Original Article

The Effect of Different Mouthwashes on Bacteremia after Debonding

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INTRODUCTION

Bacteremia is defined as the entrance of bacteria into the systemic bloodstream.^[1,2] Transient bacteremia, which may occur after bleeding or minor trauma due to procedures performed on the mucous membranes with a high bacterial density such as the oral cavity, is eliminated by the reticuloendothelial system in healthy individuals while it possesses the risk of developing bacterial endocarditis in patients with heart disease.^[3] The American Heart Association (AHA) published a joint declaration after establishing an interdisciplinary working group consisting of various councils to prevent infective endocarditis and suggested antibiotic prophylaxis to

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Objectives: This study aims to investigate the effects of various mouthwashes on bacteremia development following a debonding process, which is performed after orthodontic treatment. Subjects and Methods: The study included patients who received fixed orthodontic treatment and were indicated for debonding. A total of 40 patients in four groups were selected for the study; no mouthwash (Group 1), mouthwash containing 0.12% chlorhexidine-gluconate (Group 2), mouthwash containing essential-oils (Group 3), and mouthwash containing 7.5% povidone-iodine (Group 4). Before (T_0) and following (T_1) the debonding procedure, blood samples were obtained from the patients. Then, the blood samples were placed in blood culture bottles to investigate bacterial growth. Results: Based on the results of the study, it was determined that the blood samples obtained at T₀ did not indicate any bacterial growth. Furthermore, it was observed that the blood samples obtained at T, included Streptococcus viridans, Streptococcus oralis, Streptococcus mutans, and Staphylococcus aereus growth, respectively, in 4 patients from Group 1 while Streptococcus salivarius growth was observed in 1 patient from Group 3 in addition to *Streptococcus mitis* growth in 1 patient from Group 4. No bacterial growth was observed in Group 2. While the results obtained between Group 1 and Group 2 were statistically significant, no statistically significant difference was observed between other groups. Conclusions: Finally, it was determined that the mouthwash 0.12% chlorhexidine-gluconate was statistically significant in comparison to the control group. It can be concluded that this mouthwash can be used to decrease bacterial density in oral flora before debonding procedures.

KEYWORDS: Bacteremia, bacterial endocarditis, chlorhexidine gluconate, debonding, mouthwashes

inhibit bacterial endocarditis in high- and medium-risk patients.^[4]

One of the main causes of bacterial endocarditis is poor oral hygiene.^[5] Studies have reported that bacterial endocarditis often originates from dental procedures.^[6,7] In one study, after procedures such as tooth extraction, root canal treatment, dental scaling, and root planing as well as chewing and teeth brushing, bacteremia had been detected.^[8] However, even after dental procedures

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with no bleeding, bacteremia was detected^[9-11] and it was argued that bleeding was not necessary for the development of bacteremia.^[12] From various studies in the orthodontic literature, it has been shown that bacteremia may occur in procedures such as debonding, stripping, mini screw, and tape applications.^[11,13-21]

Our study aimed to investigate the effects of various mouthwashes on bacteremia development following a debonding process which was performed after orthodontic treatment.

SUBJECTS AND METHODS

Ethics statement

During this study, the ethical principles of medical research on human volunteers specified in the World Medical Association (WMA) Declaration of Helsinki were adhered to. Ethics Committee approval dated 21/02/2019 with meeting no. 04 and decree no. 02 was obtained from Firat University Ethics Committee for the research. Furthermore, all the participants stated their agreement to participate in the study by written consent forms.

Inclusion criteria for patients

The inclusion criteria for the patients firstly required the patients to be systemically healthy. The patients were required to refrain from using medication for a month before debonding. Almost 2 h before the schooled debonding appointment and the appointment time, the patients were required to stop eating and brushing their teeth. In addition to these criteria, the patients who agreed to follow the instructions of the study and received orthodontic treatment only with brackets and molar tubes were included in the study.

Exclusion criteria for patients

Patients with any systemic disease, especially cardiac disease, patients who have a disease that may cause immunosuppression, patients with type 1 diabetes, patients who have undergone any cardiac operation or coronary stenting, have a joint prosthesis, use any medication, receive chemotherapy or radiotherapy, dialysis patients with permanent vascular catheters, patients with gingival bleeding during debonding, hemophilia patients, and whose orthodontic treatment was conducted with molar band were excluded from the research.

