Original Article

Changes in Periodontal and Microbial Parameters after the Space Maintainers Application

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ABSTRACT

Aim: This study aims to evaluate the clinical and microbiological changes accompanying the inflammatory process of periodontal tissues during treatment with space maintainers (SMs). **Materials and Methods:** The children were separated into fixed (Group 1, n = 20) and removable (Group 2, n = 20) appliance groups. A full periodontal examination, including probing pocket depth (PPD), bleeding on probing (BOP), and plaque index (PI), was performed. Anaerobic microorganisms in the crevicular fluid were detected with the culture method. Clinical and microbial evaluations were performed before (T0) applications. as well as at three (T1), and 9 months intervals (T2) after the application of the fixed or removable appliances. **Results:** The PI, PPD, and BOP scores at the testing sites of both groups increased significantly from before treatment (T0) to the 9 months' time frame (T2) ($P < 0.05$), The presence of anaerobic bacteria in the subgingival dental plaque increased from T0 (n = 13, 65%) to T1 (n = 16, 80%) in the fixed SM group, but not statistically significant. The same values were obtained in T1 and T2 (n = 16, 80%). **Conclusion:** Although, the results of this study demonstrate that the application of fixed or removable SM appliances in children induced an increase of clinical periodontal parameters, anaerobic microbiota consisting of *Aggregatibacter actinomycetemcomitans*, *Porphyromonas gingivalis*, and *Tannerella forshia* were not observed in any of the samples in short-term. Further long-term and comprehensive investigations are necessary.

Keywords: *Anaerobic microorganism, periodontology, space maintainer*

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Introduction

Space management is an important responsibility of clinicians who are involved in monitoring developing dentition, as the loss of arch length may lead to problems such as crowding, ectopic eruption, dental impaction, crossbite formation, and dental centerline discrepancies. The use of space maintainers (SMs) might potentially obviate the need for later extractions and/or complex orthodontic treatment.[1]

SMs are fixed, or removable appliances used to preserve arch length following the premature loss or elective extraction of teeth. Retained primary teeth can also act as SMs. SM appliances are most commonly used to maintain the space created by the early loss of a first or second primary molar while awaiting the eruption of its successor.[2]

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It has been reported that local factors such as SMs or orthodontic appliances, brackets, bands, and crochets (e.g., Adams) frequently cause a bacterial retention which can lead to an inflammatory response and resultant gingival hypertrophy and possible hyperplasia.[3-5] However, no information is available on the microbiological changes that these periodontal tissues experience during treatment with fixed or removable SMs.

With the limited information available in the literature, the aims of this study were to evaluate the clinical and microbiological changes accompanying the inflammatory

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response of periodontal tissues during treatment with fixed and removal SMs.

Materials and Methods

This study included forty 6- to 9-year-old (mean age = 7.4 ± 2.7 years) referred to the Department of Pediatric Dentistry, Faculty of Dentistry, Erciyes University, Turkey, for the premature extraction of a primary maxillary or mandibular first or second molar due to caries and/or failed pulp therapy. The research protocol was approved by the Ethics Committee in Clinical Research of the Medical Faculty of Erciyes University, and informed consent was obtained from the parents of all study participants.

The following exclusionary criteria were used

Systemic illness (epilepsy, hemophilia, etc.) periodontal disease (aggressive periodontitis, etc.) bruxism, mental handicaps, abnormal breathing or oral habits, and pharmacological treatment (anticonvulsant, etc.) or antibiotic therapy during or up to 4 weeks before the study.

Before placing the appliances, all of the patients received dental hygiene instructions (bass technique). The hygiene protocol was explained using a model, after which the subjects' brushing techniques were analyzed and improved by a clinician (GH) to achieve good comprehension.[6] Oral hygiene instructions were repeated during at the three (T1) and 9 months intervals (T2) after the application of the fixed or removable appliances.

