

Research Article

Antibacterial and Anticandidal Activity of Natural and Commercial Honey - A Comparative Study



CrossMark

K. Suganthi¹, P. Saranraj²¹Department of Biochemistry, Sacred Heart College (Autonomous), Vellore, Tamil Nadu, India,²Department of Microbiology, Sacred Heart College (Autonomous), Vellore, Tamil Nadu, India

ABSTRACT

Humans have known honey and plants for many centuries and used them as sources for nutrients as well as medicine. Today there is a growing body of literature demonstrating the efficacy of honey in various health aspects and particularly as a novel agent for wound management. The potential effects of selected honeys for the treatment of particular diseases have been known for centuries as certain honeys were selected for the treatment of particular ailments; however, it was not until recently that the research has proved that certain honeys possess unusual antimicrobial properties. In the present research, Natural honey showed maximum antimicrobial activity and Antioxidant activity than commercially marketed honey. It was showed that the honey samples does not exhibit antimicrobial activity against the yeast *Candida albicans*. The pharmacological activity of Vembu honey was comparatively high when compared to the Kombu honey. All the honey samples are resistant to *C. albicans*, *Shigella flexneri*, *Enterococcus casseliflavus* and *Pseudomonas aeruginosa*. The highest inhibitory activity was observed against *Klebsiella pneumoniae* and *Staphylococcus aureus*.

Address for correspondence:

P. Saranraj, Department of Microbiology, Sacred Heart College (Autonomous), Tirupattur, Tamil Nadu, India.

E-mail: microsaranraj@gmail.com**Keywords:** Natural honey, Commercial honey, Antibacterial activity, Anticandidal activity**Received:** 25th December 2017**Accepted:** 20th January 2017**Published:** 05th February 2018

INTRODUCTION

The traditional medicinal system is the method of using various natural products such as seaweeds, plants, and plant products for treating various diseases caused by microorganisms. The traditional medicinal system was used by the peoples in the older days, but the entry of antibiotics has stopped the used of traditional medicines based treatment. The world rotation takes the peoples again to the traditional medicinal system due to the antibiotic resistance. The presence of bioactive compounds in the natural products gives an effective remedy against microbial diseases and prevents the humans from the risk of various side effects. On that line, the present study was designed to give the sweet remedy for microbial diseases using the Honey. The honey is an important part of the traditional medicinal system which was used from older days to present days by all category of peoples.^[1]

The use of honey as a traditional medicine was quoted various thousands of years ago in various religious holy books such as Holy Bible, Quran, and Bhagavat Gita. The messenger of God, Prophet Mohammad Nabi (Peace been upon him) quoted honey as a "Healer of all diseases" Honey is a sugary syrup substance produced from the supersaturated nectar, exudates of tree, and secretions of plants flower by honey bees which belongs to the *Aphis* sp. it is also referred as an alimentary product of the *Aphis* sp. Biochemically, honey is composed of water and carbohydrate sugars with the outstanding content

of 31% glucose and 38% fructose and lesser content of 1% sucrose. Honey is considered as a non-spoilage product due to the presence of mineral salts, vitamins, and beneficial microorganisms. Other common ingredients of honey are amino acids (majorly proline), organic acids, hydrogen peroxide, enzymes, polyphenols, ascorbic acid, flavonoids, bee defensin, leptosin, methylglyoxal, and phenolic acids. The mineral fractions available in the honey are iron, phosphorous, calcium, sodium, manganese, magnesium, copper, sodium, and cobalt. Some pharmacologically important enzymes such as glucose oxidase, catalase, amylase, and oxidase are present in honey.^[2,3]

