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Changes in Oral Microbial Population through Dental Scaling and Dental Polishing

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Objective: This study is intended to investigate the effectiveness of scaling and dental polishing and their correlation to oral microorganisms by examining the change in oral microbial population after scaling and dental polishing through clinical tests and real-time polymerase chain reaction.

Methods: This study examined 117 male and female adults of 20 years or older who visited the dental clinics in Daejeon, Korea. It used the real-time PCR to analyze the change of the oral microbial population before and after scaling and dental polishing.

Results: Before scaling, the prevalence rate of *Fusobacterium nucleatum* was 100.0%, meaning that it was prevalent in all examined subjects, *Parvimonas micra* 85.5%, *Prevotella intermedia* 76.1%, and *Tannerella forsythia* 72.6% while the prevalence rate of *P. micra* was 75.2% and *P. intermedia* 72.6% after scaling, and *P. micra* 45.3% and *P. intermedia* 66.7% after dental polishing. After scaling, all 11 oral bacterial pathogens including *Aggregatibacter actinomycetemcomitans*, *Porphyromonas gingivalis*, *T. forsythia*, *Treponema denticola*, *P. intermedia*, *F. nucleatum*, *P. micra*, *Campylobacter rectus*, *Eubacterium nodatum*, *Prevotella nigrescens*, and *Eikenella corrodens* showed a considerably reduced population and had statistically significant differences (p<0.05). After dental polishing, 10 oral bacterial pathogens except for *A. actinomycetemcomitans* showed a considerably reduced population and had statistically significant differences (p<0.05).

Conclusion: Considering the results that the considerably reduced prevalence rate of 11 oral pathogenic microbes related to periodontal disease after scaling and dental polishing, we concluded that regular scaling and selective dental polishing are necessary for the periodontal treatment of the adult.

Keywords: dental scaling, dental polishing, periodontal diseases

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Introduction

Periodontal disease is a gingival inflammatory disease that destroys the alveolar bone, surrounding tissues, and deep periodontal tissues. Although it is mostly chronic, and the initial symptoms are not very apparent, it is the main cause of tooth loss if it is neglected or treated promptly.

The most common cause of periodontal disease is a dental plaque and dental calculus. Dental plaque is a three-dimen-

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sional complex bacteria community formed in the teeth and surrounding tissues and attaches to the tooth, and it becomes dental calculus after calcification. Moreover, bacterial toxins generated on the dental plaque can cause gingivitis by chemical stimulation of the gingiva [1].

Proper prevention and treatment at the gingivitis stage, which is the initial periodontal disease, can prevent further advancement. Typical methods include proper brushing and use of oral hygiene products by patients and regular visits to dental clinics for the professional to remove plaque.

Scaling is the removes various deposits attached to the tooth surface. Ultrasonic instruments used in dental clinics transform the electrical energy into vibrational energy by converting high-frequency currents into mechanical vibrations through a transducer to remove dental calculus.

First developed by Zinner in the 1950s, it is currently available in various forms. Ultrasonic and sonic descalers had been used only for removing large deposits due to large size and shape limit of the operating tip. However, small and thin tips with various shapes have been developed since the late 1980s to make them as effective as hand instruments in removing dental calculus and plaque.

Moreover, dental polishing after removing dental calculus can prevent reformation of deposits.

The dental polishing is a process of lubricating the surface of rough teeth to obtain the additional aesthetic benefit. It is selectively after removal of the dental calculus and deposits since it provides a smooth tooth surface that can prevent deposition of new dental plaque [2].

Previous studies that analyzed the bacterial population in the oral cavity of periodontal diseases patients in Korea have reported that Tannerella forsythia and Campylobacter rectus bacteria are relatively high [3].

In the past, traditional methods of bacteria cultivation had the limitation in detecting, transporting, and cultivating oral microbes, but the development of the real-time polymerase chain reaction (PCR) in recent years has allowed the detection and quantitative analysis in relatively simple ways, and thus there are many ongoing studies of the pathogens causing periodontal diseases.

Since the microbial population and species increase would increase as the periodontal disease advances, we intended to compare the change of number and types of 11 oral pathogens before and after the dental scaling and before and after the dental polishing.

Materials and Methods

1. Subjects

We explained the purpose and procedure of this study to 20 years or older patients who visited dental clinics in Daejeon region for scaling between September 2015 and February 2016 and examined 117 (51 males and 66 females) patients who agreed to participate in the study. Of the study subjects, 95% had no systemic disease, and we excluded those with systemic disease if they were taking aspirin drugs and pregnant women.

Methods

This study was approved by the Institutional Review Board (IRB) of College of Natural Science, Dankook University for the purpose of bioethics and safety. The study was conducted on those subjects who agreed to the purpose of this study (IRB approval number: DKU 2015-09-002).

