

Evaluation Of Bacteremia Following Periodontal Probing In Gingivitis And Periodontitis Patients.

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Citation

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Abstract

Background and aim

: This study was done to determine whether bacteremia occurred before after periodontal probing in gingivitis and periodontitis affected patients and to identify the microorganisms present within positive blood cultures by aerobic culture.

Materials and methods

: 20 patients for each in gingivitis and periodontitis groups were selected, all with negative history of any systemic disorders. 5ml of venous blood was collected from every patient before and within 30 seconds of full mouth periodontal probing and simultaneously oral hygiene simplified, plaque index and number of sites that bleed on probing were recorded. The blood samples collected were transferred to the culture bottles containing brain heart infusion agar and brain heart infusion broth and incubated, samples signaled positive were then subjected to microbiological investigation. The positive samples were sub cultured on blood agar plates and the organisms were confirmed by gram staining method.

Results

: Probing caused bacteremia in 9 out of 20 periodontitis, and 3 out of 20 gingivitis patients. Streptococcus species was the most isolated organism in both the groups. Number of sites that bleed on probing, total probing depth and mean probing depth were significantly associated with bacteremia. Conclusion: Based on the findings of this study, it can be concluded that patients with untreated periodontitis were at greater risk of experiencing bacteremia due to periodontal probing.

INTRODUCTION

Infective endocarditis is a rare, potentially fatal disease where susceptible endocardium or a prosthetic heart valve is colonized by microorganisms such as streptococci, staphylococci, and Candida. It is well recognized that dental procedures, which induce bacteremia of substantial magnitude, have an increased potential to induce infective endocarditis in susceptible hearts¹.

The human oral cavity has a large, varied population of bacterial organisms, particularly in the gingival crevice. In healthy gingiva, a thin surface mucosa of epithelium separates potentially pathogenic organisms from the general circulation, which supplies gingiva² through the cervical plexus of capillaries. These microorganisms are considered normal inhabitants and non pathogenic of oral cavity as long as they are confined within the mouth³. But inflammation in gingiva may result in gingival ulceration around the tooth, which results in discontinuity of this mucosal barrier⁴. After

dissolution of the continuity of the mucous membrane, microorganisms may reach the blood stream and cause complications to the body. This kind of bacteremia may be associated with chewing of food, brushing of the teeth and particularly after any oral surgical procedure. In healthy persons, the episodes of bacteremia are brief due to efficient clearance by the host immune system. However in many patients with impaired resistance owing to drug usage, damaged organs or suffering from diseases such as valvular heart disease, diabetes and acquired immune deficiency syndrome, bacteremia is associated with an increased risk of serious complications like infective endocarditis³.

As prevention of infective endocarditis is desirable, due to its high rates of morbidity and mortality, there is a general consensus that all patients with predisposing heart conditions be given appropriate antimicrobials prior to operative procedures that may give rise to infective endocarditis. This is based on the fact that infective endocarditis may follow a bacteremia and that certain dental procedures can produce

bacteremia with organisms having potential to cause infective endocarditis¹. According to American Heart Association, periodontal probing is under high-risk category for infective endocarditis⁵ and recommends antibiotic prophylaxis for patients at risk since periodontal probing is done for all patient's seeking dental treatment as a part of assessment of periodontal status.

AIMS AND OBJECTIVES

To investigate the occurrence of bacteremia due to periodontal probing in a group of patients with gingivitis as compared to a group with periodontitis.

To identify the microorganisms present within positive blood cultures [by aerobic culture].

To assess factors such as age, sex, plaque index, oral hygiene index-simplified, number of teeth probed, number of sites that bleed on probing, total probing depth and mean probing depth per tooth for any association with bacteremia.

MATERIALS AND METHODS

Forty adult patients, who have attended the dental out patient ward of department of periodontics, Rajah muthiah dental college and hospital, annamalai university, aged between 20 and 70 years, including males and females were randomly selected for the study. The ethical committee for research work of Annamalai University gave clearance to carry out the present study and procedure was explained to patients and was included, only after signing the consent form. Patients who have at least 15 natural teeth were included and Patients with history of periodontal treatment of any form in the preceding 3 months, congenital or acquired cardiac defect or cardiac prosthesis, hematological disorder or immune defect and patients taking antibiotics in the previous month or who were taking corticosteroids or immunosuppressive medication were excluded from the study.

