphus* species (*Ziziphus mauritiana* L. and *Ziziphus spina christi* L.) on some microbial pathogens. *African J. Pharm. and Pharmacol.*, **4**(4): 135-139.

- [27] **Lust, J.** (1983) The Herb Book. Bantam books ISBN 0-553-23827-2.
- [28] **Duke, J., ABogenschutz-Godwin, M.J. et al.** (2002) Cloves (*Syzygium aromaticum* (L.) Merr. & L. M. Perry). "In: Handbook of Medicinal Herbs, 2nd ed., 201-202, CRC Press, Boca Raton, FL.
- [29] **Shana, B., Caia, Y.Z., Brooks, J.D. and Corke H.** (2007) The in vitro antibacterial activity of dietary spice and medicinal herb extracts. *Int. J. Food. Microbiol.*, **117**(1): 112-119.
- [30] **Bensky, D., Clavey, S., Stoger, E. and Andrew, G.** (2004) Chinese Herbal Medicine: Materia Medica, Third Edition.
- [31] **Gupta, I., Gupta, V., Parihar, A., et al.** (1998) Effects of *Boswellia serrata* gum resin in patients with bronchial asthma: results of a double-blind, placebo-controlled, 6-week clinical study. *Eur J Med Res*; **3**: 511-514.
- [32] **Gallego, H., Lewis, E.J. and Crutchfield, C.E.** (1999) "Crystal deodorant dermatitis: irritant dermatitis to alum-containing deodorant". *Cutis*, **64**(1): 65-6.
- [33] **Oyewole, O.L., Owoseniand, A.A. and Faboro, E.O.** (2010) Studies on medicinal and toxicological properties of *Cajanus cajan*, *Ricinus communis* and *Thymus vulgaris* leaf extracts. *Journal of Medicinal Plants Research*, **4**(19): 204-208.
- [34] **Hecht, S.S., Chung, F.L., Richie, J.P., Akerkar, S.A., Borukhova, A., Skowronski, L. and Carmella, S.G.** (1995) "Effects of watercress consumption on metabolism of a tobacco-specific lung carcinogen in smokers". *Cancer Epidemiol Biomarkers Prev.*, **4**(8): 877-84.
- [35] **Lee, C.F., Han, C.K. and Tsau, J.L.** (2004) In vitro inhibitory activity of Chinese leek extract against *Campylobacter* species. *Int. J. Food Microbiol.*, **94**: 169-74.
- [36] **Chehregani, A., Azimishad, F. and Alizade, H.J.** (2007) Study on antibacterial effect of some *Allium* species from Hamedan – Iran. *Int. J. Agri. Biol.*, **9**(6): 873-876.
- [37] **AL Somai, N., Coley, K.E., Molan, P.C. and Hancock, B.M.** (1994) Susceptibility of *Helicobacter pylori* to the antibacterial activity of Manuka Honey. *Russ. Med. J.*, **87**: 9-12.
- [38] **Koo, H.M. and Park, Y.K.** (1997) Investigation of flavonoid a glycones in propolis collected by two different varieties of bees in same region. *Biosci. Biotechnol. Biochem.*, **61**: 367-369.
- [39] **Amoros, M., Simoes, C.M.O., Girre, L., Sauvager, F. and Cormier, M.** (1992) Synergistic effect of flavones and flavonols against herpes simplex virus type 1 in cell culture. Comparison with the antiviral activity of propolis. *J. Nat. Prod.*, **55**: 1732-1740.
- [40] **Grange, J.M. and Davery, R.W.** (1990) Antibacterial properties (bee glue) *J. R. Soc. Med.*, **83**: 159-160.
- [41] **Kim, J., Marshall, M.R. and Wei, C.** (1995) Antibacterial activity of some essential oil components against five food borne pathogens. *J. Agric. Food Chem.*, **43**: 2839- 45.
- [42] **Davidson, P.M. and Parish, M.E.** (1989) Methods for testing the efficacy of food antimicrobials. *Food Technol.*, **52**: 148-154.
- [43] **Snowdon, J.A. and Cliver, D.O.** (1996) Microorganisms in honey. *Int. J. Food Microbiol.*, **31**: 1-26.
- [44] **Tchoumboue, J., Julius A.N., Florence, F.A., Dongock, N.D., Jonnas, P. and Ze Antoine, M.** (2007) Physico-chemical and microbiological characteristics of honey from the sudanoguinian zone of West Cameroon, *Afri. J. .Biotech.*, **6**(7): 908-913.
- [45] **Souza, R.M., de Souza, M.C., Patitucci, M.L., Silva, J.F.** (2007) Evaluation of antioxidant and antimicrobial activities and characterization of bioactive components of two Brazilian propolis samples using a pKa-guided fractionation. *Z Naturforsch [C]*, **62**: 801-807.
- [46] **Teixeira, E.W., Message, D., Negri, G., Salatino, A., Stringheta, P.C.** (2010) Seasonal variation, chemical composition and antioxidant activity of Brazilian propolis samples. *Evid Based Complement Alternat Med. ecam.*, **7**(3): 307-315.
- [47] **Jeddar, A., Kharsnay A., Ramsaroop U.G., Bhamjee A., Hafejee I.E. and Moosa A.** (1985) The Antibacterial action of honey. *South Afri. Med. J.*, **67**: 257-258.

