

Antibacterial and Antioxidant Activities of Different Varieties of Locally Produced Egyptian Honey

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THE PRESENT study investigated the physicochemical, antioxidant, and microbiological properties of six types of honey obtained from Alexandria, Egypt. Physicochemical properties showed that optical density in the tested honey samples ranged from 0.50 to 1.65, while the pH was in the acidic range. Moisture contents and electrical conductivity in honey samples ranged from 0.5 to 0.64, and 0.29-0.79 mScm⁻¹, respectively. Total phenol varied from 5500 and 14120 mg/kg in the samples. Flavonoid contents were between 926-1657 mg/kg. The greater amount of phenolic and flavonoid compounds leads to more potent radical scavenging effect as shown by Cotton, Eucalyptus, and Black seed honey samples. Microbiological characteristics showed that Black seed followed by Eucalyptus honey extracts gave the maximum antibacterial activity against *K. pneumonia*, *P. aeruginosa*, *P. mirabilis*, *E. coli* and *S. mutans* with maximum inhibition zones equal to 42.67, 39.00, 36.00, 27.33 and 18.67 mm at 100 µl, respectively, for Black seed honey, and 41, 38.33 and 35.67 mm and approximately equal inhibition zones of 13.67 mm against *E. coli* and *S. mutans* at 100 µl, respectively, for Eucalyptus honey. The minimum inhibitory concentration values against the pathogenic bacteria showed that, at 25% concentration, Black seed and Eucalyptus honey had an effect on *E. coli* and *K. pneumonia*, while Clover honey had an effect on *P. aeruginosa* and *P. mirabilis*. Thus, honey can be used for nutritional, medicinal and industrial purposes and can be an important commodity in the international market.

Keywords :Antibacterial, Antioxidant, Local natural honey, Pathogenic bacteria, Egyptian Honey

Introduction

Honey is a natural sweetener, which is widely available across the world. It is extensively used among various natural products, for various applications, some are clinical and contains approximately 200 distinct chemical compounds. Several research studies have studied many of its biological properties, such as antioxidant, anti-inflammatory, anti-bacterial, antiviral, anti-ulcer activities; and antihyperlipidemic, antidiabetic and anticancer properties (Rao et al., 2016).

It is a supersaturated solution of sugars, composed of mainly fructose and glucose, and many other minor components such as minerals, proteins, free amino acids, vitamins, enzymes-glucose-oxidase, and catalase-, phenolic acids, and flavonoids honey's composition is variable

and its properties vary greatly depending on floral source, climate, and environmental and processing conditions (Ramos et al., 2018)

The possible health benefits of consuming honey have been subjected to extensive study all over the world on its ingredients, physicochemical properties, vitamins, mineral content and quality control (Chen et al., 2012).

Numerous studies have reported that most chronic diseases such as cancer, coronary, and neurological degeneration are a consequence of oxidative damage. It is also proven that the therapeutic potential of honey is always associated with antioxidant capacity against reactive oxygen species (Ferreira et al., 2009). Components in honey reported to be responsible for its antioxidant effects are flavonoids, phenolic acids, ascorbic acid,

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catalase, peroxidase, carotenoids, and the products of Maillard reactions (Mohammed et al., 2014).

Polyphenols are currently of particular interest, mainly because of their functional properties. Besides being radical scavenger, polyphenol could be an effective immune modulator and hormone action inhibitor. Polyphenols are believed to be potent scavengers of peroxy radicals, mainly because of the presence of high mobility of hydrogens in their molecular structures. Of the polyphenols, phenolic acids are likely to be the major group in honey. They affect the flavor and physical appearance of honey, particularly honey color. Interestingly, they have been given considerable attention to be an eligible parameter for honey quality assessment, as well as for honey marker identification, with the help of the advancement of liquid chromatography and mass spectrometry technology nowadays (Chua et al., 2013).

