# Honey Compared with Some Antibiotics against Bacteria Isolated From Burn-wound Infections of Patients in Ain Shams University Hospital

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**Abstract:** Honey is an ancient remedy for the treatment of infected wounds. Four types of honey (Citurs, Clover, Nigella and eljabaly) were used. Six different species of bacteria were isolated from 120 burn- wound patients in Ain Shams University Hospital, namely *Aeromonas schubertii, Haemophilius paraphrohaemlyticus, Micrococcus luteus, Cellulosimicrobium cellulans, Listonella anguillarum and Acinetobacter baumannii.* A comparative study between the known groups of 18 antibiotics and honey was carried out to evaluate the importance of using honey in burn-wound treatment on the 6 isolated species and compared with the effect of different types of honey on the same becateria. It was found that eljabaly has strong inhibitory effect in comparison to other mentioned types. Concentration of 25% of eljablaly showed inhitbition of 4 types, whereas 30% was potent enough to destroy the 6 isolated bacteria. Our data were that its antibacterial activity was attributed to its high osmolarity and hypertonic sugar concentration and low pH values. Amino and fatty acids, total proteins patterns were significantly changed. Total lipids of bacterial species was sharply decreased. [Journal of American Science. 2010;6(10):301-320]. (ISSN: 1545-1003).

**Keywords:** Patients, honey, antibiotics, bacteria, lipids, proteins, amino and fatty acids.

## 1. Introduction

Antibacterial activity of honey against bacteria isolated from burn-wound infections of some patients was reported, identification, characterization, using traditional and Biolog microplate automated system, analysis of fatty acids, cellular protein, amino acids were performed. A comparative study between the known groups of antibiotics and honey was carried out.

Honey has been used in the treatment of wounds for over 2.000 years. However, one of the most important properties seems to be its antibiotic action. There are many reports in the clinical literature of honey being used with success in treatment of a wide range of burn wound infection, it can be seen that the effectiveness of honey in many of its medical uses is probably due to its antibacterial activity. It inhibits a broad spectrum of bacterial species (Molan, 2001).

Antibacterial activity and mode of action of honey was studied (Cooper et al., 2002; Edward and Greenwood, 2003). The role of wound management procedure, risk factors associated with infection, typical bacterial pathogens and their associated exotoxins, antibiotic resistance were discussed (Molan, 2000).

The antibacterial activity of honey refers to some bee products, presence of "inhibin" which acts as an antibacterial factor other than $H_2O_2$, several factors such as osmotic properties of honey which is saturated or super saturated solution of sugars, 84% being a mixture of fructose and glucose, so inhibition by osmotic effect of dilute solutions of honey obviously depends on the species of bacteria.

The major antibacterial activity in honey has been found to be due to hyobogen peroxide ($H_2O_2$) produced enzymatically in the honey. Its pH being between 3.2 and 4.5, which is low enough to be inhibitory to many animal pathogen, and thus the acidity is significant antibacterial factor Oryan, 1998; Abutharfeil et al., 1999; Postmes and Vandeputte, 1999; Weston et al., 1999, and Shamala et al. (2000).

It was reported that *Pseudomonas* and *Proteus* species are inhibited by honey and also Clostridium species, *Staphylococcus aureus, Pseudomonas and Escherichia coli* (Hegazi et al., 2001; Al-Jabri et al., 2003). The chemical composition of honey is about 82% carbohydrates, enzymes, 18 free amino acids, minerals, vitamins (Kathleen et al., 2004).

Honey is in fairly widespread use as a topical antibacterial agent for the treatment of wounds, burns

and skin ulcers, there are many reports of its effectiveness (Mclnerney, 1990). The observations recorded are that inflammation, swelling and pain are quickly reduced.

**2. Material and Methods**

I. Samples of honey:

Samples of monofloral honeys (citurs, clover, nigella, and eljabaly) were obtained from the apiary of the experimental station of the faculty of Agriculture, Cairo University. Chemical analysis of honey samples was performed in Chemical Analysis Lab. of Honey Products, Beekeeping Research Department, Agriculture Research center.

II. Collection and transport of bacterial samples:

The study was performed on 120 patients who suffer from burn wound infections from Ain Shams University Hospital. Samples were collected and transported to laboratory according to Chessbrough (1984). By using a sterile cotton swab, samples were collected from wound, immersed in a container of transport medium. On collecting samples from wounds, special precaution should be taken to prevent contaminating specimen with commensal organisms from the skin.

III. Isolation of bacteria:

Culture media: Samples were cultivated on different media as follows:

(1)Nutrient agar medium: This media is a basic culture medium used in the preparation of blood agar and other media. This medium used for cultivation and maintenance of all isolated bacteria.

The medium is composed according to Atals, 1993. Medium containing (per liter of distilled water): 15g agar, 5g peptone, 5g Nacl, 5g yeast extract, 1gm beef extract, at pH 7.4 $\pm$ 0.2

(2) Blood agar medium: It is a tryptic soy agar obtained from Difco, USA with 5% sterile human blood (Damron *et al*., 1986).

(3) MacConkey agar medium: MacConkey agar is a differential and low selectivity medium used to distinguish lactose fermenting from non-lactose fermenting bacteria.

a) Lactose fermenting bacteria:

These were detected as pink to red colonies surrounded by red zone due to lactic acid fermentation. This character was observed in different species, e.g. *Klebsiella pneumoniae* with mucoid colonies and E. coli.

b) Non- lactose fermenting bacteria:

These were detected as colourless colonies, medium turn to yellow colour. *Pseudomonas aeruginosa*, *Salmonella* species, *Shigella* species and *Proteus mirabilis* belongs to this group.

There are some bacteria which do not grow on MacConkey agar e.g. *Streptococcus pyogenes*, *Streptococcus penumoniae*, Viridans species and *Pasteurlla* species. MacConkey agar is the most economically prepared from the dehydrated medium available from Oxoid Ltd. (Finegold and Martin, 1982).

(4) Mannitol salt agar: Mannitol salt agar is a differential and selective plate medium used to isolate *staphylococcus aureus*. The medium is available in dehydrated form Oxoid Ltd. Mannitol is fermented by *Staphylococcus aureus* (yellow in medium) (Finegold and Martin, 1982).

(5) Cetrimide agar: Cetrimide agar is a selective plate medium used occasionally to isolate Pseudomonas species from a mixed bacterial flora. It inhibits the growth of bacteria such as *Staphylococcus aureus* and Coliform. Cetrimide agar in dehydrated form is available from Gibco Ltd. Pseudomonas agar medium (cetrimide agar) containing acetyltrimethylamine bromide (cetrimide), except *Pseudomonas aerugionsa* all bacterial species were inhibited on this selective media. *Pseudomonas aeruginosa* produced green pigment after prolonged incubation (Finegold and Martin, 1982).

IV. Identification of bacterial isolates:

- After the purification of all bacterial isolates, the selected colonies were identified according to Bergey's manual of systematic bacteriology (Krieg and Holt, 1984 and Sneath *et al*., 1986).
- Biolog automated microplate system:

The Biolog automated microplate system was performed for identification of the selected isolates (Bochner, 1989). The system is based on the reactions to a series of 95 carbon sources, including sugars, organic acids, and amino acids that were indicated by color reactions. The Biolog automated identification system consists of microstation computer, turbidimeter (optical density at 590 nm), microlog software, microplate reader, and Gram-positive and Gram-negative microplates.

