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# In Vitro Evaluation of Anti- Microbial Activity of Manuka Honey Against Oral Pathogens

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#### ABSTRACT

Introduction: Manuka honey is a monofloral honey synthesized from the nectar of the flower Leptospermum scoparium which is simply the manuka plant. The health benefits of manuka honey is uncountable. It is known from the ancient civilization that it has numerous therapeutic properties like wound healing, treatment for ulcers, epithelial regenerations. It is also found to have so many biological activities such as antioxidant, antimicrobial, and anti proliferation activities. The aim of the present study is to evaluate the antimicrobial property of manuka honey against oral pathogens.

Materials and methods: Streptococcus mutans and Enterococcus faecalis were used for the study. The bacterial strains were maintained in nutrient agar slope at 4°C in the microbiology laboratory.

Screening of antibacterial activity (Agar well diffusion assay): Broth culture of the test organisms is prepared which is compared to 0.5 Mc farland standards. The lawn culture is made on Muller Hinton Agar (MHA). The wells were cut using sterile cork borers. Different concentrations of the manuka honey ( $25\mu$ L,  $50\mu$ L,  $100\mu$ L) added to the wells. The culture plate is incubated at  $37^{\circ}$ C. Then the zone of inhibition is measured in mm diameter and the results were tabulated.

Results: The measure of the zone of inhibition of the S mutans and E faecalis is shown in table 1. As the concentration of the manuka honey increased the zone of inhibition of both the bacteria increased.

Conclusion: Within the limits of the study it can be concluded that the manuka honey is effective against pathogens. Much more in vivo studies are required to prove the efficacy of this absolutely excellent product to this world.

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How to cite this article: Padmalochini S, Geetha V R, Lakshmi T, In Vitro Evaluation of Anti- Microbial Activity of Manuka Honey Against Oral Pathogens. Journal of Complementary Medicine Research, Vol. 13, No. 5, 2022 (pp. 64-68)

## INTRODUCTION

Manuka honey is a monofloral honey synthesized from the nectar of the flower Leptospermum scoparium which is simply the manuka plant. This honey is exported only from New Zealand which is the speciality of this honey. This variety of honey is produced by the honey bees apis mellifera from the manuka plants. Interestingly Manuka honey has high levels of Methylglyoxal (MGO) which is formed from the dihydroxyacetone (DHA) which is known to be correlated with the antibacterial activity. The properties of the honey include: they are very thick and tasty. It is the most expensive honey in the world because it is very rare. Manuka honey in its purest form would cost around 99,000 for 1g. In addition to its rare conditions, it also takes honey bees around 22,700 individual trips to collect enough nectar [4-6].

KEYWORDS: Manuka honey, anti microbial activity, zone of inhibition, MHA, MGO

ARTICLE HISTORY: Received June 15, 2022 Accepted Oct 11, 2022 Published Dec 17, 2022

DOI: 10.5455/jcmr.2022.13.05.12 The health benefits of manuka honey is uncountable. It is known from the ancient civilization that it has numerous therapeutic properties like wound healing, treatment for ulcers, epithelial regenerations. It is also found to have so many biological activities such as antioxidant, antimicrobial, and anti proliferation activities. It is exploited by the researchers in the area of tissue engineering to design templates for regeneration [7,8]. The quality test for manuka honey is done by checking the UMF rating. More the UMF value, the more antibacterial activity it has. It is also widely used in the food and cosmetic industry [9].

To explore more about manuka honey it has antiviral properties. It attacks the microorganisms with the substance MGO. It helps in the healing of minor and major injuries. In US manuka honey infused bandages have been approved for over the counter and prescription sales. It is more potent particularly against the influenza virus [10]. Manuka honey is less effective against the treatment of fungal infections caused by Candida albicans and dermatophytes. This is due to the reason that it lacks hydrogen peroxide which is essential for the antifungal activity [11]. Since manuka honey has higher UMF it has a high antibacterial property [12]. Manuka honey is proven to have a good anti-inflammatory response. The inflammation associated with ulcerative colitis, inflammatory bowel disease is due to overexpression of the inflammatory cells due to increased oxidant activity which has a good effect when treated with manuka honey. It is proven that manuka honey has a high amount of phenolic compounds which reduce free radicals that contribute to antioxidant activity [13] [14].

Oral pathogens pose a great threat to mankind. It is mostly spread by aerosols released from the dental clinics that could be spread to society very easily. Some of the oral pathogens are Streptococcus mutans, Porphyromonas gingivalis, Staphylococcus, and Lactobacillus. Since Manuka honey has antibacterial activity it can be used to treat diseases caused by oral pathogens [15]. The aim of the present study is to evaluate the antimicrobial property of manuka honey against oral pathogens.

## MATERIALS AND METHODS

Streptococcus mutans and Enterococcus faecalis were used for the study. The bacterial strains were maintained in nutrient agar slope at  $4^{\circ}$ C in the microbiology laboratory.

# Screening of antibacterial activity (Agar well diffusion assay)

Broth culture of the test organisms is prepared compared to 0.5 Mc farland standards. The lawn culture of the broth culture was made on Muller Hinton Agar (MHA). The wells were cut using sterile cork borers. Different concentrations of the manuka honey ( $25\mu$ L,  $50\mu$ L,  $100\mu$ L) were added to the wells. The culture plate is incubated at  $37^{\circ}$ C overnight. Then the zone of inhibition is measured in mm diameter and the results were tabulated. The test in done in triplicate to avoid errors.

