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Evaluation of Difference in Bacterial Contamination of Toothbrushes between Patients With Gingivitis And Patients with Healthy Gingiva-A Pilot Study

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Abstract

Aim: To Evaluate the difference in bacterial contamination of toothbrushes between patients with gingivitis and patients with healthy gingiva.

Objective: 1.To determine the bacterial contamination in terms of CFU/ml after brushing for a period of two weeks in patients with healthy gingiva.

2. To determine the bacterial contamination in terms of CFU/ml after brushing for a period of two weeks in patients with gingivitis.

3. To compare the difference in bacterial contamination in terms of CFU/ml between patients with healthy gingival and patients with gingivitis.

Background: The most commonly used method to maintain oral hygiene is toothbrush. Its main goal is to remove plaque, debris and stains which are responsible for gingivitis, periodontitis , tooth decay and halitosis. While removing, toothbrush becomes contaminated with blood, saliva, bacteria and soft debris. The toothbrush itself can act as a foci of infection and retard the disease prognosis and treatment outcomes.

***Keywords**: Toothbrush, Decontamination, bacterial colonization, brushing, gingivitis, healthy gingiva.*

Introduction

The human oral cavity is invaded by a more number of bacteria flora than any other anatomic area in the body .It has been found that more than 700 species of bacteria out of which 400 species were found in the periodontal pocket adjacent to teeth(1).Maintaining good health is very important for a good quality of life. The impact of oral health on general health has been proved time and again by many studies.(2,3,4,5,6) The mouth serves as a "window" to the rest of the body, providing signals of general health disorders. Bacteria from the mouth can cause infection in other parts of the body when the immune system has been compromised by disease or medical treatments (e.g., infective endocarditis). Systemic conditions and their treatment are also known to impact on oral health (e.g., reduced saliva flow, altered balance of oral microorganisms).Periodontal disease has an impact on cardiovascular system, this statement was proved by many stuides. In 2006, Holmlund et al., periodontal disease and number of remaining teeth related to a past history of heart attack and high blood pressure or hypertension .Other study showed that both periodontal disease and overall tooth loss from any cause are closely related to cardiovascular disease. Alman et al (2011) have shown a signicant positive association between loss of bone supporting teeth due to periodontal disease and CVD(25-28).Periodontal disease is often considered the 'sixth complication' of diabetes(29).

Oral Prophylaxis is a premise for oral health and daily plaque and debris removal is considered important

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for oral health. Improper maintenance of oral hygiene leads to the accumulation of plaque around the tooth which is a primary cause for gingivitis and periodontitis. Thus, removal of plaque plays a main key role in maintaining oral hygiene.

Tooth brushing is the most commonly used, easiest and effective method of oral hygiene practice performed around the world.(7) Toothbrush plays an important role in maintaining personal oral hygiene and it is effective tool for removing the plaque. Not only the proper selection but also care should be taken in maintaining the toothbrush which is essential for good oral hygiene because the toothbrush also gets contaminated by bacteria. Toothbrushes must have the following requirements to remove the plaque; stiff bristles which is enough to remove plaque without causing trauma to the teeth and gums and small head with soft bristles. Organisms are not only associated with oral cavity but also seen in tooth brush which includes Streptococcus mutans, Staphylococcus aureus, Pseudomonas, Lactobacillus, Klebsiella, Candida species(1).

Toothbrushes also has a significant role in disease transmission and increase the risk of infection since they can serve as a reservoir for microorganisms in healthy, oral-diseased and in immunocompromised people. Contamination is the state of retention and survival of infectious organisms that occur on animate or inanimate objects.(8) Contaminated toothbrushes may play a role in both systemic and localized diseases. This toothbrush contamination is associated with transmission of severe health problems which includes cardiovascular diseases, respiratory disorders, gastrointestinal diseases,arthritis, bacteremia ,renal problems and stroke.(7,8)

Toothbrushes can become contaminated from the oral cavity, environment, hands, aerosol contamination, and storage containers and the bacteria which attach to the toothbrush gets accumulated and survive on toothbrushes will helps in transmitting the diseases. In 1920 Cobb reported that toothbrush is the cause of repeated infections in the oral cavity (9). Contaminated tooth brush acts as an environment for microbial transport, retention and growth. Toothbrush heads between the bristle tufts is a favourable medium for the growth of microorganisms. This can be the cause of reinfection of a person with pathogenic bacteria (autoinoculation) or it can acts as a significant risk of dissemination of infection for certain patients such as immunosuppressed, cardiopathic ,organ transplant recipients(11).

So, the contamination of toothbrush can be prevented by immersing it in disinfectant solutions like 0.1% Chlorhexidine gluconate and 1% Sodium hypochlorite and replacing in a regular time period. So far many studies have evaluated the contamination risk of tooth brushes, within the bias of literature search, it was inferred that, none of the studies has focussed on the difference in contamination between a patient with gingivitis against health gingiva. This difference is studied and found to be true significance, it could help in patient education and help in better treatment outcomes. Hence this study was done to investigate and compare the bacterial load on toothbrushes used by patients with healthy gingiva and gingivitis.

