



Comparative Evaluation of Bacterial Contamination and Antibacterial Efficiency in Charcoal Bristled Bamboo Tooth Brush and Charcoal Bristled Nylon Tooth Brush -An In vivo study

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Abstract

Background-Maintaining the oral hygiene reduces the load of microorganism and the oral diseases can be controlled. The use of charcoal bristled bamboo tooth brush has made a boon in oral hygienic practice owing its better cleaning efficiency **Aim and Objective**-The aim of this study was to compare and evaluate Bacterial Contamination and Antibacterial Efficiency in Charcoal bristled Bamboo Tooth Brush and charcoal bristled Nylon Tooth Brush. **Materials and Method**-A total of 50 patients met inclusion criteria were chosen for the study. Oral prophylaxis and oral hygienic instructions were given. All participants were given first with charcoal bristled nylon tooth brush and tooth paste. After a washout period of 14 weeks, it is replaced by charcoal bristled bamboo tooth brush. Bristles of both the tooth brushes sectioned separately and placed in a 5ml of saline, followed by isolating 0.1ml of saline and cultured in blood agar for 24hours of incubation. The colony forming units were counted later. To evaluate the antibacterial efficiency, the zone of inhibition was evaluated. Data collected were analysed and compared with student t-test. **Result**-The mean colony forming units was found more in charcoal bristled bamboo tooth brush. The mean zone inhibition was found around 11.32mm for charcoal bristled nylon toothbrush and 4.94mm for charcoal bristled bamboo tooth brush. **Conclusion**-The study has shown the retention of colony forming units more in charcoal bristled bamboo tooth brush. The zone of inhibition found around charcoal bristle nylon tooth brush supported better antimicrobial properties compared to charcoal bristled bamboo tooth brush

Key words- Colony forming units, Antimicrobial efficiency, Charcoal bristled Bamboo Tooth Brush, Charcoal bristled Nylon Tooth Brush

Introduction

Oral cavity is free of microorganism before birth, because the foetus develops in a well-protected environment and soon after birth, colonization of bacterial will occur and later different microorganisms creates their own habitat in the oral cavity. Proper oral hygiene maintenance helps in reducing the microbial load and thereby controls the initiation of oral diseases¹. More than 700 species of bacteria have been identified in the environment. The continues colonization of this microorganism can promote different types of oral diseases. A major chunk of microorganisms has been implicated in the pathogenesis and perpetuation of oral diseases such as periodontitis, dental caries, gingivitis,

The basic mode of maintaining oral hygiene is by proper tooth brushing and if we look back to the ancient era, the people used to practice oral hygiene by using natural product such as neem, miswak, babul etc. A revolution to the use of this has happened at 1844 by the invention of first tooth brush by Dr. Meyer L. Rheinhe has patented a tree row brush of serrated bristles with large tufts. Later in 1920 Coombs has proposed the contamination of tooth brush after use, can harbour variety of bacteria, fungi, and virus. Toothbrushes are generally stored in a common container in the bathroom, which can introduce microbes to the toothbrush. Toothbrush gets contaminated from the unhygienic oral cavity, surroundings, hands, or storage site. Bacteria that attach, accumulate & multiply on toothbrushes may be transferred to the oral cavity leading to different diseases. Thus, it is essential to frame uniform oral health guidelines to prevent toothbrush contamination. Contaminated toothbrushes may broadcast microorganisms, which could be detrimental to oral and systemic health.

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For the advancement of better oral hygienic practices, toothbrushes are coming with varying size, shape, texture, design. Recently, the bristles of tooth brush have been altered in its lengths which favour to reach interproximal areas more easily. Handles have also been ergonomically designed to accommodate multiple dexterity levels.

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A new variant of toothbrush called charcoal bristled bamboo toothbrushes, has been introduced into the market, which is prepared by blending binchotan charcoal into the bristles⁴. Manufacturers claims that they have antimicrobial properties in them, resulting in less bacterial contamination⁵.

Bamboo toothbrushes are made of bamboo, an environmentally sustainable timber. The bamboo is heat-treated to carbonize the surface of the bamboo, giving it a quality finish and good service life. A review of literature shows that microorganisms are strongly adhered to the toothbrush⁶. So, the retention and survival of microorganisms on toothbrush after brushing represents a possible cause of re-contamination of the mouth. Charcoal bristled bamboo tooth brush which is new to modern dental care will have better efficiency, but less studies have put forwarded for it⁷.