The following steps were adopted:

1- Gram stain: by Jensen's modified method (Cruickshank *et al*., 1975)

2- Preparation of inoculum:

The isolate was grown on an appropriate agar medium as either Biolog universal growth medium (BUGM) or tryptic soy agar (TSA) and stationary incubated for 4-18 hours at 28-30$^0$C (Bochner, 1991).

3- Preparation of microplate:

Wells of the microplate were filled with 150 of $\mu$ 1 of bacterial suspension of the selected isolate by the multi-channel pipette reservoir. Microplate was then covered with its lid and incubated at 28-30$^0$C for 24 hours under humid conditions to avoid the dehydration of the outer wells of the microplate. Microplate reading was taken by a 590 nm filter on a microplate reader. Results were analyzed with Biolog GN (Gram G-ve) or GP (G+ve) data base to determine the identity of the bacterial isolate.

For differentiation of genera, species and subspecies, Biolog's cluster analysis program was used, indicating relationship between various groups of bacteria.

a) Morphological characterization of isolates and physiological, biochemical characterization of isolates were studied (Cheesbrough, 1984)

V. Sensitivity and resistance of bacterial isolates to antibiotics and honey samples:

(A) Antibiotic sensitivity test

Materials:

1) Muller Hinton agar was obtained from Oxoid.

2) Antibiotic discs obtained from Oxoid.

Method:

1- Bacterial inoculation ($10^5$ c.f.u./ml) was transferred into the surface of Muller-Hinton agar medium and followed by addition of antibiotic discs.

2- The antimicrobial activity of all compounds was determined by discs diffusion method (NCCLS, 1993).

3- Inhibtion zones were measured (mm) after 24 hours incubation at 37$^0$C (Rubinstein *et al*., 1986).

The antibiotics discs were obtained from Oxoid Ltd. As shown in the following table.

Antibiotic discs used for sensitivity test:, .

| Antibiotic group | Conc. (mcg) | Scientific name | Symbol | Trade name |
|---|---|---|---|---|
| Penicillin group | 10 | Ampicillin | AM | Ampicillin |
| | 30 | Amox./clavulanate | AUC | Augmentin |
| | 10 | Penicillin | P | Penicillin |
| | 100 | Piperacilin | PPL | Pipril |
| | 100 | Carbenicillin | PY | Pyopen |
| Sulbactam | 30 | Sulbactam/Ampicillin | SAM | Unasyn |
| Cephalosporin group | 30 | Cefradine | CE | Velosef |
| | 75 | Ceforperazone | CEP | Cefobid |
| | 30 | Cefotaxime | CTX | Claforan |
| | 30 | Ceftazidime | CAZ | Fortum |
| | 30 | Ceftriazone | CRO | Rocephin |
| Monobactam group | 30 | Aztreonam | AZ | Azactam |
| Aminoglycoside | 30 | Amikacin | AK | Amikin |
| | 10 | Gentamycin | GM | Garamycin |
| | 10 | Topramycin | NN | Nebsin |
| Quinolone group | 5 | Ofloxacin | OFX | Tarivid |
| | 5 | Ciprofloxacin | CFX | Ciprofloxacin |
| Furans | 300 | Nitrofurantoin | F/M | Nitofurantoin |

(B) Sensitivity and resistance of bacterial isolates to honey samples

Eight concentrations using distilled sterile water were made from 5% to 40% in order to evaluate the antibacterial activity against the selected bacterial isolates through estimation of minimum inhibitory concentration (MIC) method according to NCCLS, 1993.

VI. Extraction and analysis of lipids

A) Extraction and purification of total lipids:

The lipids were extracted according to Kates and Eberhardt (1957)

B) Estimation of total lipids:

Estimation of total lipids of honey- treated bacterial isolates and that untreated (control) was performed according to the method adopted by Kates and Eberhards (1957).

C) Analysis of methyl esters of fatty acids by gas-liquid chromatography

Methylation of the fatty acids

To lipid sample 20 m1 of methanol. 10 m1 of benzene and 1 ml of concentrated sulfuric acid were added then refluxed for 90 minutes in a round flask on a water bath, the methyl esters obtained were extracted with petroleum ether (b.p. 40-60 C). the petroleum ether was then evaporated, the residue was dissolved in chloroform (Harbone, 1984) and the methylated samples were subjected to analysis by GLC in GVC Pyeunican gas-liquid chromatograph equipped with dual flame ionization detector and dual channel records (Faculty of Agriculture, Cairo University).

Sources of standard fatty acids:

A set of standard methyl esters of fatty acids including both saturated and unsaturated ones with a state of purity not less than 99% was purchased from Sigma Chemical Company St. Louis, Mo 63178, USA, and used as authentic materials to characterize the unknown fatty acids.

VII. Determination of total soluble protein

Total soluble protein was determined according to the method of Bradford (1976). A calibration curve was constructed using bovin serum albumin (BSA).

Analysis of the total amino acid contents by HPLC

The method applied was made according to Cohen *et al*. (1989)

VIII. SDS ploy acrylamide gel electrophoresis (SDS-PAGE)

The electrophoresis SDS gels were prepared by Laemmli, (1970) as modified by studier, (1973).

The stock solutions used for protein electrophoresis were as follows:

A. For resoving gel (100 ml)

| | |
|---|---|
| Acrylamide | 30.0g |
| Bis- Acrylamide | 0.8 g |
| $H_2O$ (dist.) | Up to 100ml |

B. for stacking gel (100 ml)

| | |
|---|---|
| Acrylamide | 30.0g |
| Bis-Acrylamide | 1.0g |
| $H_2O$ (dist.) | Up to 100 ml |

C. Resolving gel buffer (4 X Tri. PH 8.4, $4^0$C

| | |
|---|---|
| Tris | 18.15 g |
| HCI (conc.) | $3.50^0$ ml |
| $H_2O$ (dist.) | Up to 100m1 |

**3. Results and Discussion**

I. Physical and chemical prosperities of samples of honey

Tables 1 and 2 show the Physical and chemical properties of different kinds of honey used in this research. It was found that the moisture was ranging from 16% for Eljabaly to 21% for Citrus honey. Data also show that the Eljabaly honey was shown to be more viscous and more acidic than the other types. The lowest amount of total sugar content was recorded for Clover honey (68.38%), while the highest value was found in Eljabaly honey (86.16%), Similar study was carried out by Esmail et al., (1990) on a sample of Egyptian honey of unidentified floral source and found that it was containing 0.3% protein, 78.5% carbohydrate, 0.34% fat, 18.7% water, 0.17% ash, 6.6 mg/100 calcium and 0.82mg/100g iron. Also Beattite and Mayze (2000) reported that honey is produced by bees from floral nectar as a saturated solution of sugars and consists of 84% mixture of fructose and glucose. Honey has low water content (15-21% by weight) and is acidic (pH 3.2-4.5)

The study revealed that there is a marked difference in the chemical constitution of honeys from different monofloral sources as recorded by many authors who had analyzed numerous honey types. The results show that the highest value of fructose was recorded for Eljabaly honey type (40.27%), while the lowest one was 33.61% for Clover honey type. The total sugar content reaches the highest value 86% for Eljabaly honey type, while

it was 68.38% for Clover honey type. The protein values recorded in this study ranges from 5.25% in Citrus honey to 6.92% in Eljablaly. It was also observed that Nigella honey type contains the highest value of lipids (3.76%). The variation in the physical and chemical properties of honey is due to the floral source as recorded by many workers (Molan, 1999 and Kathleen *et al*., 2004). They reported that the chemical composition of honey is about 82% carbohydrates. These are fructose and glucose (70%); 9% sucrose, maltose, isomaltose, maltulose, turansoe and kojibiose, and 4% erlose, theanderose and panose. There are many more: proteins and amino acids (enzymes such as invertase, amylase, glucose oxidase, catalase and 18 free amino acids, of which the most abundant is proline.

