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Mutans Streptococci and Lactobacillus as Risk Factors for Dental Caries in 12-Year-Old Children

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Streptococcus mutans (*S. mutans*) and Lactobacillus are the pathogenic bacteria in the oral cavity causing dental caries, the mechanism of which has been demonstrated in many in vitro studies (1,2). Furthermore, some cohort studies have clinically shown an association between the incidence of dental caries and levels of salivary *S. mutans* and Lactobacillus (3,4). However, as these studies have been carried out based on medical checkups, professional interventions have not been evaluated.

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The World Health Organization (WHO) has recommended a survey at age 12 as an international standard because, at this age, all permanent teeth have erupted, except for the third molars (5). WHO is now seeking to reduce the number of decayed, missing or filled teeth (DMF) in 12-year-old children to less than three. The purpose of this study was to assess the association between the incidence of dental caries and levels of salivary *S. mutans* and Lactobacillus in patients receiving professional preventive care.

A total of 2,132 under the age of 20 visiting private dental clinics for professional preventive programs were considered in the present study. Of these patients, 187 patients were examined for dental caries and salivary *S. mutans* and Lactobacillus in the baseline study, and those 12 years of age were analyzed in this study. The study population consisted of 80 (43.0%) males and 107 (57.0%) females averaging 5.53 ± 3.92 decayed or filled teeth per person. The mean follow-up period was 3.97 ± 2.29 years.

Salivary *S. mutans* and Lactobacillus were counted by using a commercially available *S. mutans* and Lactobacillus evaluation kit, Dentocult-SM and Dentocult-LB (Orion Diagnostica Co. Ltd., Epsom, Finland). The dental states were classed according to the manufacturer's instructions, i.e., for *S. mutans*, Dentcult SM Class 0-1: <100,000 colony forming units (CFU) *S. mutans*/ml saliva; Class 2 100,000 <CFU/ml <1,000,000; and Class 3 >1,000,000 CFU/ml, and for Lactobacillus, Dentcult-LB Class 0 <1,000 CFU Lactobacillus /ml in saliva; Class 1, 1,000 <CFU/ml <10,000 CFU; Class 2, 10,000 <CFU/ml <100,000; and Class 3 100,000 <CFU/ml <1,000,000.

Data for dental caries and dental plaque conditions, salivary buffering capacity, and 5-min stimulated salivary volume were

Table 1. Crude and adjusted odds ratios for the incidence of dental caries

	Crude Odds ratio	95% CI	*P* value	Adjusted Odds ratio	95% CI	*P* value
Follow up periods	0.831	0.725-0.953	0.008			
Sex	1.308	0.716-2.389	0.382			
df	1.024	0.946-1.110	0.555			
S. mutans	1.637	1.187-2.258	0.003	1.617	1.160-2.255	0.005
Lactobacillus	1.63	1.148-2.314	0.006	1.65	1.153-2.360	0.006
Salivary buffering capacity	1.586	1.025-2.453	0.038	1.596	1.026-2.483	0.038
Saliva volume	1.071	0.718-1.589	0.738	1.058	0.705-1.587	0.786
Dental plaque	1.519	0.854-2.729	0.162	1.428	0.787-2.591	0.242
Diet	1.358	0.848-2.173	0.203	1.461	0.902-2.366	0.123
Fluoride application	1.881	1.261-2.805	0.002	1.716	1.140-2.582	0.01

Follow-up periods were considered to be potential confounders and were adjusted by multiple logistic models. The Wald test was used to circulate *P* values.

Table 2. Dose-response relationships between the incidence of dental caries and association with the events

Factor	Criteria	Odds ratio	95% CI
S. mutans			
Class 0	CFU<10^4	Reference Group	
Class 1	CFU<10^5	1.669	0.439-6.346
Class 2	10^5<CFU<10^6	3.332	1.158-9.539
Class 3	10^6<CFU	4.179	1.447-12.069
Lactobacillus			
Class 0	CFU<10^3	Reference Group	
Class 1	10^3<CFU<10^4	1.000	0.399-2.506
Class 2	10^4<CFU<10^5	3.806	1.391-10.415
Class 3	10^5<CFU<10^6	3.581	0.914-14.029
Buffering capacity			
Class 0	7.0<pH in saliva	Reference Group	
Class 1	5.5<pH<7.0	1.001	0-1.0008
Class 2	4.5<pH<5.5	1.800	1.186-12.49
Class 3	pH<4.5	8.940	2.90-27.57
Fluoride application			
Class 0	Home, Professional	Reference Group	
Class 1	Home	1.894	0.783-4.581
Class 2	Professional	2.927	0.982-8.726
Class 3	None	4.689	0.805-27.329

Home: Brushing using fluoride dentifrice was performed every day at morning and night at home.
Professional: Topical fluoride application was performed in dental clinic one time every 3 months.

obtained by regular dental checkups. The salivary buffering capacity was evaluated by the commercially available kit Dent-Buff Strips (Orion Diagnostica). The fluoride usage and the times of daily food intake were determined using questionnaires.

Before the analysis, patients were divided into two groups: patients free from dental caries and patients who had at least one decayed or filled tooth. The cross table and chi-square test were used to check the correlation between the factors investigated in this study. Only two factors, *S. mutans* and Lactobacillus, were correlated (data not shown). Logistic regression analysis was used to evaluate the crude or adjusted odds ratios and their associated 95% confidence intervals (CI). The follow-up periods were treated as potential confounders. Table 1 shows the crude and follow-up periods adjusted by the odds ratios for the incidence of dental caries at age 12. The levels of salivary *S. mutans* and Lactobacillus were found to be closely related to the incidence of dental caries. Salivary buffering capacity and fluoride application were also correlated with the incidence of dental caries. The levels of dental plaque were not statistically significant but were highly correlated with the levels of *S. mutans* and Lactobacillus at the baseline.

Dose-response relationships were explored to determine factors associated with the incidence of dental caries. As shown in Table 2, obvious dose-response relationships were found between dental caries and each of the factors, the levels of *S. mutans* and Lactobacillus, buffering activity, and fluoride application.

The course of the dental caries was found to be multifactorial, with three primary factors: host, substrate, and micro-flora (6). In this study, we confirmed the contribution of fluoride-application and salivary-buffering-capacity factors to the host. Fluoride application has been known to reduce the incidence of dental caries by changing hydroxy apatite to fluoroapatite on the tooth surface and by preventing the attachment of *S. mutans* to the tooth surface. Professional preventive programs were found to reduce the prevalence of dental caries. However, even for patients participating in professional programs, salivary *S. mutans* and Lactobacillus remain as strong risk factors for dental caries. Controlling these factors will thus lead to one powerful strategy for preventing dental caries.

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