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## Original Article

# Preventive measures to reduce the transfer of *Streptococcus mutans* from pregnant women to their babies<sup>†</sup>

Eda Güler<sup>1\*</sup>, Hülya Köprülü<sup>2</sup>

<sup>1</sup> Department of Restorative Dentistry, Faculty of Dentistry, Ondokuz Mayıs University, Samsun, Turkey

<sup>2</sup> Department of Restorative Dentistry, Faculty of Dentistry, Hacettepe University, Ankara, Turkey

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### KEYWORDS

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**Abstract** *Background/purpose:* The aim of this study was to investigate the transfer of *Streptococcus mutans* to babies from prospective mothers in whom protective measures were carried out and who were trained in dental health care.

*Materials and methods:* Pregnant women were given fluoride varnish and restoration of active caries once during their pregnancy. No application of fluoride or restoration was given to the control group. Microbiological samples were taken after delivery of the babies in the control and study groups and after the babies had completed their 8<sup>th</sup> week, to obtain *S. mutans* isolation plaque samples. Colonies of *S. mutans* were calculated as colony-forming units (cfu). Samples were determined to be positive when both tests showed the presence of *S. mutans*. After the colonies were counted, the number of colonies was graded as 0, not-visible; 1, <10<sup>3</sup> cfu; 2, 10<sup>3</sup>–10<sup>5</sup> cfu; and 3, >10<sup>5</sup> cfu. Statistical analyses were performed with the Shapiro–Wilk test, Student's *t* test,  $\chi^2$  analysis, Wilcoxon test, Spearman's correlation analysis, and Fisher's exact test using SPSS software.

*Results:* After applying the preventive treatment program in the test group, statistically significant differences in both the plaque index and the number of *S. mutans* colonies were observed ( $P = 0.001$ ). There was a significant relationship of the number of *S. mutans* colonies of the control and test groups with those of the babies ( $r = 0.336$ ,  $P = 0.009$ ).

*Conclusions:* *Streptococcus mutans* is commonly transferred from mothers to their babies, and the preventive program applied to the pregnant women reduced both the amount of plaque and *S. mutans* colonization and thus had a positive effect.

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\* Corresponding author. Ondokuz Mayıs University, Faculty of Dentistry, Department of Restorative Dentistry, Kurupelit, Samsun 55139, Turkey. Fax: +90 362 4576032.

E-mail address: [edaguler@omu.edu.tr](mailto:edaguler@omu.edu.tr) (E. Güler).

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## Introduction

Dental infections such as tooth decay and periodontal disease are perhaps the most common bacterial infections in humans. Mutans streptococci (MS; *Streptococcus mutans*) are the principal bacteria responsible for dental caries in humans.<sup>1</sup> Several studies demonstrated the association between MS and caries in humans. MS and lactobacillus species are considered to be the principal pathogens of mineralized oral tissues.<sup>2</sup> MS have the capacity to produce acids, and are therefore considered "potential cavity formers". Traditionally, MS were believed to colonize the oral cavity only when teeth are present,<sup>3</sup> and most studies reported that initial colonization occurred only after eruption of the primary teeth.<sup>4,5</sup>

Infants are thought to acquire these organisms by vertical transmission from an infected individual, primarily the mother<sup>4</sup> within a discrete period called the "window of infectivity". This was estimated to be at around 19–31 months of age.<sup>4</sup> However recent investigations showed that colonization of *S. mutans* can occur in predentate infants.<sup>6</sup> If an infant acquires MS from the mother before or after the emergence of the primary teeth, preventive interventions aimed at reducing the mother's burden of MS could prevent or reduce the vertical transmission of these organism to infants and hence reduce subsequent caries experience.<sup>7</sup> Current knowledge of the acquisition of *S. mutans* in humans indicates that maternal transfer is a real possibility.<sup>8</sup>

The hypothesis of this study was that preventive intervention in pregnant mothers affects the transfer of *S. mutans* from mothers to their predentate babies. The aim of this study was to investigate the transfer of *S. mutans* to predentate babies from their mothers to whom a protective application was given and who were trained in dental health care.