Then there are the vitamins, minerals and antioxidants (vitamins ribovlavin, niacin, folic acid, pantothenic aid and vitamin B6, ascorbic aid (vitamin C), the minerals calcium, iron, zinc, potassium, phosphorous, magnesium, selenium, chromium and manganese, and antioxidant flavonoids, of which one pniocembrin, is unique to honey

The high acidity of honey also plays an important role in the system which prevents bacterial growth. The pH of honeys may vary from approximately 3.2 to 4.5 (average $P_H$=3.9) making it inhospitable for attack by most bacteria. Honey has an antibacterial of inhibine system. Bees add an enzyme glucose oxidase to honey and this enzyme reacts with glucose to produce hydrogen peroxide and gluconic acid, both of which have an antibacterial effect.

II. Identification of bacterial isolates:

The identification of bacteria isolated from 120 patients who suffer from burn wound infections revealed that they belong to 6 species (Table 3), namely *Aeromonas schubertii, Haemophilus paraphrohaemlyticus, Micrococcus luteus, Cellulosimicrobium cellulans, Listonella anguillarum, and Acinetobacter baumannii*. The identification process was carried according to Bergey's manual systemtic bacteriology (Krieg and Holt, 1984 and Sneath *et al*., 1986)

III. Sensitivity and resistance of bacteria isolates to antibiotics and honey samples:

Table 4 shows that *Aeromonas schubertii* is resistant to the most antibiotics used in this experiment except Ceforperazone, Ofloxacin and Ciprofloxcaicn. This bacterial species were intermediately affected by Cefradine. These results also indicate that *Haemophilus paraphrohaemlyticus* is sensitive to Gentamycin, Ofloxacin, Ciprofloxacin and Nitrofurantion, but it shows an intermediately effect by only three antibiotics namely Ampicillin, Amox./Clavulanate and Sulbactam/Ampicillin.

*Micrococcus luteus* shows resistance against Sublactam/Ampicillin, Cefradine, Ceftraixone, Aztreonam and Nitrofurantoin, whereas the other rest antibiotics affect greatly on the growth of such bacterial species.

Both *Cellulosimicrobium cellulans* and *Listonella anguillarum* show sensitivity to 5 types of antibiotics, while *Acinetobacter baumannii* was sensitive to 7 antibiotics as shown in Table4:

These findings reflect to variation of effect of 18 antibiotics on the isolated bacterial species, in order to compare with the influence of the different honey types used in this study for evaluation its antibacterial activity. The tables 5, 6, 7, 8 and 9 revealed that the honey type eljabaly was the strongest as compared with the other three types of honey used in this study. It was observed that 25% of Eljabaly was enough to prevent the growth of 4 bacterial species; this means that 30% of this honey can destroy all the 6 studied bacterial species. The data obtained from this experiment explain that honey has strong antibacterial activity due to its hygroscopic proerties and low pH values as recoreded by Efem, 1988

Honey may work in clearing infection is through an activating effect on the body's immune system as it has been reported that stimulates mitogenesis in B and T lymphocytes and activates neutrophils.

The acidity of honey also help with oxygenation, as acidification of wound increases the release of oxygen from hemoglobin. Honey prevents partial-thickness burns from converting to full-thickness burns needsing skin grafts. Honey select to have good antibacterial activity provides treatment option worthy of serious consideration, especially on infected and recalcitrant wounds. On burns its antibacterial and anti-inflammatory properties allow a moist healing environment to be maintained that protects the wound from deterioration and fibrosis.

The sensitivity to honey of Gram-positive cocci of clinical significance isolated from wounds.

The mode of action of honey has not yet been fully elucidated but osmolarity, acidity, hydrogen peroxide generation and photochemical components are considered important (Molan 1992).

Honey does not adversely affect human tissue (Molan 1999) not only has it the potential to limit the growth of wound pathogen, but there is evidence that

honey has the potential to promote healing (Molan 1999, Tonks *et al*., 2001).

IV. Effect of Elijably honey type on total lipids and total proteins of the bacterial species

Table 10 shows that the total lipid of bacterial species was sharply decreased on treating the bacterial cells with honey type.

Table 11 shows the variation of total proteins as a result of treating bacterial cells with Eljabaly honey type (Molan, 1999).

V. Effect of Eljabaly honey type on Fatty acids pattern of the bacterial species

11 fatty acids were recorded for all bacterial species. It was observed from Table 12 that some fatty acids were only detected in control sample and completely disappears in the treated samples. It was also observed that the percentage of some fatty acids was decreased or completely disappers in some bacterial species these results agreed with (Bogdanov, 1997 and Al- Jabri *et al*., 2003).

VI. Effect of Eljabaly honey type on amino acids pattern of the bacterial species:

Tables 13, 14, 15, 16, 17, 18 show the variation of amino acids content of bacterial species.

VIII. Polyacrylamide gel electrophoresis of cellular proteins of control and treated cells of bacterial species:

The protein profiles obtained from electrophoretic analysis of 6 bacterial species was shown in table 19. The total number of protein bands was 37. It was observed that all the control samples of bacterial species were characterized by higher number of bands 32, 34, 29, 30, 33, and 30 bands as compared with the treated samples with honey, where they were 30, 28, 25, 27, 26 and 25 bands. This means that at least 2 to 7 protein types were failed to be synthesized on treating the bacterial cells with honey types.

It was concluded that most of the characterized bands were located in the region between M.wt. 97 and 24 k Da, where above 97 K Da the bands number were few and the intensity was faint well, Moreover, the bands below 24k Da were about 7 compressed and not well organized. It was observed that control samples of bacterial species were representing the same characterizing bands at M.wt of 102, 97 and 85, 64, 56,49, 38, 29, and 25 k Da in varying degrees. On the other hands these bands were clearly decreased in treated cells where the lowest intensity were located at M.wt. 97, 85, 71, 55, 43, 32 and 24 k Da. The most characteristic features of these bacterial isolates is the apperarnce of at least 17 bands located in the region between M.wt 116 and 9 k Da. While in the treated samples were characterized by decrease or absence of protein bands in such range between 126 and 9 K Da.

The results of this work are already providing a better understanding of the effect of honey as strong antibacterial agent against the studied bacterial species in several ways through direct effect on the metabolic activity through biosynthesis of fatty and amino acids similar study was conducted by Gilmour *et al*., 2000.