Materials and Method

Study design: A non randomized clinical trial.

Study setting: Approximately 1000 patients are visiting saveetha dental college daily.

Among them, 90% of the patients are diagnosed with poor oral hygiene and they were given a demo of modified bass brushing method followed by health education to improve their oral health.

Study Population:18 to 45 years who visited the OP of saveetha dental college were selected based on the study criteria.

Eligibility Criteria:

Ø **Inclusion Criteria**:

· Patients with age group between 18-45 years ,

· Group-A(gingivitis)-Based on gingival index by Loe and Silness.

· Group-B(Healthy gingiva)- gingiva which is firm in consistency, with pink colour and scalloped margins were included in this study.

Ø **Exclusion Criteria:** Patients with a history of systemic disease(Myocardial infarction, ischaemic heart disease, COPD, Bronchial asthma, Hyperthyroidism, Hypothyroidism, Hypercholestrolemia, hypertension, diabetes mellitus, renal disorders, blood disorders, Parkinsons disease, cushing syndrome), patients who had periodontitis and who are not willing to participate were excluded from this study.

Informed consent:

· Prior to start the study written informed consent was obtained from all the participants.

· Institution ethical committe approval was also obtained prior to the study.

Sample size: Based on the study by Taji.et.al, the sample size of this present study was

10%.

Sampling: A non probability type of sampling was used. Selective /judgemental. Patients

visiting the OP was choosen based on the inclusion and exclusion criteria until

the sample size was achieved in each group.

Armamentarium:

The following equipments/materials were used for the study:

· Steriled mouth mirror

· Surgical gloves

· Steriled containers

· Normal saline

· Cuvettes

· Micropipette

· Petri dish

· Nutrient agar

· Spirit lamp

· Metal loop

· Incubator

Method

All the gingivitis patients were selected based on gingival index given by Loe H and Silness P (1963).For assessing the severity of gingivitis, and its location by examining qualitative changes of gingival tissues. The severity of gingivitis is scored on the selected index teeth(16,36,12,32,24,44) .Tissues surrounding each tooth divided into 4 gingival scoring units which are Disto-facial papilla, Facial margin, Mesio-facial papilla and Lingual gingival margin.

Grading of the gingivitis:

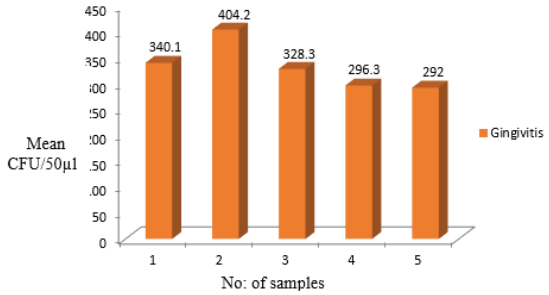
Score-0,gingival status is normal gingiva and the criteria is natural coral pink gingiva**;Score-1,gingival status is mild inflammation and the criteria is** slight changes in colour, slight edema. No bleeding on probing;Score-2 ,gingival status is moderate inflammation and the criteria is Redness, edema ,glazing and it bleeds on probing and score-3,gingival status is severe inflammation and the criteria is marked redness and edema/ ulceration/ tendency to bleed spontaneously.

All the examinees who met the criteria were informed about the study. Both were each given a new toothbrush with same brand of fluoridated tooth paste. Each subjects were given a demo of modified bass brushing method and they were requested to follow twice daily for a period of 2 weeks, since it is effecting in cleaning proximal and gingival sulcus. At the end of 2 weeks, brushes were collected in a sterile bag and processed.

Each toothbrush was then transferred into the container containing 10ml of steriled normal saline and mixed vigorously for 1 minute. After mixing,50µL of saline was transferred into the cuvette which is incubated at 37^0C for 1hr by placing in the incubator.

50µL of saline was then spread onto the plates of nutrient agar for the growth of an aerobic bacteria. Each sample was processed 3 times and incubated to minimise the manual and laboratory errors.The nutrient medium was incubated aerobically for 24hrs at 37^0C.Then total bacterial count was done. The results are tabulated which are as follows,

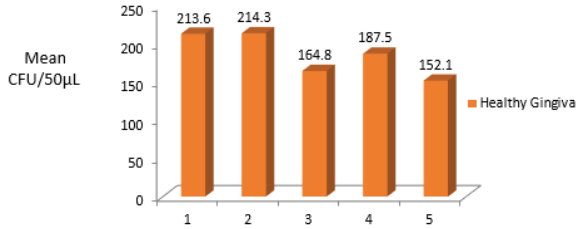
Result



Graph-1 shows the mean of bacterial colony counts of 5 samples in gingivitis patients.

Graph-1,depicts the mean of bacterial colony count in terms of CFU/50µL of all the 5 samples in gingivitis patients.Sample-1 has a mean of 340.1 cfu/50µL,sample-2 has a mean of 404.2 cfu/50µL, sample-3 has a mean of 328.3 cfu/50µL,sample-4 has a mean of 296.3cfu/50µL and sample-5 has a mean of 292cfu/50µL.

