

Antimicrobial Effect of Coconut Flour on Oral Microflora: An *in vitro* Study

¹Jamileh Bigom Taheri, ²Felipe Wilfredo Espineli, ³Hans Lu, ¹Maryam Asayesh,

¹Mahin Bakhshi and ⁴Mohammad Reza Nakhostin

¹Department of Oral Medicine, Faculty of Dentistry,

Shahid Beheshti University of Medical Sciences, Tehran, Iran

²College of Dentistry, ³College of Microbiology, Centro Escolar University, Philippines

⁴Restorative Department, Faculty of Dentistry,

Shahid Beheshti University of Medical Sciences, Tehran, Iran

Abstract: Coconut flour is made from fresh organic coconut meat. It can be used as substitute to usual flours. Although, it contains carbohydrates, it has very low glycemic index. Furthermore, it also has antimicrobial properties due to its high lauric acid content that had been used as medicaments for some oral infections such as mouth sores. More recently, lauric acid has been recognized for its unique properties in food use which are related to its antiviral, antibacterial and antiprotozoal functions. To determine its antimicrobial property, 45 plates using nutrient agar were inoculated with oral microflora sample from 15 dental students. The plates were added with saturated coconut flour at 100, 80, 60 and 40% concentrations. These were incubated for 48 h and the zone of inhibition was then measured. The results had shown that there was no zone of inhibition present on the experimental plates of all the concentrations which implies that the commercially available coconut flour had been negative for the antimicrobial test performed. However, 2% of the results had shown growths of molds around the Agar Well's rims at its pure concentration. The commercially available coconut flour has no effect on microbes of the oral cavity and no antimicrobial property at any concentrations. However, it may initiate the growth of molds at its pure concentration. To prove the effectiveness and health benefits of coconut flour in the oral cavity as an antimicrobial agent in the form of oral strip.

Key words: Coconut flour, lauric acid, antimicrobial effect, oral, antiviral, fresh organic

INTRODUCTION

The universal tendency towards the herbal medicine has been considerably high so that the financial volume of herbal medicines sold during 1998 reached to almost 4 billion dollars in the USA (Neville *et al.*, 2009). Many of herbal medicines have been consumed for 100 of years however, assessing the efficacy of plant medicines in comparison with current chemical drugs, requires the performance of accurate investigations (Ernest and Pitter, 2002). *Cocos nucifera* is commonly known as coconut widely used in the tropical zone. The most important coconut producing countries in the world are Philippines, India, Sri Lanka, Malaysia and Indonesia. Coconut is widely used in Asian countries for a variety of purposes (Srivastava and Durgaprasad, 2008). Coconut is not only known for providing meat, juice, milk and oil but it is also a good source of flour. Coconut flour can be used as substitute to usual flours. Although it contains carbohydrates, it has low glycemic index (Trinidad *et al.*, 2003). Furthermore, it also has antimicrobial properties due

to its high lauric acid content that had been used as medicaments for some oral infections such as mouth sores. Lauric acid, the major fatty acid which constitutes 50% of fat from the coconut has long been recognized for the unique properties that it lends to nonfood uses in producing soaps and cosmetics industry. More recently, lauric acid has been recognized for its unique properties in food use which are related to its antiviral, antibacterial, antifungal and antiprotozoal functions. This fatty acid is found in the largest amounts in lauric fats, especially from coconut as well as other coconut products such as the coconut flour (Ogbolu *et al.*, 2007; Rouse *et al.*, 2005; Yang *et al.*, 2009). Since, it may help improve the oral health conditions of those who can not afford to buy the expensive effective oral care products available in the market we have done this study.

MATERIALS AND METHODS

The researchers used the experimental method which involves manipulating condition and studying

effects to determine the microbial population which was based on the outcome of study.

Subjects of study: The researchers used the simple random sampling or fishbowl technique, this involves taking 15 healthy dental students of Centro Escolar University, College of Dentistry in Philippines with the age ranging from 20-24 years old.

Preparation of the cultured saliva samples: The soft tissue area including cheeks and tongue were swabbed using the cotton swabs. The cotton swabs were immersed in 15 test tubes which contained 10 mL of the prepared nutrient broth each. These were sealed sterilized plastic caps to avoid contamination. The test tubes were incubated for 48 h.

Preparation of coconut flour at different concentration: In an Erlenmeyer flask, 300 mL of distilled water was measured. Using the platform balance 50 mg of coconut flour was weighted. It was thoroughly mixed with the distilled water. Then 13 mg was added to make the solution saturated. The saturated coconut solution was the 100% concentrated. The different concentration was obtained using the procedure indicated below. The 200 mL saturated coconut flour was divided into 4 equal parts. Of 80% concentration. Out of that 50 mL saturated coconut flour, 40 mL of the solution was added to 10 mL distilled then it was thoroughly mixed in an Erlenmeyer flask.

This was stirred with a stirring rod for the 60% concentration. From the saturated coconut flour solution 30 mL of the solution was transferred to another flask and was mixed with 20 mL distilled water. To get 40% concentration, 20 mL of the saturated solution of the coconut flour was poured to a flask and was mixed with 30 mL distilled water. The flasks were labeled with 100, 80, 60 and 40% concentration, respectively. This same procedure was repeated for this trial.

Experimental procedure: About 15 subjects were used as controlled group and experimental group. In the controlled group, only 10 cc of saliva sample was poured on the 1st 15 nutrient agar plates while in the experimental group 10 cc of saliva sample and 10 cc of coconut flour were poured in the 2nd 15 plates. These plates were incubated for 24 h for the preliminary counting of the colony forming unit.