Study volunteers were selected from patients whose fixed orthodontic treatments were completed at the Department of Orthodontics in the Faculty of Dentistry at Firat University and who underwent debonding. Around 40 patients; 21 women and 19 men, who met these criteria were randomly selected for the study. All patients were scheduled for an appointment at 9:00 am for debonding. The patients were instructed to stop eating and to refrain from brushing their teeth 2 h before the appointment time and the debonding procedure. The patients were also instructed not to undergo any procedures, including examinations in other departments before the debonding appointment. At first 10 mL of blood samples (T_o) were obtained from the patients via the antecubital veins and this procedure was conducted under aseptic conditions before any procedures. Later, the patients were asked to gargle for 30 s immediately in Group 1, and in Group 2 with Kloroben (Drogsan Pharmaceuticals, Ankara, Turkey) mouthwash containing 0.12% chlorhexidine gluconate, in Group 3 with Listerine (Johnson and Johnson Healthcare Products, New Jersey, USA) mouthwash containing essential-oils (E.O.)., and in Group 4, with Biokadin (Kansuk Laboratory, Istanbul, Turkey) mouthwash containing 7.5% povidone-iodine, and then the debonding process was started. Immediately after the debonding procedure, a second 10 mL blood sample (T₁) was obtained from the patients via antecubital veins, which was also conducted under aseptic conditions.

In this study, BD BACTEC[™] FX (Becton Dickinson, Diagnostic Instrument Systems, Sparks, MD, USA) Blood Culture System and BD BACTECTM Plus (Becton Dickinson, Diagnostic Instrument Systems, Sparks, MD, USA) blood culture bottles were used [Figure 1]. All of the blood samples in the study were obtained by asepsis and antisepsis rules. Blood samples were taken and left to incubate in the microbiology laboratory after labeling them with the related data (patient, time of the study and code of the group). For the laboratory analysis, the blood culture bottles were stored on the BD BACTECTM FX device following the registration. The incubation time for the device was set as 7 days. The blood cultures were stored at 37 °C for the entirety of the incubation period and then monitored. After the reproduction signal was heard, the incubation bottles were taken out of the device and alcohol was used to wipe their plastic caps. Then, by using a sterile syringe, 2 to 3 mL of blood was obtained from the vial. The blood sample was subcultured into eosin methylene blue (EMB) agar, chocolate agar, and sheep blood agar in a biosafety cabinet and next to the burner flame and left in the incubator. Following the determination that the bacterial growth was successful, the bacterial identification process was conducted with BD PHOENIX[™] 100 (Becton Dickinson, Diagnostic Instrument Systems, Sparks, MD, USA).

Statistical analysis

In the statistical analysis of the resulting data, IBM SPSS package software (Statistical Package for Social

Sciences, version: 24.0, Chicago, IL) was used to conduct the relevant analyses. The normality of distribution of continuous variables was tested by the Shapiro Wilk test. The one-way ANOVA test (for normal data) was used for comparison of the independent groups and the Chi-square test was conducted to investigate the relationship between categorical variables. The results were interpreted at a 95% confidence interval while the *P* value <0.05 was accepted as the statistical significance level.

RESULTS

The average age of the 40 patients included in the study was 17.28 ± 1.74 years. The minimum and maximum values for the duration of treatment were 1.42 and 2.83 years, respectively and the average duration of treatment was 2.01 ± 0.36 years. In terms of age and duration of treatment, it was observed that there was no statistically significant difference between the groups (P = 0.875 and P = 0.123, respectively) [Table 1].

Of the 40 patients investigated in the study, 21 (52.5%) were females and 19 (47.5%) were males. In terms of gender, it

Table	Table 1: Distribution of age and treatment duration by								
groups									
Groups	n		Mean±Sd	Р		Mean±Sd	Р		
Group 1	10	Age	16.98 ± 2.14	0.875	Duration of	$1.94{\pm}0.30$	0.123		
Group 2	10		17.26 ± 2.04		treatment	1.95 ± 0.32			
Group 3	10		17.25 ± 1.20		(year)	$1.90{\pm}0.44$			
Group 4	10		$17.64{\pm}1.63$			2.24 ± 0.29			
Total	40		17.28 ± 1.74			2.01±0.36			

was determined that there was no statistically significant difference between the groups (P = 0.592) [Table 2].