STUDY PROCEDURE

Study procedure involved the following steps:

INITIAL BLOOD SAMPLING

Initial blood sampling was obtained by venipuncture from antecubital vein. Before obtaining blood, the area was wiped with povidone iodine and with 70% iso propyl alcohol and allowed to dry 5 ml of blood was obtained by means of disposable syringe. Immediately the blood sample was transferred into culture bottle containing biphasic medium of 50ml of brain heart infusion agar and brain heart infusion

broth.

CLINICAL EXAMINATION

During clinical examination oral hygiene index-simplified [OHI-S] - by Greene and Vermilion⁶ -1964, plaque index - by Loe and Silness⁶ -1964 were recorded:

PERIODONTAL PROBING

Periodontal probing was done for all teeth using a standard periodontal probe [Michigan 'o' probe with Williams markings].probing was done on mesial, mid, distal surfaces of buccal and lingual aspects of all the teeth. Probe was inserted parallel to the long axis of the teeth and presence or absence of bleeding, probing depth at that particular site was recorded simultaneously.

FINAL BLOOD SAMPLING

Immediately 30 seconds after periodontal probing, second blood sample was collected [5ml] under aseptic conditions and transferred to the culture bottle containing brain heart infusion agar and brain heart infusion broth.

Selected patients for the study were divided based on bleeding on probing and probing pocket depth into gingivitis [patients who had ≥ 12 sites that bleed on probing] and periodontitis [patients who had atleast six teeth with ≥ 5 mm of pocket depth] groups, with each group consisting of 20 patients [Patients later found not to fit into any category after probing and clinical examination were excluded].

MICRO BIOLOGICAL INVESTIGATIONS

Media employed was brain heart infusion agar and brain heart infusion broth. The biphasic medium of brain heart infusion agar and brain heart infusion broth was prepared as follows:

Brain heart infusion agar was prepared as per the instructions of the manufacturer, sterilized and 50 ml was poured into the sterile culture bottle. This was allowed to solidify on one side of the bottle; the following day, brain heart infusion broth was prepared as per the instructions of the manufacturer, sterilized and 50ml was added to the solid medium and stored in refrigerator until future use.

Blood samples which were collected were incubated for 48 hours at 37C. Culture bottles after 48 hours were examined for any turbidity. The bottles which signaled positive were taken and sub cultured on to blood agar plates. The streaked plates were incubated at 37C for 24-48 hours and observed for growth of any colonies[Fig 3,4]. The organisms were

confirmed by Gram's staining method. The bottles were observed for a period of seven days for growth. If there was no growth a blind sub culture was done from the bottles on to blood agar, before being discarded as negative. Smears were prepared from the suspected colonies on a new glass slides which were air dried and heat fixed. The smears were then subjected to Gram staining.

The morphology of the bacteria like shape, arrangement was observed and grouped into gram positive cocci and gram negative bacilli. Gram positive cocci in chains with the property of alpha-hemolysis on blood agar plates were confirmed as streptococcus viridians [Fig 3]. Gram positive cocci in clusters with the property of Beta-hemolysis on blood agar plates were confirmed as staphylococcus aureus by coagulase test [Fig 4].

STATISTICAL ANALYSIS

Statistical analysis was done for the data collected from parameters-age, sex, number of teeth probed, oral hygiene index-simplified, plaque index, number of sites that bleed on probing, total probing depth, mean probing depth. The statistical package used for analysis was SPSS [statistical package for social science]. Median, range were estimated for parameters age, number of teeth probed, oral hygiene index-simplified, plaque index, number of sites that bleed on probing, total probing depth, mean probing depth. Median test was employed to compare the median values for the above parameters. Since the values were not equally distributed, proportion was estimated for the parameter sex. Proportions were estimated from the samples for each group. Proportions were compared by pearson's chi- square test with Yates continuity correction / fisher's exact [2 tailed] appropriately. In the present study, $P < 0.05$ was considered as the level of significance.

RESULTS

Out of 20 gingivitis patients, none had exhibited positive bacteremia before probing, while 3 [15%] had exhibited positive bacteremia after probing. Out of 20 periodontitis patients, none had exhibited positive bacteremia before probing, while 9 [45%] had exhibited positive bacteremia after probing [Table 1]. Microorganisms recovered were Streptococcus Viridans [10 out of 12 patients], staphylococcus aureus [2 out of 12 patients] [Table 2]

In the present study, median values for age [p value < 0.0001], oral hygiene index-simplified [p value 0.0005], plaque index [p value 0.0007], number of sites that bleed on

probing [p value 0.03], total probing depth [p value 0.005], mean probing depth [p value < 0.0005], were significantly higher in periodontitis group than in gingivitis group. Number of teeth probed [p value 0.004] is significantly higher in gingivitis group than in periodontitis group in the present study. Median test was employed to estimate the P value. The proportion of males - 12 out of 20, females - 8 out of 20 in gingivitis group is statistically not significant when compared to males - 14 out of 20, females-6 out of 20 in periodontitis group. Pearson's chi-square test with Yates continuity correction was used to estimate the P value [table 3].