Clinical inclusion criteria

Premature loss of more than one primary molars; presence of indication for least two band or crochet application; presence of teeth on the mesial and distal sides of the extraction space; and angles Class I occlusion and normal primary molar relation. The radiographic inclusion characteristics were: No root resorption of abutment teeth; presence of a succedaneous tooth bud; presence of the bone crypt over the succedaneous tooth bud; succedaneous tooth root development; and absence of pathology on the eruption track of the succedaneous tooth. Removable SM and fixed appliances could be applied on patients in the study group. Thus, children were randomly divided into two groups, according to the type of SM used for treatment, as follows: Group 1 (n = 20) – fixed SM; and Group 2 (n = 20) – removable SM. The fixed appliances were made up of bands and loops, whereas the removable appliances were constructed with Adams crochets. Fixed SMs were cemented with a glass ionomer cement releasing fluoride (Rely X; 3M ESPE, St. Paul, MN, USA). All band selection, cementation, and examinations were performed by the same clinician (GH). Although examinations were carried out by the same clinician, data were recorded on the form by another investigator and thereby the examiner was blinded to previous scores.

A full periodontal examination, including probing pocket depth (PPD), bleeding on probing (BOP), and plaque index (PI), and microbial assessments were performed before (T0) and at three (T1) and 9 months (T2) after the application of the fixed or removable appliances.

Clinical and microbial evaluation

PPD and BOP were obtained at four sites per tooth, and PI was determined for the labial and lingual sites separately. The PI and BOP were measured using the Löe[7] index. The amount of plaque was scored according to the following parameters: Score 0: No plaque on the tooth; Score 1: Plaque covering up to one-third of the surface; Score 2: Plaque covering more than one-third but less than two-thirds of the tooth surface and Score 3: Plaque covering more than two-thirds of the tooth. The BOP was scored according to the following measures: Score 0: No bleeding on blunt probing; Score 1: Bleeding on blunt probing up to 30 s later; Score 2: Immediate bleeding on blunt probing; Score 3: Spontaneous bleeding. The periodontal evaluations were carried out in all patients by the same clinician with a marked periodontal probe (WHO-DMS probe; Deppeler, Rolle, Switzerland). The examiner was trained to apply the correct index used during the investigation. Calibration and reliability assessments were performed in a group of five children examined twice on two successive days. By comparison of the results of the examinations, the degree of agreement between examiners was achieved. The clinical parameters recorded included BOP and PI as measured by a blunt periodontal probe (WHO-DMS probe; Deppeler, Rolle, Switzerland). To record the PPD; however, a millimeter probe (HU-friendly Pc puns, Chicago, IL, USA) was inserted in the gingival sulcus. PPD was measured to the nearest 0.5 mm on the scale. In the fixed and removable appliance groups, the indices were recorded for the teeth on which the bands and crochets were to be applied.

Anaerobic microorganisms in the crevicular fluid were detected with the culture method. After isolating the teeth from saliva with cotton rolls and gently drying them to prevent contamination, a supragingival plaque was carefully removed using sterile curettes, without traumatizing the gingiva, at the labial and lingual sites of the another tooth on which the bands and crochets. This procedure was carried out before and at 3 and

9 months intervals after the band and loop or removable Adams crochets SMs were applied.

Statistical analyses

Power analyses were calculated for sample size determination using nQuery Advisor 5.0 (Statistical Solutions, Saugus, MA, USA). All documentation and evaluation of data were processed using the Statistical Package for Social Science Statistical software version 16 (SPSS Inc., Chicago, IL, USA). The Kolmogorov–Smirnov test was used to determine the normality of the distribution of the data. Clinical parameters were evaluated according to repeated times variance analyses and two-way variance analyses. Microbial parameters were evaluated according to the Chi-square and Fisher's exact tests.

Results

Clinical parameters

Plaque index

The PI scores at the testing sites of both groups increased significantly from before treatment (T0) to 3 months later (T1) ($P < 0.05$), whereas no significant changes in PI scores were recorded in either group between T1 and 9 months after treatment (T2) ($P > 0.05$) [Table 1]. No significant differences in PI scores were found between the fixed and removable groups in the same time assessments.

