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# Feeding frequency in infancy and dental caries in childhood: a prospective cohort study

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**Objectives:** Feeding patterns in infancy are plausible contributors to dental caries later in childhood, yet relatively few cohort studies have examined potential dietary risk factors at this age. This study aimed to investigate the associations between feeding frequency at age 12 months and caries prevalence at age 3 years. **Methods:** In this prospective birth cohort of 345 Brazilian children, all foods and drinks consumed at age 12 months, including bottle-use and breastfeeding, were recorded using two 24-hour infant dietary recalls with mothers. The prevalence of early childhood caries (ECC) and severe ECC (S-ECC) at age 38 months were compared in groups defined according to 12-month feeding frequency, using regression models to adjust for sociodemographic characteristics and total carbohydrate intake. **Results:** Independent of other variables, compared with children with infrequent bottle-use and breastfeeding at 12 months, at 38 months the ECC prevalence was 1.8-times higher in children bottle-use higher with combined high frequency of bottle and breastfeeding together (P = 0.04), but the association with consumption of other foods or drinks more than five times/day [risk ratio (RR) = 1.2; P = 0.10] was not statistically significant. Prevalence of S-ECC was significantly associated with frequent breastfeeding (RR = 2.4; P < 0.001) and with greater frequency of consumption of other foods or drinks (RR = 1.7, P = 0.001). **Conclusions:** High-frequency feeding in late infancy, including both bottle use and breastfeeding, were positively associated with dental caries in early childhood, suggesting possible early-life targets for caries prevention.

Key words: Dental caries, preschool child, nutrition, breastfeeding, feeding practices

#### INTRODUCTION

Consistent evidence associates dental caries in childhood with diminished quality of life for children and their families<sup>1,2</sup>. Given the cost of restorative dental treatment and the relatively high disease prevalence already reached by the time children achieve school age, there is considerable interest in effective caries prevention further upstream. Infant feeding practices are an attractive intervention target, especially given that inadequate feeding, specifically a high-sugar and low-fibre infant diet, has been associated with childhood overweight, obesity and micronutrient deficiency<sup>3,4</sup>, in addition to dental caries<sup>5</sup>. Defining specific aspects of early-life feeding that may set a child on a dietary course that heightens caries risk could inform effective, early prevention.

It is highly plausible that food-intake frequency is a caries risk factor in children<sup>6,7</sup>. However, measurement and methodological challenges face any study of infant-feeding behaviours. Feeding habits, particularly during the first year of life, involve a number of simultaneous and interrelated behaviours, such as breastfeeding, bottle-feeding and the introduction of complementary foods<sup>8</sup>. Some feeding behaviours established at this age may be maintained in later years, potentially impacting health over the course of life<sup>9,10</sup>. However, most studies of dental caries and feeding practices among preschool children have been cross-sectional, impeding the ability to assess feeding

behaviours that were in place earlier. Additionally, feeding information is collected using non-validated questionnaire items or food frequency instruments, which may fail to capture important details regarding daily behaviours<sup>11</sup>. Moreover, many studies have categorised breastfeeding and bottle-feeding mutually exclusively and/or dichotomously<sup>12,13</sup>, missing information about the frequency of these behaviours, separately or in combination. To our knowledge, no prospective studies using 24-hour dietary recalls have assessed infant-feeding behaviours in relation to later dental caries occurrence and severity. Better understanding of the relationship between food-intake frequency in infancy and tooth decay in childhood adds to the body of evidence potentially informing specific guidelines and future interventions.

The present study aims to measure associations between infant feeding frequency, feeding methods (e.g. breast, bottle or both), and dental caries in early childhood among a birth cohort of children, about whom feeding habits were recorded prospectively using multiple 24-hour infant diet recalls with mothers. Study objectives were to calculate the independent associations of breastfeeding frequency, baby bottleuse frequency and other food-intake frequency at age 12 months with the prevalence of early childhood caries (ECC) and severe early childhood caries (S-ECC) at age 3 years. In addition, we calculated associations of total feeding frequency (all food and drink sources) with ECC, S-ECC and the number of decayed, missing and filled teeth (dmft) at age 3 years.