## RESULTS

Anti bacterial activity of Manuka honey was evaluated using agar well diffusion assay against Streptococcus mutans and Enterococcus faecalis. The zone of inhibition is recorded (in mm diameter) and shown in table 1. As the concentration of the manuka honey increased the zone of inhibition of both the bacteria increased. Manuka honey showed significant inhibitory effect against Streptococcus mutans (21mm), and Enterococcus faecalis (19 mm) at 100µL concentration,

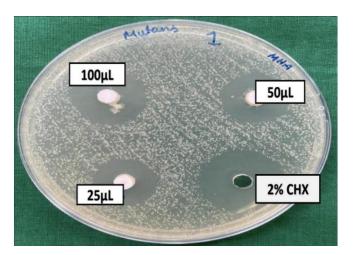


Fig.1: represents the zone of inhibition exhibited by the Streptococcus mutans at different concentrations of the manuka honey.

concentrations	S mutans	E faecalis
25µL	9mm	-
50µL	14mm	11mm
100µL	21mm	19mm
2%CHX	26mm	24mm

<b>Table 1:</b> showing the tabulation of the values of the zone of inhibition in mm diameter	·.
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# DISCUSSION

The reason to study manuka honey and its antimicrobial activity is that many microorganisms have become resistant to the specific antibiotics prescribed by the physicians. So, there arises a need to switch for an alternative which is safe and does not have any side effects [16]. It is reported in a study that gram negative bacteria has become the serious antibiotic resistant ones. Among them is Pseudomonas aeruginosa, a serious nosocomial pathogen posing threat to humans [17]. Now here comes the role of manuka honey as it can be an alternative in the treatment protocols. It is widely being used in treating diabetic ulcers, burns, and non healing wounds. It also has shown that it can combat even the MRSA ( methicillin resistant staphylococcus aureus). Among the available honey, manuka honey has the ability to release hydrogen peroxide which is helpful in eradicating bacterial activity [18].

The antibacterial activity of the manuka honey is due to MGO and hydrogen peroxide [1] as it was described earlier. Other important factors which contribute for the antibacterial activity are  $P_h$ , osmotic pressure, low protein content hyperosmolality effect, different grades of flavonoids etc. There are many studies reporting the antibacterial activity in terms of MIC [2,3]. In the present study we used the zone of inhibition as a parameter to measure the antibacterial activity. We found that at high concentration of the manuka honey it was able to eliminate the growth of the pathogens.

Some of the unique features which differentiates manuka honey from other honey is that its antibacterial activity is independent in inducing the inflammatory cytokines during an immune response. Depending upon the species of a bacteria it can even alter the shape and size of the bacteria also [19]. Manuka honey can be effective against the S aureus biofilms also. It may oppose the growth at low concentrations which is a much needed weapon against several pathogens. The mechanism of the biofilm destruction is by the reduction in the biofilm matrices. This mechanism is due to the complexity of the honey components which work alone or in groups to prevent the resistance of the pathogens against the honey [20].

Synergistic activities of manuka honey with the available antibiotics can be a better option as it is more effective against the pathogens. For example oxacillin can be combined with manuka honey to act against antibiotic resistant bacteria such as MRSA. This was experimented because as we know manuka honey can be effective against biofilms. If this is combined with the conventional antibiotics good antibacterial activity can be explored. Some of the antibiotics which can be given synergistically are oxacillin, clindamycin and rifampicin [21]

### [12].

Therefore, manuka honey can be implicated not only in the field of medicine but also in dentistry. There are several researches made on the use of manuka honey to treat oral pathogens. So many researchers incorporated manuka honey in dental treatment protocols and in some of the dental products. Nayak et al incorporated manuka honey in mouthwash and found that there was reduction in plaque formation and preventing the formation of the oral biofilms compared to the conventionally used xylitol chewing gums. It is moreover a safer option than xylitol because it is a natural product [22]. It can be used in the periodontal applications and demineralization as a topical agent [23]. It can also be used as a root canal disinfectant, infusing suture threads in manuka honey and wound dressing material in chronic non healing discharging wounds as it has a high wound healing property [24] [25]. Our team has extensive knowledge and research experience that has translated into high quality publications[26-35].

Since manuka honey is not cost effective and not easily available, difficulties might arise in incorporation of the product commercially. Further much more in vivo studies are required to check the acceptance of the human immune system and other issues in introducing manuka honey in the pharmaceutical world.

# CONCLUSION

Within the limits of the study it can be concluded that the manuka honey is effective against pathogens. Much more in vivo studies are required to prove the efficacy of this absolutely excellent product to this world.

# ACKNOWLEDGEMENT

The authors would like to thank orange lab, center for infectious diseases, Saveetha Dental College and Hospitals, Saveetha Institute of medical and technical Sciences, Saveetha University for providing research laboratory facilities to carry out the study.

### **Funding Source**

The present project was funded by Saveetha Institute of Medical and Technical sciences Saveetha Dental college and hospital Saveetha University Veeramalai Associates

### CONFLICT OF INTEREST

The author declares that there was no conflict of interest in the present study.

Ethical approval-NA Informed Consent- None declared

#### Authorship contribution

PS compiled the manuscript RVG conducted the study LT designed the study

#### **Authors Contribution**

PADMALOCHINI S: Literature search, data collection, analysis, manuscript drafting.

Dr. R. V. GEETHA: Data verification, manuscript drafting.

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