Antimicrobial activity of honey is due to peroxide and non-peroxide factors. As to the first ones, some investigators have estimated that the major one is hydrogen peroxide, resulting of the oxidation of glucose by glucose oxidase during the ripening of honey. While, the nonperoxide is due to antimicrobial factors, physicochemical characteristics-high osmolarity, acidity, peptides, lysozyme, phenolic acids, and flavonoids (Ramos et al., 2018).

In the present study, antibacterial activities of six different Egyptian honeys collected and tested against six pathogenic bacteria in different conditions. Physical, chemical and antioxidant properties of local honeys were also studied to investigate and evaluate of functional properties of local to confirm its economical and nutritional importance.

Materials and Methods

Bacterial strains and cultivation

Six pathogenic bacterial species including Gram-positive and Gram-negative were used. These bacteria were kindly provided from The National Institute of Oceanography and Fisheries, and The High Institute of Public Health, Egypt. The Gram-negative bacteria were *E. coli*, *P. mirabilis*, *K. pneumoniae*, and *P. aeruginosa*, while the Gram-positive bacteria were *Staph. aureus*, and *S. mutans*.

Prior to the experiment, the strains were inoculated onto the surface of nutrient agar media; the inoculums suspensions were obtained by taking five colonies from 24 h cultures. The colonies were suspended in 5 ml of sterile saline (0.85% NaCl) and shaken for 15 sec. The density was adjusted to the turbidity of a 0.5 McFarland Standard (equivalent to $1-5 \times 10^8$ cfu/ml).

Honey samples

Six locally produced honey samples were purchased from Al-Dakhkhny Honey Market in Alexandria, Egypt. These samples were Cotton, Citrus, Sidr, Black seed, Eucalyptus, and Clover honey.

Physicochemical honey properties

The optical density of honey was determined by dissolving one gram of honey with 9 ml of distilled water and centrifuging for 10 min at 3000 rpm. The absorbance of the filtrate supernatant at 530 nm against distilled water as a blank using a computerized UV-Visible Spectrophotometer (T70 Split-Beam UV/VIS Spectrophotometer, England). The pH of honey filtrate was determined by pH meter as previously described (Egan et al., 1987). The water content of honey was estimated using an Abbe refractometer ($A = 589$ nm), that measures the refractive index of samples according to the method previously recommended (William, 1980). The electrical conductivity was estimated using conductometer (WTW. D8120 Weilheim LF. 42) from a solution containing 10 g of honey in 75 ml of distilled water. The result is expressed in milliSiemens per centimeter (mS. cm^{-1}) (Vorwohl, 1964).

Antioxidant activity

Total phenolic content

Phenolic compounds were determined according to Singleton et al. (1999), using Folin-Ciocalteu reagent and saturated sodium carbonate solution (75 g/L). The tubes were left in the dark at room temperature for two hours for color development, and then absorbance was read at 765 nm by a spectrophotometer. A calibration curve was made by using a solution of gallic acid.

Total flavonoid content

Honey solution (100 mg ml) was prepared with methanol 50% and previously homogenized and filtered through quantitative filter, 5 ml of honey solution was mixed with 5 ml of AlCl_3 (2%) in methanol. The mixture was homogenized

and allowed to stand for 30 min. The absorbance was measured at 415 nm. Quercetin was chosen as a standard, using a seven point standard curve (0–50 mg/L), the data were expressed as milligram quercetin equivalents (QE)/g lyophilized powder (Chang et al., 2002).

Determination of DPPH (2, 2-Diphenyl-1-picryl hydrazyl) scavenging activity

The DPPH assay was done as described by Koleva et al. (2002). Different concentrations of each honey sample or quercetin (test or standard, respectively) were added, at an equal volume, to methanolic solution of DPPH (100 µM). The positive control was a mixture of equal volumes of methanol and DPPH solution while the blank was methanol alone. After 15 min at room temperature, the absorbance was recorded at 517 nm. The IC_{50} values denote the concentration of sample, which is required to scavenge 50% of DPPH free radicals.