The 3rd plates were punched with 5 holes using sterile cork bores. The 4 holes were punched in a position that it would represent the 4 corners of a square and the 5th hole was placed at the center of it. Using the serological

pipette, 5 cc each coconut flour concentration were placed in the four holes and 5 cc distilled water at the center for the placebo effect.

Incubation of the plates for colony forming unit: The 15 plates with the inoculated saliva samples and the other 15 plates which contained different coconut flour concentration were placed in the incubator for 24 h for data gathering and recording. The experimental procedure was done in 2 trials to get an accurate result of the effectiveness of the coconut flour as an antimicrobial agent. The result of the zone of inhibition calculated by subtracting the diameter of the placebo from the diameter of the agar well which contains the coconut flour concentration.

RESULTS AND DISCUSSION

The results showed CFU mean value of 36.4 colonies in saliva of control group and 37.6 in saliva of experimental group. It had no significant difference by statistical analysis. Table 1 shows that after 2 experimental trials, a high number of experimental plates exhibit no zone of inhibition but there were few plates presents a zone of inhibition the 40 and 60% concentration. Among the 2 concentrations which present a zone, 40% concentration is greater. There have been some changes that occurred on the 2% of the agar well plates. There was a growth of molds around the rim of the 100% concentration. The results had shown that there was no zone of inhibition present on the experimental plates of all the concentrations which implies that the commercially available coconut flour had been negative for the antimicrobial test performed.

However, 2% of the results had shown growths of molds around the Agar Well's rims at its pure concentration. Coconut flour is a high fiber, high protein, low fat, gluten free and low glycemic index product. The fatty acid content of coconut flour is mostly lauric acid that causes some of the antifungal, antiviral and antibacterial properties of coconut. So, it is a medium chain fatty acid which has the additional beneficial function of being formed into monolaurin in the human or animal body. Monolaurin is the antiviral, antibacterial and antiprotozoal monoglyceride used by the human or animal to destroy lipid coated viruses, including *Listeria monocytogenes* (Rouse *et al.*, 2005). Department of Food

Table 1: Mean value of inhibition zone in different concentration of coconut flour after 24 h incubation

| Concentration of coconut flour (%) | Zone of inhibition (mm) |
|------------------------------------|-------------------------|
| 100 | 6 |
| 80 | 6 |
| 60 | 6 |
| 40 | 6 |

Science and Human Nutrition, Clemson University, South Carolina, USA studied in EDTA, lauric acid, nisin and combination of 3 antimicrobial agents on *Listeria* and *Salmonella* that can grow during food transport and storage and they can demonstrate bactericidal and bacterio static effect of them (Raybaudi-Massilia *et al.*, 2006). In the study, slight changes of the mean value of the CFU count gave no statistical significance. Therefore, there had been no evidence of decrease or increase in number of colony forming unit on experimental plates compared to the control group of plates, this implied that coconut flour has no effect on the CFU count of the microbes in the oral cavity. In other researches, different concentration had not been studied and we assayed efficacy of different concentration of coconut in oral microflora.

In a clinical trial sodium hypochlorite and coconut soap compared as disinfecting agents in the reduction of denture stomatitis, *Streptococcus mutans* and *Candida albicans*. The association of coconut soap and 0.05% sodium hypochlorite significantly reduced clinical signs of denture stomatitis but not significantly and the association of coconut soap and 0.5% sodium hypochlorite was effective in controlling denture biofilm (Barnabe *et al.*, 2004). A cross-sectional laboratory study to determine the *in vitro* sensitivity and resistance of organisms in culture isolates from skin infections and mechanisms of action of monolaurin, a coconut lauric acid derivative, compared with 6 common antibiotics: penicillin, oxacillin, fusidic acid, mupirocin, erythromycin and vancomycin.

It was concluded that monolaurin has statistically significant *in vitro* broad-spectrum sensitivity against gram-positive and gram-negative bacterial isolates from superficial skin infections. Most of the bacteria did not exhibit resistance to it. Monolaurin needs further pharmacokinetic studies to better understand its novel mechanisms of action, toxicity, drug interactions and proper dosing in order to proceed to *in vivo* clinical studies (Carpo *et al.*, 2007). Researchers found that there was no change on diameter of the regional measurement of the wells which contains the flour coconut flour at 80, 60 and 40% concentration. It depicted that there was no zone of inhibition that occurred on the experimental plates. However, there have been some changes that occurred on the 2% of the agar well plates.

There was a growth of molds around the rim of 100% concentration. This evidently showed the growth of molds due to the contamination of these samples. These may be avoided if the researchers added streptomycin to the agar plates but this may deviate the experiment from its main purpose which is to determine the antimicrobial

property of coconut flour but not its susceptibility to microbes. Based on the conclusions, the researchers came up with the following recommendation, further study on the microbial effect on specific organisms and pathologic disease in the oral cavity must be done and further experiments using coconut flour as a medium for producing improved oral stripes due to its purely organic characteristic must be done by future researchers.

CONCLUSION

The commercially available coconut flour has no effect on microbes of the oral cavity and no antimicrobial property at any concentrations. However, it may initiate the growth of molds at its pure concentration.

RECOMMENDATIONS

Further study on the microbial effect on specific organisms and pathologic disease in the oral cavity must be done. Further study on other properties of the coconut flour their tannic acid, lauric acid and capric acid content, should be made.

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