According to the blood culture results of the study, 40 patients had no bacterial growth in T_0 blood samples while among the collected T_1 blood samples, bacteremia was detected in 6 patients [Table 3]. *Streptococcus viridans, Streptococcus oralis, Streptococcus mutans,* and *Staphylococcus aereus* were detected in four different patients in Group 1, *Streptococcus salivarius* was detected in 1 patient in Group 3 and *Streptococcus mitis* was detected in 1 patient in Group 4, respectively. In Group 2, no bacteria were detected [Table 4]. Although the results between Group 1 and Group 2 were found to be statistically significant (P = 0.010),



Figure 1: Blood culture bottle used in the study

Table 2: Distribution of gender by groups											
Gender		Group 1	Group 2		Group 3			Group 4	Total		P
	п	Percentage	n	Percentage	n	Percentage	n	Percentage	n	Percentage	
Male	3	30.0	6	60.0	5	50.0	5	50.0	19	47.5	0.592
Female	7	70.0	4	40.0	5	50.0	5	50.0	21	52.5	

			Table	3: Ba	cterial grow	th b	y time and n	ulti	ple comparis	sons	of groups			
			Group 1		Group 2		Group 3		Group 4		Total		Р	
		n	Percentage	n	Percentage	n	Percentage	n	Percentage	n	Percentage	2 vs. 1	3 vs. 1	4 vs. 1
T ₀	Positive	0	0	0	0	0	0	0	0	0	0	1.000	1.000	1.000
	Negative	10	100	10	100	10	100	10	100	40	100			
Τ,	Positive	4	40.0	0	0	1	10.0	1	10.0	6	15	0.010	0.112	0.112
1	Negative	6	60.0	10	100	9	90.0	9	90.0	34	85			

	Table 4: Bacteria detected in groups by time							
	Group 1	Group 2	Group 3	Group 4				
T ₀	-	-	-	-				
	Streptococcus viridans (1)							
T ₁	Streptococcus oralis (1)	-	Streptococcus salivarius (1)	Streptococcus mitis (1)				
	Streptococcus mutans (1)							
	Staphylococcus aereus (1)							

there was no statistically significant difference between the other groups. Detection of these bacteria in the blood, which should normally be sterile, suggested the possibility of causing bacterial endocarditis.

DISCUSSION

Many researchers have wondered whether orthodontic applications cause bacteremia and many studies have been conducted in this field.^[11,13-21] However, no study has been conducted on how different antiseptic mouthwash affects bacteremia development after debonding until now. Thus, this study aimed to investigate the effects of various mouthwashes on bacteremia development following a debonding process, which is performed after orthodontic treatment.

Blood culture systems are the gold standard in the diagnosis of bacteremia while they are the most commonly used methods in microbiology laboratories.^[22] Despite current advances in diagnostic methods, blood cultures are still the most reliable way to diagnose fungemia and bacteremia.^[23] For these reasons and based on the literature, blood culture systems were adopted in the current study.

Due to the damage of the vacuum effect resulting from the negative pressure in blood culture bottles, direct blood collection is not recommended in the performance of these studies. The recommended method is to take the necessary blood sample from the patient first and then place it in the blood culture bottle after changing the needle tip.^[24] Therefore, an indirect blood collection procedure was preferred in this study.