Median values for Age [p value 0.08], number of teeth probed [P value 0.73], oral hygiene index-simplified [P value 0.08], plaque index [P value 0.98] were statistically not significant when compared to bacteremia positive [n = 12] and bacteremia negative [n = 28] patients. The inference is that, these parameters have got no influence on incidence of bacteremia among the above groups in the present study. Median values for number of sites that bleed on probing [P value < 0.0001], total probing depth [P value 0.0004], mean probing depth [P value 0.0001] were significantly higher in bacteremia positive patients when compared to bacteremia negative patients. The inference is that the incidence of bacteremia increases with number of sites that bleed and total probing depth, among the above patients in the present study. Out of 12 [75%], females - 3 out of 12 [25%], in bacteremia positive patients was statistically not significant [P value 0.48], when compared to proportion of males - 17 out of 28 [60.7%], females - 11 out of 28 [39.3%] in bacteremia negative patients. The inference is that the parameter sex has no influence on incidence of bacteremia among the above patients. Fisher's exact test [2 tailed] was employed to determine the P value [table 4].

Figure 1

Table 1: Incidence of bacteremia before and after probing in gingivitis and periodontitis groups

GROUP	BEFORE PROBING	
	POSITIVE BACTEREMIA	NEGATIVE BACTEREMIA
GINGIVITIS [n = 20]	0	20 [100%]
PERIODONTITIS [n = 20]	0	20 [100%]
	AFTER PROBING	
	POSITIVE BACTEREMIA	NEGATIVE BACTEREMIA
GINGIVITIS [n = 20]	3 [15%]	17 [85%]
PERIODONTITIS [n = 20]	9 [45%]	11 [55%]

Figure 2

Table 2: Microorganisms recovered by aerobic culture in positive patients in gingivitis and periodontitis groups.

Type of organism	Gingivitis group	Periodontitis group
Streptococcus Viridans	2	8
Staphylococcus aureus	1	1

Figure 3

Table 3: Median, Range, and Test of Significance of Median Values between Gingivitis and Periodontitis Groups

PARAMETER	GINGIVITIS [n = 20]		PERIODONTITIS [n = 20]		VALUE
	Median	Range	Median	Range	
Age [in years]	24.5	21 – 32	43.5	35 – 70	<0.0001
Number of teeth probed	32	28 – 32	28.5	22 – 32	0.004
Oral hygiene index-simplified	1.63	0.59 – 3.0	2.66	2.0 – 4.0	0.0005
Plaque index	0.67	0.5 – 1.33	1.11	0.87 – 2.20	0.0007
Number of teeth that bleed	89	72 – 116	104.5	70.0 – 178.0	0.03
Total probing depth [in mm]	437.5	361 – 604	574	393 – 714	0.0005
Mean probing depth [in mm]	13.71	12.3 – 18.75	19	17.4 – 24.1	<0.0001
Sex	Males	12 [60%]	14 [70%]		0.74
	Females	8 [40%]	6 [30%]		

Figure 4

Table 4: Median, Range, and Test of Significance of median Values Between Positive and Negative Results

PARAMETER	BACTEREMIA POSITIVE [n =12]		BACTEREMIA NEGATIVE [n =28]		P VALUE
	Median	Range	Median	Range	
Age	40.5	22 – 70	29	21 – 58	0.08 [n sig]
Number Of Teeth probed	30	22 – 32	31.5	22 – 32	0.73 [n sig]
Oral Hygiene Index	2.49	1.6 – 4.0	2.0	0.59-3.60	0.08 [n sig]
Plaque index	1.01	0.66-2.2	0.96	0.50-1.60	0.98 [n sig]
Number of sites that bleed on probing	159	111-178	84	70-104	<0.0001 [sig]
Total probing depth	640	531-714	454	361-590	0.0001[sig]
Mean probing depth	22.4	18-24.1	14.47	12.3-19.9	0.0004 [SIG]
Sex	Male	9 [75%]	17 [60.7%]		0.48 [n sig]
	Female	3 [25%]	11 [39.3%]		

Figure 5

Fig 1: Alpha haemolytic colonies on blood plate agar.

