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# ASSESSMENT OF ANTIBACTERIAL ACTIVITY OF SOME UNIFLORAL EGYPTIAN HONEYS

BY

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## ABSTRACT

***This*** work include three research items, the first one to study some physicochemical properties of three different unifloral Egyptian honeys; Citrus, Clover and Cotton honeys and also assess *in vitro* antibacterial activity (AA) and its affect by storage, dilution with water (33% w/v) or autoclaving (121°C for 15 min). Generally, all tested properties of fresh honeys were within the international honey standards. In samples of honeys stored for 12 or 24 month, OD, HMF and acidity increased while RI, Wa, TSS, EC and pH almost remained unaffected and $H_2O_2$ values decreased. The results show that the different three types of honey exhibited various degrees of AA against different indicator bacteria (IB) as indicated by reduction percentage of initial bacterial count, depending on type of honey and the IB, where the highest AA was for clover honey followed by citrus and cotton honeys, respectively. Results also showed that different species of bacteria differ in their sensitivity to honey, *Salmonella enteritidis* was the most sensitive followed by *Staphylococcus aureus*, *Listeria monocytogenes* and *Escherichia coli*, respectively. Assessment the AA of the tested honeys as affected by storage up to 24 months at room temperature was slightly reduce the AA. The average reduction percentages were 2.6 % and 4.6 %, after 12 and 24 months, respectively. While, diluting honeys with water increase the AA by 8.3 % in average. Also autoclaving treatment decreased the AA by 13.5 % in average. The second research item aiming to investigate the relative contribution of the peroxide and non peroxide honey components in the total AA of fresh honeys. The AA of the three unifloral Egyptian honeys is mainly attributed to non- peroxide antibacterial agents which their contribution is 88% in average, while the contribution of hydrogen peroxide is only 12%. The results also indicate that the contribution of the thermostable antibacterial components in honey is 86.8% in average, while the contribution of the thermolabile components is only 13.2% to the total AA. The third research item was to compare AA of the fresh clover honey with the effect of 16 antibiotics on the IB. The fresh clover honey exhibited AA comparable to that exhibited by the tested antibiotics. Moreover, AA of water diluted fresh clover honey was generally higher compared to sme of the tested antibiotics. In conclusion, all the three unifloral Egyptian honeys tested in this study especially clover honey exhibited high AA against all the four pathogenic bacteria. Among these honeys, clover honey appeared to deserve further investigations, where it my have an important economical value, since it may prove to be a promise natural food preservative or/ and a valuable therapeutic honey.

**Key words:** Honey, physicochemical properties, antibacterial activity, *Escherichia coli*, *Salmonella enteritidis*, *Listeria monocytogenes*, *Staphylococcus aureus*, Antibiotics.

## INTRODUCTION

Honey is nectar collected from many plants and produced by honeybees *(Apis mellifera)*. The composition of honey is variable, owing to the differences in plant types, climate, environmental conditions (Azeredo *et al.*, 2003, Kücük *et al.*, 2007). Honey has been reported to contain about 200 substances and is considered as an important part of traditional medicine (White, 1979). The antimicrobial action of honey was reported for the first time by Van Ketel in 1892 (Molan, 1992a). The different aspects of the antibacterial properties of honey have been extensively reviewed (Molan, 1992a, b). The reasons for the antimicrobial activity of honey are so far controversial. There are two sorts of antibacterial agents or so called "inhabines". One of them is heat-and light-sensitive and has its origin in the $H_2O_2$, produced by honey glucose oxidase (White *et al.*, 1963, White and Subers, 1964 and Dustmann, 1972). Some workers believe that hydrogen peroxide is the main antimicrobial agent (Dustmann, 1979 and Morse, 1986). Other authors find that the non-peroxide activity is the more important one. The argument of the latter is that in ripe honey the glucose oxidase is inactive and honey contains only a small peroxide amount, not sufficient to inhibit bacterial growth. The non-peroxide antimicrobial activity is insensitive to heat and light and remains intact after storage of honey for longer periods (Bogdanov, 1984 and Roth *et al.*, 1986). It has been documented that honey has a bacteriostatic and bacteriocidal effect against various species of both gram positive and gram negative bacteria, as well as an anti-fungal effect (Molan and Betts 2000). Extensive review on the antibacterial activity of honey showed that, pure honey has bactericidal activity against many enteropathogenic organisms, including those of the *Salmonella* sp., *Shigella* sp., *E. coli* and was also found is more effective as an antibacterial agent against several *Pseudomonas, Mycobacterium, and Staphylococcus* strains (Jeddar *et al.*, 1985).