Probing pocket depth

There were significant increases in PPD scores between T0 and T1 in both the fixed and removable groups ($P < 0.05$)

Table 1: Periodontal parameters at before and 3 and 9 months after application of space maintainers

Parameters	Fixed SM group				Removable SM group			
	T0	T1	T2	*P*	T0	T1	T2	*P*
PI	0.8±0.6[a]	1.2±1.1[b]	1.1±0.9[b]	0.03	0.6±0.5[a]	1.1±06[b]	1.3±0.8[b]	<0.01
PPD	2.6±0.9[a]	3.6±1.3[b]	3.0±1.0[c]	0.01	2.8±0.8[a]	3.8±1.3[b]	3.3±1.1[c]	0.01
BOP	0.3±0.2[a]	0.8±0.6[b]	1.2±09[c]	<0.01	0.5±0.3[a]	1.0±0.7[b]	0.9±0.8[b]	0.03

Means followed by distinct letters are statistically different. SM=Space maintainer; PI=Plaque index; PPD=Probing pocket depth (mm); BOP=Bleeding on probing (%); T0=Baseline; T1=3 months after; T2=9 months after

Table 2: Prevalence of the anaerobic pathogenesis, before and 3 and 9 months after fixed space maintainer application

		Fixed SM group		
Patient	Age	T0	T1	T2
1	7	-	-	-
2	8	*P. oralis*	*P. oralis*	*P. oralis*
3	7	*P. melaninogenica*	*P. melaninogenica*	*P. melaninogenica*
4	6	-	-	-
5	7	*P. oralis, Clostridium bifermentans*	*P. oralis*	*P. oralis, C. bifermentans*
6	6	-	*P. oralis*	*P. buccae*
7	7	*P. oralis*	*P. oralis*	*Prevotella* spp., *Fusobacterium* spp., *Peptostreptococcus* spp.
8	8	*P. oralis*	*P. oralis, A. odontoluticum*	*P. oralis, A. odontoluticum*
9	6	*P. denticola*	*P. denticola*	*P. denticola*
10	5	-	*P. oralis*	*Prevotella* spp.
11	6	*P. oralis, P. intermedia*	*C. tyrobutyricum*	*Prevotella* spp.
12	7	*P. oralis*	*P. oralis, Clostridium* spp.	*P. oralis, Clostridium* spp.
13	4	*P. oralis*	*P. oralis, F. nucleatum, Clostridium* spp.	*Prevotella* spp., *Peptostreptococcus* spp.
14	9	-	*P. oralis*	*Prevotella* spp., *A. meyeri*
15	5	-	-	-
16	9	*P. oralis*	*P. oralis*	*P. oralis*
17	8	*P. intermedia*	*P. intermedia*	*P. intermedia*
18	4	*P. oralis, P. Denticola*	*Prevotella* spp., *Clostridium* spp.	*Prevotella* spp., *Clostridium* spp.
19	4	*P. oralis, P. denticola*	*Prevotella* spp., *Clostridium* spp.	*Prevotella* spp., *Clostridium* spp.
20	7	-	-	-

P. melaninogenica=Prevotella melaninogenica; *P. oralis=Prevotella oralis*; *P. buccae=Prevotella buccae*; *A. meyeri=Actinomyces meyeri*; *A. odontoluticum=Actinomyces odontoluticum*; *C. bifermentans=Clostridium bifermentans*; *F. nucleatum=Fusobacterium nucleatum*; T0=Baseline; T1=3 months after; T2=9 months after; SM=Space maintainer; *P. denticola=Prevotella denticola*; *P. intermedia=Prevotella intermedia*

Table 3: Prevalence of the anaerobic pathogenesis, before and 3 and 9 months after removable space maintainer application