Figure-1 depicts the bacteria in agar plate of gingivitis patients.



Graph-2 shows the mean of bacterial colony counts of 5 samples in patients with healthy gingiva.

Graph-2,depicts the mean of bacterial colony count in terms of CFU/50µL of all the 5 samples in patients with healthy gingiva. Sample-1 has a mean of 213,6 cfu /50µL,sample-2 has a mean of 214,3cfu/50µL, sample-3 has a mean of 164.8 cfu/50µL,sample-4 has a mean of 187.5 cfu/50µL and sample-5 has a mean of 152.1 cfu/50µL.

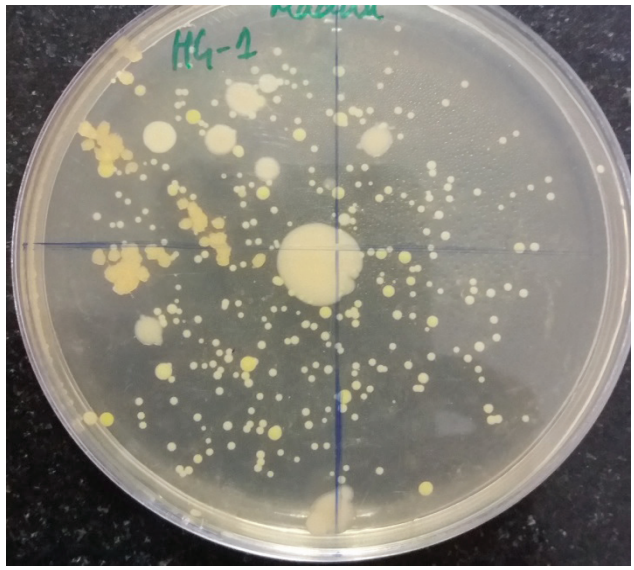


Figure-2 depicts the bacteria in agar plate of patients with healthy gingiva.

Discussion

The result of this study showed that the bacterial contamination was more in toothbrush used by gingivitis patients than the patients with healthy gingiva and the predominant microorganisms isolated were S. aureus, and S.mutans.

In the present study, microbial contamination was seen in all the 10 toothbrushes (100%) and this finding was consistent with some previous studies found microbes on all of the tested toothbrushes[12-15].But in one of the previous studies, microbial contamination was seen in 7 out of 10 toothbrushes (70%)[16].Bunetel et al. found that toothbrushes used by patients with existing oral disease quickly became contaminated [18].Several of the studies found that toothbrushes were contaminated before use[17-20]. Caudry et al. found that toothbrushes are heavily contaminated with normal use[8].

In the present study, Predominant microorganisms isolated were S. aureus, and S. mutans and this finding was consistent with most similar studies[12,13,14].In other

study, Microbial growth was detected on almost all of the brushes tested in this study (>90%), with development of streptococci observed on the vast majority of the brushes, which shows that toothbrushes are an excellent means of transport for bacteria. Nearly half of the brushes showed growth of mutans streptococci, members of the oral microflora, that are currently considered to be major cariogenic agents [24]. Other study reported that toothbrushes are heavily infected with escheriachia coli followed by klebsiella pneumonia , streptococcus pyogenes, staphylococcus auerus [31].Glass found that toothbrushes from both healthy patients and patients with oral disease contained potentially pathogenic bacteria and viruses such as Staphylococcus aureus, E. coli, Pseudomonas, and herpes simplex virus[17].Svanberg M. found that toothbrushes could be heavily infected with microorganisims especially mutans streptococci[30].

In the present study, the mean of bacterial colony count in gingivitis patients ranges from 10^2 to 10^5 Colony forming units /50µL and in patients with healthy gingiva the mean ranges from10^1 to 10^3 colony forming units/50µL. In one of the previous studies, the total microbial load per tooth- brush was found to be $10^4$to 10^6colony forming units[15].

The American Dental Association recommends a routine change of toothbrushes every 3 months [7]. According to the reports of Denny and glass[23,24] healthy patients replace their toothbrush every two weeks. Patients who are sick should change their toothbrushes at the beginning of an illness, when they first feel better, and when they are completely well. Chemotherapy or immune-suppressed patients should change their toothbrushes every three days, and persons submitted to major surgery should change their toothbrushes every day. So, the replacement of toothbrush in regular time periods is very essential to prevent the continuation of reinfection of oral diseases.

Conclusion

1.The result of this study showed that the bacterial contamination was more in toothbrush used by gingivitis patients than the patients with healthy gingiva and the predominant microorganisms isolated were S. aureus, and S.mutans.

2.Toothbrushes have an important role in transferring microorganisms which increases the risk of infection. So, the dentist should be more responsible in order to aware the patients for the issue of choosing, keeping and maintaining the hygiene of the toothbrushes, as well as their replacement in regular period of time.

Limitations of the study:

- A first limitation was the time constraint .
- A second limitation was the small sample size**.**

Direction of the future research: It is the follow up of this present study, assessment of progression and prognosis of a disease by using decontaminant solutions.

Source of Funding- Self

Conflict of Interest -Nil

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