Objectives of this work were: (I) to assess *in vitro* antibacterial activity (AA) and determine some physicochemical properties of three different unifloral Egyptian honeys, Citrus, Clover and Cotton honeys, (II) to investigate the relative contribution of the peroxide and non peroxide honey components in the total AA of fresh honeys, and (III) to compare AA of the honey type showed the highest AA with the those of some antibiotics.

## MATERIALS AND METHODS

### 1. Material

#### 1.1. Honey samples

Nine honey samples from unifloral sources; Citrus (*Citrus spp.*), Egyptian clover (*Trifolium alexandrium*) and Cotton (*Gossypium vitifolium*) were obtained by ordinary beekeeping practices during three successive seasons. Citrus honeys were obtained from an apiary situated in Banha, Qalyoubia government, while clover and cotton ones were obtained apiaries situated in Fayoum government, Egypt. Bee colonies in these apiaries were situated in Langstroth's standard hives and headed with local hybrid Carniolan, *Apis mellifera carnica*, queens. Honey samples were collected in sterile screwed brown bottles, and samples of the first and second seasons were stored for 24 and 12 months, respectively at room temperature (25 ± 10°C) in the dark until to be tested. Samples of the third season were used as fresh honeys.

#### 1.2. Indicator bacterial strains

Two Gram negative bacteria (*Escherichia coli* ATCC 25922 and *Salmonella enteritidis* ATCC 13076 and two Gram positive bacteria (*Listeria monocytogenes* ATCC 15313 and *Staphylococcus aureus* ATCC 8095) were used as indicator bacteria for determination the antibacterial activity. The strains were obtained from the culture collection of Agricultural Microbiology Department, Faculty of Agriculture, Fayoum University.

#### 1.3. Antibiotics

In *vitro* diagnostics discs (Pasteur LAB, Egypt) of 16 antibiotics listed in Table 4

were used to compare their antibacterial activity with that of fresh clover honey.

## 2. Methods

### 2.1. Physicochemical properties of honey

Moisture content in honey was determined by a refractometer, ash content, total acidity, pH, refractive index (RI), color, total soluble solid (TSS) and electrical conductivity (EC) were determined according to the official methods of analysis of the Association of Official Analytical Chemists (A.O.A.C. 1990). Hydroxymethyl furfural content was determined by the UV spectrophotometer method (White, 1979). Screening for peroxide accumulation as an indicator of glucose oxidase activity was carried out according to López-Sabater *et al.* (1993). Water activity ($w_a$) was measured according to Beckh *et al.* (2004).

### 2.2. Preparation of bacterial inocula

An isolated pure colony of an overnight grown bacterial strain was picked carefully using a sterile transfer loop, inoculated to LB broth in an Erlemeyer flask and grown overnight at the optimum temperature for each bacterial strain. About 50 µl of the overnight culture were inoculated to 20 ml of LB broth and grown further for about 3-4 h until an OD. of 0.6 (564 nm) was achieved. The suspension was then diluted 1:50 with LB broth in order to obtain the standard inoculum.

### 2.3. Assessment of antibacterial activity

**(A) Plate count assay**

The antibacterial activity of honey samples was done using standard plate count method. Before applying, 100 µl of inoculums containing a known initial counts of each test microorganism were thoroughly mixed with one ml of crude honey (CH), honey diluted with water (DH), diluted honey with water treated by catalase (CDH), 10 µl $ml^{-1}$ (329300 U/ml) of bovine liver catalase (Fluka), and row honey autoclaved by temperature at 121°C for 15 min (ADH). All treatments incubated at room temperature for 45-230 min depending upon honey type and storage period. The percentage of survived viable counts of each test microorganism were determined by applying one ml of each honey treatment onto sterile Petri dish, followed by mixing with agar media. The plates were leaved to solidify at room temperature for 1h, the plates were incubated for 48 h at the optimum temperature for each bacterial strain. The inhibition was expressed as decreasing percentage of initial counts.