#### METHODS

#### Study design and participants

The present investigation is a prospective cohort study nested within a clinical trial in Porto Alegre, a city in southern Brazil with 1.4 million inhabitants and a fluoridated community water supply (0.7 ppm F). The trial recruited pregnant women at 20 municipal health centres (Basic Health Units, UBS) in 2008. The UBS system primarily serves low-income individuals, but services are available to all city residents. Health centres were eligible for inclusion if they recorded  $\geq 100$  infant visits annually and were not involved in any other nutritional guidance programme. The goal of the trial was to investigate the effect on child health outcomes of a health professional training programme that provided evidence-based healthy feeding recommendations to new mothers (randomisation at health centre level). As reported earlier, the intervention was associated with infant feeding habits closer to guideline recommendations<sup>14</sup> but there was no statistically significant reduction in tooth decay among children of mothers recruited from intervention health centres<sup>15</sup>.

From April 2008 to December 2008, 736 pregnant women (exclusion criteria: human immunodeficiency virus positive) were invited to participate in the trial, and 715 provided written informed consent and were enrolled. A team of trained and calibrated researchers administered standardised maternal questionnaires during pregnancy and at child ages 6, 12 and 38 months. A dental examination was completed for children who remained in the cohort at 38 months (n = 458), with no statistically significant difference between the children who entered the study and who were examined, with the exception of maternal age (mothers lost to follow-up were 1.3 years younger, on average). The present analysis is based on the subsample of children who had both a completed dental examination and two 24-hour recalls recorded on separate days at the 12-month visit (n = 345). Based on confidence intervals set at 95%, a 2:1 ratio of 'unexposed' children (lower feeding frequency) to 'exposed' (higher feeding frequency), and an expected prevalence of ECC among unexposed children of 38%, this study would have 80% power to detect a risk ratio of 1.42, comparable with association strengths reported in similar populations<sup>16</sup>.

#### Sociodemographic variables

Field researchers collected sociodemographic data during pregnancy, including: maternal age and education (years of study); family structure (nuclear, children living with father and mother, *vs.* non-nuclear); family income (monthly from all sources, later categorised as above or below three times the minimum wage); and social class ('A' through 'E,' where 'A' indicates higher class, based on Brazilian Association of Economic Research Institutes classification). Anthropometric data at birth (weight and length) were obtained from the child's birth identification card. Anthropometric data at 6, 12 and 38 months (weight, height, body mass index) were obtained during home visits according to standard protocols and converted into standard World Health Organization (WHO) Z-scores<sup>17</sup>.

#### Infant feeding habits

Feeding habits at the 12-month visit were assessed via two 24-hour dietary recalls using the multiple pass method and were conducted via face-to-face interviews with mothers on random days of the week, at least 7 days apart. Mothers were asked to recall all breastfeeding episodes and foods and drinks consumed by the child the day before, including details about the food types, quantities, brands and preparation methods. Recall of portion sizes was aided using food photographs compiled specifically for research. A detailed review of all reported items was made at the end of each interview to correct errors or omissions.

The mean daily frequencies of breastfeeding and baby-bottle use (including all liquids, such as milk, water, juice, soda or tea) over the two recalls were each later categorised as 0, 1–3 and >3. The three times/day cut-off point was based partly on dietary guidelines that recommend two servings of milk per day for children 1–3 years of age<sup>18</sup>. Among children breastfed more than three times/day, the mean  $\pm$  standard deviation number of daily breastfeeding episodes was 9.6  $\pm$  5.5 (median = 7.5). As a sensitivity check, we repeated the main analyses using a cut-off point of more than five times/day to define the highest breastfeeding frequency category.

The mean daily frequency of all other foods and drinks was categorised as  $\leq 5$  and >5, based on the Ten Steps to Healthy Feeding, a Pan American Health Organization guideline, which recommends five meals a day from age 6 months<sup>19</sup>. Total daily feeding frequency was calculated as the sum of the average number of daily episodes of breastfeeding, bottle use and food or drink consumption, subsequently categorised into quintiles.