Nowadays, honey is considered as a "Sweet Medicine" and it does not have any medicine hatters. Honey is the product with tremendous antimicrobial activity due to the presence of phenolic compounds, and it is administered orally or topically. In older days, the honey was dipped in the dressing materials and used for dressing the various types of wounds. From older days to present days, the honey was mixed with coconut oil and used for treating peptic ulcer which was caused by *Helicobacter pylori*. In human diet, honey is playing a dual role. Honey in combination with the water was used for reducing the weight and in contrast, honey in combination with milk was helped to increase the weight. Initially, at the time of birth, a drop of honey was given to the infants to give the sweetness and nutrients. Various researchers reported the microcidal and microstatic activities of honey samples against pathogenic



bacteria and fungi. Honey was also used as a preservative for the preservations of various food products which are easily prone to microbial spoilage.^[4,5] The present study was aimed to study the comparative analysis of antibacterial activity and anticandidal activity of the two natural honey (Kombu honey and Vembu honey) and two commercially marketed honey.

MATERIALS AND METHODS

Collection of Honey Samples

The Natural (Kombu honey and Vembu honey) and Commercial honey (Commercially marketed honey - 1 and Commercially marketed honey - 2) samples selected for the present study was collected from Alangayam, Vellore district, Tamil Nadu, India.

Collection of Bacterial and Fungal Cultures

Nine different bacterial cultures and one fungal yeast culture was procured from Department of Biochemistry and Microbiology, Sacred Heart College (Autonomous), Tirupattur, Tamil Nadu, India. Gram-positive bacteria - *Staphylococcus aureus*, *Bacillus cereus* and *Enterococcus casseliflavus*; Gram-negative bacteria - *Escherichia coli*, *Salmonella typhi*, *Proteus mirabilis*, *Klebsiella pneumoniae*, *Shigella flexneri* and *Pseudomonas aeruginosa*; Yeast - *Candida albicans*.

Culture Maintenance and Inoculum Preparation

Maintenance of test bacterial and yeast cultures

The test bacterial isolates were sub-cultures and maintained on Nutrient agar slants, and the yeast isolate *C. albicans* were subcultures and maintained on Sabouraud's dextrose agar slants. The microbial slants were stored in a refrigerator at 4°C.

Bacterial inoculum preparation

Bacterial inoculums were prepared by inoculating a loopful of test organisms in 5 ml of Nutrient broth and incubated at 37°C for 24 h until moderate turbidity was developed.

Yeast inoculum preparation

The inoculums of Yeast *C. albicans* was prepared by inoculating a loopful of test organisms in 5 ml of Sabouraud's dextrose broth and incubated at room temperature for 48 h until a moderate turbidity was developed.

Determination of Antibacterial and Anticandidal Activity of Natural and Commercially Marketed Honey

Mueller Hinton agar plates were prepared and inoculated with test bacterial and yeast isolates by spreading the bacterial inoculum on the surface of the media. Wells (6 mm in diameter) were punched in the Mueller Hinton agar. Honey samples with 75 µl/ml concentrations were mixed with 1 ml of dimethyl sulfoxide (DMSO), mixed well and added to the well. Well-containing DMSO alone act as a Negative control. The bacteria inoculated plates were incubated at 37°C for 24 h. The yeast inoculated plates were incubated at room temperature for

48 h. The antibacterial and anticandidal activity was assessed by measuring the diameter of the zone of inhibition (in mm).

RESULTS AND DISCUSSION

The antimicrobial activity and antioxidant activity of the four different honey samples (two Natural honeys - Vembu honey and Kombu honey and two Commercially marketed honeys - Commercially marketed honey 1 and Commercially marketed honey 2) was studied in this present research. The antimicrobial activity of honey was studied against nine bacterial cultures (three Gram-positive bacteria and six Gram-negative bacteria) and one fungal yeast culture (*C. albicans*) by Agar well diffusion method which was proposed by Kirby and Bauer. The DPPH free radicals scavenging activity of the honey samples also studied in this present research. The findings of the present research were given and discussed below.