**(B) Well- diffusion bioassay (Torres *et al.*, 2004)**

Sterilized LB agar medium was cooled to 48°C, 5 ml of each standard inoculum were mixed with 1litter LB agar medium and 20 ml of each inoculum poured into sterile Petri dishes. When the agar was solidified three holes of 8 mm diameter were bored per plate. Each hole was then filled with 200µl honey and the plates were placed in refrigerator for 2 h, giving the honey enough time to diffuse. Finally, the plates were incubated for 24 h at the optimum temperature for each bacterial strain. This method was used to assess the AA of honey when compared with antibiotics. The AA was assessed by measuring the zone of inhibition (mm) against the indicator bacteria.

**(C) Disc diffusion assay (Patton *et al.*, 2006)**

LB agar plates were inoculated by swabbing overnight cultures onto the surface of agar plates which allowed standing at room temperature for 3 h before antibiotic discs were applied. The plates were incubated for 24 h at the optimum temperature for each bacterial strain. The AAs of antibiotics were assessed by measuring the zone of inhibition (mm) against the indicator bacteria.

## RESULTS AND DISCUSSION

The three honeys of different floral origin have been studied for their physicochemical properties and the results are shown in Table 1. Generally, all tested properties of fresh honeys were within the international honey standards (Anonymous, 1999). Regarding fresh honey samples, except values of HMF content, values of some tested properties

such as Ash, EC and the acidity were found the highest in cotton honey compared with clover and citrus honeys. Clover honey had a higher HMF content than that in citrus and cotton honeys. However, values of HMF content in the three honeys were much lower the upper limit (40 mg/kg), which indicates the freshness of these honey samples. Some European bee federations market a part of their honey as 'quality honey', having a maximum of 15 mg/kg (Anon.,1999). In samples of honeys stored for 12 or 24 month, OD, HMF and acidity increased while RI, Wa, TSS, EC and pH almost remained unaffected and $H_2O_2$ values decreased. In literature, studies on correlation between the antibacterial activity of honey and their physicochemical properties are rare. However, Vorlova *et al.* (2005) found a statistically significant relation between the electrical conductivity of honey, the higher conductivity, the higher the antibacterial activity. The present study showed opposite trend which may be due to number of honeys used compared to the 20 honey types used in the study of those authors. They also found that the pH values do not have a significant influence on AA.

The antibacterial activity (AA) of honey is one of the characteristics that make it beneficial to human health, but some factors can affect of this character. In the present study, the AA of three Egyptian honeys of different floral origin, fresh or stored, crude or treated was *in vitro* assessed. The results in Table 2 revealed that all the four indicator bacteria (IB), regardless of Grame reaction, were sensitive to all the three honeys tested. However, the different honeys exhibited various degrees of AA against different IB as indicated by reduction percentage in initial bacterial count. However, no honey exhibited a complete inhibition of bacterial growth at any the honey treatments tested, the exception was clover honey when diluted with water against both *Salmonella enteritidis* and *Staphylococcus aureus*. In a study by Efem (1988) *S. aureus* was the most sensitive to the antibacterial action of honey.

When the results in Table 2 were pooled and illustrated in Fig. 1, the overall values indicated that the three unifloral Egyption honeys exhibited differing antibacterial activities against the four indicator pathogenic bacteria used. Clover honey showed the highest AA followed by citrus and cotton honeys, respectively. This result highlights the finding that the floral origin of honey may have contributed to differences in their antibacterial activities (Allen *et al.*, 1991; Willix *et al.*, 1992). Results also showed that different species of bacteria differ in their sensitivity to honey, *Salmonella enteritidis* was the most sensitive followed by *Staphylococcus aureus*, *Listeria monocytogenes* and *Escherichia coli*, respectively.

Assessment the AA of the tested honeys as affected by storage up to 24 months at room temperature (25± 10°C) in dark (Table 2 and Fig. 1) revealed that these storage conditions slightly reduced the AA and the reduction was increased by increasing period of storage. The average reduction percentages were 2.6 % and 4.6 %, after 12 and 24 months, respectively. This result is in agreement with that of Bogdanov (1997) who found that the non- peroxide AA of honey was only slightly affected by storage at room temperature (20-25°C) for 15 months in dark.