Similar results were reported by Kathleen *et al*., 2004 who concluded that the chemical composition of different types of honey indicates the abundance of proline beside other 18 amino acids and enzymes such as invertase, amylase, glucose oxdase and catalase that may affect greatly the formation of antibacterial system inhibin that can interfere the amino acids pathway of bacterial species. It can be concluded that honey has been found to control wound infections and accelerate wound healing.

**Figure (1):** SDS-page of total protein isolated from 6 species (1,3,5,7,9,11) as a control and (2,4,6,8,10 and 12) as treated, M refers as protein standard.

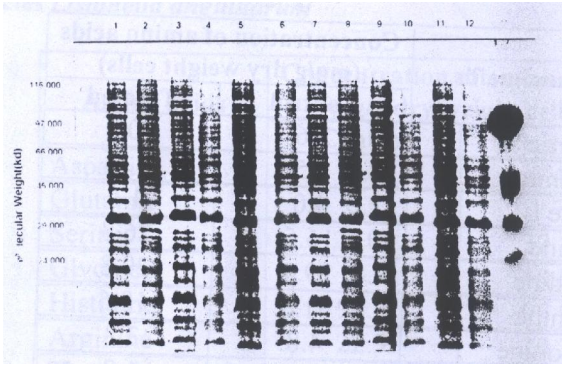

**Table (l):** Physical properties of samples of Honey

| Parameter | Honey Type | | | |
|---|---|---|---|---|
| | Citrus | Clover | Nigella | Eljabaly |
| Moisture | 21% | 19.51% | 17.52% | 16% |
| Specific gravity | 1.32 | 1.39 | 1.43 | 1.42 |
| Viscosity | 13.83 poise | 13.62 poise | 1521 poise | 18.81 poise |
| Granulation | Few | Few | Few | Few |
| *E.C | 0.022% | 0.014 % | 0.020% | 0.026% |
| **T.S.S | 71% | 80% | 83.52% | 85.31% |
| fermentation | 17% (safe) | 18% (safe) | 17% (safe) | 17% (safe) |

*E.C = Electrical Conductivity
**T.S.S = Total Soluble Solid

**Table (2):** Chemical composition of samples of Honey

| Parameter | Honey Type | | | |
|---|---|---|---|---|
| | Citrus | Clover | Nigella | Eljabaly |
| PH | 3.61 | 3.72 | 3.46 | 2.93 |
| Protein | 5.25 | 5.84 | 6.26 | 6.92 |
| Lipid% | 2.93 | 2.78 | 3.76 | 3.48 |
| Fructose% | 35.19 | 33.61 | 37.41 | 40.27 |
| Glucose% | 27.13 | 24.76 | 28.13 | 32.18 |
| Sucrose% | 3.54 | 5.17 | 4.51 | 7.23 |
| Maltose% | 5.48 | 4.84 | 4.85 | 6.32 |
| *H.M.F mg/kg | 9.88 | 12.71 | 7.68 | 23.04 |
| Fe mg/kg mg/lOOg | 24.89 | 33.17 | 27.22 | 91.21 |
| Na mg/kg mg/lOOg | 30.13 | 32.12 | 37.77 | 41.2 |
| K mg/kg mg/lOOg | 59.17 | 77.13 | 62.26 | 93.16 |
| Ca mg/kg mg/lOOg | 72.82 | 66.57 | 70.49 | 75.41 |
| P mg/kg mg/lOOg | 22.18 | 27.74 | 21.14 | 34.19 |

*H.M.F = Hydroxy Methyle Furfural

**Table (3):** Bacterial species isolated from burn wound infection according to biology microplate system.

| Sample No. | Gram-stain type | Bacterial species |
|---|---|---|
| 1 | + ve | Micrococcm luteus |
| 2 | - ve | Aeromonas schubertii |
| 3 | - ve | Haemophilus paraphrohaemlyticus |
| 4 | + ve | Micrococcus luteus |
| 5 | + ve | Micrococcus luteus |
| 6 | + ve | Cellulosimicrobium cellulans |
| 7 | - ve | Listonella anguillorum |
| 8 | - ve | Acinetobacter baumannii |

| | | |
|---|---|---|
| 9 | + ve | Cellulosimicrobium cellulans |
| 10 | - ve | Acinetobacter baumannii |

Table (4): Antibiotic bioassay of different bacterial species

| Antibiotic | Symbol | Bacterial species | | | | | |
|---|---|---|---|---|---|---|---|
| | | I | II | III | IV | V | VI |
| Penicillin | | | | | | | |
| Ampicillin | AM | R | I | S | R | S | R |
| Amox./Clavulanate | AUC | R | I | S | I | S | R |
| Penicillin | P | R | R | I | R | R | R |
| Piperacillin | PPL | R | R | I | R | R | R |
| Carbenicillin | PY | R | R | I | R | R | R |
| Sulbactarn/ Ampicillin | SAM | R | I | R | S | I | R |
| Ccphalasporins: | | | | | | | |
| Cefradine | CE | I | R | R | R | R | S |
| Cefoperazone | CEP | S | R | S | I | I | R |
| Cefotaxime | CYX | R | R | S | R | I | R |
| Ceftazidime | CAZ | R | R | S | S | R | R |
| Ceftriaxone | CRO | R | R | R | R | R | S |
| Monobactams: | | | | | | | |
| Aztreonam | AZ | R | R | R | S | R | S |
| Aminoglcosides: | | | | | | | |
| Amikacin | AK | R | R | S | S | R | S |
| Gentamycin | GM | R | S | I | I | I | R |
| Tobramycin | NN | R | R | I | S | S | S |
| Quinolones: | | | | | | | |
| Ofloxacin | OFX | S | S | S | R | S | R |
| Ciprofloxacin | CFX | S | S | S | R | I | S |
| Furans: | | | | | | | |
| Nitrofurantion | F/M | R | S | R | R | R | S |

R = Resistant I = Intermediate S = Sensitive
I = Aeromonas schubertii
II =Haemophilus paraphrohaemlyicus
III=Micrococcus luteus
IV =Cellulosimicrobium cellulans
V = Listonella anguillarum
VI =Acinetobacter baumannii

**Table (5):** Evaluation of the Minimum Inhibitory concentration (MIC) of citrus honey against bacterial species.

| Bacterial species | Concentration of honey | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 5% | 10% | 15% | 20% | 25% | 30% | 35% | 40% |
| Aeromonas schubertii | R | R | R | R | R | R | R | S |
| Haemophilus paraphrohaemlyticus | R | R | R | R | R | R | S | S |
| Micrococcus luteus | R | R | R | R | R | R | S | S |
| Cellulosim icrobium cellulans | R | R | R | R | R | R | S | S |
| Listonella anguillorum | R | R | R | R | R | R | R | S |
| Acinetobacter baumannii | R | R | R | R | R | R | R | S |

