# Mutans Streptococci Oral Colonization in 12–30-month-old Brazilian Children over a One-year Follow-up Period

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## Abstract

***Objectives:*** *The infection levels of mutans streptococci were investigated during a one-year follow-up in children aged 12 to 30 months attending school nurseries where a sucrose-rich diet was provided.* ***Methods:*** *Oral levels of mutans streptococci obtained from 101 children at baseline and after a one-year follow-up were compared by age, number of teeth, feeding habits, and presence of visible plaque at baseline. Baseline predictors and changes in mutans streptococci levels during the study were compared to caries incidence after one year.* ***Results:*** *Fluctuations in mutans streptococci levels during the follow-up period were not related to feeding habits or presence of visible plaque. Mutans streptococci levels increased after one year among children aged 12 to 24 months, while a significantly higher proportion of those aged 25–30 months showed a decrease in mutans streptococci levels during the study. Multiple logistic regression analysis suggested that high levels of mutans streptococci (≥100 cfu) at baseline were associated with a higher caries increment, while reduction in mutans streptococci was negatively associated with caries incidence.* ***Conclusions:*** *Our data suggest that despite early mutans streptococci infection and high exposure to sucrose, mutans streptococci may achieve relatively stable levels after 2 years of age. Heavy colonization by mutans streptococci in an early age was related to an extremely high caries incidence during childhood, while decreasing levels of mutans streptococci can be associated to the decrease in caries activity. [J Public Health Dent 2001;61(3):161-67]*

*Key Words: children, mutans streptococci, dental caries, diet, dental plaque.*

The early oral colonization by mutans streptococci has been related to a high caries incidence in childhood (1,2). The understanding of factors related to the initial infection and establishment of mutans streptococci in the mouth is important for the development of preventive strategies to control infection by these organisms. Studies have shown that mothers are the main source of mutans streptococci transmission (2,3); nevertheless, factors involved in the initial establishment of mutans streptococci in the mouth are not completely clear. Caufield et al. have postulated a discrete "window of infectivity" for mutans streptococci acquisition occurring between 19 to 31 months of age (4). However, the time of infection may vary depending on factors modulating transmission from mothers or other caregivers to the children, immunological status, oral hygiene habits, and sucrose consumption (2,5,6). For example, prolonged sucrose consumption in baby bottles has been associated with heavy infection with mutans streptococci and a distinct development of caries lesions on the smooth surfaces of upper incisors shortly after eruption of these teeth (6,7). In a Brazilian population of children attending school nurseries and exposed to a high sucrose challenge, we have described a prevalence of mutans streptococci of 70.8 percent among children who were between 12 to 19 months of age (8). In the present study, we investigated the variations in the infection levels of mutans streptococci during a one-year follow-up in the same population and its associations with factors studied at baseline that could be important for mutans streptococci establishment. We also analyzed the caries incidence in the same follow-up period with respect to initial levels of mutans streptococci, changes in their levels during the follow-up, and other predictors observed at baseline.

## Methods

**Study Group.** The study group included 101 infants and toddlers (55 girls and 46 boys) who were between 12 and 30 months of age. The group was a subset from 142 children previously studied (8). These children were examined clinically and microbiologically at baseline and after a one-year follow-up. Children undergoing antibiotic therapy were not included in the study. To explore factors involved in mutans streptococci progression during one year, infection levels of mutans streptococci were measured at baseline and after a one-year follow-up. These data were then analyzed with respect to baseline variables including age, number of erupted teeth, presence of visible plaque, and sucrose consumption in baby bottles at night. The infection levels of mutans streptococci observed after one year were also compared to the initial levels of mutans streptococci. Baseline predictors and changes in mutans streptococci levels were analyzed with respect to caries inci-

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dence during the follow-up period.