1) Dental cleaning

- (1) The visually apparent supragingival calculus was removed with an ultrasonic scaler (Electro Medical Systems [EMS], Zurich, Switzerland).
- (2) A paste-type dental abrasive agent, which has excellent affinity with teeth and contained fluorine, was sprayed to a rubber cup (Radent prophy paste; Pascal, Bellevue, WA, USA).

The tooth surface was divided into three sections of centrifugal, central, and mesial sections, and the rubber cup was applied in painting method to each section for 2 to 3 seconds [4].

2) Real-time PCR

To quantify the oral microbes, we collected saliva samples of the subjects under the same conditions and requested a periodontal analysis (Cytogen, Seoul, Korea).

The DNA of oral microbes was extracted according to the manufacturer's guide using Exgene Clinic SV mini Kit (Gene-All, Seoul, Korea). For oral microbial DNA extraction, the study subjects gargled for about 20 seconds three times before and after the dental scaling and after the dental polishing, and the saliva was then collected in a plastic container which was kept in a refrigerator.

For the reference curve for quantification in the real-time PCR analysis, genomic DNA was extracted from the microbes in pure culture using the Exgene Clinic SV mini Kit.

The genomic DNA was diluted in the factor of 1:10 from 105 to 100 copies and was used as the template DNA by the real-time PCR for quantification analysis [5].

3. Statistical analysis

The collected data were statistically analyzed using IBM SPSS 23.0 (IBM Co., Armonk, NY, USA) with the statistical significance level of p<0.05.

We compared the change of microbial population before and after the dental scaling to investigate its effectiveness and performed the t-test of the samples.

We also compared the change of microbial population before and after the dental polishing to investigate its effectiveness and performed the t-test of the samples.

Results

 Prevalence rate of oral microbes before and after dental scaling and after dental polishing

Before scaling, the prevalence rate of *Fusobacterium nucleatum* was the highest at 100.0%, followed by *Parvimonas micra* at 85.5%, *Prevotella intermedia* at 76.1%, and *T. forsythia* at 72.6% while the prevalence rate of *P. micra* was 75.2% and *P. intermedia* 72.6% after scaling, and *P. micra* 45.3% and *P. intermedia* 66.7% after dental polishing (Table 1).

Table 1. Prevalence rate of oral microorganism (n = 117)

Bacteria –	Before dental scaling		After dental scaling		After dental polishing	
	Yes	No	Yes	No	Yes	No
Aa	20 (17.1)	97 (82.9)	10 (8.5)	107 (91.5)	8 (6.8)	109 (93.2)
Pg	62 (53.0)	55 (47.0)	50 (42.7)	67 (57.3)	44 (37.6)	73 (62.4)
Tf	85 (72.6)	32 (27.4)	72 (61.5)	45 (38.5)	59 (50.4)	58 (49.6)
Td	75 (64.1)	42 (35.9)	64 (54.7)	53 (45.3)	58 (49.6)	59 (50.4)
Pi	89 (76.1)	28 (23.9)	85 (72.6)	32 (27.4)	78 (66.7)	39 (33.3)
Fn	117 (100.0)	0 (0.0)	117 (100.0)	0 (0.0)	117 (100.0)	0 (0.0)
Pm	100 (85.5)	17 (14.5)	88 (75.2)	29 (24.8)	53 (45.3)	13 (11.1)
Cr	51 (43.6)	66 (56.4)	27 (23.1)	90 (76.9)	14 (12.0)	103 (88.0)
En	63 (53.8)	54 (46.2)	46 (39.3)	71 (60.7)	35 (29.9)	82 (70.1)
Pn	82 (70.1)	35 (29.9)	59 (50.4)	58 (49.6)	33 (28.2)	84 (71.8)
Ec	79 (67.5)	38 (32.5)	45 (38.5)	72 (61.5)	34 (29.1)	83 (70.9)

Values are presented as number (%). Aa: Aggregatibacter actinomycetemcomitans, Pg: Porphyromonas gingivalis, Tf: Tannerella forsythia, Td: Treponema denticola, Pi: Prevotella intermedia, Fn: Fusobacterium nucleatum, Pm: Parvimonas micra, Cr: Campylobacter rectus, En: Eubacterium nodatum, Pn: Prevotella nigrescens, Ec: Eikenella corrodens.

