



Honey as a Natural antibiotic adjuvant

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Abstract:

Since the ancient times, the medicinal importance of honey has been documented in the world's oldest medical literatures and it has been known to have antimicrobial and wound-healing activity. The current study focuses on the adjuvant property of honey. The antibiotics Tetracycline and Ampicillin used in this study are not showing or showing negligible effect against *Klebsiella pneumoniae* and *Streptococcus* alone but, when the antibiotics are mixed with honey is showing significant effect. This indicates honey is enhancing the antimicrobial property of the antibiotics tetracycline and ampicillin. The antimicrobial activity in most honeys is due to the enzymatic production of hydrogen peroxide, low pH level and its high sugar content (high osmolarity). In the current study comparison between the antimicrobial properties of natural and artificial honey also done. Natural honey is showing good antibacterial activity when compared with artificial honey. This may be due to decreasing the consistency and adulteration of artificial honey.

Thus, current work provides valuable information on the quality and possible therapeutic potential use and the medicinal property of honey with emphasis on their antibacterial activities.

Key words: Honey, Adjuvant, Antibacterial Activity, Antibiotics,

1. INTRODUCTION

The use of honey as a drug for the treatment of disease dates back to 2100-2000 BC. For instance, pale honey was described by Aristotle (384-322 BC) as being “good for sore eyes and wounds” (Mandal and Mandal, 2011; Vallianou et al., 2014). Many reports have shown that honey has antimicrobial activity against microorganisms such as protozoa, fungi, and bacteria, including viruses (Carter et al., 2016).

The beneficial properties of honey have been explored and studied in modern times, and there is evidence to suggest that some parts of its historical reputation may hold truth. Antimicrobial agents are essentially important in reducing the global burden of infectious diseases. However, as resistant pathogens develop and spread, the effectiveness of the antibiotics is diminished. This type of bacterial resistance to the antimicrobial agents poses a very serious threat to public health,

and for all kinds of antibiotics, including the major last-resort drugs, the frequencies of resistance are increasing worldwide. Therefore, alternative antimicrobial strategies are urgently needed, and thus this situation has led to a re-evaluation of the therapeutic use of ancient remedies, such as plants and plant-based products, including honey (Mandal et al. 2010 and Basualdo et al. 2007).

Ampicillin is not the agent of first choice for gram-negative infections, because the high inhibitory concentrations necessary are not always reached in the aqueous humor (David J. Maggs, 2008). Abd-El Aal et al. (2007) showed that honey had a more pronounced inhibitory effect (85.7%) on Gram negative bacteria (*Pseudomonas aeruginosa*, *Enterobacter spp.*, *Klebsiella*) in comparison to commonly used antimicrobial agents. A 100% inhibition was observed in the case of Gram positive methicillin-resistant *Staphylococcus aureus* in comparison to the use of antibiotics alone. The antimicrobial properties of honey have been well documented, and honey has been used from ancient times as a method of accelerating wound healing. Its potential to assist wound healing has been demonstrated repeatedly (Vallianou et al., 2014). A possible reason behind its activity relies on its ability to generate hydrogen peroxide by the bee-derived enzyme glucose oxidase (Jing et al., 2014).

The antimicrobial activity of honey has been reported to be due to osmotic effect, acidity, hydrogen peroxide and phytochemical factors (Babacan, 2007). Antibiotic-resistant pathogenic microorganisms posture a very stern risk to public health. Resistance not only is problematic in hospitals; resistant bacteria are now documented among numerous groups in the

public (Carattoli, 2008). Frequencies of bacterial antibiotics resistance are growing wide-reaching while very few new antibiotics are being advanced (Fischbach, 2009). Therefore marginal antibacterial, antifungal and antiviral tactics are needed⁵⁰. Honey has broad-spectrum action against pathogenic bacteria and fungi (Taormina, 2001). Researchers showed effectiveness of natural honey in handling of chronic wound infections not responding to antibiotic therapy (Cooper, 2002).

Therefore, this study seeks to assess the physicochemical properties, antimicrobial activity and adjuvant property of the natural honey.

2. MATERIALS AND METHODS

2.1 Collection of honey

The honey sample used for this experiment was collected from the known people of Gulyam village, Ballari District, Karnataka. The vegetation of the various sample sites was noted and recorded. The sample was stored in transparent glass bottle at room temperature during the period of the experiment.

2.2 Physical and chemical properties of honey:

The physical properties like color and texture and chemical properties like moisture content and pH were measured as per the International Honey Commission (Bogdanov, 1984 and Shahnawaz et al., 2013)

2.3 Bacterial cultures used:

All the bacterial cultures were obtained from the clinical isolates of VIMS, Ballari includes *Klebsiella oxytoca*, *Escherichia coli*, *Klebsiella pneumonia* and *Streptococcus*. All the cultures were subcultured on nutrient agar and maintained carefully for experimental purpose.