**Table (6):** Evaluation of the Minimum Inhibitory concentration (MIC) of Clover honey against bacterial species.

| Bacteria1 species | Concentration of honey | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 5% | 10% | 15% | 20% | 25% | 30% | 35% | 40% |
| Aeromonas schubertii | R | R | R | R | R | R | R | S |
| Haein ophilus paraph roll aem lyticus | R | R | R | R | R | R | S | S |
| Micrococcus luteus | R | R | R | R | R | R | R | S |
| Cellulosimicrobium cellulans | R | R | R | R | R | R | R | S |
| Listonella anguillorum | R | R | R | R | R | R | S | S |
| Acinetobacter baumannii | R | R | R | R | R | R | R | S |

**Table (7):** Evaluation of the Minimum Inhibitory concentration (MIC) of NigelJa honey against bacterial species.

| Bacterial species | Concentration of honey | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 5% | 10% | 15% | 20% | 25% | 30% | 35% | 40% |
| Aeromonas schubertii | R | R | R | R | R | R | R | S |
| Haemophilus paraphrohaemlyticus | R | R | R | R | R | R | R | S |
| Micrococcus luteus | R | R | R | R | R | R | R | S |
| Cellulosim icroblum cellulans | R | R | R | R | R | R | S | S |
| Listonella anguillorum | R | R | R | R | R | R | S | S |
| Acinetobacter baumannii | R | R | R | R | R | R | R | S |

**Table (8):** Evaluation of the Minimum Inhibitory concentration (MIC) of Eljabaly honey against bacterial species.

| Bacterial species | Concentration of honey | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | 5% | 10% | 15% | 20% | 25% | 30% | 35% | 40% |
| Aeromonas schubertii | R | R | R | R | S | S | S | S |
| Haemophilus paraphrohaemlyticus | R | R | R | R | R | S | S | 's |
| Micrococcus luteus | R | R | R | R | R | S | S | S |
| Cellulosim icrobium cellulans | R | R | R | R | S | S | S | s |
| Listonella anguillorum | R | R | R | R | S | S | S | s |
| Acinetobacter baumannii | R | R | R | R | S | S | S | s |

R = Resistance S = Sensitive

**Table (9):** Evaluation of the Minimum Inhibitory concentration (MICs) of different types honey against bacterial species.

| Bacterial species | MIC of honey | | | |
|---|---|---|---|---|
| | Cirtrus | Clover | Nigella | Eljabaly |
| Aeromonas schubertii | 40% | 40% | 40% | 25% |
| Haemophilus paraphrohaemlyticus | 35% | 35% | 40% | 30% |
| Micrococcus luteus | 35% | 35% | 40% | 30% |
| Cellulosimicrobium cellulans | 35% | 40% | 35% | 25% |
| Listonella anguillorum | 40% | 35% | 35% | 25% |
| Acinetobacter baumannii | 40% | 40% | 40% | 25% |

**Table (l0):** Effect of honey types on the total lipids (mg/g of fresh weight of cells) of bacterial species.

| Bacterial species | Concentration of lipids (mg/g of fresh weight of cells) | |
|---|---|---|
| | Control | Treated |
| Aeromonas schubertii | 6.44 | 1.07 |
| Haemophilus paraphrohaemlyticus | 10.00 | 1.50 |
| Micrococcus luteus | 28.88 | 1.19 |
| Cellulosimicrobium cellulans | 17.69 | 6.25 |
| Listonella anguillorum | 67.25 | 43.81 |
| Acinetobacter baumannii | 17.63 | 1.36 |

**Table (11):** Effect of honey types on the total lipids (mg/g of fresh weight of cells) of bacterial species.

| Bacterial species | Concentration of lipids (mg/g of fresh weight of cells) | |
|---|---|---|
| | Control | Treated |
| Aeromonas schubertii | 311.34 | 252.74 |
| Haemophitus paraphrohaemlyticus | 347.98 | 300.36 |
| Micrococcus luteus | 339.66 | 245.42 |
| Cellulosimicrobium cellulans | 336.98 | 252.74 |
| Listonella anguillorum | 278.38 | 194.12 |
| Acinetobacter baumannii | 238.08 | 201.46 |

**Table (12):** Constituent fatty acids of the isolated bacterial species

| Fatty acid | Fatty acid percent/ total fatty acid contents | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | (1) | | (2) | | (3) | | (4) | | (5) | | (6) | |
| | C | T | C | T | C | T | C | T | C | T | C | T |
| Caprylic | 5.379 | 0.039 | 5.491 | 0.048 | 0.398 | 14.975 | 0.018 | 2.256 | 0.084 | 1.106 | 0.049 | 0.641 |
| Capric | 0.097 | 0.265 | 0.078 | - | 0.294 | 1.269 | - | 0.058 | 0.198 | 0.173 | 0.072 | 0.156 |
| La uric | 1.820 | 3.148 | 1.197 | . | - | 27.331 | - | 2.841 | 0.840 | 2.823 | 0.229 | - |
| Mvrisfic | 47.959 | 6.482 | - | . | 31.092 | . | 0.072 | 27.569 | 53.391 | ,33.008 | . | . |
| Pentadecylic | 4.007 | 2.761 | 2.994 | - | 0.237 | . | - | 3.815 | 0.199 | 2.743 | - | . |
| Palmitic | 4.098 | 3.876 | 0.971 | 1.414 | 0.236 | 51.253 | 0.670 | 6.328 | 1.521 | 3.305 | . | 6.765 |
| Margaric | 19.403 | 18.330 | 21.773 | 57.866 | . | - | - | - | 32.372 | 21.228 | . | 1.744 |
| Stearic | 12.335 | - | . | - | . | . | - | - | - | - | . | . |
| Linolenic | - | 0.425 | - | . | . | - | . | . | . | . | - | 1.510 |
| Olcic | . | - | 0.773 | - | 1.333 | . | . | 1.079 | . | 0.284 | . | . |
| Arachidic | - | - | - | - | - | - | - | 0.276 | - | 0.177 | - | - |

C = Control T = Treated

1- Aeromonas
2- Haemophilus paraphrophaemylticu
3- Micrococcus luteus
4- Cellulosimicrobium cellulans
5- Listonella anguillarum
6- Acinotobacter baumann

**Table (13):** Total amino acid content of the control and treated species Aeromonas schubertii

| Amino acids | Concentration of amino acids (mg/g dry weight of cells) | |
|---|---|---|
| | Control | Treated |
| Aspartic | 24.1 | 16.6 |
| Glutamic | 7.3 | 13.7 |
| Serine | 9.6 | 8.8 |
| Glycine | 72.4 | 5.4 |
| Histidine | 103.5 | 19.1 |
| Arginine | 9.7 | 6.8 |
| Theronine | 5.0 | 9.2 |
| Alanine | 0.7 | 6.5 |
| Proline | 3.7 | 16.13 |
| Tyrosine | 11.1 | 16.2 |
| Valine | 6.8 | 12.5 |
| Methionine | 10.1 | 21.9 |
| Cystine | 15.0 | 31.3 |
| Isoluecinc | 8.2 | 9.2 |
| Luecine | 7.7 | 14.8 |
| Phenylalanine | 4.2 | 21.7 |
| Lysine | 12.1 | 23.2 |