#### **Dental examinations**

At the 38-month home visit, one of two trained and calibrated dentists, masked to the child's infant feeding habits, completed dental examinations according to WHO guidelines, modified to record non-cavitated  $(d_1)$ lesions<sup>15</sup>. Teeth were brushed, dried with gauze and visually evaluated with the aid of a mirror-mounted intra-oral light. Tooth-level inter-examiner kappa was 0.75, and intra-examiner kappa was 0.83 for both examiners<sup>15</sup>. The number of decayed (white spot or cavity), missing from decay and filled teeth (d<sub>1</sub>mft) and surfaces (d<sub>1</sub>mfs) were calculated for each child. ECC was considered present when there was at least one caries-affected surface ( $d_1$ mfs  $\geq 1$ ), while S-ECC was defined as the presence of one or more affected maxillary incisors,  $d_1$ mfs  $\geq 4$  or, for children <36 months of age, one or more affected smooth surfaces<sup>20</sup>.

#### Data analysis

The prevalence of ECC and S-ECC in children at 38 months of age was compared according to frequency of feeding at 12 months (breastfeeding, bottle-use and other food/drink consumption) using chi-square tests. Considering the strong negative correlation between the frequency of breastfeeding and bottle-feeding (Spearman r: -0.717; P < 0.001), a variable was created with four mutually exclusive categories for breastfeeding and bottle-use frequency (*Figure 1*): highfrequency breastfeeding (breastfeeding more than

		Daily bottle-feeding frequency						
		>3	1 – 3	0				
Daily breastfeeding frequency	>3	<i>n</i> = 9	n = 45	n = 87				
	1 – 3	n = 7	<i>n</i> = 16	<i>n</i> = 10				
	0	<i>n</i> = 101	n = 68	n = 2				
High-frequency bottle-feeding only High-frequency breastfeeding only Moderate high frequency mixed feeding								

High-frequency breastfeeding only Moderate/high frequency mixed-feeding Low-frequency bottle or breastfeeding

*Figure 1.* Categories of breastfeeding and bottle-use frequency at 12 months of age. Children (n = 345) were classified into four mutually exclusive categories of daily breastfeeding and bottle-use frequency, based on two maternal-reported 24-hour dietary recalls at the time of the 12-month home visit.

three times/day; bottle = 0 times/day); high-frequency bottle-use (bottle more than three times/day; no breastfeeding); moderate/high-frequency mixed-feeding (both breastfeeding and bottle-feeding once or more times/ day); and low-frequency exclusive feeding (no breastfeeding; bottle three times/day or fewer or no bottle; breastfeeding three times/day or fewer). Poisson regression with robust variance in SPSS software was used to obtain unadjusted and multivariable adjusted risk ratios (RR) and 95% confidence intervals (95% CI) for ECC and S-ECC in children at 38 months of age, according to categories of feeding frequency at 12 months of age. We refer to 'risk ratios' because caries prevalence at age 38 months reflects the cumulative incidence of caries from infancy to follow-up. Adjustment variables included plausible confounders: child age; sex of child; maternal age; maternal education; household social class; and allocation status in the nesting trial. Total carbohydrate intake was also included in models to estimate the associations with feeding frequency, independent of total sugar and starch consumption. Unadjusted and adjusted RRs and d1mft ratios were similarly obtained according to quintile of total feeding frequencies. Ratios were considered statistically significant if the 95% CI excluded the null value (ratio: 1.00).

#### **Ethical considerations**

Internal Review Boards at the Federal University of Health Sciences of Porto Alegre (UFCSPA) and the

University of California Berkeley (with reliance at the University of California San Francisco) approved the present study, which was conducted following the ethical guidelines of the Declaration of Helsinki. All mothers provided written informed consent for child participation at both enrolment and the 3-year visit. Children with dental caries, anaemia, obesity or who were underweight were referred for care at the nearest municipal health centre (UBS).

#### RESULTS

The characteristics of the analytic sample (n = 345)were similar to those of all children examined at the 38-month visit (Table 1). Half (50%) of the children in the analytic sample were female, with a mean  $\pm$ SD age of  $38.1 \pm 2.4$  months. Nearly half of the mothers had attained fewer than 9 years of formal education, approximately half the children lived in nuclear families and over two-thirds of households reported family income lower than three times the minimum wage (Table 1). Overall, the prevalence of ECC and S-ECC at the 38-month visit was 54.8% 32.5% (189/345)and (112/345),respectively (Table 1).