- [48] **Radwan, S.S., El-Essawy, A.A. and Sarhan, M.M.** (1984) Experimental evidence for the occurrence in honey of specific substances active against microorganisms. *Zentralbl. Mikrobiol.*, **1139**: 249-455.
- [49] **Ibrahim, A.S.** (1985) Antibacterial action of honey. *Bull Islam Med.*, **1**: 363-365.
- [50] **Brady, N.F., Molan, P.C., Harfoot, C.G.** (1997) The sensitivity of dermatophytes to the antimicrobial activity of manuka honey and other honey. *Pharm. Sci.*, **2**: 1-3.
- [51] **Marcucci, M.C.** (1995) Propolis: chemical composition, biological properties and the therapeutic activity. *Apidologie*, **26**: 83-99.
- [52] **Özan, F., Sümer, Z., Polat, Z.A., Er, K., Özan, Ü. and Değer, O.** (2007) Effect of mouthrinse containing propolis on oral microorganisms and human gingival fibroblasts. *Eur. J. Dent.*, **1**: 195-201.
- [53] **Alghalibi, S.M.S.** (2009) Effect of propolis ethanolic extract on phytopathogenic fungi, dermatophytes and Bacteria. *Sixth international conference of the Arab Federation of Beekeepers.*, Abha. Saudi Arabia.
- [54] **Hasanain, F.M.** (1997) Evaluation of antibacterial activity of some bee products against certain phytopathogenic bacteria and fungi. *Alex J. Agric. Res.*, **42**: 239-250.
- [55] **Singh, G. and Kachroo, P.** (1976) *Forest Flora of Srinagar*. Bishen Singh Mahendra Pal Singh.
- [56] **Salamah, A.A., Hassan, H.M. and Nassar, T.M.** (1989) Antibacterial Wild Flowering Plants in Saudi Arabia. *J. King Saud Univ. Science*, **1**(1, 2): 5-19.
- [57] Encyclopedia Britannica., Web. Apr, 19, (2011). From <http://www.britannica.com/EBchecked/topic/17885/alum>

دراسة عن النشاط المضاد للنمو الميكروبي لمنتجات عسل النحل وبعض مستخلصات النباتات الفلكلورية السعودية

نيفين بهاء الدين غانم

قسم علوم الغذاء والتغذية، كلية العلوم الزراعية والأغذية
جامعة الملك فيصل، الأحساء - المملكة العربية السعودية
nevineghanem@yahoo.com

المستخلص. تم استخدام مستخلصات مائية وأخرى كحولية مستخلصة من ١٧ من النباتات الفلكلورية المستخدمة في الطب الشعبي في المملكة العربية السعودية، ويتم استخدامها كمضادات ضد الكائنات الدقيقة المسببة للأمراض. تم ذلك باختبار فحص منطقة التثبيط وتعيين التركيز الأدنى المثبط لنمو الميكروبات المستخدمة. أظهر المستخلص الكحولي من البروبوليز بالمقارنة مع شمع العسل وعسل النحل (الصدر) أعلى منطقة تثبيط (٢٣mm) ضد البكتريا المكورة العنقودية الذهبية *Staphylococcus aureus* ATCC 255923. بينما أدى استخدام المستخلص المائي للشبة فعالية عالية ضد جميع السلالات الميكروبية المختبرة، وكانت مناطق التثبيط الميكروبي تتراوح ما بين ٢٥ حتى ٢٩ مم، يليها *Juglans regia* stricta Rhazya (٢٨mm) مع *S. epidermidis* ATCC12228، ثم *Commiphora* (٢٤mm) مع *Strep. pyogenes* ATCC19615 يليها *S. aureus* ATCC255923. كما أظهر استخدام خليط من مستخلصات الايثانول للبروبوليز وشمع النحل فعالية أكثر من استخدام كل منهما على حده بزيادة ١,٥ ضعف في منطقة التثبيط ضد بكتريا *S. aureus* ATCC255923 وخميرة NCTC2708

C. albicans. تم اختبار المواد الأكثر تثبيطاً وفعالية ضد الكائنات الدقيقة المستخدمة لتحديد أدنى تركيزها مثبط للنمو. وجد أن قيمة المتوسط العام للشبة المثبطة للنمو هي ٠,٢٩ (w/v)، يليها *Rhazya stricta* والبروبوليز بقيمة (٣,٧٥، ٢,٦٦، ٠,٤) على التوالي.

الكلمات الدالة: التركيز الأدنى المثبط، النباتات الطبية الفولكلورية، بروبوليز، عسل، فحص منطقة التثبيط، منتجات نحل العسل.