Determination of nitric oxide (NO) radical scavenging activity

Different concentrations of honey samples (50 µL) were mixed with equal volume of sodium nitroprusside (10 mM) in phosphate buffer saline for 150 min at room temperature using methanol as a blank. After incubation, 125 µL of Griess reagent was added to each tube and incubated for another 30 min. The absorbance of chromophore formed was measured at 546 nm on UV-visible spectrometer. Ascorbic acid was used as positive control. Scavenging/Reduction % = $[\text{Absorbance of control} - \text{Absorbance of test sample} / \text{Absorbance of control}] \times 100$. The IC_{50} values for each test compounds as well as standard preparation were calculated (Natalia et al., 2014).

Determination of ferric reducing antioxidant power (FRAP) assay

One ml of honey of different concentrations was mixed with the same volume of sodium phosphate buffer and potassium ferricyanide in separate test tubes. The reaction mixtures were incubated in a temperature-controlled water bath at 100°C for 20 min followed by addition of 1 ml of trichloroacetic acid. The mixtures were then centrifuged for 10 min at room temperature. The supernatant obtained (1 ml) was added with 1 ml of deionized water and 200 µL of $FeCl_3$. The blank was prepared as the samples except that potassium ferricyanide was replaced by distilled water. The absorbance of the reaction mixture was measured at 700 nm. IC_{50} values denote the concentration of

sample, which is required to scavenge/reduction 50% of FRAP free radicals (Sutharsingh et al., 2011).

Screening of antibacterial activity

The six types of honey were tested for antibacterial activity by well diffusion method and minimum inhibitory concentration (MIC) against pathogenic strains.

Agar well-diffusion method

Bacterial isolates were prepared to match 0.5 McFarland standards using the micropipette, 100 µL of organisms. Muller Hinton Agar plates were swabbed with 100 µL of each strain of pathogenic organisms. Approximately 5-mm diameters of wells were punched in solid agar with a cork borer. Using a micropipette, different types of honey (5 mg/ml) were loaded at different concentrations including 25, 50, 75 and 100 µL, then the plates were incubated at 37°C for 24 h. Antibacterial activity was expressed as the diameter of the clear zone inhibition, measured in millimeters (mm) (Mehrotra et al., 2010).

Minimum inhibitory concentration (MIC)

The MIC of the honey was determined using the serial micro dilution method as described by Samie et al. (2005). Control wells contained broth only (negative or sterility control) or bacteria and broth (positive control). One hundred microliter of the nutrient broth was added into each well of micro titration plate except row one of the positive control. Then, 100 µL of the honey was then added into each well of micro titration plate except row one of the negative control. A twofold serial dilution was done by mixing the contents in each well of the first row and transferring 100 µL to the second well of the same column and the same was done up to the last well of the same column and the last 100 µL from the last well was discarded; 50 µL of an overnight culture of the strain was mixed with the prepared extract dilutions and were incubated at 37°C for 24 h. Data were analyzed according to Patton et al. (2006). Briefly, the optical density was determined just prior to incubation (T_0) and again after 24 h incubation (T_{24}) at 600 nm by IgG-ELISA Kit of Bioactivia-Germany. The OD for each replicate at T_0 was subtracted from the OD for each replicate at T_{24} . The adjusted OD of each control well was then assigned a value of 100% growth. The percent inhibition of growth was thus determined using the formula Percent Inhibition = $1 - (\text{OD test well} / \text{OD of corresponding control})$

well) $\times 100$. The MIC is reported as the lowest concentration of test material which results in 100% inhibition of growth of the test organism. The MICs were tested for their antibacterial activity against the pathogenic bacteria which were *E. coli*, *Proteus mirabilis*, *K. pneumoniae*, *P. aeruginosa*, *Staph. aureus*, and *S. mutans*.