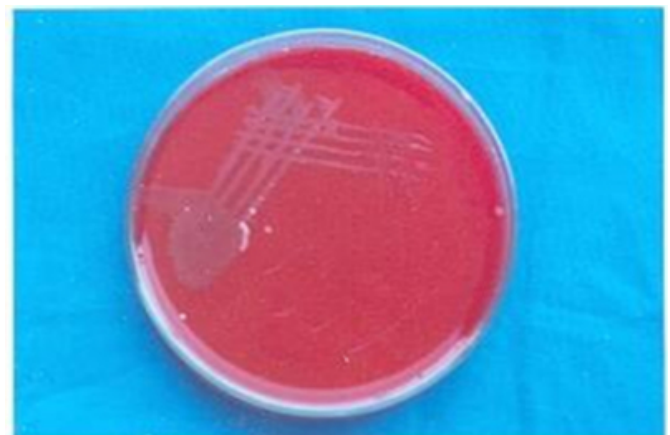


Figure 6

Fig 2: Beta haemolytic colonies on blood plate agar



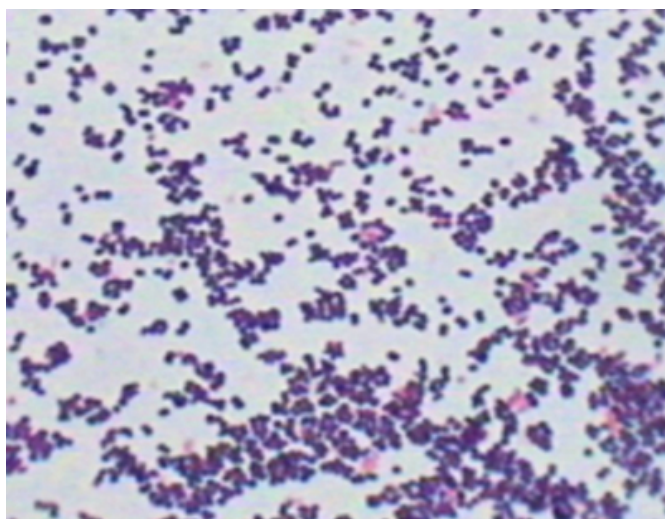
Figure 7

Fig 3: Photomicrograph [100x] showing Gram positive cocci in chains [streptococci]



Figure 8

Fig 4: Photomicrograph [100x] showing Gram positive cocci in clusters [staphylococci]



DISCUSSION

Infective endocarditis is a disease in which microorganisms colonize the damaged endocardium on heart valves. The incidence of infective endocarditis in the developing countries is 1 to 5 new cases per lakh per year¹. Certain dental procedures are capable of producing bacteremia, which can eventually lead to infective endocarditis. The micro flora of the oral cavity can be a potential source of bacteremia after disruption of capillaries in the gingival tissues⁷. This kind of transient bacteremia is well documented following tooth extraction, following treatments

for periodontal diseases such as gingivectomy, supra and sub gingival scaling⁸. Based on available literature about 15 to 40 percent of endocarditis cases could be due to dental treatment⁸. Generally such bacteremia poses little threat to a healthy patient however, patients at a high risk for cardiovascular infection because of various cardiac lesions, such as rheumatic heart disease, congenital defects, valvular disease, or prosthetic implants could be seriously compromised by this kind of bacteremia⁹.

In the present study, the presence of microorganisms following probing was identified by aerobic culture since, aerobic/gram positive bacteria were mostly implicated in the pathogenesis of infective endocarditis.

The incidence of bacteremia in gingivitis group was 15%, this is in accordance with the study done by Max Winslow et al¹⁰ who showed 9.5% of bacteremia in gingivitis group. The incidence of bacteremia in periodontitis group was 45%, this finding was in accordance with findings of Christopher G Daly et al¹¹-43%, Max Winslow et al¹⁰- 42% , John Iofthus¹²- 30%, Aldona et al¹³- 61%, James Felix et al¹⁴- 50%, and Thomas Witzemberger et al¹⁵- 55%. In the present study microorganisms recovered were *Streptococcus Viridans* [10 out of 12 patients], *staphylococcus aureus* [2 out of 12 patients]. These microorganisms were the same types that were the suspicious causative agents as per the American Heart Association standard antibiotic regimens for prophylaxis for sub acute bacterial endocarditis¹⁶. When the parameters were compared between gingivitis and periodontitis groups, age was significantly higher in periodontitis group than in gingivitis group, which was not in accordance with the study done by Christopher G. Daly¹¹. Number of teeth probed in the present study was significantly higher in gingivitis group than in periodontitis group which also differs from the study done by Christopher G. Daly¹¹ which could be due to the difference in age groups of patients involved in both the studies.

