

Associations of Dental Caries with Salivary Mutans Streptococci/Lactobacilli and Plaque pH in 7-9 Years Old Children in Rural of Diyarbakir-Turkey¹

TÜRKİYE-DİYARBAKIR'IN KIRSALINDA 7-9 YAŞINDAKİ ÇOCUKLARDA TÜKÜRÜK MUTANS STREPTOKOK/LAKTOBASİL VE PLAK pH'SI İLE DIŞ ÇÜRÜĞÜ ARASINDAKİ İLİŞKİLER

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Summary

Purpose: The microbial flora of the mouth is highly complex, containing a wide variety of bacterial species. The most common types of oral diseases, such as dental caries and periodontal diseases, are both related to dental plaque and seem to occur when the normal balance between the microorganisms and the host is disturbed in some way. Mutans Streptococci and Lactobacilli are cariogenic since they are able to produce acid rapidly from fermentable carbohydrates. Therefore, estimation of salivary levels of these organisms may be useful for assessing caries risk in patients and for monitoring their response to preventive measures. This study aimed to evaluate the relation between plaque pH, MS, LB and DMF-T and/or df-t.

Materials and Methods: Seventy students were examined for dental caries. The levels of Mutans Streptococci and Lactobacilli in saliva were estimated by Dentocult system. Plaque pH of students was measured with scraping/harvesting method.

Results: Significant difference was determined between MS and LB while no significant association was found between plaque pH and dental caries.

Conclusion: The methods of preventive dentistry should not be confined by national boundaries, and should continue to be promoted throughout the world.

Key Words: Dental Caries, Streptococcus Mutans, Lactobacilli, Resting Plaque pH

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Özet

Amaç: Ağız mikrobiyal florası geniş bakteri türlerini içeren oldukça karmaşık bir yapıya sahiptir. Diş çürüğü ve periodontal hastalıklar gibi ağız hastalıklarının en yaygın tipleri, hem diş plağı ile ilişkilidir hem de mikroorganizma ile konakçı arasındaki normal balans herhangi bir yolla bozulduğu zaman meydana geldiği görülmektedir. Mutans streptokokları ve Laktobasiller, fermente edilebilir karbonhidratlardan hızlı bir şekilde asit üretebildikleri için karyojeniktirler. Bu nedenle, bu organizmaların tükürük seviyelerinin belirlenmesi, hastaların çürük riskini değerlendirmek ve koruyucu önlemlere karşı cevaplarını gözlemek için faydalı olabilir. Bu çalışma, plak pH, Mutans Streptokokları, Laktobasiller ve DMF(T)/df(t) arasındaki ilişkiyi değerlendirmek için amaçlandı.

Materyal ve Metod: 70 öğrenci çürük açısından değerlendirildi. Tükürük Mutans streptokokları ve Laktobasil değerleri Dentocult sistem ile hesaplandı. Öğrencilerin plak pH değerleri, Scraping/Harvesting metodu ile ölçüldü.

Bulgular: Sonuçta, plak pH ve diş çürüğü arasında önemli bir ilişki olmadığı gözlemlendi. Fakat tükürük Mutans Streptokokları ve Laktobasil seviyeleri ile çürük arasında ise önemli derecede ilişki gözlemlendi.

Sonuç: Sonuç olarak, koruyucu dişhekimliğinin metodları ülkemizle sınırlı kalmamalıdır ve dünyanın her yerinde ilerleyerek devam etmelidir.

Anahtar Kelimeler: Diş Çürüğü, Streptokokus Mutans, Laktobasil, Dinlenme Halindeki Plak pH

Mutans streptococci and lactobacilli were known with their more acidogenic and acidophilic traits than those of other oral bacteria have a cariogenic potential in animals and humans (1,2). Additionally, it has been clearly shown in many in-vivo and in-vitro studies that a process starting with the colonisation of MS and continuing with contribution of LB in susceptible individuals, leads eventually to caries initiation and progression (3-6). An acidogenic environment is crucial to

maintain the hemostasis of these organisms in dental plaque and to initiate the demineralisation on enamel surface (7,8).

Although there are conflicting reports regarding the relationship between caries incidence and the acidogenicity of dental plaque, based upon the clear evidence presented by Stephan et al. (9), the patients having caries and/or extreme caries activity have more acidogenic pH than those of caries free and/or caries-inactive counterparts in

resting plaque pH as well as in minimum plaque pH after sugar rinse (10).