Results and Discussion

Physicochemical characteristics of honey

The physicochemical properties of the different samples of local honey are reported in Table 1. The tested honey showed no sign of fermentation before initiating the physicochemical analysis. Physicochemical study showed that all the honey types were different in properties

except that all were acidic. The color of honey types varies from golden yellow to dark brown, which is due to the presence of some different chemical constituents as reported by Taormina et al. (2001). The optical density in tested honey samples ranged from 0.50 to 1.65. While the pH was in the acidic range and approximately equal in all honey types. These results are in agreement with Salonen et al. (2017) who mentioned that honey is acidic, and its pH ranges between 3.5 and 5.2. The pH of honey is low enough to inhibit the growth of many bacterial species, but this acidity may be neutralized in the body by the buffering liquid fluids. Radwan et al. (1984) reported that the acidity of honey is primarily to the content of gluconolactone/gluconic acid present as a result of enzyme action in the ripening nectar.

TABLE 1. Physicochemical properties of different varieties of honey

Honey type	Color	Optical density	pH	Water activity (a_w)	Electrical conductivity ($mScm^{-1}$)
Cotton	Brown	0.63	4.74	0.52	0.70
Orange	Golden yellow	0.50	4.17	0.5	0.34
Sidr	Brown	1.65	4.95	0.64	0.51
Black seed	Brown	0.59	4.52	0.5	0.88
Eucalyptus	Dark brown	1.05	4.20	0.52	0.91
Clover	Golden yellow	0.54	4.43	0.61	0.29

The moisture contents in tested honey samples ranged from 0.5 to 0.64, which are very similar to some studies which reported the moisture content of honey between 0.56 and 0.58 (Ruagg & Blanc, 1981). The highest moisture content was found in Sidr honey. Moisture content variation can be explained by the composition and floral origin of honey samples. The low moisture content of honey forms an important part of the system which protects honey from attack by microorganism and therefore affects its storage life (Adenekan et al., 2010). Honey is a sugar solution of high osmolarity which inhibits bacteria. The high sugar concentration tie-up water molecules, so that bacteria would have insufficient water to support their growth (Malika et al., 2005).

The electrical conductivity (EC) values varied from 0.29- 0.79 $mScm^{-1}$. The lowest was obtained from Clover honey samples and this was significantly different from the highest

value of EC obtained from Eucalyptus. The EC is a good criterion related to botanical origin of honey and this is very often used in routine honey control instead of the ash content. The electrical conductivity may also be explained by taking into account the ash and acid content of honey, which reflects the presence of ions and organic acid; the higher their content, the higher the resulting conductivity (Adenekan et al., 2010).

Antioxidant activity of honey

Total phenolic and total flavonoid content

The phenolic contents of the honey samples were between 5500 and 14120 mg/kg. The total phenolic content in Cotton honey (14120 mg/kg) and Eucalyptus honey (13690 mg/kg) were the maximum. While low values were reported by Sidr and Orange honey samples. Honey samples from Brazil ranged from 870.83-1110.37 mg/kg (Bueno-Costa et al., 2016). The results were higher than the commercial Slovenian

honey which ranged from 44.8 to 233.9 mg kg⁻¹ (Bertoncelj et al., 2007).

Flavonoids are low molecular weight phenolic compounds responsible for the aroma and the antioxidant potential of honey. Total flavonoid content among honey samples ranged between

926-1657 mg/kg (Table 2), and the maximum total flavonoid content was reported by Cotton (1657 mg/kg) followed by Eucalyptus (1415 mg/kg). The results are in agreement with those of several previous studies, in which it was found that honey samples with higher polyphenol content will also yield high flavonoid levels (Mohammed et al., 2014).