Approximately half of the children (174/345; 50.4%) were breastfeeding at the time of the 12month visit, while 71.3% (246/345) used bottles. *Table 2* shows the average daily frequencies of breastfeeding, bottle-feeding, other food/drink intake and total (combined) feeding episodes obtained from the two, 24-hour dietary recalls. Breastfeeding featured the greatest variation feeding frequency: while nearly half the children were no longer breastfeeding at 12 months, 15.1% (52/345) of children were breastfed  $\geq 10$  times/day. The daily frequency of breastfeeding and bottle-feeding were negatively correlated: high-frequency breastfeeding was least common among children bottle-fed more than three times/day, and vice versa. Only two children in the sample were neither bottle-fed nor breastfed at the 12-month visit.

Relative to the low/moderate-frequency exclusive feeding group, caries prevalence was statistically significantly higher in all other feeding bottle-use and breastfeeding frequency groups (Table 3). In multivariable adjusted models, compared with the reference of low/moderate-frequency feeding at 12 months, the 38-month prevalence of ECC was 1.8-fold greater with high-frequency exclusive breastfeeding, 1.4-fold greater with high-frequency exclusive bottle-feeding and 1.5-fold greater with high/moderate-frequency mixed feeding (Table 3). ECC prevalence was not associated with greater frequency of other foods in adjusted models (Table 3), although S-ECC prevalence was 1.7-fold higher (Table 4). S-ECC prevalence was elevated approximately two-fold in the high-frequency exclusive breastfeeding group (Table 4). Although not statistically significant for both ECC and S-ECC in all feeding groups, the adjusted results were consistent with a pattern of increased caries prevalence in early childhood among children whose

Table 1 Sociodemographic characteristics, feeding behaviours and dental-caries status of the study population

Variables	3-year	follow-up sample $(n = 458)$	Analytic sample $(n = 345)$		
	<i>n</i> *	Value	<i>n</i> *	Value	
Sociodemographic characteristics					
Male child	458	233 (50.9)	345	172 (49.9)	
Age (in months) at 3-year visit	458	$38.1 \pm 2.4$	345	$38.1 \pm 2.4$	
Maternal age (in years) at delivery date	458	$26.4 \pm 6.7$	345	$26.2\pm6.8$	
Mother has <9 years of formal education	458	214 (46.7)	345	156 (45.2)	
Household income <three minimum="" salary<sup="" times="">†</three>	444	304 (68.5)	338	232 (68.6)	
Family composition is nuclear	458	226 (49.3)	345	170 (49.3)	
Anthropometry					
Birth weight <2,500 g	444	30 (6.8)	340	25 (7.4)	
Length at birth <48 cm	435	110 (25.3)	337	84 (24.9)	
Length-for-age Z-score at 12 months	409	$-0.04 \pm 0.92$	344	$-0.05 \pm 0.90$	
Weight-for-length Z-score at 12 months	408	$0.93 \pm 1.00$	344	$0.92 \pm 1.00$	
BMI Z-score >2 at 12 months	408	67 (16.4)	344	54 (15.7)	
Feeding habits		× ,		. ,	
Exclusive breastfeeding duration <4 months	439	331 (75.4)	339	255 (75.2)	
Sugar introduction before age 6 months	408	216 (52.9)	344	177 (51.2)	
Dental caries experience at age 38 months		X ,		( )	
Early childhood caries	458	250 (54.6)	345	189 (54.8)	
Severe early childhood caries	458	157 (34.3)	345	112 (32.5)	
d <sub>1</sub> mft	458	$2.1 \pm 3.0$	345	$2.1 \pm 3.0$	

Values are given as n (%) or mean  $\pm$  standard deviation.

BMI, body mass index; d<sub>1</sub>mft, decayed (including non-cavitated lesions), missing, filled (restored), primary tooth index.

\*Sample size for some variables may be less than the total due to missing values.

<sup>†</sup>Monthly income of <1,245 Brazilian reais; approximately US \$750 in 2008.

Table 2 Daily frequencies of breastfeeding, bottle-use and intake of other food or drink, and the sum of all feeding episodes in children at 12 months of age

Feeding practice	Daily frequency						
	Range	$Mean \pm SD$	Median (Q1–Q3)				
Breastfeeding Bottle feeding Other food/drink Total	$0-21.0 \\ 0-7.5 \\ 0-10.5 \\ 4.0-28.0$	$\begin{array}{c} 4.1 \pm 5.8 \\ 2.3 \pm 1.8 \\ 4.6 \pm 1.5 \\ 11.0 \pm 5.1 \end{array}$	1.0 (0-6.5) 2.5 (0.5-3.5) 4.5 (3.5-5.5) 9.0 (7.5-12.5)				

Q1–Q3, interquartile range; SD, standard deviation.

mothers reported higher frequencies of breastfeeding, bottle-feeding or both at 12 months.