Therefore, the aim of the study is to compare and evaluate the bacterial contamination and antibacterial efficiency of charcoal bristled bamboo tooth brush and charcoal bristled nylon tooth brush.

Materials and method

A total of 50 students aged between 6-13 years, was chosen based on the following inclusion criteria

1-Patient with fully erupted primary tooth,

2-Patient who are willing to come for follow up

The exclusion criteria were patients with open carious lesions, poor plaque scores (plaque index scores of >2), severe gingivitis (gingival index score >2), throat infections, irregular brushing frequency, those using mouthwash and/or antibacterial toothpastes, smokers, or those who were medically compromised.

After choosing the proper sample population, Oral prophylaxis was done and, brushing technique and oral hygienic instructions were given for students who are participating in the study. The participants were advised not to use any type of mouth wash, and wash the tooth brush bristles under running water without using their fingers to clean the bristles. They were also advised not to cover the tooth brush bristles with a cap and keep the bristles side upright, Also the tooth brush should kept 2 feet way from the commode area.

In this study, all the participants were first given charcoal bristled bamboo tooth brush and instructed to follow the instructions. On the 14th day all the tooth brushes were collected back and given charcoal bristles nylon tooth brush to the same participants and advised to follow the same instructions. So, all samples got collected within 28 days.

Assessment of colony formation unit on tooth brush

It was done on nylon and bamboo toothbrush after 14 days of the use, The participants received individual sterile pouches into which the tooth brush has to return. The head of the tooth brush is cut apart, and one-third of the bristles were cut and collected on separate sterile Petri dishes. Using sterile forceps, toothbrush bristles were placed in separate test tubes containing a 5 ml of saline swirled. A sterile pipette was used to extract 0.1 ml of saline, which was poured onto an agar plate. These agar plates were then placed in gas pack jar for anaerobic culture. The agar plates with gas pack jar were then placed in the incubator for 48 hrs ,after which the colony forming units (CFU) present on each agar plate of microbial growth were noted⁹.

To evaluate antibacterial efficacy of, the zone of inhibition was evaluated for both after 24 h of incubation¹⁰.





Statistical analysis

Data were entered in Microsoft Excel spreadsheet and data were analysed using statistical package of social sciences 25.0 software (SPSS Inc., Chicago, USA). The data were analysed for probability distribution. Inter group comparison was done using Paired t-test. For all statistical purposes, a p-value of ≤ 0.05 was considered significant

Results

A total of 100 toothbrushes were collected from the participants (50-charcoal bristled bamboo tooth brush and 50-charcoal bristled nylon tooth brush). So, a total of 100 agar plate were made to evaluate the colony forming units. (Figure 1)

All the 100 plates were identified with bacterial colony formation

Table 1 presents the results of the paired sample t-test comparing the number of CFUs between the 2 types of bristles. The mean CFUs for charcoal bristles bamboo tooth brush was 99.88 and mean CFUs for bamboo charcoal nylon tooth brush was 51.52 and the difference between the groups was statistically significant (p-0.013).

Table 2 presents the comparison of mean inhibition zone between the groups. The mean zone of inhibition for charcoal bristles bamboo tooth brush was 4.94mm and mean zone of inhibition for charcoal bristles nylon tooth brush was 11.32 mm and the difference between the groups was statistically highly significant (p-0.000).(Figure 2)

Study Groups	n	Colonies forming units (CFUs) Mean±SD	Standard error of the mean	95% Confidence Interval (lower-upper)	p-value
Group 1 (Bristles of used charcoal bamboo tooth brush)	50	99.88±1307	18.49142	10822 85897	0.013*
Group 2 (Bristles of used charcoal nylon tooth brush)	50	51.52±1212	17.1506		

Table 1- Comparison of the number of colonies forming units (CFUs) between bristles of charcoal bamboo tooth brush and charcoal Nylon toothbrush (n-100)

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Study Groups	n	Zone of inhibition (mm)	Standard error of the mean	95% Confidence Interval (lower-upper)	p-value
Group 1 (Bristles of used charcoal bamboo toothbrush)	50	4.94±1.47	0.2088	-5.0520	0.000*
Group 2 (Bristles of used charcoal nylon toothbrush)	50	11.32±4.33	0.6130	7.7079	

