Salivary anti-oxidant levels on consumption of natural honey and commercially available honey

Nikita Lolayekar¹*, Amitha M. Hegde¹, Swathi Shetty² and Shanaya Khadkhodayan³

¹Department of Pedodontics and Preventive Dentistry, A. B. Shetty Memorial Institute of Dental Sciences, Nitte University, Derlakatte, Mangalore, Karnataka, India
²Department of Pedodontics and Preventive Dentistry, A J Institute of Dental Sciences, Mangalore, Karnataka, India
³A. B. Shetty Memorial Institute of Dental Sciences, Nitte University, Derlakatte, Mangalore, Karnataka, India

ABSTRACT

Literature shows that honey has therapeutic features indicating it’s potential for the treatment of various oral diseases. Natural honey sourced from honeycombs and commercially available honeys have been shown to differ in their composition, which includes the concentration of antioxidants in them. Studies evaluating the antioxidant levels of saliva on consumption of these honeys are sparse, hence the need for the study. Saliva samples of 30 caries free children were obtained and evaluated for their antioxidant levels using a spectrophotometer. Following which, they were given a tablespoon of natural honey and saliva samples were collected again to measure the antioxidant levels. On another occasion, the same procedure was repeated with commercially available honey. After assessing the antioxidant levels, a statistical analysis using the Students Paired ‘t’ test, revealed the following results: i) There was a significant (<0.001) increase in the antioxidant levels on consumption of either types of honey. ii) When comparing the antioxidant levels on consumption of both types of honey, the antioxidant levels were higher with commercially available honey than with natural honey. Although honey by itself may not serve as a major source of dietary antioxidants, it demonstrates the potential for honey to play a role in providing antioxidants in a highly palatable form, as was seen in this study.

Keywords: antioxidants, honey, saliva

INTRODUCTION

Honey is an ancient remedy for the treatment of infected wounds, which has recently been ‘rediscovered’ by the medical profession, particularly where conventional modern therapeutic agents are failing [1]. An alternative medicine branch, called apitherapy, has developed in recent years, offering treatments based on honey and other bee products against many diseases [2]. The large volume of literature reporting its effectiveness indicates that honey has the potential for treatment of periodontal disease, mouth ulcers and other problems of oral health. Being concentrated plant secretions, honey contains various phytochemicals, some of which are antioxidants [3]. Although honey has been used since a long time, only recently has its antioxidant property come into the limelight [4].

Natural honey sourced from honeycombs and commercially available honey has been shown to differ in their composition. Studies with honey selected to have a high level of antibacterial activity have been established but selection of honeys with high levels of antioxidants is currently just at the research stage [3]. Studies evaluating the antioxidant levels of saliva after consumption of natural honey and commercially available honey are sparse, hence the need for the study.

Aims of the study:

i. To assess the antioxidant levels of saliva on consumption of natural honey.
ii. To assess the antioxidant levels of saliva on consumption of commercially available honey.
iii. To compare the antioxidant levels of saliva obtained, on consumption of natural honey and commercially available honey.

EXPERIMENTAL SECTION

Source of data:
Fresh pure honey extracted from honeycombs around Mangalore was used as the natural honey. The honey did not contain artificial preservatives or diluents, nor was it heated by any artificial method. The honey was stored in stainless steel containers and in the dark, at room temperature. A commonly consumed brand of commercially available honey sourced from local stores was used for comparative evaluation in this study.

30 children in the age group of 6-12 years visiting the Department of Pedodontics and Preventive Dentistry at A.B. Shetty Memorial Institute of Dental Sciences, Mangalore were selected for the purpose of this study. After explaining the procedure, informed consent was obtained from each of them. Ethical clearance was obtained from the concerned institutions prior to conducting the study.

Inclusion criteria:
• Healthy caries free children, in the age group of 6-12 years.

Exclusion criteria:
• Children below the age of 6 years and above 12 years of age.
• Children with caries.
• Children with systemic illness, including allergies and on any medications.

Saliva collection:
The saliva was collected without any stimulus under standard temperature and humidity conditions in the morning. All subjects were asked to refrain from eating or drinking anything for a minimum of 2 hours before saliva collection. Subjects were comfortably seated and after a few minutes of relaxation were trained to avoid swallowing saliva and asked to lean forward and spit all the saliva they produced for 10 minutes into a graduated test tube, through a glass funnel. The saliva was then assessed to estimate the total salivary antioxidant level of the volunteer.

Estimation of total salivary anti oxidant concentration
Saliva was collected in an effendrop tubes and levels of total antioxidants levels were estimated in ul/ml as follows:

Collection of unstimulated saliva

Maintenance of sample at 4°C until evaluation

Analytical procedure
1. Mixing of chemicals including NaBenzoeate, acetic acid and ethanol to prepare the reagent.
2. Mix the reagent with the saliva

Incubation at 90°C for 90 minutes

Estimation of TAC using spectrophotometer
Estimation of total salivary antioxidant concentration on consumption of natural honey
The volunteers were asked to consume a tablespoon of natural honey, after which the salivary antioxidant level was again evaluated following the above mentioned methodology.

Estimation of total salivary antioxidant concentration on consumption of commercially available honey
On the following day, under the same conditions, the same group of volunteers was asked to consume a tablespoon of commercially available honey, after which the salivary antioxidant level was again evaluated following the above-mentioned methodology.

Statistical analysis
The results obtained were evaluated using the Student’s Paired ‘t’ test to compare the total salivary antioxidant levels on consumption of natural and commercially available honey.

RESULTS

i. There was a significant increase (<0.001) in the salivary antioxidant levels with the consumption of both, the natural honey as well as the commercially available honey (Table 1).