The children studied were from families of low socioeconomic level and attended nine public school nurseries in the city of Piracicaba, São Paulo, randomly selected from a total of 21 units of the city. Children attended the school nurseries for five days per week, where they received a very homogeneous diet during a day care period of 10 hours, including four sucrose-rich meals (Table 1). This population consumed fluoridated drinking water (0.7 ppm). After the children reached 2 years of age, their teeth were brushed daily with fluoridated toothpaste by the health agents of the school nurseries. Written informed consent was obtained from the parents of all children participating in the study. All consent and experimental procedures were approved by the institutional Ethical Committee of the University of São Paulo, School of Dentistry.

**Bacteriological Examination.** Oral samples were collected with sterile tongue blades, which were introduced in the mouth until moistened with saliva. Excess saliva was removed by pressing both sides of the tongue blade against the dorsum of the tongue (9). The tongue blades were then pressed on the surface of MSB agar (Difco) containing 2 UI/ml of bacitracin (Sigma, USA) and sucrose 15 percent (Difco) in contact plates (Nunc, Denmark). The plates were incubated in plastic bags with expired air at 37°C for 48 hours (10). The number of mutans-like colonies was measured in a predetermined area of the tongue blade impression (1.5 $cm^2$) using a stereoscopic microscope. Children were then classified in four different categories of mutans streptococci levels: 0 cfu, 1–20 cfu, 21–99 cfu, and ≥100 cfu.

**Clinical Examination.** After sample collection, children were clinically examined with the aid of a portable light for evaluation of the presence of visible plaque on labial surfaces of upper incisors (11). Accumulated plaque was considered present when visually detected in one or more of the examined surfaces (8). After plaque registration, the teeth were carefully brushed and the proximal surfaces cleaned with dental floss. Tooth surfaces were dried with gauze and then visually examined with the aid of a mouth mirror under artificial light to detect initial (white-spot) and manifest caries lesions. A manifest lesion was defined as a cavity detected by visual inspection. The criteria used for the diagnosis of initial and manifest lesions were the same used at the baseline (8). All clinical examinations were carried out by one of the authors (ROMG) whose intraexaminer reliability was determined for tooth surfaces at baseline (kappa=0.68 and 0.79 for initial and manifest lesions, respectively) and after one-year follow-up (kappa=0.81 and 0.95 for initial and manifest lesions, respectively).

**TABLE 1**
**Dietary Schedule Offered in Public School Nurseries for Children 12–36 Months Old**

| Time | Meal Content |
|---|---|
| 8:00 AM | Milk with chocolate formula*; bread with butter |
| 9:30 AM | Fruit juice† |
| 10:30 AM | Egg or chicken or beef; boiled vegetable; rice and bean or pasta; salad |
| 11:30 AM | Candy |
| 2:00 PM | Cereal porridge or milk with chocolate formula* and biscuits |
| 4:00 PM | Soup |

*Total of 125 g of chocolate formula more 50 g of sucrose per liter of milk.
†Total of 75 g of sucrose per liter of juice.

**Interviews.** At baseline, the health agents of each nursery interviewed the mothers of 79 of the 101 children, using a standardized questionnaire with open-ended questions regarding feeding habits (8). For example, the evaluation of sucrose consumption at night in baby bottles was based on the following question: "What is the recipe used to prepare the baby bottle which you offer to your child at night?" The use of bottle with sucrose was considered when sucrose, chocolate formula, or other formulas containing sucrose were added to the milk offered at night.

**Statistical Analysis.** Bivariate analysis was used to assess the impact of factors that could be related to mutans streptococci detection and changes in categorical levels of mutans streptococci, and crude odds ratio was calculated.

Multiple logistic regression analysis was employed to evaluate the independent relationship between caries incidence and other explanatory variables including age, number of erupted teeth, sucrose consumption in baby bottles, plaque on upper incisors and oral levels of mutans streptococci. The presence of caries lesions (initial and manifest) at baseline was also included as an explanatory variable in the regression model. In addition, the change in mutans streptococci levels after one year, including three categories (decrease, no change, or increase in mutans streptococci levels), was analyzed as an explanatory variable for caries incidence in the multiple logistic regression model. All variables that achieved $P<.02$ in a bivariate analysis for caries incidence were tested in the multivariate model using stepwise forward selection procedure. The independent variable was kept in the final model when $P<.05$ or adjusted the parameter estimates for the other variables. Variables that were not included in the final regression model were tested in the same categories used for bivariate analysis. The Hosmer-Lemeshow goodness-of-fit test was used to verify the sensitivity of the final model.