Table 2- Comparison of zone of inhibition between bristles of charcoal bamboo tooth brush and charcoal Nylon toothbrush (n-100)

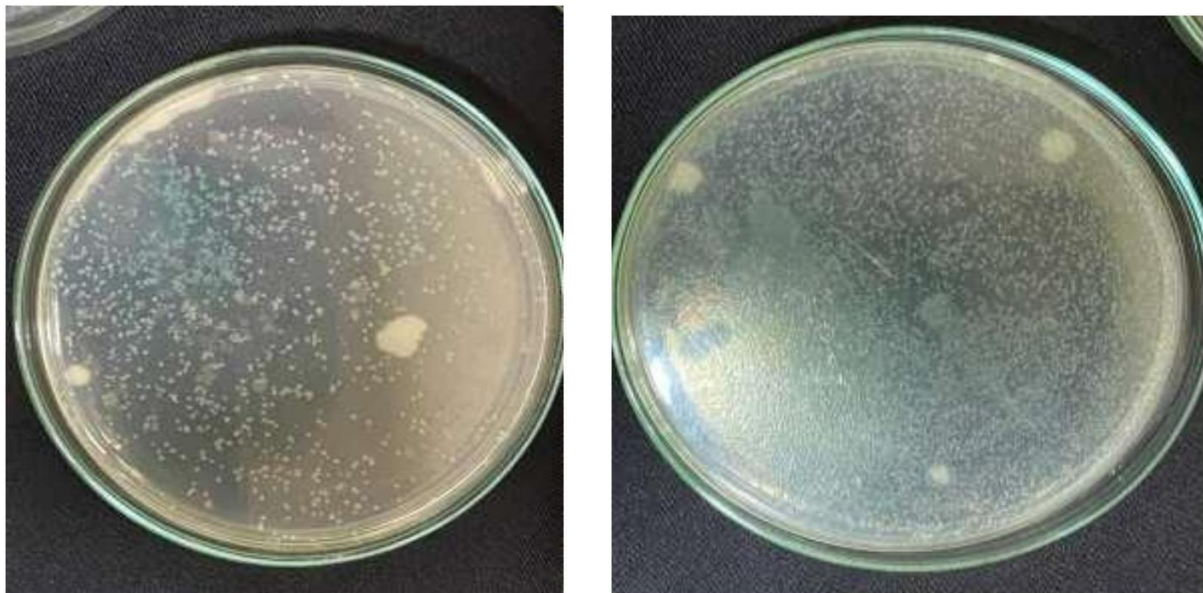


Figure 1

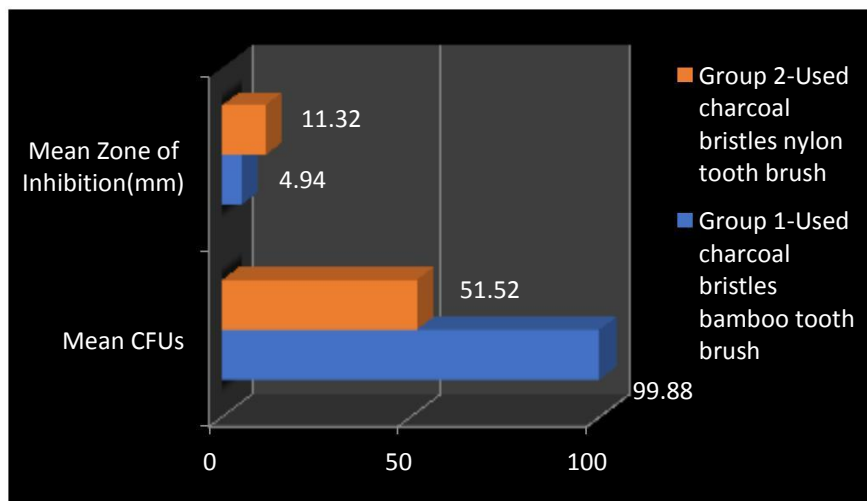


Figure 2- Comparison of the number of colonies forming units (CFUs) and zone of inhibition between bristles of charcoal bamboo tooth brush and charcoal Nylon toothbrush