The antimicrobial activity of Natural Kombu Honey was studied against microbial pathogens, and the findings are furnished in Table 1. The Natural Kombu Honey exhibited more antimicrobial activity against bacterial pathogens and did not show any inhibitory activity against the fungal pathogenic yeast *C. albicans*. It was observed that the Gram-negative bacteria have showed highest inhibitory activity when compared to the Gram-positive bacteria. The Natural Kombu Honey showed highest inhibitory activity against the Pneumonia causing bacteria *K. pneumoniae* (35 mm in dm) followed by the Gram-positive Urinary tract infection causing cocci *S. aureus* (32 mm in dm), Typhoid causing *S. typhi* (22 mm in dm), Coliform bacteria *E. coli* (20 mm in dm), Urinary tract infection causing bacilli *P. mirabilis* (20 mm in dm) and Food borne disease causing Gram-positive bacilli *B. cereus* (18 mm in dm). The Natural Kombu

Table 1: Antimicrobial activity of natural kombu honey against pathogenic microorganisms

S. No	Microorganisms	Zone of inhibition (mm in dm)	
		Natural kombu honey	DMSO
Gram-positive bacteria			
1	<i>S. aureus</i>	32	NZ
2	<i>B. cereus</i>	18	NZ
3	<i>E. casseliflavus</i>	NZ	NZ
Gram-negative bacteria			
4	<i>E. coli</i>	20	NZ
5	<i>S. typhi</i>	22	NZ
6	<i>K. pneumoniae</i>	35	NZ
7	<i>P. mirabilis</i>	20	NZ
8	<i>S. flexneri</i>	NZ	NZ
9	<i>P. aeruginosa</i>	NZ	NZ
Fungi - Yeast			
10	<i>C. albicans</i>	NZ	NZ

*NZ: No zone of inhibition. DMSO: Dimethyl sulfoxide, *S. aureus*: *Staphylococcus aureus*, *B. cereus*: *Bacillus cereus*, *E. casseliflavus*: *Enterococcus casseliflavus*, *E. coli*: *Escherichia coli*, *S. typhi*: *Salmonella typhi*, *K. pneumoniae*: *Klebsiella pneumoniae*, *P. mirabilis*: *Proteus mirabilis*, *S. flexneri*: *Shigella flexneri*, *P. aeruginosa*: *Pseudomonas aeruginosa*, *C. albicans*: *Candida albicans*

Honey was completely resistant to *E. casseliflavus*, *S. flexneri*, *P. aeruginosa* and fungal yeast *C. albicans*. The negative control DMSO also does not show any inhibitory activity.

The antimicrobial activity of Natural Vembu Honey was tested against microbial pathogens, and the results are tabulated in Table 2. Like Natural Kombu Honey, the Natural Vembu Honey also exhibited good antimicrobial activity against bacterial pathogens and not against the fungal pathogenic yeast *C. albicans*. As same as Natural Kombu Honey, it was observed that the Natural Vembu Honey have showed highest inhibitory activity against Gram-negative bacteria when compared to the Gram-positive bacteria. The Natural Vembu Honey have showed highest zone of inhibition against the Pneumonia causing bacteria *K. pneumoniae* (38 mm in dm) followed by the Gram-positive clustery arranged cocci *S. aureus* (35 mm in dm), Typhoid causing *S. typhi* (25 mm in dm) and Coliform bacteria *E. coli* (20 mm in dm). The Natural Vembu Honey was completely resistant to *B. subtilis*, *P. mirabilis*, *E. casseliflavus*, *S. flexneri*, *P. aeruginosa* and fungal yeast *C. albicans*. The negative control DMSO also does not showed any inhibitory activity.