We stratified by age and used the Mantel-Haenszel summary odds ratio to evaluate the relationship between baseline levels of mutans streptococci and caries development. The Mann-Whitney U-test and chi-square test were used to evaluate differences between groups.

## Results

Among 142 infants and toddlers examined at baseline, 41 children did not remain in the prospective study because they relocated or were not present on the days of the examinations. Comparative analysis between the 101 participants and 41 dropouts did not detect significant differences in the mean age or number of erupted teeth observed at baseline (Mann-Whitney

U-test: $P<.05$). No differences were detected between the groups in the frequencies of mutans streptococci detection, baby bottle usage, presence of visible plaque, or caries prevalence when initial lesions were considered. However, the 41 dropouts showed a significantly higher prevalence of manifest lesions (34.1%) than was observed in the 101 children who remained in the study (12.9%; chi-squared: $P<.01$). The nine clusters of children from each school nursery included in the study were compared regarding the frequency of mutans streptococci detection and frequency of heavy infection at both phases of the study, and regarding the number of children who developed new manifest lesions during the follow-up period. Differences between these clusters

**FIGURE 1**
**Frequencies of Children with Different Oral Levels of Mutans Streptococci (cfu) at Baseline and after 1-year Follow-up by Months of Age at Baseline**

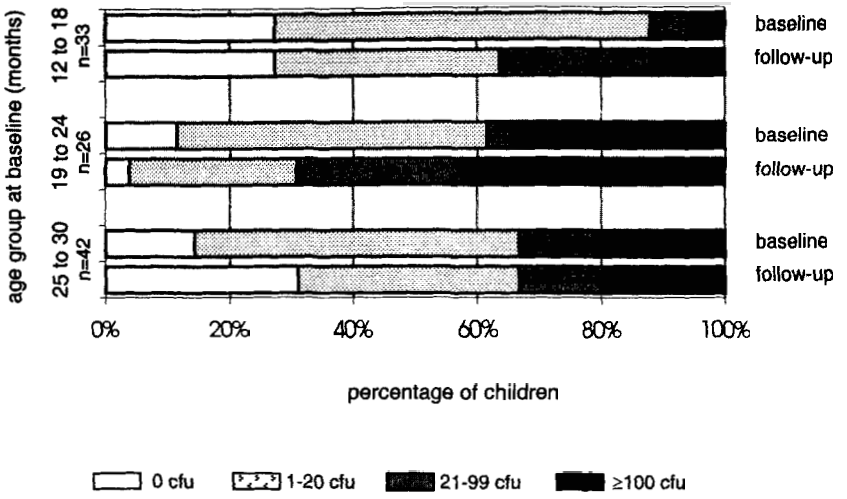


**TABLE 2**
**Bivariate Analysis of Mutans Streptococci Detection at Baseline and After 1-year Follow-up by Selected Baseline Characteristics and Frequencies of Children with Detectable Levels of Mutans Streptococci at Baseline But Not after 1 Year**