TABLE 2. Total phenolic and total flavonoid content of different varieties of honey

Honey type	Total Phenolic content (mg/kg)	Total Flavonoid content (mg/kg)
Cotton	14120	1657
Orange	5500	926
Sidr	6460	1119
Black seed	11150	1383
Eucalyptus	13690	1415
Clover	9520	126

The total flavonone and dihydroflavonol content ranged from 1706 to 2606 mg of pinocembrin kg⁻¹ in the Brazilian honey as investigated by Pontis et al. (2014). The flavonoid content results in this study were higher than the results obtained for honey from the northeast of Brazil, which ranged from 2.5 to 83.8 mg of quercetin kg⁻¹ of honey (Liberato et al., 2011). Also, Meda et al. (2005) evaluated twenty-seven Burkina Faso honey samples and determined their flavonoid content using the aluminum chloride method, and their results ranged from 17 to 83.5 mg of quercetin kg⁻¹ of honey.

The distinctive differences between different types of honey from different countries are caused by different locations, especially the climatic and vegetation conditions, and the flowering period of nectariferous plants (Lachman et al., 2010)

DPPH scavenging activity test

Table 3 shows the scavenging ability of honey samples expressed as IC₅₀ with respect to the DPPH[•] radical, which ranged from 24.07 to 95.07 mg/ml. Cotton honey had the highest antioxidant capacity (24.07 mg/ml). Possibly, observed high radical scavenging activity of the Cotton honey is due to the high phenolic and flavonoid contents in it, thus indicating high antioxidant potential.

Honey from the northeast of Brazil had IC₅₀ values ranging from 4.2 to 106.72 mg/ml although most values were above 20 mg/ml (Liberato et al., 2011). These results showed that

the honey samples collected in the present study have greater antioxidant potential compared to the results reported in the other literatures. Furthermore, it was observed that the highest radical scavenging activity is linked with the highest honey concentration tested of all honey samples. Recently, similar findings were reported by Khalil et al. (2010) who found the highest scavenging activity at highest honey concentrations.

Nitric oxide (NO) scavenging activity test

Results in Table 3 shows the scavenging ability of honey samples expressed as IC₅₀ with respect to the NO, which ranged from 5.52-83.07 mg/ml, with maximum NO scavenging activity reported by Cotton honey (5.52 mg/ml) followed by Eucalyptus and Black seed.

As free radical scavenger, it could be attributed to presence of conjugated ring structures and carboxylic group which have been reported to inhibit lipid peroxidation (Rice-Evans et al., 1995). NO is a free radical generated from sodium nitroprusside in aqueous solution at physiological pH and reacts with oxygen to form nitrite. It is well known that NO play an important role in various inflammatory processes such as carcinomas, juvenile diabetes, arthritis, multiple sclerosis and ulcerative colitis (Hazra et al., 2008). Lower values of NO radical scavenging activity expressed as IC₅₀, represents high antioxidant activity as investigated by Jain & Agrawal (2008).

TABLE 3. Antioxidant activities of different varieties of honey

Honey type	IC ₅₀ (mg/ml)		
	DPPH	NO	FRAP
Cotton	24.07	5.52	1.24*(10 ³)
Orange	95.07	83.07	9.72*(10 ³)
Sidr	57.4	51.34	4.85*(10 ³)
Black seed	37.03	20.06	3.13*(10 ³)
Eucalyptus	37.62	19.46	2.67*(10 ³)
Clover	39.44	48.81	4.77*(10 ³)

Ferric reducing antioxidant power (FRAP) assay test

Higher FRAP values give higher antioxidant capacity where it directly estimates the presence of either antioxidants or reductants, depending on the ability of the analyte to reduce the Fe³⁺/Fe²⁺ redox couple (Rabeta & Nur Faraniza, 2013). In this study, values of FRAP were expressed as IC₅₀ parameter. The FRAP values for the investigated honey samples ranged from 1.24*(10³) - 9.72*(10³) as reported in Table 3. Significant differences among the FRAP values for the different types of honey, suggesting that they have different antioxidant potentials.

Cotton and Eucalyptus honey gave the highest values than others. The mean FRAP value of the investigated honey samples from Egypt is higher than that of Bangladesh honey (Mohammed et al., 2014), suggesting the high quality of Egyptian honey, as represented by their high antioxidant potentials.