Despite the general decrease in caries prevalence in young children in recent years, many children with lesions at an early age have been noted in some developing countries. For example in Brazil, caries has been reported in 46% of 6 to 36-months-old children (11). Although many significant differences in caries prevalence have been observed among the children living different areas of the world, major etiologic factors of dental caries are well established. However, many of the studies make clear that these variables have been performed in the developed countries, and the impact of each factor may be different in each population due to cultural, social and economic differences, and may vary in any of population as the time passes. For example in south-east of Turkey, the mean DMF(T) for rural districts was 1.63 in 1980's (12). On the other hand, in a recent field-trial, it was interestingly appeared as 3.09 (13). This observation suggests that the impact(s) of a (or other) factor(s) has to be considered.

With the help of this findings, the aim of this study was to investigate the relationship between df/DMF(T) score and salivary levels of Mutans Streptococci(MS), Lactobacilli(LB), and resting plaque pH values in 7 to 9 year old Turkish children from rural Diyarbakır of south-eastern Turkey.

Materials and Methods

This study was conducted in a village of Diyarbakır city, on 70 primary school children whose age were ranging 7 to 9 years.

The children were examined by the same examiner according to the WHO criteria (14). Subjects were reclined on a padded table so that oral cavity could easily be illuminated by day-light. Caries status were recorded by using the df(t) and/or DMF(T) index. A caries lesion was determined as a cavitation permitting probe penetration, rather than colour changes on enamel surface.

Paraffin-stimulated whole saliva samples were collected over 5 min. in a sterile tube. A CRT

bacteria test (CRT Bacteria Standard Package VIVADENT Ets., FL-9494 Schaan/Liechtenstein in Germany), a dip-slide method, was used to determine salivary levels of MS and LB. In this examination, one surface of the specifically designed test tube was used to spread the saliva for the MS count, and other surface was used for the LB count.

Bacterial levels were scored according to manufacturer's scoring-card. In this procedure, four different growing examples of both bacteria were scored as the range of 0 to 3 (0 and 1= $< 10^5$, 2 and 3= $\geq 10^5$).

For resting pH measurements, the plaque samples were taken 3.5-4 h. after breakfast by means of a proper hand instrument. Samples were taken from the mesial approximal sites of the upper first permanent molars. When the plaque was too sparse to be sampled, the same procedure was repeated on the mandibular counterparts. When the first molars were absent in the mouth, plaque samples were obtained from the buccal sites of second primary molars, and in the cases where plaque material could not be detected on the teeth sites mentioned above, samples were taken from the buccal sites of upper incisors.

Before sampling, the teeth were isolated with cotton-rolls. Immediately after the sampling procedure, plaque materials were transported to a tube containing 5 ml. distilled water (buffered in pH=6.5). According to a scraping/harvesting method protocol (15) resting plaque pH was measured by a pH-meter (EDT Instruments BA 350).

In the statistical method, for the comparison of the means, Tukey's HSD was used. Variance analysis (ANOVA) was used to explore significance of the differences between df/DMF(T), salivary MS/LB levels, and resting plaque pH values.

Results

None of 70 children examined in the context of this study were caries-free and mean df(t) and DMF(T) and df(t) and/or DMF(T) scores were 3.31 ± 1.05 , 1.27 ± 0.65 and 4.71 ± 1.85 , respectively.

Table 1. Summary of the salivary concentrations of MS and LB, mean resting plaque pH, and mean df/DMF(T) score.

	Mean	SD
Mutans Streptococci	2,75	±0,49
Lactobacilli	2,37	±0,68
Resting plaque pH	5,04	±0,42
df/DMF(T)	4,71	±1,86

The most affected teeth were the mandibular molars, which accounted for 56.4% of all recorded lesion in primary dentition, while 98.8% of all lesions recorded were upper first molars in permanent dentition.

In Table 1, mean values of salivary MS and LB levels, resting plaque pH, df(t) and/or DMF(T) had been presented.

Detecting the MS values, only one of the children showed the score 0, while the rest of children showed the scores 2 and 3.

Detecting the LB values in all children, it was seen that 8 of them had scores of 0 and 1, and 62 of them had scores 2 and 3. In the children with high MS levels (scores 2,3), higher mean caries were observed than those of low MS levels (scores 0,1) This observation was statistically significant for MS levels ($p < 0.001$). This difference also can be seen for LB levels ($p < 0.001$).

Same significant correlation had been observed between the salivary MS levels and resting plaque pH ($p < 0.001$), and the salivary LB levels and resting plaque pH ($p < 0.001$), whereas there had been no correlation between the resting plaque pH and mean df(t) and/or DMF(T) score ($p > 0.005$).

Discussion

In spite of the fact that dental caries is significantly declining over the world, as observed in our study, it is still present and continues to affect the majority of the world population, especially in less developed or developing countries.