| | Mutans Streptococci | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| | At Baseline | | | After 1-year Follow-up | | | Detected at Baseline But Not after 1 Year | |
| Observations at Baseline | Detectable Levels | | | Detectable Levels | | | | |
| | *n* | % | OR (95% CI) | *n* | % | OR (95% CI) | *n* | % |
| Age group (months): | | | | | | | | |
| 12–18 (*n*=33) | 24 | 72.7 | 1.00 | 24 | 72.7 | 1.00 | 6 | 18.2 |
| 19–24 (*n*=26) | 23 | 88.5 | 2.88 (0.69, 11.97) | 25 | 96.2 | 9.38 (1.10, 79.72) | — | — |
| 25–30 (*n*=42) | 36 | 85.7 | 2.25 (0.71, 7.14) | 29 | 69.0 | 0.84 (0.30, 2.29) | 12 | 28.6 |
| Number of erupted teeth: | | | | | | | | |
| 1–8 (*n*=16) | 8 | 50.0 | 1.00 | 12 | 75.0 | 1.00 | 2 | 12.55 |
| 9–19 (*n*=69) | 60 | 87.0 | 6.67 (2.00, 22.24) | 54 | 78.3 | 1.20 (0.34, 4.26) | 10 | 18.2 |
| 20 (*n*=16) | 15 | 93.8 | 15.00 (1.58, 142.07) | 12 | 75.0 | 1.00 (0.20, 4.95) | 6 | 20.0 |
| **Baby bottle at night:** | | | | | | | | |
| **No or without sucrose (*n*=17)** | 14 | 82.4 | 1.00 | 14 | 82.4 | 1.00 | 2 | 11.8 |
| **With sucrose (*n*=62)** | 51 | 82.3 | 0.99 (0.19, 4.73) | 48 | 77.4 | 0.73 (0.14, 3.33) | 14 | 22.6 |
| Plaque on upper incisors: | | | | | | | | |
| Visible (*n*=55) | 47 | 85.5 | 1.00 | 42 | 76.4 | 1.00 | 10 | 18.2 |
| Not visible (*n*=46) | 36 | 78.3 | 0.61 (0.22, 1.71) | 36 | 78.3 | 1.11 (0.44, 2.84) | 8 | 17.4 |
| Oral levels of mutans strep. | | | | | | | | |
| 0 cfu (*n*=18) | | | | 13 | 72.2 | 1.00 | | |
| 1–20 cfu (*n*=55) | | | | 38 | 69.1 | 0.86 (0.22, 3.18) | 17 | 30.9 |
| 21–99 cfu (*n*=8) | | | | 8 | 100.0 | — | — | — |
| ≥100 cfu (*n*=20) | | | | 19 | 95.0 | 7.31 (0.67, 186.09) | 1 | 5.0 |

OR=odds ratio; CI=confidence interval; cfu=colony-forming units.

were not statistically significant (chi-squared: $P>.05$).

The frequencies of children with detectable levels of mutans streptococci at baseline and follow-up by different observations at baseline are shown in Table 2. There was a group of 18 children with detectable levels of mutans streptococci at baseline from whom mutans streptococci could not be recovered after the follow-up period. Multiple logistic regression analysis was not performed because only the number of the erupted teeth was significant in the bivariate analysis for mutans streptococci at baseline and only age group was significant in the bivariate analysis for mutans streptococci after a one-year follow-up. Figure 1 illustrates the fluctuations in the categorical levels of mutans streptococci during the follow-up period among different age groups. While 45 children remained in the same category of mutans streptococci levels detected at baseline after one year, fluctuations in the mutans streptococci levels were observed among 56 children (Table 3). Comparisons of the frequencies of children with increasing ($n$=30) and decreasing ($n$=26) levels of mutans streptococci during one year by different variables observed at baseline are shown in Table 3. Reduction of the categorical levels of mutans streptococci during the follow-up period was significantly more frequent among children with initial age of 25 to 30 months. Of the 20 children heavily infected (≥100 cfu) at baseline, seven (35%) experienced a decrease in mutans streptococci levels during the follow-up. Among these seven children, four were classified as having 21–99 cfu, two as having 1–20 cfu of mutans streptococci, and one had undetectable levels of mutans streptococci after one year.

Table 4 shows the number of children who developed new manifest lesions and the mean caries increment in a one-year follow-up by baseline mutans streptococci levels. Nine (45%) of the heavily infected children at baseline did not develop new manifest lesions during the follow-up. Among these nine children, six showed lower levels of mutans streptococci after one year. Multiple logistic regression analysis for the development of manifest lesions suggested that children who experienced a decrease in mutans streptococci levels during the follow-up period had a lower likelihood of developing new manifest lesions (Table 5). Heavily infected children at baseline had a significantly higher risk for developing new manifest lesions than those with lower levels of mutans streptococci (Table 5). The other variables listed in Table 2 were tested in the multivariate analysis, but they did not achieve statistical significance.