Regarding the effect of different treatments on the AA of honeys, the results (Table 2 and Fig. 1) indicate that it was slightly increased by diluting honeys with water (33%, w/v) and the effect percentage varied according the floral origin of honey, was the highest in cotton honey and the lowest in clover honey, and was 8.3 % in average. The effect of dilution with water is expected as honey when diluted with water; the bee-derived glucose oxidase enzyme persent in honey is activated and catalyzed slow generation of hydrogen peroxide (Molan, 1992b). On the other hand, when the diluted honey treated with catalase, AA was reduced which indicate that hydrogen peroxide contributes to AA of the tested honeys.

**Table (1): Physicochemical properties of three popular unifloral Egyptian honeys (citrus, clover and cotton) produced during three successive seasons.**

| Properties | Citrus | | | Clover | | | Cotton | | | IHS |
|---|---|---|---|---|---|---|---|---|---|---|
| | Fresh | 1 year | 2 years | Fresh | 1 year | 2 years | Fresh | 1 year | 2 years | - |
| Refractive index (RI) | 1.50 | 1.50 | 1.50 | 1.50 | 1.50 | 1.50 | 1.49 | 1.49 | 1.50 | - |
| Colour (OD) | 0.04 | 0.05 | 0.09 | 0.04 | 0.06 | 0.11 | 0.11 | 0.15 | 0.22 | - |
| Water activity ($w_a$) | 0.56 | 0.56 | 0.52 | 0.56 | 0.56 | 0.52 | 0.55 | 0.55 | 0.52 | - |
| TSS (%) | 82.0 | 82.0 | 84.0 | 84.00 | 82.00 | 84.00 | 82.00 | 82.00 | 84.00 | - |
| Ash (%) | 0.13 | 0.18 | 0.15 | 0.16 | 0.16 | 0.20 | 0.42 | 0.41 | 0.44 | $\leq 0.6$ |
| HMF ($mg/kg^{-1}$) | 10.4 | 73.82 | 186.6 | 12.10 | 47.66 | 73.62 | 6.12 | 65.05 | 135.17 | $\leq 40$ |
| EC ($mScm^{-1}$) | 0.40 | 0.44 | 0.34 | 0.42 | 0.41 | 0.50 | 0.88 | 0.85 | 0.91 | $\leq 0.8$ |
| Acidity ($meq/kg^{-1}$) | 10.83 | 14.60 | 15.27 | 11.01 | 11.50 | 11.68 | 28.06 | 33.74 | 37.71 | $\leq 50$ |
| pH | 4.47 | 4.50 | 4.60 | 4.64 | 4.76 | 4.73 | 4.66 | 4.70 | 4.74 | - |
| $H_2O_2$ (ppm) * | 2.23 | 1.98 | 0. 92 | 2.40 | 2.16 | 1.25 | 3.98 | 3.04 | 1.94 | - |

TSS =Total soluble solids HMF = Hydroxy methyl furfural
EC = Electrical conductivity * = In water diluted honey (33%, W/V)
IHS = International honey standard values

**Table (2): Antibacterial activity of three popular Egyptian unifloral honeys against four pathogenic bacteria as affected by storage period, heating, dilution and catalase treatment.**