Antibacterial activity test

Agar well-diffusion method

Antibacterial potency of honeys was investigated against various Gram negative and Gram positive pathogenic bacteria. All the honey types showed high antibacterial activity against most of the tested bacterial strains at 100 µl concentration as shown in Fig. 1-6. *K. pneumonia*, *P. aeruginosa*, *P. mirabilis*, *E. coli* and *S. mutans* showed the maximum sensitivity to Black seed honey extracts with maximum inhibition zones equal to 42.67 mm, 39.00, 36.00, 27.33 and 18.67 at 100 µl, respectively (Fig. 4). This was followed by extracts of Eucalyptus honey against *K. pneumonia*, *P. aeruginosa* and *P. mirabilis* which gave inhibition zones equal to 41, 38.33 and 35.67 mm at 100µl respectively and approximately equal inhibition zones equal to 13.67 mm against both *E. coli* and *S. mutans* (Fig. 5). In addition,

Clover honey extracts showed antibacterial effect against *K. pneumonia*, *P. aeruginosa*, *P. mirabilis*, *E. coli* and *S. mutans* by inhibition zones equal to 39.33, 31.00, 31.00, 30.67 and 25 mm at 100 µl respectively (Fig. 6). The Orange, Sidr, and Cotton honey extracts recorded the lowest antibacterial effect among the six types. Orange honey had shown antibacterial effect against only *K. pneumonia*, *P. mirabilis* and *E. coli* giving inhibition zones equal to 36.33, 34.33 and 27 mm at 100 µl respectively (Fig. 2). Results revealed a higher inhibition zone value for Gram negative bacteria comparing to the other tested Gram positive. This may be due to the differences in bacterial cell walls, since Gram negative bacteria have thinner cell wall comparing to Gram positive one (Rai et al., 2009).

The conflict in the observed antibacterial activity can be due to several reasons. It might be related to the differences in susceptibility of each species of microorganism to the antibacterial activity of honey used (Taormina et al., 2001). Moreover, it could also possibly be due to the different floral sources utilized by the bees and the geographical factors like temperature, humidity where the honey was produced and harvested. Some studies claimed that the antibacterial activity of honey is related to its acidity and other physiochemical parameters. Other possible explanation is the utilization of hydrogen peroxide and non-peroxide antioxidant components as reported by Melissa et al. (2004).

The differences in results may be also due to attributable to the method by which activity was assessed and variations in protocols between different research groups. In particular, the method for inoculating the agar, whether it is surface inoculation such as was used in the current study or incorporation of the organisms into the agar plate itself may influence results (Irish et al., 2008).