The antimicrobial activity of Commercially Marketed Honey - 1 was determined against pathogenic microorganisms and the findings are given in Table 3. Like Natural Kombu Honey and Natural Vembu Honey, the Commercially Marketed Honey - 1 also exhibited good antimicrobial activity against bacterial pathogens and not against the fungal pathogenic yeast *C. albicans*. In contrast to the Natural Honey, it was noticed that the commercially marketed Honey - 1 have showed highest inhibitory activity against Gram-positive bacteria when compared to the Gram-negative bacteria. The commercially marketed Honey - 1 have showed the highest zone of inhibition against the *S. aureus* (22 mm in dm) followed by the *K. pneumoniae* (21 mm in dm), *S. typhi* (12 mm in dm) and *E. coli* (8 mm in dm). No zone of inhibition was recorded against *B. subtilis*, *P. mirabilis*, *E. casseliflavus*, *S. flexneri*, *P. zaeruginosa* and *C. albicans*. The negative control DMSO also does not show any inhibitory activity.

The antimicrobial activity of Commercially Marketed Honey - 2 was studied against pathogenic bacteria and yeast, and the results are shown in Table 4. Like Natural Kombu Honey, Natural Vembu Honey and Commercially Marketed Honey - 1, the Commercially Marketed Honey - 2 also exhibited an increased antimicrobial activity against bacterial pathogens and not against the fungal pathogenic yeast *C. albicans*. The antimicrobial activity of the Commercially Marketed Honey - 2 is more when compared to Commercially Marketed Honey - 1. In contrast to the Commercially Marketed Honey - 1 and similar to Natural Honey, it was recorded that the Commercially Marketed Honey - 2 have showed the highest inhibitory activity against Gram-negative bacteria when compared to the Gram-positive bacteria. The Commercially Marketed Honey - 1 have showed the highest zone of inhibition against the *K. pneumoniae* (25 mm in dm) followed by the *S. aureus* (23 mm in dm), *S. typhi* (15 mm in dm) and *E. coli* (14 mm in dm). No zone of inhibition was recorded against *B. subtilis*, *P. mirabilis*, *E. casseliflavus*, *S. flexneri*, *P. aeruginosa* and *C. albicans*. The negative control DMSO also does not show any inhibitory activity.

The pharmacological activity of honey was studied by various researchers in various microorganisms like bacteria

Table 2: Antimicrobial activity of natural Vembu honey against pathogenic microorganisms

S. No	Microorganisms	Zone of inhibition (mm in dm)	
		natural vembu honey	DMSO
Gram-positive bacteria			
1	<i>S. aureus</i>	35	NZ
2	<i>B. cereus</i>	NZ	NZ
3	<i>E. casseliflavus</i>	NZ	NZ
Gram-negative bacteria			
4	<i>E. coli</i>	22	NZ
5	<i>S. typhi</i>	25	NZ
6	<i>K. pneumoniae</i>	38	NZ
7	<i>P. mirabilis</i>	NZ	NZ
8	<i>S. flexneri</i>	NZ	NZ
9	<i>P. aeruginosa</i>	NZ	NZ
Fungi - Yeast			
10	<i>C. albicans</i>	NZ	NZ

*NZ: No zone of inhibition. DMSO: Dimethyl sulfoxide, *S. aureus*: *Staphylococcus aureus*, *B. cereus*: *Bacillus cereus*, *E. casseliflavus*: *Enterococcus casseliflavus*, *E. coli*: *Escherichia coli*, *S. typhi*: *Salmonella typhi*, *K. pneumoniae*: *Klebsiella pneumoniae*, *P. mirabilis*: *Proteus mirabilis*, *S. flexneri*: *Shigella flexneri*, *P. aeruginosa*: *Pseudomonas aeruginosa*, *C. albicans*: *Candida albicans*

Table 3: Antimicrobial activity of commercially marketed honey - 1 against pathogenic microorganisms

S. No	Microorganisms	Zone of inhibition (mm in dm)	
		Commercial Honey - 1	DMSO
Gram-positive bacteria			
1	<i>S. aureus</i>	22	NZ
2	<i>B. cereus</i>	NZ	NZ
3	<i>E. casseliflavus</i>	NZ	NZ
Gram-negative bacteria			
4	<i>E. coli</i>	8	NZ
5	<i>S. typhi</i>	12	NZ
6	<i>K. pneumoniae</i>	21	NZ
7	<i>P. mirabilis</i>	NZ	NZ
8	<i>S. flexneri</i>	NZ	NZ
9	<i>P. aeruginosa</i>	NZ	NZ
Fungi - Yeast			
10	<i>C. albicans</i>	NZ	NZ