Caries prevalence (ECC and S-ECC) and experience (d<sub>1</sub>mft) at 38 months were higher over rising quintiles of total 12-month feeding episodes (breastfeeding, bottle-use and other foods/drinks combined) (Table 5). In adjusted models, S-ECC prevalence and d1mft were about two times greater in the second and third quintiles of total feeding frequency compared with the first, and were three to four times greater in the fourth and fifth quintiles (Table 5). Notably, most of the variation in the number of total daily feeding episodes was the result of daily breastfeeding frequency. Therefore, in an exploratory step, we repeated this analysis with breastfeeding events excluded from the calculation of 12-month feeding episode quintiles (but included breastfeeding frequency as an adjustment variable in regression models). In this exploratory analysis, S-ECC prevalence was about 1.5-times greater in the fourth and fifth quintiles of bottle-use and other food/drink frequency compared with the first quintile (not statistically significant), and there was no meaningful or statistically significant associations with ECC or d<sub>1</sub>mft.

A sensitivity analysis was performed that included all children with at least one completed 24-hour recall (n = 400; 55 additional children). There was very little difference in the estimated measures of association. For ECC, compared with a reference of lowfrequency bottle or breastfeeding, the RRs for high-frequency exclusive breastfeeding (RR = 1.86; 95% CI: 1.36-2.55), high-frequency exclusive bottlefeeding (RR = 1.36; 95% CI: 1.01-1.83) and moderate/high-frequency mixed-feeding (RR = 1.50; 95%CI: 1.11-2.03) were nearly identical to values from the smaller analytic sample, as was the case for S-ECC, which also featured similar RRs for highfrequency exclusive breastfeeding (RR = 1.98; 95%CI: 1.27–3.08), high-frequency exclusive bottlefeeding (RR = 1.13; 95% CI: 0.73-1.74) and high/ moderate-frequency mixed feeding (RR = 1.40; 95%) CI: 0.90-2.12). Likewise, estimated ratios for frequency of other food intake and for quintiles of total feeding frequency did not differ meaningfully in the sensitivity analysis.

In the sensitivity analysis that used a cut-off point of more than five breastfeeding episodes/day to define the highest frequency category, the magnitude of the associations between higher frequency feeding and dental caries were attenuated but maintained statistical significance in the high-frequency breastfeedingonly category. Specifically, for S-ECC, compared with a reference of low-frequency bottle or breastfeeding, adjusted RRs were 2.16 (95% CI: 1.36–3.43) for high-frequency exclusive breastfeeding, 1.01 (95% CI: 0.63–1.61) for high-frequency exclusive bottle-feeding and 1.22 (95% CI: 0.75–1.98) for moderate/high-frequency mixed-feeding.

#### DISCUSSION

In the present evaluation of early-life feeding habits and dental caries experience of a birth cohort of Brazilian children, more frequent feeding at age 12 months, which included more frequent episodes of daily bottle-use, breastfeeding and intake of other

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Variable	п	ECC	Unadjusted		Adjusted*	
		n (%)	RR (95% CI)	Р	RR (95% CI)	Р
Breast and bottle feeding						
Low-frequency bottle or breastfeeding	80	30 (37.5)	Reference		Reference	
Moderate/high frequency mixed-feeding	77	43 (55.8)	1.50(1.05 - 2.10)	0.02	1.45 (1.02-2.07)	0.04
High-frequency bottle-use only	101	57 (56.4)	1.51 (1.08-2.09)	0.02	1.37 (0.98–1.92)	0.07
High-frequency breastfeeding only	87	59 (67.8)	1.81 (1.32-2.48)	< 0.001	1.82 (1.28-2.57)	0.001
Other foods/drinks		. ,	× ,		. ,	
≤5 times/day	252	133 (52.8)	Reference		Reference	
>5 times/day	93	56 (60.2)	1.14 (0.93–1.40)	0.20	$1.19\ (0.97 - 1.45)$	0.10

Table 3 Early childhood caries (ECC) prevalence (in children at 38 months of age), according to daily frequency of breastfeeding, bottle-use and consumption of other foods when the children were 12 months of age

95% CI, 95% confidence interval; ECC, early childhood caries; RR, risk ratio.