Discussion

There has been growing consciousness about the environment worldwide, where consumers demand products that are made from materials that are sourced in a sustainable manner and have a low environmental footprint. Bamboo is an eco-friendly timber substitute for nondegradable materials. The past couple of decades have witnessed rapid growth in commercial applications of bamboo⁸. According to world bank report, the use bamboo toothbrushes is reported to have gone up exponentially. According to the literature reports, the toothbrush contamination in healthy individuals happens early after the first use, and it gets higher with its further usage. Oral cavity is a place with the highest concentration of different microbial populations (more than bacterial 700 species⁹, from which 400 are found in the periodontal pockets); it is especially colonized by *Staphylococcus* sp., *Streptococcus* sp., *Neisseria* sp., *Bacteroides* sp., *Actinomyces* sp, *Treponema* sp., *Mycoplasma* sp. These microorganisms can settle the toothbrushes, as well, including the organisms which are not normally associated with the oral flora, like the family of Enterobacteriaceae. Studies have indicated that the area between the tufts of bristles on a toothbrush can be a breeding ground for microorganisms, as well as a source of their growth¹¹.

A study conducted in vitro revealed that toothbrushes were heavily contaminated with a range of microorganisms, some of which, such as *S.mutans*, are responsible for the onset of dental caries, while others, like lactobacilli, are linked to the advancement of dental caries and *Candida* can cause candidiasis. Microorganisms can reproduce and their population can grow on toothbrushes, which could be a potential risk, particularly for people with weakened immune systems, diabetes, vascular diseases, and the elderly.

The findings of the case described Nayak L et al., 2016 stated that bamboo toothbrushes have antimicrobial properties, sustainability and biodegradable¹², whereas plastic toothbrushes made of organic and inorganic materials had a positive result with the present study.

Al-Ahmad et al, 2015 studied the antimicrobial effect of silver-coated toothbrush heads in-vitro, he concluded that there was no significant reduction in the CFUs by silver-coated toothbrushes for the above-mentioned tested organisms¹³.

This study found that bamboo toothbrushes had higher bacterial retention, which may be attributed to property of absorption of a significant amount of moisture at standard humidity conditions. The moisture-rich environment in the toothbrush head can be a harbours bacterial growth.

The results showed that with in the duration of use, the toothbrush bamboo tooth brush has shown high bacterial contamination and also its shows variation in the antibacterial efficiency.

Caudry et al. found that bacteria are strongly adhered to the toothbrush filaments and the retention of moisture, epithelial and oral debris in the filament bundles raise the bacterial survive¹⁴

Raiyani CM et al. 2015 pointed that toothbrush head had heavy contamination when used for 3 months period compared to 1 month used toothbrush⁸, the usage of the same toothbrush in an extended period of time makes it a reservoir of microorganisms despite the fact that it is used to lower the present flora in the dental plaque, so the contaminating microorganisms may be imported in the mouth again. The time period of using the toothbrush may also be connected with its contamination, hence not only the proper cleaning will reduce the microbe

volume, but the replacing of the used toothbrush with a new one is a condition for better oral health in individuals, as well

Taghreed Almutairi et al., 2014 depicted those contaminated toothbrushes and their bristles during their everyday use plays a key role in the transmission and survival of microorganisms¹⁵.

Glass and Shapiro pointed that contaminated toothbrushes may have a role in developing of local and systemic diseases. The possibility these toothbrushes to be associated with the influence to some systemic conditions, like heart diseases, arthritis, bacteremia and brain stroke has been already documented¹⁶.

It is obvious that there is insufficient information about the contamination of charcoal bristled bamboo toothbrush as a factor that can influence the oral health. Our expectations are that the dentists will become more active in including the advices and recommendations for proper toothbrush for the oral hygiene methods, because of the high bacterial contamination that dynamically increases with the time and the type of toothbrush used, which might increase the potential for worsening the patients' health

Conclusion

Our study has shown that the number of colony forming units in charcoal bristled bamboo toothbrushes were higher as compared to charcoal bristled nylon toothbrushes after 2 weeks of usage. Further studies should be conducted with a larger sample size, longer duration of use, and with identification of specific micro-organisms in the bristles.

Ethical clearance

Ethical clearance number (KDC/20/411P).

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Nil.

Conflicts of interest

There are no conflicts of interest

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