## Discussion

The studied population was characterized by a very early mutans streptococci infection and high sucrose challenge (Tables 1 and 2). We have previously reported that 70.8 percent of children between 12 and 18 months of age had detectable levels of mutans streptococci, and 10 percent of this age group were heavily infected (8). Among 16 children with only incisors erupted (1–8 teeth), 50 percent had detectable levels of mutans streptococci

**TABLE 3**
**Bivariate Analysis of Changes in Categories of Mutans Streptococci Oral Levels after One-year Follow-up Period, by Selected Baseline Characteristics**

| Observations at Baseline | Same Levels at Baseline $n$ (%) | Changes in Categories of Mutans Streptococci Oral Levels | | |
|---|---|---|---|---|
| | | Increase $n$ (%) | Decrease $n$ (%) | OR* (95% CI) |
| Age group (months) | | | | |
| 12–24 ($n$=59) | 29 (49.2) | 20 (33.9) | 10 (16.9) | 1.00 |
| 25–30 ($n$=42) | 16 (38.1) | 10 (23.8) | 16 (38.1) | 3.20 (1.07, 9.57) |
| Number of erupted teeth | | | | |
| 1–8 ($n$=16) | 7 (43.8) | 7 (43.8) | 2 (12.5) | 1.00 |
| 9–19 ($n$=69) | 31 (44.9) | 20 (29.0) | 18 (26.1) | 3.15 (0.58, 17.17) |
| 20 ($n$=16) | 7 (43.8) | 3 (18.8) | 6 (37.5) | 7.00 (0.86, 56.89) |
| Baby bottle at night | | | | |
| No or w/o sucrose ($n$=17) | 5 (29.4) | 7 (41.2) | 5 (29.4) | 1.00 |
| With sucrose ($n$=62) | 28 (45.2) | 18 (29.0) | 16 (25.8) | 1.24 (0.33, 4.71) |
| Plaque on upper incisors | | | | |
| Not visible ($n$=55) | 25 (45.5) | 15 (27.3) | 15 (27.3) | 1.00 |
| Visible ($n$=46) | 20 (43.5) | 15 (32.6) | 11 (23.9) | 0.73 (0.25, 2.11) |
| Oral levels of mutans strep. | | | | |
| 0 cfu ($n$=18) | 5 (27.8) | 13 (72.2) | — | † |
| 1–20 cfu ($n$=55) | 25 (45.5) | 13 (23.6) | 17 (30.9) | |
| 21–99 cfu ($n$=8) | 3 (37.5) | 3 (37.5) | 2 (25.0) | |
| ≥100 cfu ($n$=20) | 13 (65.0) | — | 7 (35.0) | |

*Comparisons between groups with increased and decreased levels of mutans streptococci.
†Not calculated because of empty cells.

**TABLE 4**
**Frequency of Infants Who Developed New Manifest Lesions in One-year Follow-up, and Number of Lesions (Initial and Manifest) Developed by Oral Levels of Mutans Streptococci (cfu) Detected at Baseline in Different Age Groups**

| Baseline Mutans Streptococci Levels by Age Group | Infants with New Manifest Lesions | | | # of Lesions Developed in 1 Year | |
|---|---|---|---|---|---|
| | *n* (%) | OR* | 95% CI | Mean (SD) | *P*-value† |
| 12–18 months | | | | | |
| 0–99 cfu (*n*=31) | 4 (12.9) | | | 1.00 (1.18) | |
| ≥100 cfu (*n*=2) | — | 1.00 | — | — | |
| 19–24 months | | | | | |
| 0–99 cfu (*n*=17) | 4 (23.5) | | | 1.12 (1.62) | |
| ≥100 cfu (*n*=9) | 5 (55.6) | 4.06 | 0.54–34.62 | 4.78 (3.83) | .013 |
| 25–30 months | | | | | |
| 0–99 cfu (*n*=33) | 6 (18.2) | | | 1.70 (3.26) | |
| ≥100 cfu (*n*=9) | 6 (66.7) | 8.39 | 1.35–67.66 | 4.56 (3.47) | .006 |