**Table (14):** Total amino acid content of the control and treated species Haemophilus paraphrohaemlyticus

| Amino acids | Concentration of amino acids (mg/g dry weight of cells) | |
|---|---|---|
| | Control | Treated |
| Aspartic | 3.5 | 33.6 |
| Glutamic | 17.2 | 7.4 |
| Serine | 6.0 | 10.6 |
| Glycine | JL5.0 | 10.8 |
| Histidine | 52.5 | 13.4 |
| Arginine | 27.6 | 12.9 |
| Theronine | 5.2 | 24.4 |
| Alanine | 4.1 | 16.2 |
| Proline | 14.2 | 30.6 |
| Tyrosine | 49.0 | 13.4 |
| Valine | 30.5 | 22.3 |
| Methionine | 24.5 | 20.7 |
| Cystine | 35.1 | 9.4 |
| Isoluecinc | 15.4 | 31.7 |
| Luecine | 16.4 | 16.6 |
| Phenylalanine | 9.5 | 1.3 |
| Lysine | 25.1 | 25.0 |

**Table (15):** Total amino acid content of the control and treated species Micrococcus luteus

| Amino acids | Concentration of amino acids (mg/g dry weight of cells) | |
|---|---|---|
| | Control | Treated |
| Aspartic | 6.2 | 7.4 |
| Glutamic | 16.1 | 7.2 |
| Serine | 13.3 | 19.7 |
| Glycine | 10.6 | 37.4 |
| Histidine | 40.6 | 99.2 |
| Arginine | 13.8 | 37.2 |
| Theronine | 9.1 | 6.9 |
| Alanine | 9.5 | 2.9 |
| Proline | 58.3 | 3.5 |
| Tyrosine | 21.1 | 1.2 |
| Valine | 21.5 | 2.0 |
| Methionine | 18.2 | 3.1 |
| Cystine | 10.2 | 5.9 |
| Isoluecinc | 11.9 | 1.4 |
| Luecine | 21.9 | 2.3 |
| Phenylalanine | 16.8 | 1.2 |
| Lysine | 39.7 | 6.5 |

**Table (16):** Total amino acid content of the control and treated species Cellulosimicrobium cellulans

| Amino acids | Concentration of amino acids (mg/g dry weight of cells) | |
|---|---|---|
| | Control | Treated |
| Aspartic | 10.8 | 7.0 |
| Glutamic | 11.8 | 10.5 |
| Serine | 3.7 | 4.2 |
| Glycine | 20.9 | 10.0 |
| Histidine | 59.7 | 16.3 |
| Arginine | 58.5 | 21.1 |
| Theronine | 9.0 | 15.3 |
| Alanine | . 2.5 | 9.7 |
| Proline | 9.8 | 6.3 |
| Tyrosine | 15.6 | 30.3 |
| Valine | 30.5 | 17.3 |
| Methionine | 12.9 | 22.9 |
| Cystine | 56.2 | 39.9 |
| Isoluecine | 10.1 | 12.4 |
| Luecine | 9.8 | 16.1 |
| Phenylalanine | 6.8 | 10.0 |
| Lysine | 8.4 | 4.5 |

**Table (17):** Total amino acid content of the control and treated species Listondla anguillomm Listonella anguillorum

| Amino acids | Concentration of amino acids (mg/g dry weight of cells) | |
|---|---|---|
| | Control | Treated |
| Aspartic | 0.8 | 3.4 |
| Glutamic | 0.5 | 1.7 |
| Serine | 4.0 | 32.1 |
| Glycine | 14.9 | 10.0 |
| Histidine | 96.2 | 96.9 |
| Arginine | 22.9 | 20.2 |
| Theronine | 10.6 | 3.3 |
| Alanine | 5.7 | 2.4 |
| Proline | 8.58 | 3.2 |
| Tyrosine | 12.5 | 1.1 |
| Valine | 28.5 | 4.6 |
| Methionine | 16.5 | 2.0 |
| Cystine | 29.4 | 1.3 |
| Isoluecine | 8.9 | 2.4 |
| Luecine | 3.5 | 3.9 |
| Phenylalanine | 8.9 | 2.6 |
| Lysine | 6.5 | 2.7 |

**Table (18):** Total amino acid content of the control and treated species Listonella Acinetobacter baumannii

| Amino acids | Concentration of amino acida (mg/g dry weight of cells) | |
|---|---|---|
| | Control | Treated |
| Aspartic | 2.6 | .04 |
| Glutamic | 0.5 | 1.5 |
| Serine | 4.0 | 4.1 |
| Glycine | 16.5 | 26.8 |
| Histidine | 29.4 | 29.8 |
| Arginine | 51.0 | 18.6 |
| Theronine | 4.8 | 3.8 |
| Alanine | 2.5 | 1.9 |
| Proline | 6.2 | 11.5 |
| Tyrosine | 12.3 | 12.3 |
| Valine | 22.0 | 22.4 |
| Methionine | 14.1 | 6.2 |
| Cystine | 37.4 | 29.6 |
| Isoluecine | 15.8 | 15.1 |
| Luecine | 3.1 | 7.8 |
| Phenylalanine | 6.4 | 7.8 |
| Lysine | 9.1 | 2.9 |

**Table (19):** Molcular weight results of SDS- PAGE from ultrascan analysis of 6 species (1,3,5,7,9,11) as a control samples and (2,4,6,8,10 and 12) as treated samples, lane 13 refers as protein standard.