| Treat-ment | Storage period (year) | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Fresh | | | | 1 | | | | 2 | | | |
| | Citrus | | | | | | | | | | | |
| | *Ec* | *Se* | *Lm* | *Sa* | *Ec* | *Se* | *Lm* | *Sa* | *Ec* | *Se* | *Lm* | *Sa* |
| CH | 87.5 | 90.1 | 85.4 | 89.1 | 86.7 | 88.0 | 85.0 | 87.3 | 85.1 | 86.6 | 84.8 | 88.3 |
| DH | 93.9 | 96.3 | 93.0 | 95.9 | 92.8 | 93.5 | 92.0 | 93.8 | 89.4 | 91.9 | 91.1 | 94.8 |
| CDH | 82.3 | 89.7 | 79.2 | 86.9 | 82.6 | 84.0 | 78.0 | 83.5 | 78.0 | 81.2 | 76.2 | 82.0 |
| ADH | 81.5 | 89.5 | 78.3 | 86.6 | 80.1 | 83.6 | 77.3 | 82.2 | 77.4 | 80.3 | 75.3 | 81.1 |
| | Clover | | | | | | | | | | | |
| CH | 94.2 | 99.7 | 95.4 | 95.2 | 89.4 | 97.8 | 93.2 | 92.1 | 87.9 | 97.8 | 93.0 | 91.0 |
| DH | 98.0 | 100.0 | 98.5 | 100.0 | 92.6 | 98.0 | 95.0 | 95.6 | 91.0 | 97.9 | 94.2 | 95.0 |
| CDH | 87.5 | 98.7 | 92.0 | 90.7 | 81.9 | 97.6 | 89.2 | 87.3 | 80.0 | 97.5 | 88.4 | 83.3 |
| ADH | 86.2 | 98.4 | 91.3 | 89.9 | 80.3 | 97.5 | 89.0 | 86.6 | 78.7 | 97.2 | 88.0 | 82.1 |
| | Cotton | | | | | | | | | | | |
| CH | 78.0 | 82.2 | 80.0 | 78.1 | 79.4 | 82.7 | 77.1 | 77.6 | 77.1 | 82.1 | 74.6 | 79.0 |
| DH | 94.9 | 94.0 | 96.4 | 93.6 | 92.3 | 93.0 | 92.3 | 91.0 | 89.4 | 91.1 | 87.5 | 88.5 |
| CDH | 76.2 | 78.3 | 79.4 | 75.1 | 78.2 | 77.5 | 74.2 | 73.1 | 73.1 | 75.2 | 70.9 | 71.9 |
| ADH | 75.1 | 76.6 | 79.0 | 73.0 | 78.0 | 76.2 | 72.7 | 72.0 | 71.6 | 73.9 | 70.0 | 70.4 |

CH =Crude honey DH =Water diluted honey (33% w/v)
CDH =Catalase-treated, diluted honey ADH =Autoclaved, diluted honey
*Ec=Escherichia coli, Se=Salmonella entretidis, Lm=Listeria monocytogenes, Sa=Staphylococcus aureus*

As concerns the effect of heating, when samples of the tested honeys were autoclaved at 121°C for 15 minutes, the antibacterial activities of these honeys were partly decreased, and the decreasing percentage was the highest in cotton honey and the lowest in clover honey, and was 13.5 % in average. In this respect, White and Subers (1964) found that heating honey at 70°C for 15 minutes had no or very little effect on the non- peroxide AA, whereas the peroxide accumulation capacity is severely damaged. This finding may explains the results in Table 3 which show that the tested honeys retain most of their antibacterial activities (87% in average) after heating in autoclave.