<table>
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<tr>
<th>Pair 1</th>
<th>Mean</th>
<th>Std. Deviation</th>
<th>t</th>
<th>df</th>
<th>Sig. (2-tailed)</th>
</tr>
</thead>
<tbody>
<tr>
<td>CONTROL Total Antioxidant capacity</td>
<td>0.973333</td>
<td>0.466819</td>
<td>-4.895</td>
<td>29</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Natural honey Total Antioxidant capacity</td>
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<table>
<thead>
<tr>
<th>Pair 2</th>
<th>Mean</th>
<th>Std. Deviation</th>
<th>t</th>
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<tr>
<td>CONTROL Total Antioxidant capacity</td>
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<td>Commercial honey Total Antioxidant capacity</td>
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<td>1.573944</td>
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</tr>
</tbody>
</table>

ii. On comparing the salivary antioxidant levels of natural and the commercially available honey used, there were higher levels of antioxidant levels seen on consumption of the commercially available honey (Graph 1).

Graph 1: Comparison of salivary antioxidants levels on consumption of natural honey & commercially available honey

DISCUSSION
Honey is the natural sweet substance produced by honey bees from nectar or blossoms or from the secretion of living parts of plants or excretions of plants, which honey bees collect, transform, and combine with specific substances of their own to ripen and mature. Honey is most commonly consumed in its unpreserved state, i.e. liquid,
crystallized or in the comb. In these forms, it is taken as medicine, eaten as food or incorporated as an ingredient in various food recipes [1].

The term “oxidative stress” describes the lack of equilibrium between the production of free radicals and the antioxidant protective activity in a given organism [5]. Natural and synthetic antioxidants have a long history as preservatives in the food supply. Their role in the human body has yet to be fully elucidated but much evidence indicates a role in counteracting the effects of naturally occurring free radicals that are associated with a number of diseases and the aging process. Antioxidants, such as those found in honey, play a role in combating damage caused by oxidizing agents in foods and in the human body [6,7].

Previous research has shown that even a single dose of honey can boost antioxidants, which is why we asked our volunteers to consume a tablespoon of either honey in this study. In a clinical trial to study the effect of topical application of pure honey on radiation-induced mucositis, patients were instructed to take 20 ml of honey 15 minutes before radiation therapy, then again at intervals of 15 minutes and six hours after radiation. The results showed the application of natural honey was effective in managing radiation-induced mucositis [8].

Among the compounds found in honey, vitamin C, phenol compounds, catalase, peroxides, glucose oxidase enzymes have antioxidant properties. Honey also contains flavonoids and carotenoids. High levels of these indicators ensure a high level of antioxidants in honey [9]. This may explain the significant increase (<0.001) in the salivary antioxidant levels with the consumption of natural and commercially available honey in this study.

In vitro experiments on the inhibition of oxidation in a model system using various honeys have demonstrated a wide variation in the antioxidant capacity among floral sources. Honey made by bees fed herbal extracts exhibit greater antioxidant activity than normal honey [10]. In general, higher antioxidant content was also found in darker honeys and in honeys with higher water content [11]. This may explain why we found higher levels of antioxidants on consumption of the commercially available honey.

There is a significant correlation between the antioxidant activity, the phenolic content of honey and the inhibition of the in vitro lipoprotein oxidation of human serum [12]. The influence of honey ingestion on the antioxidative capacity of plasma was tested in two studies [13,14]. In the first one, the trial persons were given maize syrup or buckwheat honeys with a different antioxidant capacity in a dose of 1.5 g/kg body weight. In comparison to the sugar control, honey caused an increase of both the antioxidant and the reducing serum capacity. In the second study humans received a diet supplemented with a daily honey serving of 1.2 g/kg body weight. Honey increased the body antioxidant agents: blood vitamin C concentration by 47%, β-carotene by 3%, uric acid by 12%, and glutathione reductase by 7%.

**ADVANTAGES OF HONEY:**
- Even though the potency maybe highly varied, its properties are said to remain consistent.
- Anti-inflammatory, anti-bacterial and anti-oxidative are the three main properties of honey. Hence it can be used to treat oral diseases and conditions like periodontitis, mouth ulcers and radiation mucositis.
- Apart from its above-mentioned benefits, it happens to be a natural commodity with nearly zero side effects. This would certainly increase patient compliance as it plays to the human psyche, i.e. natural is better than artificial.
- The pleasing taste of honey makes it highly palatable, allowing individuals reluctant to include other plant-derived antioxidants in their diet, to more readily consume it.
- Also, it is highly economical and would hence, be an option for treating even the lower strata of society, as it would be well within their means. It is already on its way to replace refined sugar in the commercial market.
- Technology has been developed to produce honey in the form of candy, lozenges, gels and syrups that would make it suitable for various medical and dental therapeutic applications. It is only a few trials and tests away from replacing certain artificial drugs too.

**DRAWBACKS OF HONEY:**
- Due to the diversity in the natural source of its ingredients (flowers), it has a huge variation in its potency depending on where, when and how it has been prepared.
- Honey is a dietary reservoir of C botulinum spores for which there is both microbiological and epidemiological evidence [15]. In order to minimise the risk of infantile botulism, it is recommended not to give honey to an infant who is less than 1 year old [16].
CONCLUSION

The medicinal advantages of this simple commodity, honey, is astounding and has given it a firm position in the research section of the dental and medical field as a therapeutic agent. Honey itself may not serve as a major source of dietary antioxidants but it has the potential to play a role in providing antioxidants in a highly palatable form, as was seen in our study. Even though further studies are required to determine its exact potential in this vast field, it has been successful in proving itself to be safe and potent. Certainly while considering a child’s diet, compared to sucrose, which is cariogenic and has no antioxidant value, honey can be a flavorful, supplementary source of antioxidants in the diet.

REFERENCES