In this study, the status of bacteremia development was investigated by taking blood samples at various times from volunteers, who were completely healthy and did not have any systemic disease. The patients were required to be free of any recent systemic disease, not to use any medication, especially antibiotics, and not to undergo even the smallest oral procedures that could result in bacteremia. They were instructed to refrain from brushing their teeth or eating anything for 2 h before the appointment time. The purpose of these instructions was to exclude the influences of other elements on the bacteremia investigated in the study. The initial blood samples (T_0) were obtained to determine if there was any non-study-based bacteremia before commencing the procedure. Then, the patients were asked to gargle for 30 s immediately in Group 1, and in Group 2 with a mouthwash containing 0.12% chlorhexidine gluconate, in Group 3 with a mouthwash containing essential oils, and in Group 4 with a mouthwash containing 7.5% povidone-iodine, and the debonding process was then started. The second

blood samples (T_1) were obtained immediately after the debonding procedures. The reason why blood was taken immediately is that the reticuloendothelial system cells of a healthy person destroy the bacteria within 20 min.^[25]

According to the results, none of the blood samples obtained at T₀ showed bacterial growth while among the blood samples taken at T₁ S. viridans, S. oralis, S. mutans, and S. aereus were detected in four different patients in Group 1, respectively, S. salivarius was detected in 1 patient in Group 3, and S. mitis was detected in 1 patient in Group 4. In Group 2, no growth was detected. The results between Group 1 and Group 2 produced statistically significant differences (P = 0.010) while there was no statistically significant difference between the other groups. Most of the detected bacteria (83.33%) was streptococcus. Particularly, the detection of S. viridans is a significant subject that requires attention. In many studies, S. viridans was the most common agent of bacterial endocarditis and the primary cause of bacterial endocarditis.[7,26-28] Other studies on bacteremia have identified similar bacteria. Akbulut et al.[11] stated that they had determined S. viridans, S. mitis, S. salivarius, S. oralis, and S. aereus; McLaughlin et al.[13] stated that they had determined S. mitis; Erverdi et al.[14] stated that they had determined S. salivarius and S. mitis; Erverdi et al.^[15] stated again that they had determined S. aereus in their another study; Burden et al.[16] stated that they had determined S. mitis; Uysal et al.[17] stated that they had determined S. sanguis; Yagci et al.[18] stated that they had determined S. sanguis; Lucas et al.[20] stated that they had determined S. viridans; and Gürel et al.[29] stated that they had determined S. aereus. According to these results, the bacteria causing bacteremia after band application, band removal, rapid maxillary expansion device removal, orthodontic mini screw placement, and stripping procedures were found to be similar to the bacteria causing bacteremia in this study.

McLaughlin *et al.*^[13] researched bacteremia in 30 patients before and after band application in their study in 1996. Blood samples were taken from the patients twice, which were conducted before the band application and 60 s and immediately after the band placement. Bacteria were found in one of the samples that were taken before the band was applied and in three of the samples that were taken after the band was applied. In both samples, *S. sanguis* and *S. mitis* bacteria were found. The bacteria identified in this study and the bacteria in the current study are similar in terms of characterization. Besides, these bacteria are important because they are the most common agents of bacterial endocarditis. Lucas et al. could not detect a significant relationship between the bacteremia and the procedures performed in the bacteremia studies they had carried out in 2002^[19] before band application and in 2007^[20] after band removal. Erverdi et al.[14] studied bacteremia in a sample consisting of 30 patients before and after debonding and detected bacteremia both before and after the procedure in their study in 2000. S. salivarius and S. sanguis were observed in the samples taken before the procedure and S. sanguis and S. mitis were observed in the samples taken after the procedure. Further, Burden et al.[16] studied bacteremia in 30 patients after debonding and observed bacteremia in 1 patient before the procedure and 4 patients after the procedure in their study in 2004. S. viridans, S. mitis, S. sanguis, and S. mutans were observed in these patients. Although the bacteria observed in these studies were similar to the bacteria observed in the current study, the presence of bacteremia before the debonding process in these studies suggests that other factors as the cause of bacteremia could not be eliminated. Furthermore, it is thought that the absence of bacteremia before debonding in the current study makes this study particularly valuable.

CONCLUSION

To conclude, in the present study, the effects of different mouthwashes on the development of bacteremia after debonding, which was conducted after orthodontic treatment, were investigated. The results obtained in the study were as follows;

- 1. Debonding can result in bacteremia.
- 2. No significant relationship was found between bacteremia and age, sex, and duration of treatment in our study.
- 3. In patients who require prophylaxis, prophylaxis may be required before debonding.
- Mouthwash containing 0.12% chlorhexidine gluconate can be used to decrease the bacterial density in the oral flora before debonding procedures.

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Conflicts of interest

There are no conflicts of interest.

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