\*Adjusted for child age, child sex, maternal age, maternal education, household social class, allocation status in the nesting trial, and total carbohydrate intake.

#### Feldens et al.

Variable	n	S-ECC	Unadjusted		Adjusted*	
		n (%)	RR (95% CI)	Р	RR (95% CI)	Р
Breast and bottle feeding						
Low-frequency bottle or breastfeeding	80	18 (22.5)	Reference		Reference	
Moderate/high frequency mixed-feeding	77	23 (29.9)	1.33 (0.78-2.26)	0.30	1.43 (0.84-2.42)	0.18
High-frequency bottle-use only	101	29 (28.7)	1.28(0.77-2.12)	0.35	1.18(0.71 - 1.97)	0.53
High-frequency breastfeeding only	87	42 (48.3)	2.15 (1.35-3.41)	0.001	2.43 (1.49-3.96)	< 0.001
Other foods/drinks						
≤5 times/day	252	72 (28.6)	Reference		Reference	
>5 times/day	93	40 (43.0)	1.51 (1.10-2.04)	0.01	1.66 (1.23-2.25)	0.001

 Table 4 Severe early childhood caries (S-ECC) prevalence (in children at 38 months of age), according to daily frequency of breastfeeding, bottle-use and consumption of other foods when the children were 12 months of age

95% CI, 95% confidence interval; RR, risk ratio; S-ECC, severe early childhood caries.

\*Adjusted for child age, child sex, maternal age, maternal education, household social class, allocation status in the nesting trial and total carbohydrate intake.

 Table 5 Child dental status (in children at 38 months of age), according to quintiles of total daily feeding episodes in children at 12 months of age

Total feeding frequency	п	Mea	n daily feeding	; episodes	ECC	Unadjusted	Adjusted*
		Breastfeeding	Bottle use	Other food/drink	n (%)	RR (95% CI)	RR (95% CI)
1st quintile	55	0.16	2.76	3.37	22 (40.0)	Reference	Reference
2nd quintile	90	0.62	3.12	4.22	39 (43.3)	1.08 (0.73-1.62)	1.09 (0.73-1.62)
3rd quintile	62	1.56	2.92	4.93	39 (62.9)	1.57 (1.08-2.29)	1.62 (1.11-2.35)
4th quintile	72	4.72	1.76	5.37	43 (59.7)	1.49 (1.03-2.17)	1.50 (1.03-2.18)
5th quintile	66	13.87	1.06	4.83	46 (69.7)	1.74 (1.21–2.50)	1.75 (1.21–2.52)
Total feeding frequency	n	Mean daily feeding episodes			S-ECC	Unadjusted	Adjusted*
		Breastfeeding	Bottle use	Other food/drink	n (%)	RR (95% CI)	RR (95% CI)
1st quintile	55	0.16	2.76	3.37	7 (12.7)	Reference	Reference
2nd quintile	90	0.62	3.12	4.22	26 (28.9)	2.27 (1.06-4.88)	2.44 (1.13-5.27)
3rd quintile	62	1.56	2.92	4.93	18 (29.0)	2.28 (1.03-5.05)	2.62 (1.19-5.77)
4th quintile	72	4.72	1.76	5.37	31 (43.1)	3.38 (1.61-7.11)	3.79 (1.80-7.97)
5th quintile	66	13.87	1.06	4.83	30 (45.5)	3.57 (1.70-7.50)	3.94 (1.84-8.44)
Total feeding frequency	п	Mean	Mean daily feeding episodes			Unadjusted	Adjusted*
		Breast-feeding	Bottle-use	Other food/drink	$Mean \pm SD$	RR (95% CI)	RR (95% CI)
1st quintile	55	0.16	2.76	3.37	$0.84 \pm 1.3$	Reference	Reference
2nd quintile	90	0.62	3.12	4.22	$1.61 \pm 2.5$	1.93 (1.13-3.28)	1.96 (1.13-3.40)
3rd quintile	62	1.56	2.92	4.93	$1.77 \pm 2.1$	2.12 (1.27-3.55)	2.30 (1.35-3.91)
4th quintile	72	4.72	1.76	5.37	$2.94 \pm 3.9$	3.52 (2.09-5.92)	3.57 (2.09-6.10)
5th quintile	66	13.87	1.06	4.83	$3.02\pm3.6$	3.61 (2.17-6.00)	3.52 (2.07-6.00)

95% CI, 95% confidence interval; d1mft, decayed (including non-cavitated lesions), missing, filled (restored), primary tooth index ECC, early childhood caries; RR, risk ratio; S-ECC, severe early childhood caries; SD, standard deviation.