Plaque index, number of sites that bleed on probing, total probing depth, mean probing depth per tooth were significantly higher in periodontitis group than in gingivitis group, which were in accordance with the study done by Christopher G. Dal¹¹.

Oral hygiene index-simplified which was not included in any of previous studies, but included in the present study was significantly higher in periodontitis group than in gingivitis group.

In the present study the median of age in bacteremic patients was 40.5 years as against 29 years in non bacteremic patients, which was statistically insignificant. The association of age with bacteremia could not be evaluated with certainty by different authors. The studies conducted by Thomas Witzemberger et al¹⁵, James Felix et al¹⁴, Luther lineberger et al¹⁷, could not find any association between age and bacteremia. However Okabe et al¹⁸ based on his study observed that the incidence of bacteremia tends to increase with age of the patient.

There was no significant difference in the occurrence of bacteremia between males and females in the present study, which is in accordance with the study done by Christopher G. Daly et al¹¹, K.okabe et al¹⁸, Luther lineberger et al¹⁷.

There was no significant difference found for number of teeth probed, plaque index in occurrence of bacteremia which was in accordance with the study done by Christopher G. Daly et al¹¹. Studies done by Thomas Witzemberger et al¹⁵, Victoria Lucas et al¹⁹, coulter W.A et al²⁰ showed that plaque index was not associated with bacteremia which is similar to the present study. Oral hygiene index-simplified was also not statistically significant in occurrence of bacteremia in the present study.

Since the factors age, sex, number of teeth probed, plaque index, and oral hygiene index-simplified showed no significant difference between positive bacteremic patients and negative bacteremic patients, it seems that these factors do not predict which patients will show bacteremia from periodontal probing.

The lack of association between bacterial plaque and presence of bacteremia was most likely due to fact that plaque assessments were made with naked eye. It is nearly impossible for small levels of plaque to be detected in this way. The tooth surface would always be colonized by some plaque which is not visible to the naked eye. Such sub ocular levels of plaque nevertheless are scooped down by the tip of periodontal probe as it is traversing the tooth surface in the region of gingival crevice²¹.

Number of sites that bleed on probing, mean probing depth per tooth, were significantly associated with occurrence of bacteremia in the present study and this is in accordance with the study done by Christopher G. Daly et al¹¹. Total probing depth is also significantly associated with bacteremia which differs from Christopher G. Daly et al¹¹.

The probe tip is known to penetrate through the epithelial lining of the pocket when inflammation is present and bleeding on probing is indicative of an inflammatory lesion in the underlying connective tissue. Bleeding sites, but not non bleeding sites, show histologic ulceration of the pocket epithelium. Thus whenever bleeding on probing occurred, any microorganisms dislodged from the plaque biofilm by the probe tip may have had chance to spread into vascular connective tissue and then into circulation system¹¹. This may explain why the number of sites which bleed on probing was significantly associated with bacteremia.

Since in the present study, the incidence of bacteremia was found to be more in periodontitis group, and the factors which were associated with bacteremia i.e. number of sites that bleed on probing, total probing depth, mean probing depth per tooth were also significantly higher in periodontitis group than in gingivitis group, patients with untreated periodontitis were at greater risk of experiencing bacteremia due to periodontal probing. Since these factors cannot be measured until after probing was performed, they are of no use to the clinician as predictive factors for bacteremia prior to probing, and the only factor which was significantly predictive for the occurrence of bacteremia prior to probing was the presence of interproximal bone loss on dental panoramic or intra oral periapical. radiographs. This method of screening for alveolar bone loss has been shown to correlate well with the presence of periodontitis⁸. In the previous study done by Christopher G. Daly et al¹¹ with untreated periodontitis and who exhibited bone loss on dental panoramic radiographs, experienced 43% of bacteremia following periodontal probing, which coincides well with the present study.

Thus for an individual adult with periodontitis who has not received any periodontal treatment within the previous year, may have at least 45% chance of bacteremia occurring following periodontal probing.

CONCLUSION

Based on the findings of this study, it can be concluded that patients with untreated periodontitis were at a greater risk of experiencing bacteremia due to periodontal probing. This study also indicates the importance of history taking to determine the presence of or past history of rheumatic fever, rheumatic heart diseases or any other valvular heart diseases. In individuals evidencing such background, it would be appropriate to advocate prophylaxis even before periodontal probing was done.

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