2.4 Antibiotics:

The antibiotics Tetracycline and Ampicillin were purchased from Prashanth Marketing, Haveri.

2.5 Preparation of honey and antibiotic samples for testing

Honey		
1	Purity	100% pure
2	Colour	Deep brown
3	Suspended particles	few suspended particles were present
4	Moisture contents	17.5%
5	pH	4.13

Honey is diluted to 1:1 (w/v)

) ratio and the Tetracycline was mixed with the honey in 1:1 ratio (w/v) and the Ampicillin also mixed with the honey in 1:1 ratio (w/v). Then labeled as

H – Honey only,

H1– Honey + Tetracycline

H2 – Honey + Ampicillin

1 – Tetracycline (1mg/ml)

2 – Ampicillin (1mg/ml)

2.6 Determination of antimicrobial activity

An antibacterial activity of the honey and antibiotics was determined by an agar diffusion assay procedure (Gulfranz et al., 2011). Nutrient agar plates were prepared and inoculated with test bacterial isolates by spreading the bacterial inoculum on the surface of the media. 6 mm in diameter wells were punched in the nutrient agar. Each of the wells was labelled to avoid mixing up the positions of the different honey and antibiotic concentrations.

20µl of the each sample was placed in the respective wells and incubated at 37°C for 24 hours and then observed for inhibition zone and the zone of inhibition was measured.

3. RESULTS AND DISCUSSION

3.1 Physico-chemical properties

The obtained honey was 100% pure, deep brown in colour, few suspended particles were present, further the percentage of moisture contents per 100 g of honey was 17.5% and the pH was 4.13 and were listed in table:1

Table: 1 Physico-chemical properties of honey

3.2 Anti bacterial effect:

Antibacterial properties of honey and antibiotics were analyzed. Honey was exhibited good antibacterial property comparatively to antibiotics alone. The antibiotic Ampicillin alone was not

S. No	Bacterial strain	Inhibition zone in mm				
		H	1	2	H1	H2
1	<i>Klebsiella oxytoca</i>	1	2	4	9	5
2	<i>Escherichia coli</i>	6	14	13	16	15
3	<i>Klebsiella pneumoniae</i>	2	7	1	9	3
4	<i>Streptococcus</i>	13	1	2	16	14

showing any effect on *Klebsiella pneumoniae* and *Streptococcus*. Tetracycline alone has less effect on *Klebsiella oxytoca* and *Streptococcus*. Tetracycline with honey was shown increased effect and the inhibitory zone was increased from 2mm to 9mm against *Klebsiella oxytoca* and 1mm to 16mm against *Streptococcus*. Same way Ampicillin alone has no effect on *Klebsiella pneumoniae* and *Streptococcus* but, when Ampicillin was mixed with honey the antimicrobial activity was amplified and the inhibitory zone was increased from 1 to 3mm against *Klebsiella pneumoniae* and significantly from 2 to 14mm against *Streptococcus*. The

results were shown in table 2. This indicates Honey has a good adjuvant property.

Table: 2 Zone of inhibition (mm) of different samples against bacterial strains tested

Note: H – Honey only, H1– Honey + Tetracycline, H2 – Honey + Ampicillin, 1– Tetracycline (1mg/ml), 2 – Ampicillin (1mg/ml)

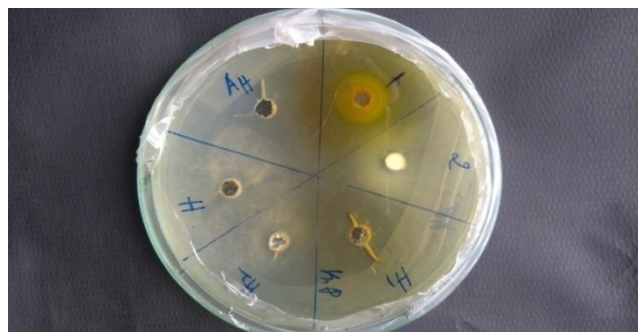


Fig:1 Antimicrobial activity against *Streptococcus*

It is reported that proteins, minerals, phytochemicals and antioxidants present in honey are responsible for medical and biological activities of honey in the treatment of infections, burns, wounds and ulcers (Moumbe et al., 2013).

The acidity is likely to contribute to the antibacterial potency of the honey (Boateng and Diunase, 2015). Moreover, bee honey is a solution of supersaturated sugar; these sugars prevent the thriving of microorganisms (bacteria and yeast) due to their high affinity for water molecules, thereby leaving little or no water to support their growth. As a result microbes become dehydrated and die in the end (Israili, 2014). The unique antimicrobial properties of honey may involve in the enhancement of antibiotic activity of ampicillin and tetracycline.

4. Conclusion:

It is concluded that, the natural honey is an interesting and promising alternative to classical antibiotics also it enhances the activity of antibiotics and honey should be more seriously considered as therapeutic agents.

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