*NZ: No zone of inhibition. DMSO: Dimethyl sulfoxide, *S. aureus*: *Staphylococcus aureus*, *B. cereus*: *Bacillus cereus*, *E. casseliflavus*: *Enterococcus casseliflavus*, *E. coli*: *Escherichia coli*, *S. typhi*: *Salmonella typhi*, *K. pneumoniae*: *Klebsiella pneumoniae*, *P. mirabilis*: *Proteus mirabilis*, *S. flexneri*: *Shigella flexneri*, *P. aeruginosa*: *Pseudomonas aeruginosa*, *C. albicans*: *Candida albicans*

and fungi. Broadly, in our study, we found that the Natural honey was good for the treatment of bacterial diseases than the Commercial honey. The same was also observed in the

Table 4: Antimicrobial activity of commercially marketed honey - 2 against pathogenic microorganisms

S. No	Microorganisms	Zone of inhibition (mm in dm)	
		Commercial Honey - 1	DMSO
Gram-positive bacteria			
1	<i>S. aureus</i>	23	NZ
2	<i>B. cereus</i>	NZ	NZ
3	<i>E. casseliflavus</i>	NZ	NZ
Gram-negative bacteria			
4	<i>E. coli</i>	14	NZ
5	<i>S. typhi</i>	15	NZ
6	<i>K. pneumoniae</i>	25	NZ
7	<i>P. mirabilis</i>	NZ	NZ
8	<i>S. flexneri</i>	NZ	NZ
9	<i>P. aeruginosa</i>	NZ	NZ
Fungi - Yeast			
10	<i>C. albicans</i>	NZ	NZ

*NZ: No zone of inhibition. DMSO: Dimethyl sulfoxide, *S. aureus*: *Staphylococcus aureus*, *B. cereus*: *Bacillus cereus*, *E. casseliflavus*: *Enterococcus casseliflavus*, *E. coli*: *Escherichia coli*, *S. typhi*: *Salmonella typhi*, *K. pneumoniae*: *Klebsiella pneumoniae*, *P. mirabilis*: *Proteus mirabilis*, *S. flexneri*: *Shigella flexneri*, *P. aeruginosa*: *Pseudomonas aeruginosa*, *C. albicans*: *Candida albicans*

case of antioxidant activity. Both the natural honey and commercial honey has not showed any inhibitory activity against *C. albicans*. The Natural honey samples have showed best antioxidant activity when compared to the commercially marketed honey. Both the Natural Kombu Honey and Natural Vembu Honey have showed more or less similar activity against pathogenic microorganisms. The Natural Kombu Honey has showed broad spectrum activity against pathogenic bacteria by providing inhibitory activity against many tested bacterial isolates. The Natural Vembu Honey has showed Narrow spectrum activity against pathogenic bacteria by providing inhibitory activity against limited tested bacterial isolates. The Natural Kombu Honey has showed less inhibitory activity against many bacterial isolates but in contrast, the Natural Vembu Honey more inhibitory activity against selected bacterial isolates. Honey is the only food material which cannot be spoiled for hundreds of years and does not allow its container also to spoil. However, the commercial honeys are available in marked with an expiry date. This one character is enough to propose the natural honey as the best one in all the ways than the Commercial honey.