## Materials and methods

Ethical clearance was obtained from the Ethics Committee of Ondokuz Mayıs University. All pregnant women were informed about the aim of the study, and signed informed consent was obtained. Pregnant women were chosen

according to the following criteria; 25–30 years old, and a first-time mother without the smoking habit or systemic disorders. Mothers in the control group were in the third trimester of pregnancy, whereas mothers in the study group were in the second trimester of pregnancy. The procedures applied to the study and control groups are given in Table 1. The plaque indices of the control and study groups were recorded as the Silness–Loe plaque index.<sup>9</sup> After determining the status, a mechanical plaque-removal procedure was applied to those women who needed periodontal care.

After delivery by the pregnant women in the control and study groups and after the babies had reached their 8<sup>th</sup> week, microbiological samples were taken to obtain *S. mutans* isolation plaque samples. A sterile cotton tip was rubbed on the dorsum of the tongue and in the toothless crest of babies and on all teeth surfaces of mothers. In order to have a uniform sample spread, the cotton tips were completely immersed in water; they were kept at room temperature for 15 min. Five milliliters of liquid was taken and put into trypticase yeast-extract cystine sucrose basitracin (TYCBS) (LabM, Lancashire, UK) including 0.25 units of basitracine and 20% sucrose, thus enabling a cultivation line in the loop and medium. This was incubated in a 5% CO<sub>2</sub> atmosphere at 37°C for 72 hours. Colonies were calculated as colony forming units (cfu) for *S. mutans*. To identify the bacteria, a biochemical analysis (Rapid Strep ID 32 API, BioMericux Vitek, Marcy-l Etoi'le, France) and gram staining were done to randomly selected colonies in petri dishes. The colonies were determined to be positive when both tests showed the presence of *S. mutans*. After the colonies were counted, the number of colonies was graded as 0, not visible; 1, <10<sup>3</sup> cfu; 2, 10<sup>3</sup>–10<sup>5</sup> cfu; and 3, >10<sup>5</sup> cfu.

Statistical analyses were performed with the Shapiro–Wilk test, Student's *t* test,  $\chi^2$  analysis, Wilcoxon test, Spearman's correlation analysis, and Fisher's exact test using SPSS software (SPSS, Chicago, IL, USA).

## Results

The average age of the study group was 24.57 years, and the average decayed, missing, and filled permanent teeth

**Table 1** Treatment procedures of the study and control groups.

Procedure	Study group		Control group	
	Mothers ( <i>n</i> = 30)	Babies ( <i>n</i> = 30)	Mothers ( <i>n</i> = 30)	Babies ( <i>n</i> = 30)
DMFT evaluation	First appointment		First appointment	
Plaque index	First appointment and after treatment		First appointment	
Diet analysis	+		+	
Oral hygiene education	+		+	
Mechanical plaque control	+		+	
Restoration of active caries	+		–	
Fluoride varnish	+		–	
Microbiological evaluation	First appointment and after treatment	8 <sup>th</sup> week	First appointment	8 <sup>th</sup> week

DMFT, index of decayed, missing, and filled permanent teeth.

**Table 2** Effect of the preventive treatment program on the plaque index and the number of *Streptococcus mutans* colonies in mothers of the study group.

	Before the preventive treatment program				After the preventive treatment program				Z	P
	n	Mean	Minimum	Maximum	n	Mean	Minimum	Maximum		
No. of <i>S. mutans</i> colonies <sup>a</sup>	30	2.7	2	3	30	1.4	1	2	-4.79	0.001
Plaque index <sup>b</sup>	30	1.5	0	3	30	0.73	0	2	-4.07	0.001

<sup>a</sup> 0, undetectable; 1, <10<sup>3</sup> colony-forming units (cfu); 2, 10<sup>3</sup>–10<sup>5</sup> cfu; 3, >10<sup>5</sup> cfu.