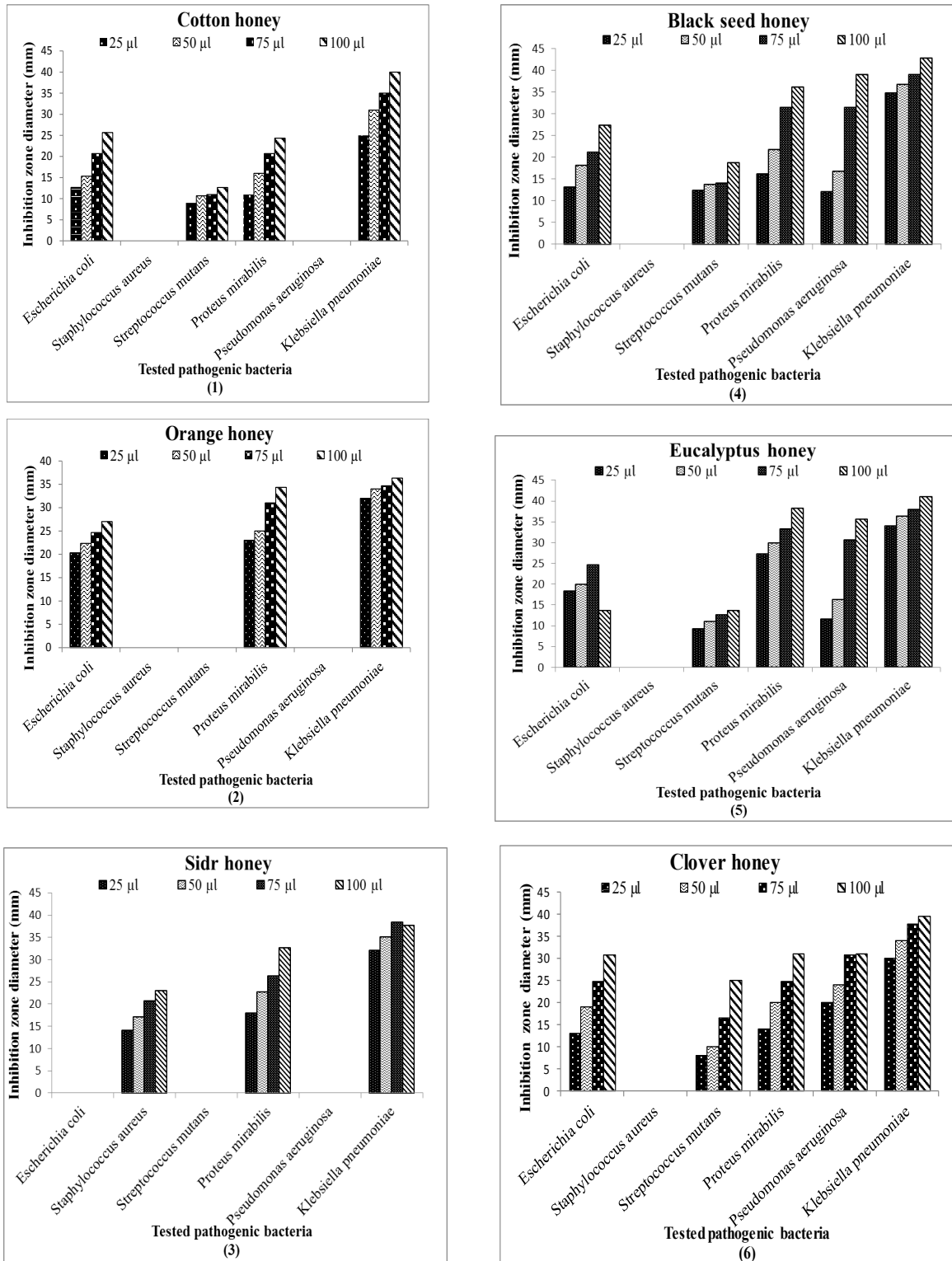


Fig. 1-6. The inhibition zone of different types of honey against various pathogenic microorganisms by well diffusion method.

Minimum inhibitory concentration (MIC)

The MIC of the different types of honey against the pathogenic bacteria *S. aureus*, *S. mutans*, *P. aeruginosa*, *E. coli*, *P. mirabilis*, *K. pneumonia* is shown in Table 4. It was found that *E. coli*, *P. mirabilis* and *K. pneumonia* were affected by all types of honey compared with *P. aeruginosa* and *S. mutans*. While *S. aureus* showed high resistance for all honey types except for 100% Sidr honey

only, and this was contradicted by Alvarez-Suarez et al. (2012) who reported that *S. aureus* is more sensitive to the action of honey than *P. aeruginosa*. At 25% concentration, Black seed and Eucalyptus honey had an effect on *E. coli* and *K. pneumonia*, while Clover honey had an effect on *P. aeruginosa* and *P. mirabilis*. In some studies, resistant and sensitive *P. aeruginosa* was resistant to Sidr honey (Alaa et al., 2015).

TABLE 4. Minimum inhibitory concentration (MIC) of different types of honey against the most susceptible bacteria. method.