When compared with previous studies (12,13), our results showed the increasing rate of dental caries and MS infection. Although they were randomly selected from only two different primary school of the village, the available results from this study indicates the hazardous spreading of caries infection over the rural areas of Diyarbakır. Moreover, these results are almost confirmed the similar evidence from our other field trial and also agreed with the our previous results that caries infection was rapidly spreading among young population at the beginning from 1990' in rural Diyarbakır (13).

In our study, the mean DMF(T) of (1.27) the rural children is higher than the values of 0.71 in Kenyan's 12-13-years old children living in urban areas (16), and is approximately similar to those reported as 1.19 by Warren et al (17). in 12-year-old Taiwanese children. Also the mean DMFT/dft (4.71) score obtained from our study is lower than the 6.3 reported by Carlsson et al. in Mozambique (18).

The production of acid by acidogenic and aciduric bacteria in dental plaque is an absolute prerequisite for the development of dental caries from the well organised study by Stephan in 1944 (9), it was obviously proved that the lower the pH profile measured in resting period and/or in a period immediately after sugar rinse, the higher the caries activity; and the longer the retativity of the lower pH profile, the greater the likelihood of tooth demineralization. However, later studies have collectively indicated that the most dramatic decrease in the range of the pH profile occurs with initial white spot or deep advanced caries lesions rather than with the resting plaque from caries active individuals (7,19).

In the results of our study, we found an insignificant relation between the plaque pH profile and the mean df(t)/DMF(T). Similar with many in vivo and in vitro studies (20-23). In addition, Kashket and Yaskell (24) have recently shown that enamel demineralization does not necessarily persist throughout the period of low pH. They found that demineralization was more likely to be limited by other factors, such as the

presence or high concentrations of calcium and phosphate ions in the plaque.

The present study is consistent with many others confirming that MS, together with LB, could be shown to be highly associated with caries in persons in epidemiologic studies. This finding is in accordance with the results of Nishimura et al (25), who investigated the relationship between the caries incidence and plaque and salivary MS and LB levels, with a new caries activity test in Japan; of Kreulen et al (26), who explored the risk factors of infant caries in Netherlands, and of Carlsson et al (18), who studied the relationship between the MS in the saliva and dental caries in children in Mozambique.

In our findings, the increasing number of MS has indicated the increasing number of caries lesion. This is in line with the clear evidence from the studies by Mattos-Graner et al (27), in Brasil and by Alaluusua and Renkonen (28) in Sweden that children with high numbers of MS develop a significantly higher caries incidence than children with low numbers.

Considering the observation that the most affected teeth is first molars in permanent dentition, our study is highly parallel to Garcia-Godoy et al.'s (29) conclusion that even though caries incidence seems to a steady declining trend over the world, the occlusal surfaces of the first permanent molars are still caries prone sites.

Even though second molars were the last ones erupted in primary dentition, they were the most decayed as observed in the present study. On the other hand, this observation is in contradiction with the basic philosophy of the classical caries etiology that dental caries is of an increasing potential with time. However, in many studies performed in primary dentition, the occlusal surface of second molars had been found to be the most commonly affected by caries, in spite of the fact that the fissure system in the primary teeth was less pronounced than in the permanent molars (30-32). Consequently, it could be emphasized by considering our findings that in the children having a cariogenic potential and living in a population with high MS infection or high caries prevalence

such as in rural districts of Turkey, preventive measures aiming to reduce infection level should and/or could firstly be directed to occlusal fissures of primary molars in where the proper stagnation areas for MS colonization exist.

As observed in this study, caries incidence is rapidly increasing among young population in rural Diyarbakır. Although, we did not investigate in the present study why this increasing trend was observed in rural Diyarbakır over two decades, recent social changes may be the explanation of this development. One of these changes, of course, was the migration from rural district to urban and/or sub-urban areas. This occurrence had also led to some habitual differences as in feeding habits and sugar consumption, especially for children. Many of the children and infants living in same region for the first time met with the sweet and sticky products and sugar drinks such as chocolate, cola, twix and ice-cream after the migration to the urban and near-urban areas. In those children who are not aware of any oral hygiene procedure, the equilibrium which had been set by simple feeding habit between the oral bacteria and host defence until that time could be broken down.

So, it was believed in this study until right now that the dental caries in rural sites of Turkey is less prevalent than in urban districts. On the contrary, the number of observations have surprisingly been carried out by us that dental caries rapidly spreads in rural areas as much as in urban counterparts.

Consequently, as pointed out in this study, children from some part of the world countries including Turkey, have been appearing to have caries experience at high rate since the first day of their life. Thus, the methods of preventive dentistry should not be confined by national boundaries, and should continue to be promoted throughout the world.

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