| | | Lane 2 | | | Lane 3 | | | Lane 4 | | | Lane 5 | | | Lane 6 | | | Lane 7 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Peak | MW (kd) | Band | Peak | MW (kd) | Band | Peak | MW (kd) | Band | Peak | MW (kd) | Band | Peak | MW (kd) | Band | Peak | MW (kd) | Band | Peak |
| 7.5 | 125.598 | 1 | 71.82 | 129.824 | 1 | 77.35 | 131.413 | 1 | 57.18 | 127.105 | 1 | 91.88 | 125.556 | 1 | 70.85 | 124.15 | 1 | 74.24 |
| 4.09 | 115.027 | 2 | 81.53 | 125.185 | 2 | 74.79 | 124.781 | 2 | 59.32 | 119.062 | 2 | 88.53 | 116.973 | 2 | 75.68 | 114.714 | 2 | 80.56 |
| 6.59 | 109.276 | 3 | 84.35 | 113.885 | 3 | 86.32 | 120.25 | 3 | 64.91 | 112.342 | 3 | 106.38 | 110.531 | 3 | 76.62 | 108.305 | 3 | 83.24 |
| 5.47 | 102.303 | 4 | 87.47 | 108.228 | 4 | 89.65 | 113.34 | 4 | 67.09 | 103.716 | 4 | 107.97 | 101.527 | 4 | 76.56 | 101.522 | 4 | 82.88 |
| 8.91 | 97.904 | 5 | 90 | 101.367 | 5 | 86.06 | 106.04 | 5 | 76.32 | 93.689 | 5 | 115.68 | 97.996 | 5 | 81.56 | 97.238 | 5 | 94.97 |
| 46.26 | 90.987 | 6 | 90.18 | 97.036 | 6 | 101.71 | 102.19 | 6 | 79.12 | 88.4 | 6 | 153.85 | 91.297 | 6 | 77.94 | 84.221 | 6 | 143.59 |
| 3.71 | 85.807 | 7 | 113.91 | 90.225 | 7 | 146.74 | 94.903 | 7 | 84.91 | 82.208 | 7 | 108.09 | 85.056 | 7 | 92.79 | 76.709 | 7 | 90.44 |
| 4.32 | 81.518 | 8 | 85.03 | 85.122 | 8 | 88.26 | 89.45 | 8 | 90.68 | 78.702 | 8 | 120.15 | 80.942 | 8 | 96.06 | 61.391 | 8 | 118.62 |
| 20.29 | 76.877 | 9 | 94.32 | 80.308 | 9 | 97 | 84.936 | 9 | 86.76 | 73.189 | 9 | 132.74 | 77.576 | 9 | 104.65 | 56.319 | 9 | 87.59 |
| 1.06 | 71.446 | 10 | 105.94 | 76.319 | 10 | 126.53 | 80.055 | 10 | 81.32 | 67.569 | 10 | 117.35 | 71.763 | 10 | 110.74 | 49.485 | 10 | 122.59 |
| 9.15 | 68.876 | 11 | 88.18 | 71.481 | 11 | 94.09 | 74.899 | 11 | 96.56 | 61.93 | 11 | 109.15 | 66.857 | 11 | 104.12 | 46.721 | 11 | 143.56 |
| 2.82 | 64.955 | 12 | 98.35 | 65.982 | 12 | 84.35 | 70.595 | 12 | 106.15 | 57.592 | 12 | 134.82 | 61.411 | 12 | 91.44 | 44.111 | 12 | 94.21 |
| 26.79 | 60.81 | 13 | 105.76 | 60.464 | 13 | 83.09 | 66.539 | 13 | 107.97 | 50.168 | 13 | 153.12 | 56.809 | 13 | 99.79 | 39.038 | 13 | 156.35 |
| 46.91 | 56.102 | 14 | 114.41 | 55.812 | 14 | 126.79 | 63.181 | 14 | 103.5 | 47.337 | 14 | 160.26 | 50.011 | 14 | 74.09 | 37.66 | 14 | 131.29 |
| 8.06 | 53.689 | 15 | 120 | 48.96 | 15 | 149.03 | 58.243 | 15 | 91.32 | 44.341 | 15 | 148.44 | 47.257 | 15 | 113.85 | 31.695 | 15 | 107.26 |
| 58.29 | 49.171 | 16 | 107.09 | 46.191 | 16 | 100.59 | 55.304 | 16 | 111.41 | 39.477 | 16 | 128.88 | 44.026 | 16 | 93.71 | 27.849 | 16s | 136.09 |
| 35.76 | 46.712 | 17 | 100.44 | 43.578 | 17 | 160.41 | 50.982 | 17 | 119.53 | 32.684 | 17 | 136.76 | 39.59 | 17 | 92.65 | 26.293 | 17 | 66.74 |

**Table (19)** continued:

| | Lane 2 | | | Lane 3 | | | Lane 4 | | | Lane 5 | | | Lane 6 | | | Lane 7 | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **MW (kd)** | **Band** | **Peak** | **MW (kd)** | **Band** | **Peak** | **MW (kd)** | **Band** | **Peak** | **MW (kd)** | **Band** | **Peak** | **MW (kd)** | **Band** | **Peak** | **MW (kd)** | **Band** | |
| 43.412 | 18 | 103.79 | 38.788 | 18 | 137.44 | 48.41 | 18 | 85.18 | 27.858 | 18 | 104.24 | 36.883 | 18 | 91.41 | 24.47 | 18 | 2 |
| 38.61 | 19 | 119.35 | 32.336 | 19 | 114.41 | 44.958 | 19 | 91.85 | 26.477 | 19 | 196.97 | 32.241 | 19 | 106.62 | 20.01 | 19 | 6 |
| 31.681 | 20 | 118.18 | 28.992 | 20 | 142.82 | 39.939 | 20 | 98.41 | 24.267 | 20 | 118.03 | 27.983 | 20 | 89.76 | 17.331 | 20 | 9 |
| 29.014 | 21 | 97 | 26.957 | 21 | 75.85 | 37.367 | 21 | 80.59 | 21.921 | 21 | 120.71 | 26.63 | 21 | 105.18 | 14.691 | 21 | 9 |
| 27.163 | 22 | 96.32 | 24.524 | 22 | 211.56 | 32.467 | 22 | 94.53 | 19.801 | 22 | 124.24 | 24.635 | 22 | 79.91 | 12.909 | 22 | 1 |
| 25.617 | 23 | 78.68 | 21.67 | 23 | 70.38 | 29.93 | 23 | 78.62 | 17.249 | 23 | 103.79 | 22.31 | 23 | 99.18 | 11.757 | 23 | 7 |
| 23.633 | 24 | 98.15 | 19.429 | 24 | 106.76 | 28.002 | 24 | 95.29 | 14.702 | 24 | 141.76 | 20.204 | 24 | 83.91 | 10.709 | 24 | 1 |
| 21.174 | 25 | 97.71 | 16.92 | 25 | 108.53 | 26.393 | 25 | 71.26 | 12.9 | 25 | 117.03 | 17.413 | 25 | 100.91 | 9.614 | 25 | 9 |
| 19.391 | 26 | 102.68 | 14.417 | 26 | 115.62 | 24.331 | 26 | 79.32 | 11.738 | 26 | 133.94 | 15.007 | | | | 26 | 1 |
| 16.748 | 27 | 75.19 | 12.555 | 27 | 84.03 | 21.775 | 27 | 76.03 | 10.837 | 27 | 98.91 | 13.211 | | | | 27 | 8 |
| 14.151 | 28 | 93.94 | 11.505 | 28 | 126.21 | 19.779 | 28 | 96.62 | 9.509 | 28 | 119.74 | 11.796 | | | | 28 | 1 |
| 12.403 | 29 | 76.58 | 10.466 | 29 | 101.09 | 17.059 | | | | 29 | 142.82 | 9.743 | | | | 29 | 8 |
| 11.442 | 30 | 98.21 | 9.384 | 30 | 128.68 | 14.39 | | | | | | | | | | 30 | 1 |
| 10.403 | | | | 31 | 85.82 | 12.689 | | | | | | | | | | | |
| 9.185 | | | | 32 | 116.91 | 11.612 | | | | | | | | | | | |
| | | | | 33 | 88.38 | 10.547 | | | | | | | | | | | |
| | | | | 34 | 125.26 | 9.37 | | | | | | | | | | | |

**Table (19):** Molcular weight results of SDS- PAGE from ultrascan analysis of 6 species (1,3,5,7,9,11) as a control samples and (2,4,6,8,10 and 12) as treated samples, lanc 13 refers as protein standard.