Tested pathogenic bacteria	MIC (%)					
	Cotton	Orange	Sidr	Black seed	Eucalyptus	Clover
<i>Staphylococcus aureus</i>	-	-	100	-	-	-
<i>Streptococcus mutans</i>	50	-	-	50	50	50
<i>Pseudomonas aeruginosa</i>	-	-	-	50	50	25
<i>Escherichia coli</i>	50	50	-	25	25	50
<i>Proteus Mirabilis</i>	50	50	50	50	50	25
<i>Klebsiella pneumoniae</i>	50	50	50	25	25	50

Many researchers investigated the role of the non-peroxide activity as part of the antimicrobial properties of honeys in ripe honeys, in which only a small amount of peroxide, is not sufficient for bacterial growth inhibition. And though it was found that the antibacterial activity of honeys was related to the polyphenols found therein. Non-peroxide compounds could mostly be related to the antibacterial effect which was by the studied honey samples, because the samples with the highest phenolic contents revealed the smallest MICs against the tested bacterial strains (Sousa et al., 2016).

Conclusion

The antioxidant and antimicrobial properties of locally produced honeys in Egypt make them highly added valuable products. The differences between honey samples in terms of antibacterial and antioxidant activity could be attributed to the natural variations in floral sources of nectar and the different locations. Honey has potential to play a role in providing antioxidants in a highly palatable form against reactive oxygen species causing most chronic diseases such as cancer, coronary and neurological degeneration. Moreover, the results revealed that the antibacterial effect of different types of honey is type and concentration dependent.

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(Received 28 / 4 /2017;
accepted 8 / 1 /2018)

النشاطات المضادة للبكتيريا و للأكسدة لأنواع مختلفة من العسل المصري المنتج محليا

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تم من خلال البحث دراسة الخصائص الفيزيوكيميائية، النشاطات المضادة للأكسدة و للميكروبات لستة أنواع عسل أبيض تم الحصول عليهم من الأسكندرية، مصر. وقد أظهرت العوامل الفيزيوكيميائية، أن الكثافة الضوئية لعينات العسل المختبرة تراوحت بين 0.50 إلى 1.65. بينما كان أس الهيدروجين يدل على حامضية العينات المختلفة من العسل. و تراوحت قيم محتوى الرطوبة والتوصيل الكهربائي بين 0.5 إلى 0.64 و 0.29 إلى 0.79 على التوالي. و قد تراوح محتوى الفينول فى العينات بين 5500 و 14120 مجم/كج. بينما كان محتوى الفلافينويد بين 926 و 1657 مجم/كج. و قد أظهر المحتوى العالى من الفينول و الفلافينويد كفاءة عالية فى النشاط المضاد للأكسدة كما هو واضح من عينات عسل القطن، الكافور و حبة البركة. بدراسة النشاط المضاد للميكروبات، أوضحت النتائج أن عينات العسل من حبة البركة بلبه عسل الكافور، قد أظهرت أعلى نشاط مضاد للبكتيريا ضد كل من بكتيريا الكليبسيلا نيمونيا، سودومونس اريجينوزا، بروتيس ميرابيليس، اشيريشيا كولاي و ستربيتوكوكس ميوتانس بأعلى مناطق تثبيط تقدر بحوالى 42.67، 39.00، 36.00، 27.33 و 18.67 مم عند 100 ميكرو لتر، على التوالي لعسل حبة البركة و 41.00، 38.33 و 35.67 مم و منطقة تثبيط تساوى تقريبا 13.67 مم ضد كل من الاشيريشيا كولاي و الستربيتوكوكس ميوتانس عند 100 ميكرو لتر، على التوالي لعسل الكافور. وقد أوضحت نتائج أقل التركيزات المثبطة للبكتيريا الممرضة، أن عند تركيز 25% كان عسل حبة البركة و عسل الكافور له تأثير على الاشيريشيا كولاي و الكليبسيلا نيمونيا، بينما كان عسل البرسيم له تأثير على السودومونس اريجينوزا و البروتيس ميرابيليس. لذلك يوصى البحث بأهمية استخدام العسل كغذاء، لأغراض طبية و صناعية و يوضح أهميته كسلعة مهمة فى السوق العالمى.