Table 2. Results of t-test of samples for effectiveness of dental scaling and dental polishing (n = 117)

Bacteria	Dental scaling		p-value*	Dental polishing		
	Before dental scaling	After dental scaling	p-value*	Before dental scaling	After dental polishing	p-value*
Aa	1.29 ± 2.87	0.59 ± 1.96	0.001	0.59 ± 1.96	0.46 ± 1.70	0.063
Pg	3.60 ± 3.44	2.76 ± 3.23	0.000	2.76 ± 3.23	2.28 ± 2.98	0.000
Tf	4.61 ± 2.92	3.58 ± 2.91	0.000	3.58 ± 2.91	2.72 ± 2.78	0.000
Td	4.17 ± 3.24	3.34 ± 3.12	0.000	3.34 ± 3.12	2.88 ± 2.98	0.001
Pi	5.33 ± 3.16	4.51 ± 2.93	0.000	4.51 ± 2.93	3.87 ± 2.86	0.000
Fn	6.92 ± 0.99	6.36 ± 1.03	0.000	6.36 ± 1.03	6.06 ± 1.09	0.000
Pm	5.97 ± 2.57	4.82 ± 2.83	0.000	4.82 ± 2.83	3.89 ± 2.91	0.000
Cr	2.98 ± 3.45	1.44 ± 2.66	0.000	1.44 ± 2.66	0.71 ± 1.96	0.000
En	3.89 ± 3.67	2.69 ± 3.39	0.000	2.69 ± 3.39	1.97 ± 3.06	0.000
Pn	4.60 ± 3.08	3.04 ± 3.07	0.000	3.04 ± 3.07	1.57 ± 2.55	0.000
Ec	3.63 ± 2.63	1.88 ± 2.45	0.000	1.88 ± 2.45	1.35 ± 2.19	0.000

Values are presented as mean ± standard deviation. Aa: Aggregatibacter actinomycetemcomitans, Pg: Porphyromonas gingivalis, Tf: Tannerella forsythia, Td: Treponema denticola, Pi: Prevotella intermedia, Fn: Fusobacterium nucleatum, Pm: Parvimonas micra, Cr: Campylobacter rectus, En: Eubacterium nodatum, Pn: Prevotella nigrescens, Ec: Eikenella corrodens. *p-value determined from t-test.

Change of oral microbe population before and after dental scaling and after dental polishing

The analysis results showed that all 11 oral bacterial pathogens including *Aggregatibacter actinomycetemcomitans*, *Porphyromonas gingivalis*, *T. forsythia*, *Treponema denticola*, *P. intermedia*, *F. nucleatum*, *P. micra*, *C. rectus*, *Eubacterium nodatum*, *Prevotella nigrescens*, and *Eikenella corrodens* showed a considerably reduced oral microbe population after the dental scaling and had statistically significant differences (p<0.05).

After dental polishing, 10 oral bacterial pathogens except for *A. actinomycetemcomitans* showed a considerably reduced population and had statistically significant differences (p<0.05) (Table 2).

Discussion

Periodontal disease is usually a chronic inflammatory disease and can easily reoccur since there are no detectable symptoms, unlike acute disease. Therefore, it is important to prevent periodontal disease before the disease can be affected. From 2012 to 2016, using statistics from The National Health Insurance Corporation analyzed the statistics of the use of dental care institutions using the big data between 2012 and 2016 and reported that the periodontal disease as the cause of the visiting the institutions increased by an average of 12% per year [6].

Cooperation and willingness of patient are important for preventing periodontal disease, but patients' cooperation and their willingness are important, but it is necessary to have the help by oral health professionals to continue oral health care.

It has been reported that the change of microbes in oral cavity can be confirmed among the population group with low knowledge of oral hygiene by inducing them to change the behavior with oral hygiene products in addition to tooth brushing [7].

Dental clinics perform removal of dental calculus to prevent dental caries and periodontal disease.

The purpose of this study was to investigate the changes of oral microbes before and after dental scale and after dental polishing of 117 patients who visited dental clinics in Daejeon region for dental scaling and agreed to participate in this study.

It is known that more than 500 types of bacteria are present in the saliva. The analysis results of the change of 11 oral microbes including *A. actinomycetemcomitans*, *P. gingivalis*, *T. forsythia*, *F. nucleatum*, and *E. corrodens*, which were known to be the pathogen bacteria to cause periodontal diseases [8], showed that the population of all microbes decreased, and the

population decrease of statistically significant.

P. intermedia, *F. nucleatum*, and *P. micra* bacteria, which showed high prevalence rate before the removal of the dental calculus, are some of the major pathogens of periodontal disease. They are present in the paradental cyst of adult periodontitis patients and are also associated with acute necrotizing ulcerative gingivitis [9].

The reduction of *T. forsythia*, which has been reported to be found more frequently in active lesions than inactive lesions [10], was also statistically significant.

It has been reported that the ratio of motile bacteria in the paradental cyst decreased after scaling [11], and thus regular dental scaling can alleviate gingivitis and prevent progression to periodontal disease and should be regarded as an important basic preventive procedure.