*Mantel-Haenszel summary chi-squared (*n*=101): 6.81; *P*=.009.
Crude odds ratio: 5.85. Mantel-Haenszel weighted odds ratio: 4.94; 95% CI: 1.66–14.81.
†Mann-Whitney U-test.

**TABLE 5**
**Multiple Logistic Regression Analysis for Development of New Manifest Lesions During One-year Follow-up**

| Independent Variables | $OR_{crude}$ | $OR_{adjusted}$ | 95% CI | *P*-value |
|---|---|---|---|---|
| Presence of caries lesions at baseline | | | | |
| No caries lesions | 1.00 | 1.00 | | |
| With caries lesions | 6.67 | 6.17 | 1.80, 21.08 | .004 |
| Oral levels of mutans streptococci at baseline | | | | |
| 0–99 cfu | 1.00 | 1.00 | | |
| ≥100 cfu | 5.85 | 8.79 | 1.63, 47.39 | .012 |
| Changes in mutans streptococci levels after 1 year | | | | |
| No change | 1.00 | 1.00 | | |
| Increase | 0.66 | 1.44 | 0.40, 5.25 | .580 |
| Decrease | 0.07 | 0.03 | 0.00, 3.34 | .005 |
| Age at baseline | | | | |
| 12–18 months | 1.00 | 1.00 | | |
| 19–30 months | 0.31 | 1.13 | 0.28, 4.49 | .862 |

Constant –2.0777. Hosmer-Lemeshow goodness-of-fit test: *P*=.6.

at baseline (Table 2). The mutans streptococci prevalence in this population was much higher than that reported in previous studies carried out in developed countries (4,12,13). Mutans streptococci was not detected before 19 months of age in samples of pooled plaque or saliva collected with swabs from a total of 46 children in the United States (4). It is noteworthy that those American children were considered to be at high risk for mutans streptococci acquisition because their mothers exhibited high mutans streptococci levels and a high number of caries lesions (4). Although the mother is considered the main source for infection (2,3), little is known about mutans streptococci transmission in day care nurseries, where the transmission of various infectious agents might increase. Mutans streptococci was detected in 20 percent of saliva samples collected with cotton rolls in 12- to 18-month-old children in Japan (12) and in 6 percent of oral samples collected by drawing swabs on the anterior two-thirds of the dorsum of the tongue from 12-month-old infants in Sweden (13). Variations in methods for sampling and culturing could influence the detection of mutans streptococci in the mouth (14).

The increase in levels of mutans streptococci with increasing age and number of teeth has been described previously (15,16). We observed an increase in mutans streptococci levels during the follow-up period in 33.9 percent of children who were 12–24 months of age. On the other hand, a significantly higher proportion of children older than 2 years of age experienced a reduction of mutans streptococci levels during the follow-up (Table 3). These data are consistent with previous findings of Caufield et al. (4) suggesting that there is a decreasing risk for mutans streptococci acquisition after 2 years of age. The frequencies of increasing levels of mutans

streptococci were also negatively related to the number of teeth erupted at baseline, although the differences did not achieve statistical significance (Table 3). However, it is important to consider that the relativly small sample sizes may have limited the statistical analysis for some variables, which were evidenced by wide confidence intervals for odds ratio estimates.