<sup>b</sup> Score 0, visible (-) and probing (-); 1, visible (-) and probing (+); 2, visible (+) and probing (+); 3, dense plaque.

(DMFT) score was 6.63. The average age of the control group was 24.43 years, and the average DMFT score was 7.83. Student's *t* test showed that there were no statistically significant differences ( $P > 0.05$ ) in the age ( $t = 0.170$ ,  $df = 58$ ,  $P = 0.865$ ) or DMFT score ( $t = -1.575$ ,  $df = 58$ ,  $P = 0.121$ ) between the study and control groups. There was no statistically significant difference in the plaque index ( $P = 0.836$ ) or the number of *S. mutans* colonies ( $P = 0.785$ ) between the control and study groups before the application of the preventive treatment program.

The results of the Wilcoxon test showed that there were statistically significant differences in the plaque index ( $P = 0.001$ ) and the number of *S. mutans* colonies ( $P = 0.001$ ) before and after application of a preventive treatment program in the study group (Table 2).

There was no statistically difference in the number of *S. mutans* colonies ( $P = 0.056$ ) between babies of the study and control groups by comparison with the  $\chi^2$  test (Table 3).

According to the Spearman's correlation analysis, there was a significant relation between the numbers of *S. mutans* colonies in the control and study groups to which the conservative/preventive program had been applied compared with their babies ( $r = 0.336$ ,  $P = 0.009$ ). The *S. mutans* colony number being greater or less than 10<sup>5</sup> in the pregnant women was seen to influence the *S. mutans* colony number of their babies according to Fisher's exact test (Table 4).

## Discussion

According to the results of our study, the null hypothesis was accepted. Because dental caries is an infectious disease, a plausible method of prevention is to remove the cariogenic bacteria from the mouth. In this regard,

knowledge of the exact time when *S. mutans* colonizes the mouths of infants is important to determine the optimal period for preventive and interceptive treatments.<sup>10</sup>

Berkowitz and Jordan found that mother-infected baby pairs had the some bacteria by serotyping.<sup>3</sup> Köhler and Bratthal reported that transmission of bacteria from mother to infant occurs by means of spoons.<sup>11</sup> Li and Caufield showed that mothers were the main source of bacteria in their study.<sup>5</sup> In a similar study, Wan et al. showed the transmission of MS from mothers to their predate infants.<sup>12</sup> Many studies demonstrated a strongly increased risk of MS colonization in children whose mothers have a high level of salivary MS. In fact, the large number of bacterial cells in the mother leads to implantation of MS in the child just after eruption of the first teeth.<sup>4,13</sup> Köhler et al. demonstrated that early infection with MS increases the risk of caries developing in the primary dentition.<sup>14</sup> According to findings of the above studies, the focus of preventive efforts on mothers with high levels of salivary MS was examined in the present study to minimize the caries risk in their children. Köhler et al. used preventive measures including: (1) information about the study, (2) dietary counseling, (3) professional tooth cleaning, (4) fluoride treatment, and (5) excavation of large caries.<sup>14</sup> The same protocols were carried out in the present study.

According to the results of the present study, there were no statistically significant differences in the age or DMFT scores between the study and control groups. Subjects in the control and study groups were homogeneous with respect to age and DMFT status.

It is important to decide the time of dental treatment of pregnant women. Usually dentists prefer the second trimester to perform dental procedures.<sup>5,15</sup> The second trimester is the most suitable time for pregnant women in terms of the emotional situation and physical comfort.<sup>16</sup> In the present study, pregnant women in the second trimester

**Table 3** Comparison of the number of *Streptococcus mutans* colonies between babies in the study and control groups.

Group	Number of <i>S. mutans</i> colonies (colony-forming units) <sup>a</sup>			Total
	0	1		
Control	n	26	4	30
	%	86.7	13.3	100
Study	n	30	0	30
	%	100	0	100

<sup>a</sup> 0, undetectable; 1, <10<sup>3</sup> colony-forming units.