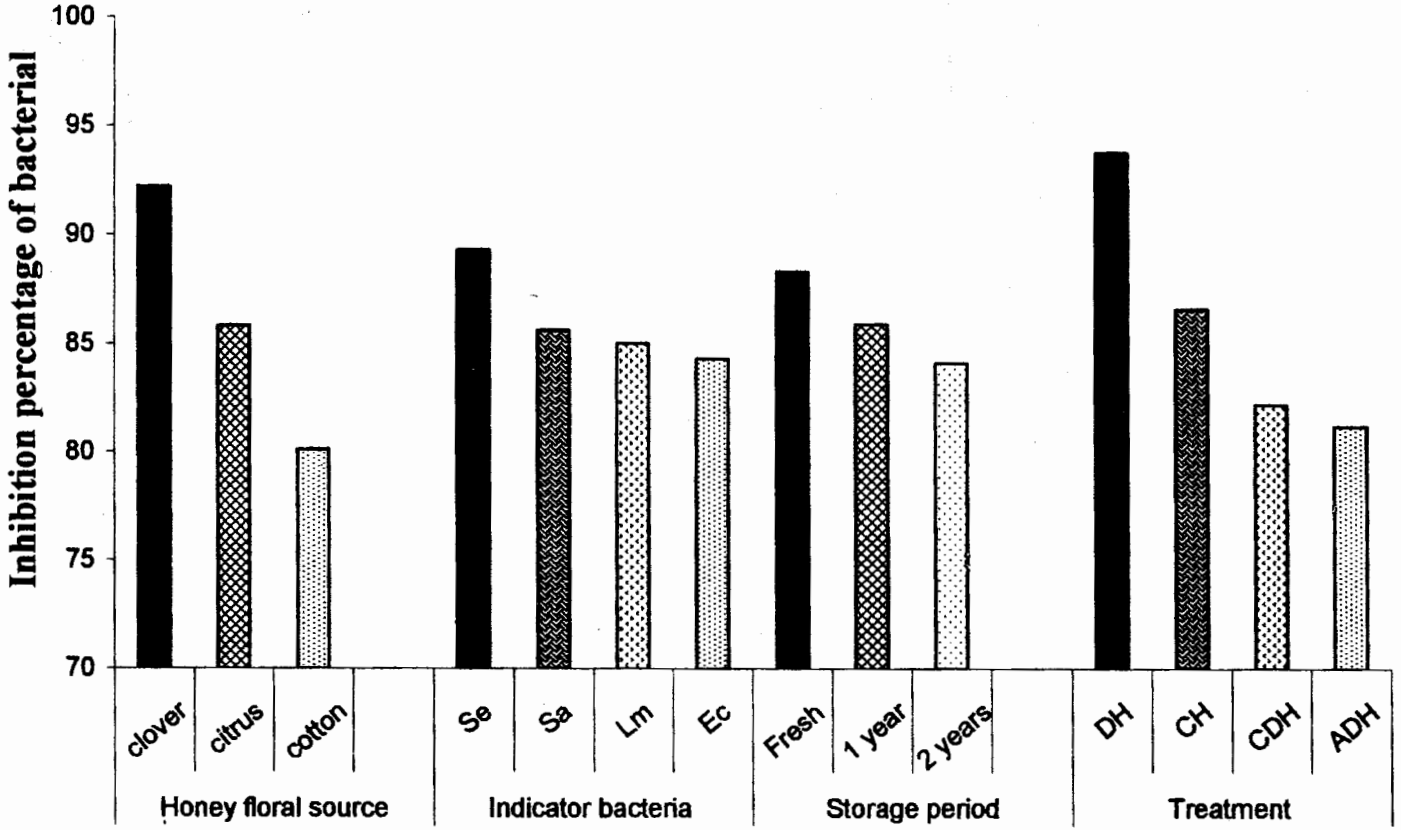


**Fig (1): Antibacterial activity of each factor regardless of other factors or treatments.**
***See Table [2] for abbreviations***

**Table (3): Relative contribution of peroxide and nonperoxide antibacterial agents in the total antibacterial activity (AA) of three fresh unifloral Egyptian honeys**

| | Antibacterial activity of water diluted honey (decreasing in initial viable count %) | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| | Total AA[1] | | Contribution of: Peroxide AA[2] | | Non peroxide[3] | | Heat sensitivity: Thermolabile AA[4] | | Thermostable AA[5] | |
| | AA | % | AA | %[6] | AA | % | AA | % | AA | % |
| Citrus | 94.8 | 100 | 10.0 | 10.5 | 84.8 | 89.5 | 11.3 | 11.9 | 83.5 | 88.1 |
| Clover | 99.1 | 100 | 6.9 | 7.0 | 92.2 | 93.0 | 7.6 | 7.7 | 91.5 | 92.3 |
| Cotton | 94.7 | 100 | 17.4 | 18.4 | 77.3 | 81.6 | 18.8 | 19.9 | 75.9 | 80.1 |
| Average | | 100 | | 12.0 | | 88.0 | | 13.2 | | 86.8 |

See Table 2 for abbreviations

(1) AA of DH (2) AA of DH-AA of CDH (3) AA of CDH

(4) AA of DH-AA of ADH (5) AA of ADH (6) related to total AA

It was reported that AA of most honeys depends mainly on the enzymatic generation of hydrogen peroxide, and the phytochemical (non- peroxide) components make only a minor contribution to the AA of honey (Molan, 1992a). It was also reported that for a few honeys, unidentified non-peroxide components make a major contribution (Molan, 1992b). The manuka honey from the plant *Leptospermum scoparium* grown in New Zealend had AA being of phytochemical origin and it was suggested that this honey had specific AA due to non-peroxide agents (Molan, 1992b; Cooper *et al.*, 1999; Snow and Manley- Harris, 2004).