Removable SM group				
Patient	**Age**	**Baseline**	**T1**	**T2**
1	7	*P. denticola*	*Clostridium* spp.	*Clostridium* spp.
2	8	*P. oralis, C. bifermentan*	*P. oralis*	*P. oralis, C. bifermentans*
3	8	*P. denticola, P. oralis, P. intermedia*	-	*P. oralis*
4	6	*P. oralis*	*P. oralis*	*P. oralis*
5	9	-	*Actinomyces* spp.	*Actinomyces* spp.
6	8	-	*Actinomyces* spp.	*Actinomyces* spp.
7	7	-	-	-
8	7	*P. oralis*	*P. oralis*	*P. oralis*
9	9	*P. oralis*	*P. buccae*	*P. oralis*
10	8	*P. oralis*	*P. buccae*	*P. oralis*
11	7	*P. oralis*	*C. tyrobutyricum*	*C. tyrobutyricum, F. nucleatum*
12	8	*P. oralis*	*P. oralis*	*P. oralis*
13	10	*P. oralis, P. intermedia*	-	-
14	7	*P. oralis*	*P. oralis, Bacteroides* spp., *Clostridium* spp.	*P. oralis*
15	6	*P. oralis*	*P. oralis, Bacteroides* spp., *Clostridium* spp.	*P. oralis*
16	7	-	-	-
17	7	*P. oralis*	*P. oralis*	*P. oralis*
18	5	*P. oralis*	*P. oralis, Bacteroides* spp., *Clostridium* spp.	*P. oralis*
19	6	*P. intermedia*	*P. intermedia*	*P. intermedia*
20	7	-	-	*P. oralis*

P. oralis=Prevotella oralis; *P. buccae=Prevotella buccae*; *C. bifermentans=Clostridium bifermentans*; *F. nucleatum=Fusobacterium nucleatum*; T0=Baseline; T1=3 months after; T2=9 months after; SM=Space maintainer; *P. denticola=Prevotella denticola*; *P. intermedia=Prevotella intermedia*

[Table 1]. Between T1 and T2, the PPD scores decreased significantly ($P < 0.05$), but the T2 scores remained significantly higher than the T0 scores ($P < 0.05$). No significant differences in PPD scores were found between the fixed and removable groups ($P > 0.05$).

Bleeding on probing

BOP at the testing sites increased significantly between T0 and T1 in both the fixed and removable groups ($P < 0.05$) [Table 1]. In the fixed group, there were a significantly higher number of tested sites with BOP at T2 than at T1. In the removable group, however, the difference in BOP scores between T1 and T2 was not significant ($P > 0.05$). Significant differences in BOP scores between the fixed and removable groups were found at the T2 assessment ($P < 0.05$).

Microbiology

The anaerobic microorganisms detected in the analyses of the subgingival microbiota are presented in Tables 2 and 3. The most frequently isolated bacterial species were *Prevotella* phylotypes, especially *Prevotella oralis*.

The presence of anaerobic bacteria in the subgingival dental plaque increased from T0 (n = 13, 65%) to T1 (n = 16, 80%) in the fixed SM group, but not significantly ($P > 0.05$), and the same values were obtained in T1 and T2 (n = 16, 80%). In the removable group, however, the presence of anaerobic bacteria in the subgingival dental plaque was detected at the same rate (n = 15, 75%) in T0 and T1, and it increased at T2 (n = 17, 85%), but not significantly ($P > 0.05$). The most important bacteria that cause periodontal tissue loss – *Aggregatibacter actinomycetemcomitans* (Aa), *Porphyromonas gingivalis* (Pg), *Tannerella forshia* and *Prevotella nigrescens* – were not detected in any patients. However, *Prevotella intermedia* and *Fusobacterium nucleatum*, which are other important bacteria in terms of causing periodontal disease, were detected in five children (two in the fixed SM group and three in the removable SM group) and two children (one fixed SM and one removable SM), respectively. Although all of *P. intermedia* were determined before application of the appliances, *F. nucleatum* were only determined after the application.

Discussion

SMs are widely used in pediatric dental practice. Although the change in microbiota, which involved the growth of periodontogenic bacteria, was associated with the gingival inflammation found around the bands of the fixed SMs or the crochets of the removable SMs, microbial change during or after the SM treatment is unclear. This prospective study was carried out because microbial and clinical periodontal data during SM treatment are largely lacking.