**Table 4** Comparison of the numbers of *Streptococcus mutans* colonies in babies of the control and study group women for whom a preventive treatment program was applied when *S. mutans* colony numbers were less or greater than  $10^5$  colony-forming units (cfu).

No. of <i>S. mutans</i> colonies in the control and study groups of women which received the preventive treatment program (cfu)		No. of <i>S. mutans</i> colonies in babies		
		Undetectable	Detectable	Total
$\leq 10^5$	<i>n</i>	41	0	41
	%	100	0	100
$> 10^5$	<i>n</i>	15	4	19
	%	78.9	21.1	100
Total	<i>n</i>	56	4	60
	%	93.3	6.7	100

were chosen for the study group. However the control group was constituted of pregnant women who had a shorter time before giving birth (e.g., 1 week). For this reason, the only preventive treatment applied was oral hygiene education.

Fluoride is the most widely known and accepted anti-carries agent.<sup>17</sup> The advantages of this regimen include its general acceptance by subjects, its low cost, and easy application. In the present study 0.05% sodium fluoride was chosen as a minimal preventive protocol. Topical fluoride application was carried out after excavation of large caries and plaque control in the study group.

The time of the initial acquisition of MS by babies was investigated in different studies. Berkowitz et al. noted that the initial colonization in babies occurred at a mean of 3.8 months of age.<sup>8</sup> In Gripp and Schlagenhaut's study, infant age at the baseline was 20–137 days, and all of them were edentulous.<sup>18</sup> Wan et al. researched the colonization of *S. mutans* before tooth eruption in 6-month-old babies.<sup>12</sup> In Turkey, mothers have only an 8-week leave of absence after the birth of a baby. After 8 weeks, the contact of the mother with the baby may decrease because of such factors as care being provided by a babysitter or grandmother. To eliminate these factors on the effect of mothers on babies, colonization of *S. mutans* was evaluated in babies at 8 weeks old, similar to what was done in the above studies.

Microbiological samples were taken from mothers and infants by swabbing the dorsum of the tongue and all surfaces of the teeth of the mothers and the toothless crest of babies, using sterile cotton tips as described by Wan et al.<sup>19</sup>

Wan et al. showed that TYCBS was the most selective and sensitive medium for *S. mutans* in the laboratory and clinical research.<sup>6</sup> For this reason, TYCBS was used in the present study as a medium.

Berkowitz et al. classified *S. mutans* levels as  $<10^3$ ,  $10^3$ – $10^5$ , and  $>10^5$  cfu in their study.<sup>8</sup> In the present study *S. mutans* levels were classified as undetectable,  $<10^3$ ,  $10^3$ – $10^5$ , and  $>10^5$  cfu.

Results of the present study demonstrated a reduction in salivary MS levels in experimental group subjects that became significant ( $P < 0.001$ ) 3 months after the study began. Using a risk threshold of  $10^5$  cfu/mL, a 3-month treatment regimen resulted in a significant reduction in caries risk in experimental group subjects.

Brambilla et al. reported that there was a statistically significant difference in plaque and *S. mutans* levels between mothers in whom preventive application was carried out and those in whom preventive application was not carried out.<sup>17</sup> The treatment protocols applied and findings of the present study paralleled those of that study.

## Conclusions

Data from our study showed that *S. mutans* infection in prenatate infants can occur especially in those with mothers who have poor oral hygiene. Pregnant mothers who had numbers of *S. mutans* exceeding  $10^5$  cfu affected *S. mutans* acquisition by their babies. Our data support preventive approaches impeding the early acquisition of *S. mutans*. *Streptococcus mutans* is transferred to babies from their mothers, and the preventive program applied to prospective mothers reduced both the amount of plaque and *S. mutans* colonization and thus had a positive effect. In order to avoid the transfer of MS from mother to child, the extent of the mother's infection needs to be assessed.

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