| **Lane 8** | | | **Lane 9** | | | **Lane 10** | | | **Lane 11** | | | **Lane 12** | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| **Band** | **Peak** | **MW (kd)** | **Band** | **Peak** | **MW (kd)** | **Band** | **Peak** | **MW (kd)** | **Band** | **Peak** | **MW (kd)** | **Band** | **Peak** | **MW (kd)** |
| 1 | 59.82 | 130.995 | 1 | 75.79 | 126.914 | 1 | 49.91 | 124.198 | 1 | 83.74 | 124.3 | 1 | 48.91 | 124.277 |
| 2 | 74.76 | 124.554 | 2 | 77.53 | 121.57 | 2 | 61.94 | 109.3 | 2 | 79.79 | 117.506 | 2 | 64.71 | 109.534 |
| 3 | 85.06 | 108.622 | 3 | 97.09 | 110.751 | 3 | 67.94 | 97.565 | 3 | 98.74 | 109.532 | 3 | 65 | 100.69 |
| 4 | 88.47 | 101.803 | 4 | 90.32 | 103.83 | 4 | 90.74 | 90.88 | 4 | 101.41 | 100.675 | 4 | 70.88 | 97.22 |
| 5 | 87.65 | 97.497 | 5 | 109.5 | 99.458 | 5 | 77.71 | 84.652 | 5 | 109.88 | 97.2 | 5 | 69.5 | 90 |
| 6 | 112.21 | 90.72 | 6 | 138.38 | 93.242 | 6 | 82.74 | 81.123 | 6 | 150.32 | 90.604 | 6 | 75.59 | 83.316 |
| 7 | 90.85 | 80.844 | 7 | 96.94 | 86.171 | 7 | 88.94 | 77.19 | 7 | 102.88 | 83.865 | 7 | 82.21 | 79.881 |
| 8 | 102.18 | 76.869 | 8 | 106.88 | 82.542 | 8 | 86.12 | 71.901 | 8 | 115.94 | 80.403 | 8 | 80.76 | 76.053 |
| 9 | 94 | 65.555 | 9 | 132.47 | 79.067 | 9 | 81.06 | 66.974 | 9 | 128.15 | 77.083 | 9 | 70.65 | 71.903 |
| 10 | 102.06 | 61.044 | 10 | 112 | 74.125 | 10 | 97.26 | 61.505 | 10 | 114.47 | 71.35 | 10 | 65.82 | 67.031 |
| 11 | 112.68 | 56.394 | 11 | 100.29 | 69.993 | 11 | 106.03 | 56.885 | 11 | 103.94 | 66.043 | 11 | 83.5 | 60.761 |
| 12 | 120.97 | 49.536 | 12 | 94.03 | 66.091 | 12 | 109.79 | 50.062 | 12 | 132.15 | 61.13 | 12 | 92.5 | 56.284 |
| 13 | 106.12 | 46.762 | 13 | 128.26 | 62.407 | 13 | 104.97 | 47.298 | 13 | 151.76 | 56.583 | 13 | 95.56 | 50.276 |
| 14 | 99.38 | 43.826 | 14 | 151.38 | 58.089 | 14 | 92 | 44.371 | 14 | 158.03 | 49.861 | 14 | 94.79 | 47.2 |
| 15 | 100.85 | 39.055 | 15 | 117.71 | 54.851 | 15 | 109.88 | 39.607 | 15 | 146.53 | 47.135 | 15 | 89.44 | 44.002 |
| 16 | 111.74 | 32.855 | 16 | 159 | 51.056 | 16 | 131.65 | 32.468 | 16 | 129.65 | 43.937 | 16 | 96.5 | 39.866 |
| 17 | 115.5 | 29.49 | 17 | 149.74 | 48.21 | 17 | 73.56 | 30.243 | 17 | 136.82 | 39.542 | 17 | 117.32 | 31.866 |

Lane 13

| Band | Peak | MW(kd) |
|---|---|---|
| 1 | 49.94 | 116 |
| 2 | 251.09 | 97 |
| 3 | 65.62 | 66 |
| 4 | 157.26 | 45 |
| 5 | 142.47 | 29 |
| 6 | 112.29 | 24 |

**Table (19)** continued:

| | | | | | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| 18 | 97.88 | 27.44 | 18 | 129.53 | 45.197 | 18 | 84.24 | 28.17 | 18 | 105.91 | 36.858 | 18 | 89.76 | 28.283 |
| 19 | 98.15 | 24.987 | 19 | 91.65 | 43.294 | 19 | 90.09 | 26.805 | 19 | 210.06 | 32.252 | 19 | 95.82 | 27.117 |
| 20 | 80.12 | 21.949 | 20 | 140.56 | 40.01 | 20 | 96.12 | 24.442 | 20 | 125.76 | 28.024 | 20 | 88 | 24.928 |
| 21 | 98.41 | 19.701 | 21 | 88.29 | 37.78 | 21 | 78.53 | 22.287 | 21 | 127.26 | 26.867 | 21 | 78.85 | 22.755 |
| 22 | 98.21 | 17.305 | 22 | 211.18 | 32.968 | 22 | 94.5 | 20.036 | 22 | 129.91 | 24.868 | 22 | 94.09 | 20.339 |
| 23 | 102.18 | 14.663 | 23 | 112.53 | 28.36 | 23 | 78.41 | 17.508 | 23 | 104.76 | 22.538 | 23 | 93.38 | 15.043 |
| 24 | 76.41 | 12.787 | 24 | 117.21 | 26.779 | 24 | 95.03 | 15.083 | 24 | 144.18 | 20.284 | 24 | 92.29 | 11.85 |
| 25 | 95.85 | 11.813 | 25 | 118.62 | 24.748 | 25 | 78.97 | 12.104 | 25 | 118.41 | 17.501 | 25 | 98.24 | 9.806 |
| 26 | 77.79 | 10.757 | 26 | 95 | 22.224 | 26 | 95.38 | 9.852 | 26 | 138.09 | 15.1 | | | |
| 27 | 99.59 | 9.586 | 27 | 136 | 20.392 | | | | 27 | 100.15 | 13.213 | | | |
| | | | 28 | 112 | 17.668 | | | | 28 | 125.26 | 11.891 | | | |
| | | | 29 | 138.56 | 14.982 | | | | 29 | 91.68 | 11.084 | | | |
| | | | 30 | 95 | 13.263 | | | | 30 | 149.44 | 9.836 | | | |
| | | | 31 | 120.09 | 12.169 | | | | | | | | | |
| | | | 32 | 97.35 | 11.086 | | | | | | | | | |
| | | | 33 | 125 | 9.814 | | | | | | | | | |
| | | | | | | | | | | | | | | |

## 4. Conclusion

1- Six different species of bacteria were isolated from burn-wound patients in Ain Shams University Hospital , namely *Aeromonas schubertii, Haemophilius paraphrohaemlyticus, Micrococcus luteus, Cellulosimicrobium cellulans, Listonella anguillarum and Acinetobacter baumannii*

2- Four types of honey (Citurs, Clover, Nigella and Eljabaly). Comparative study between the above types of honey and the known groups of 18 antibiotics to evaluate the importance of using honey in burn-wound treatment on the 6 isolated species of bacteria.

3- It was found that Eljabaly honey has strong inhibitory effect in comparison to other mentioned types. Concentration of 25% of Eljabaly showed inhibition of 4 types of isolated bacteria, whereas 30% was potent enough to destroy the 6 isolated bacteria.

4- The antibacterial activity of honey was attributed to its high osmolarity and

hypertonic sugar concentration and low PH values.

5- Total lipids of bacterial species were sharply decreased. Amino and fatty acids, total proteins patterns were significantly changed.

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4/18/2010