The prevalence rates of oral microbes before scaling were found to be 100.0% for *F. nucleatum*, 85.5% for *P. micra*, and 76.1% for *P. intermedia*, and the figures were not different from the result of prevalence rates of oral microbes in previous studies, and the number of oral microbes decreased after the dental scaling.

In addition, a study by Suzuki et al. to assess prevalence rates of *T. forsythia* and *F. nucleatum* in paradental cyst reported that both bacteria grew well in the paradental cyst. These studies have shown that the bacteria that cause periodontal diseases can coexist with each other well [12,13].

Although the patients of systemic disease accounted for less than 5% of subjects of this study, it is necessary for the patients to be aware of the importance of oral health management since a previous study has reported that the possibility of detecting *P. intermedia* in the oral cavity of patients of systemic disease was 4 times higher [14].

Dental polishing of teeth roughened after removal of dental calculus can prevent redeposition and improve aesthetics. The use of a hand instrument was necessary in the past due to the lack of diversity of ultrasonic descaler. However, it has been reported that using a hand instrument, a curet, in particular, could leave severe instrument marks depending on the stroke direction [15].

Since a wide range of tip sizes and shapes are available in ultrasonic descalers nowadays, a dental polishing using the abrasive agent after removing the calculus can result in a smooth surface of teeth [16], and this study also found that the reduction of population of 10 types of oral microbes after a dental polishing was statistically significant.

A previous study on changes of microbes in the furcation areas of multi-rooted cadavers and the single-rooted cadavers after a dental calculus removal procedure or root planing procedure also reported the reduction of plaque index and gingival index and the reduction of the ratio of motile bacteria, in particular [17].

Since the study of correlation between modified-patient hygiene performance, gingival index, Pocket depth, papillary marginal attachment index, and oral microbes has been confirmed that *P. gingivalis*, *P. micra*, *C. rectus*, and *E. nodatum* had a high correlation with the oral test indices, controlling bacterial population in the in the mouth is very important for preventing periodontal diseases [18].

In this study, there was no statistically significant decrease in *A. actinomycetemcomitans* after dental polishing, and it was consistent with the previous study of Korean chronic periodontitis patients that reported a low occurrence in the test of seven types of bacteria. It may be due to differences in experimental methods and sampling methods, but it has been reported that there was a possibility of the low bacterial population in Korean periodontal disease patients [19].

The purpose of this study was to investigate the change of prevalence rate of oral microbes before and after dental scaling and after dental polishing and the efficiency of dental scaling and dental polishing among the adults in Daejeon region. However, there is a limitation to generalizing it since the subjects were only the local residents in Daejeon region, and the additional studies of patients of systemic diseases and various oral bacterial analysis. We expect that the results of this study can be a reference to improve awareness for scaling among the general population as well as the periodontal disease patients.

Conclusion

This study was carried out to quantify the prevalence of oral microbes before and after a dental scaling and after a dental polishing of 117 volunteers who visited dental clinics in Daejeon region for scaling. Three tests were performed before and after dental scaling and after dental polishing. The saliva was collected with special gargle solution to examine the change of the microbial population in the oral cavity. The following conclusions were obtained.

- 1. Before scaling, the prevalence rate of *F. nucleatum* was 100.0%, meaning that it was prevalent in all examined subjects, *P. micra* 85.5%, *P. intermedia* 76.1%, and *T. forsythia* 72.6% while the prevalence rate of *P. micra* was 75.2% and *P. intermedia* 72.6% after scaling, and *P. micra* 45.3% and *P. intermedia* 66.7% after dental polishing.
- 2. After scaling, all 11 oral bacterial pathogens including *A. actinomycetemcomitans*, *P. gingivalis*, *T. forsythia*, *T. denticola*, *P. intermedia*, *F. nucleatum*, *P. micra*, *C. rectus*, *E. nodatum*, *P. nigrescens*, and *E. corrodens* showed a considerably reduced population and had statistically significant differ-

ences (p<0.05).

3. After the dental polishing, 10 oral bacterial pathogens including *P. gingivalis*, *T. forsythia*, *T. denticola*, *P. intermedia*, *F. nucleatum*, *P. micra*, *C. rectus*, *E. nodatum*, *P. nigrescens*, *E. corrodens* showed a considerably reduced population and had statistically significant differences (p<0.05).

The results of the testing of 11 pathogen microbes confirmed that the oral microbial population considerably decreased after scaling and dental polishing, and thus we concluded that regular scaling and selective dental polishing are necessary for the periodontal treatment of the adult.

Conflict of Interest

No potential conflict of interest relevant to this article was reported.

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