In the present study, the relative contribution of both peroxide and non-peroxide antimicrobial agents to the total AA of the fresh honeys was determined and the results are given in Table 3. These results suggest that the AA of the three unifloral Egyptian honeys is mainly attributed to non-peroxide antibacterial agents which their contribution is 88% in average, while the contribution of hydrogen peroxide is only 12% to the total AA of these honeys. To the authors' knowledge, this fending was not reported in the previous studies on Egyptian honeys. The results also indicate that the contribution of the thermostable antibacterial components in honey is 86.8% in average, while the contribution of the thermolabile components is only 13.2% to the total AA. According to Postmes *et al.* (1993) and Cooper *et al.* (1999), honeys with non-peroxide AA are likely to be more effective *in vivo* as compared with honeys with hydrogen peroxide AA which would be partly inactivated by the catalase in tissues and blood.

The emergence of multi – antibiotic resistant bacteria created a lot of concern in the medical field; hence there is a need to find an alternative to counter these multi – antibiotic resistant bacteria. In the present study, the AA of the fresh clover honey, which showed the highest antibacterial activity, was compared with the AA of 16 different antibiotics. The results in Table 4 show that the clover honey exhibited AA comparable to that exhibited by the tested antibiotics. Moreover, AA of water diluted honey was generally higher compared to that of the tested antibiotics. However, these results should be regarded as indicative rather than conclusive since two different methods were used to assay AA and different doses were applied. In a study by Farouk *et al.*, (1988), honey was found to be more effective as antibacterial agent against *Pseudomonas* and *Staphylococcus* strains than the antibiotic, gentamicin. Karayil *et al.* (1998) also found that honey at concentrations of 30-50% was superior to cephaloridine and gentamicin in inhibiting growth of nine pathogenic bacteria.

In conclusion, all the three unifloral Egyptian honeys tested in this study exhibited high AA against all the four pathogenic

bacteria used, and this activity is due mainly to non- peroxide antibacterial agents. In addition, the AA of these honeys can with stand storage at room temperature for 24 months and autoclaving at 121°C for 15 minutes. Among these honeys, clover honey appeared to deserve further investigations, since it may prove to be a promise natural food preservative or/ and a valuable therapeutic honey. According to Lusby *et al.* (2005), in Australia, two honeys, Medihoney and Manuka are marketing as therapeutic honeys suitable for use in remedy of ulcers, infected wounds, and burns.

**Table (4): Antibacterial activity of fresh clover honey in comparison to those of 16 antibiotics**

| Antibacterial agent | Inhibation zone diameter (mm) | | | |
|---|---|---|---|---|
| | *E.coli* | *S. entretides* | *L. monacytogenes* | *S. aureus* |
| **a) Clover honey*** | | | | |
| **Crude** | 38 | 31 | 35 | 36 |
| **Water diluted (33% w/v)** | 42 | 38 | 43 | 40 |
| **Autoclaved** | 35 | 28 | 31 | 29 |
| **b) Antibiotics**** | | | | |
| **Streptomycin 10µg** | 28 | 30 | 19 | 36 |
| **Ampicillin 10µg** | 28 | 30 | 28 | 38 |
| **Erthryomycin E 10µg** | 20 | 28 | 13 | - |
| **Neomycin 30µg** | - | 28 | 14 | 32 |
| **Chloramphenicol 30µg** | 7 | 25 | 22 | 25 |
| **Zinnnat cmx 30µg** | 20 | 16 | - | 26 |
| **Germycin 10µg** | 24 | 28 | - | 32 |
| **Augmentin 30µg** | 30 | 35 | 30 | 46 |
| **Rocephen 30µg** | 28 | 18 | 7 | 30 |
| **Pyopen 100µg** | 36 | 22 | 12 | 22 |
| **Rifadn 30µg** | 8 | 24 | 17 | 16 |
| **Colistin sulfate 10µg** | 18 | 24 | 15 | 32 |
| **Claforan 30µg** | 18 | - | - | 18 |
| **Amiks 30µg** | 30 | 22 | - | 38 |
| **Penicillin 10µ** | - | - | - | 20 |
| **Negram 30µg** | 19 | 25 | - | 8 |