Kumar *et al.*^[6] revealed the activity of Honey samples against wound infections and its management. Srisayam and Chantawannakul^[7] investigated the antifungal activity of honey samples against two fungal yeast isolates *viz.*, *C. albicans* and *Saccharomyces cerevisiae*. They showed that the honey samples are completely resistant to *C. albicans* and *Saccharomyces cerevisiae*. The finding of the research of Srisayam and Chantawannakul^[7] was similar with the results of our present study. In the present study, we studied

the antimicrobial activity of Natural and Commercial honey samples against the fungal yeast *C. albicans*. We also found that the *C. albicans* was totally resistance to both Natural and Commercial honey samples. According to the studies of Kaur *et al.*,^[8] it was found that Tualang honey had an important role in bactericidal effects and bacteriostatic effects in the treatment of burns and several wounds by using dressings soaked with this honey.

Recently, Kalidasan *et al.*^[9] investigated the antibacterial activity of two Malan honey, Kombu honey and two commercial honeys. They found that the Natural honey has showed more antibacterial activity than the commercial honey. The finding of the present research was similar with the results of Kalidasan *et al.*^[9] They also concluded that the Natural honey was the best source for treating bacterial infection than the Commercial honey.

CONCLUSION

Honey is the only food material which cannot be spoiled for hundreds of years and does not allow its container also to spoil. However, the commercial honeys are available in marked with expiry date. This one character is enough to propose the natural honey as the best one in all the ways than the commercial honey. Natural honey showed maximum antimicrobial activity and antioxidant activity than commercially marketed honey. It was showed that the honey samples does not exhibited antimicrobial activity against the yeast *C. albicans*. The pharmacological activity of Vembu honey was comparatively high when compared to the Kombu honey. All the honey samples are resistant to *C. albicans*, *S. flexneri*, *E. casseliflavus* and *P. aeruginosa*. Highest inhibitory activity was observed against *K. pneumoniae* and *S. aureus*. In conclusion, honey is effective against the bacterial pathogens which are frequently causing Urinary tract infection and Neonatal sepsis, and it is the “sweet medicine” for bacterial infections. We also recommend the Women and Infants to take the Vembu honey regularly for preventing them from the Urinary tract infection and Neonatal sepsis.

REFERENCES

- Adams CJ, Harris MM, Molan PC. The origin of methylglyoxal in New Zealand manuka (*Leptospermum scoparium*) honey. *Carbohydrate Res* 2009;344:1050-3.
- Sanchez V, Baeza R, Ciappini C, Zamora MC, Chirife J. Comparison between Karl Fischer and refractometric method for determination of moisture in honey. *Food Control* 2010;21:339-41.
- Saranraj P, Sivasakthi S, Feliciano GD. Pharmacology of honey-a review. *Adv Biol Res* 2016;10:271-89.
- Devi VD, Kalpana G, Saranraj P. Antibacterial activity of Essential oils against human pathogenic bacteria. *Adv Biol Res* 2017;11:357-64.
- Karthikeyan M, Kanchana D, Saranraj P, Karunya SK. Quantification of metabolic products from lactic acid bacteria isolated from traditional fermented dairy products. *Afri Educ Indices* 2015;8:1-15.
- Kumar KP, Bhowmik D, Chiranjib, Biswajit, Chandira MR. Medicinal uses and health benefits of Honey: An overview. *J Chem Pharm Res* 2010;2:385-95.
- Srisayam M, Chantawannakul P. Antimicrobial and antioxidant

properties of honeys produced by *Apis mellifera* in Thailand. J Apiprod Apimed Sci 2010;2:77-83.

8. Kaur BS, Tan HT, Boukraa L, Gan SH. Different solid phase extraction fractions of Tualang (*Koombassia excelsa*) honey demonstrated diverse antibacterial properties against wound and enteric bacteria. J Apiprod Apimed Sci 2011;3:59-65.
9. Kalidasan G, Saranraj P, Ragul V, Sivasakthi S. Antibacterial activity of natural and commercial honey-a comparative study. Adv Biol Res 2017;11:365-72.

Cite this article: Suganthi K, Saranraj P. Antibacterial and Anticandidal Activity of Natural and Commercial Honey - A Comparative Study. Asian J Appl Res 2018;4(3):37-41.

Source of Support: Nil, **Conflict of Interest:** None declared.