There was a group of 18 children with detectable levels of mutans streptococci at baseline from whom mutans streptococci could not be recovered after a one-year follow-up (Table 2). Among these children, 17 carried low levels of mutans streptococci at baseline (1–20 cfu) and one was heavily infected (≥100 cfu). Fluctuations in mutans streptococci detection were particularly frequent among children with low mutans streptococci levels (16,17). Substantial reduction in mutans streptococci levels with the increasing age among children initially with high mutans streptococci oral levels was already described in the literature (5). Given the large number of children studied, subsequent samples were not tested to strengthen the first detection of mutans streptococci. Thus, it is possible that some mutans streptococci detected in low levels at baseline were transient or were not detected at one of the examinations due to natural sampling variations.

Among nine children heavily infected at baseline who did not develop new manifest lesions during the follow-up, six (66.7%) experienced a decrease in their levels of mutans streptococci, although three of them still carried levels considered high for their age (from 51 to 81 cfu ) (data not shown). The multiple logistic regression model suggested that the reduction of mutans streptococci levels after one year was associated with a lower risk for caries development (Table 5). The study of factors related to these fluctuations during the initial establishment of mutans streptococci would be of considerable interest. For example, the immune response to mutans streptococci antigens by the host and the expression of different virulence factors by mutans streptococci strains not able to persist in the mouth should be further explored.

Mutans streptococci were detected in 60 percent of samples collected with tongue blades from 15-month-old children using nursing bottles in the United States (18). A previous study suggested an increased risk for mutans streptococci colonization in children consuming sugar-containing beverages at night (13). We could not detect a relationship between sucrose consumption in baby bottles at night and the mutans streptococci detection at baseline (Table 2), nor with the changes in mutans streptococci levels during the follow-up period (Table 3). The small number of infants and toddlers who did not consume sucrose in baby bottles may have created difficulties in the analysis of these data. Alternatively, the children in this study received at least four sucrose-rich food intakes during a daily period of 10 hours (Table 1), a diet that might favor mutans streptococci colonization. Roeters et al. (17) could not detect a positive relationship between sucrose consumption and levels of mutans streptococci in children from 2 to 5 years of age evaluated through the 24-hour recall method. They suggested that variations in mutans streptococci infection because of differences in sucrose intake could be better identified in populations with low sucrose consumption (17). In contrast, rampant caries in children aged 1 to 2.5 years has been associated with high mutans streptococci levels in the absence of nursing bottles in Tanzania (19), which might be related to the presence of enamel linear hypoplasia (20). A complex group of factors related to the general health of children may influence the individual susceptibility to mutans streptococci colonization and caries development by affecting tooth development, salivary glands function, and immunological defenses (21). Despite high sucrose consumption and early mutans streptococci infection, the children in the present study did not demonstrate patterns of rampant caries in that most of the caries affected occlusal surfaces (data not shown). The exposure to fluoride may account for this observation.

Children older than 24 months of age had their teeth brushed daily with fluoride toothpaste. The role of this procedure in fluctuations of mutans streptococci infection levels should be further investigated. Despite the additional exposure to fluoride, children aged 25–30 months who were heavily infected at baseline showed a higher caries increment in one year than those of the same age group with lower levels of mutans streptococci (Table 4). No statistically significant relationship was detected between accumulation of dental plaque on upper incisors and mutans streptococci infection recorded in either phase of the study (Table 2). Moreover, the presence of visible plaque was not associated with caries development during the one-year follow-up in the multiple logistic regression analysis.

The fact that children with manifest caries lesions dropped out more frequently than those who did not have manifest lesions might limit the representativeness of the studied group. However, there were no significant differences in baseline mutans streptococci infection levels between participants and dropouts. Multiple logistic regression analysis suggested that heavily infected children have significantly higher risk for caries development (Table 5). This observation supports the evidence that the early establishment of mutans streptococci is related to a higher caries activity during childhood (1,2,12). Because mutans streptococci could be detected when only incisors were erupted and achieved high levels at an early age, our data highlight the need for early preventive measures to control mutans streptococci infection in this population. Additional studies are needed to evaluate factors related to mutans streptococci infection in school nurseries. These institutions have an important role in public health systems in developing countries, such as Brazil, because they provide care for a significant part of the low-income population.

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