* Well-difusion assay (8mm well with 200 µl honey)
** Disc diffusion assay

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# تقييم النشاط المضاد للبكتريا لبعض أنواع من عسل النحل أحادي المصدر النباتي

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تضمنت هذه الدراسة ثلاث نقاط بحثية، الأولى تهدف إلى تقدير بعض الخصائص الفيزيوكيميائية، وكذلك تقييم النشاط المضاد للبكتريا لثلاث انواع مصرية من عسل النحل أحادي المصدر النباتي (الموالح، البرسيم، القطن) ومدى تأثر هذا النشاط بتخزين العسل أو بتخفيفه بالماء (٣٣% وزن/حجم) أوبتعقيمه في الأوتوكلاف (١٢١ °م لمدة ١٥ دقيقة). بصفة عامة كانت الخصائص المختبرة لانواع العسل الطازجة مطابقة للمواصفات القياسية الدولية، ولكن تخزين العسل لمدة ١٢-٢٤ شهرا أدى الى زيادة كل من اللون، الحموضة والهيدروكسي ميثيل فورفورال، بينما لم يتأثر كل من معامل الإنكسار، الماء النشط، المواد الصلبة الذائبة، التوصيل الكهربي والأس الهيدروجيني. وقد أظهرت النتائج أن أنواع العسل الثلاثة ثبطت نمو بكتريا الاختبار الأربع بدرجات متفاوتة وذلك يعتمد على نوع العسل وسلالة البكتريا، وكان أعلى نشاط لعسل البرسيم يلية عسل الموالح ثم عسل القطن على التوالى، وكانت بكتريا *Sa. antertidis* هي الأكثر حساسية يليها بكتريا *Staph. aureus*، *L. monocytogenes* و *E.coli* على التوالى. وقد تبين ان تخزين العسل في الظلام عند درجة حرارة الغرفة (٢٥ ± ١٠ م) لم يسبب الا انخفاضا طفيفا في النشاط المضاد للبكتريا مقدارة ٢,٦ % بعد التخزين لمدة ١٢ شهرا، ٤,٦% بعد التخزين لمدة ٢٤ شهرا، بينما أدى تخفيف العسل بالماء الى زيادة في نشاطه المضاد للبكتريا مقداره ٨,٣% في المتوسط، كما أن التعقيم في الأوتوكلاف لم يسبب فقدا كبيرا في النشاط المضاد للبكتريا حيث كان مقداره ١٣,٥% في المتوسط.

والنقطة البحثية الثانية كانت تهدف إلى دراسة المساهمة النسبية لكل من فوق أكسيد الهيدروجين ومكونات العسل غير الفوق اكسيدية في النشاط المضاد للبكتريا في العسل الطازج. وقد أوضحت النتائج أن نشاط هذه الأنواع من العسل يعتمد أساسا على نشاط مكونات العسل غير فوق اكسيدية حيث بلغت نسبة مساهمتها ٨٨% بينما مساهمة فوق أكسيد الهيدروجين كانت ١٢% فقط في المتوسط، وكذلك تبين أن مكونات العسل المضادة للبكتريا والمتحملة للحرارة تساهم بنسبة ٨٦,٨% من نشاط العسل المضاد للبكتريا في المتوسط، بينما المواد المضادة للبكتريا والحساسة للحرارة تساهم بنسبة ١٣,٢% فقط.

والنقطة البحثية الأخيرة كانت مقارنة النشاط المضاد للبكتريا لعسل البرسيم الطازج مع تأثير ستة عشر مضادا حيويا، وقد أظهرت نتائج هذه المقارنة أن عسل البرسيم الطازج خاصة بعد تخفيفه بالماء قد لايقل فى تأثيره عن تأثير المضادات الحيوية المختبرة وقد يزيد تأثيره عنها في بعض الحالات. والخلاصة أنه من بين أنواع العسل المصري المختبرة فإن عسل البرسيم يستحق المزيد من البحث حيث يمكن أن يكون له قيمة اقتصادية كبيرة عند استخدامه كمادة حافظة طبيعية لبعض الاغذية وتكون لة قيمة أكبر عند استخدامه كمستحضر لعلاج بعض الأمراض.