The clinical results of this study demonstrated a significant increase in plaque formation after the application of both the fixed and removable appliances. These results are in accordance with another clinical study,[5] which reported that regardless of the quality of plaque control, fixed, and removable SMs can compromise periodontal health by increasing plaque accumulation. Higher amounts of biofilm are associated with an increase in PPD and BOP.[8] In this study, this correlation was found in both the fixed and removable groups, as PBD and BOP scores were significantly higher at T1 and T2 compared with T0.

Although the patients were instructed and motivated by a dental professional before the applications of the appliances, and none showed clinical signs of periodontal problems, *P. intermedia* was found in five patients at T0. The positive findings of *P. intermedia* and other *Prevotella* species at T0 can be explained by the higher prevalence of periodontopathogenic microbiota in young people.[9] On the other hand, some authors have suggested that groups of organisms, including *Fusobacterium* spp, *P. intermedia,* and *P. nigrescens,* are detected early in the disease process, preceding, and coexisting with the later colonizing pathogens, including Aa, *T. forshia*, Pg, and *Treponema denticola*.[10,11] This study shows that none of the samples of subgingival plaque were positive for Aa, Pg, or *T. forshia,* before and after the applications. We mainly detected *Prevotella* species, both before and after the application of the SMs. This study shows that 60% and 75% of the samples before the applications, and 80% and 85% of the samples after the application were positive for *Prevotella* species in the fixed and removable groups, respectively. These data could indicate that SM appliances have a detrimental influence on the microbial population of the surrounding tissues, as *Prevotella* species have been identified as contributing pathogens and are associated with early signs of polymicrobial oral infections, especially periodontal problems.[12]

We were not able to compare the results of the present study with the results of other studies, as there is a lack of documentation regarding the effects of SMs on the microbiota in the gingival tissue of children, and most of the studies were performed on children receiving orthodontic treatment. The increased pathogenicity of the dental plaque and the concomitant periodontal changes during orthodontic treatment have been described by several authors.[13-16] Contrary to our results, these studies reported signs of increased gingival inflammation after the band application used for fixed orthodontic treatment applications. Sallum *et al*.[17] reported on the microbial and periodontal changes such as PI, BOP, and PPD after bracket removal. The samples were taken twice: First during the final phase of orthodontic treatment and second 30 days after bracket removal and professional prophylaxis. Those authors concluded that the periodontal signs of gingival inflammation decreased significantly after bracket removal. This improvement in periodontal health 30 days after bracket removal was accompanied by a reduction of the number of sites positive for Aa and *T. forshia.* Similarly, several studies[9,15,16,18,19] reported the increased inflammation in orthodontic patients was accompanied by an increase in the number of Aa and *Bacteroides forsythus* (formerly name of *T. forshia*) which are known to be associated with some of aggressive forms of periodontitis[20,21] and refractory periodontitis,[22] respectively. The difference in results between our study and previous studies might be explained by the fact that the mean age of the subjects in our study group is lower than those of other studies, which were carried out on orthodontic patients. Although our study population consisted mostly of children, the majority of orthodontics study groups are composed of adolescents and young adults.

Although oral hygiene education was given before the application of the SMs, the present study showed that the plaque control was considered insufficient and that there were putative periodontal pathogens in the patients undergoing SM treatment. In accordance with our results, Unkel *et al*.[23] reported that the tooth brushing technique of children under the age of ten is not effective, due to their inefficiency in manipulation and lack of motivation. On the other hand, the World Health Organization has reported that training children in the 7–9 age group about oral hygiene methods is more important and effective for preventive practices than training the other age groups.[24]

Based on the methodology and follow-up period, the limitations of this study were microbial evaluation based on detection of microorganism-prevalence of microorganism not considered, and it was designed as short-term with 9-month follow-up.

Conclusion

The findings of the present study emphasize the importance of developing new oral hygiene education

and plaque control programs during SM treatment. The long-term effects of SMs on clinical and microbial parameters should be investigated in future studies with larger cohorts.

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Conflicts of interest

There are no conflicts of interest.

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