\*Adjusted for child age, sex of child, maternal age, maternal education, household social class, allocation status in the nesting trial and total carbohydrate intake.

foods and/or drinks, was associated with a higher prevalence of dental caries approximately 2 years later. The results suggest that identifiable patterns early in life may influence caries formation as one potential contributor among a multifactorial aetiology that includes fluoride exposure and hygiene behaviours, possibly by setting the stage for dietary habits at later ages. Notably, associations persisted after adjustment for total intake of carbohydrates, suggesting that feeding frequency may have a role in caries development, independent of the amount of potentially cariogenic foods consumed. The identification of feeding-related risk-indicators at such an early age suggests that efforts to prevent caries in children via diet modification may be most effective if beginning early in life, typically before school age.

Infant feeding (baby) bottles are widely used in Brazil<sup>16</sup>, and their use, particularly for non-milk contents and at night, has been associated with dental caries in investigations worldwide<sup>16,21,22</sup>. Bottles delivering sugar-containing liquids or used over long periods during the day or night exposes the tooth surface to acidic conditions for lengthy periods of time. Bottle use and breastfeeding may be inversely correlated, as high-frequency breastfeeding has been observed in children who do not use the bottle<sup>13,16,23</sup>. A strength of the present study was to consider separate categories of high-frequency breastfeeding, bottle-use or both, all of which were associated with higher ECC prevalence than using both infrequently.

High daily frequency of breastfeeding was associated with later ECC and S-ECC, including after adjustment for total carbohydrate intake and stratification for bottle-use frequency, consistent with findings from other studies in diverse settings<sup>13,24</sup>. A systematic review found that the association of breastfeeding beyond age 12 months was strongest when nursing was frequent or nocturnal<sup>25</sup>. Similarly, a recent study reported no overall association between breastfeeding duration and caries among Thai children, but did report a positive dose-response relationship between caries and frequency of nocturnal bottleuse or breastfeeding<sup>26</sup>. Importantly, the cut-off point of more than three daily breastfeeding episodes to define 'high' frequency was not intended to imply a threshold on which to base nutritional or dental health recommendations; some children in this category were breastfed many more times per day. Under a sensitivity analysis using a cut-off point of more than five daily breastfeeding episodes to define highfrequency breastfeeding, a statistically significant positive association with dental caries was maintained.

Given the undisputed and critical role of breastfeeding in assuring maternal and infant health<sup>27</sup>, dental professionals should encourage mothers to initiate exclusive breastfeeding from birth, followed by continued breastfeeding as complementary foods are added to the diet. Promotion of exclusive breastfeeding early in life does not increase caries risk<sup>28,29</sup>. However, the results of the present study and others<sup>25</sup> suggest that frequent breastfeeding at later ages might have negative implications for dental health.

A higher daily frequency of consuming other foods and drinks at age 12 months was also associated with childhood tooth decay in this population. Of note, frequent food intake in very young children has been associated with the consumption of 'non-foods', which are foods with low-nutrient but high-energy density, such as soft drinks, cookies and other sweets<sup>3</sup>. These foods may displace more nutritious items from the diet<sup>30</sup>, with potential consequences for tooth decay and obesity. Reminiscent of the conclusions of the Vipeholm trial<sup>31</sup>, that sugar consumption frequency was a better predictor of caries risk than total amount consumed, here, associations with daily food intake were robust to adjustment for total daily carbohydrate intake. However, this finding does not total sugar consumption in caries exonerate

development. Recent evidence suggests a stronger association in adults between caries experience and amount of sugar consumed than sugar consumption frequency<sup>32</sup>.

The associations reported in the present study were prospective and adjusted for plausible confounding variables; however, any causal mechanisms are likely to include the interplay of several related feeding behaviours. For instance, feeding frequency and/or sweet snack consumption in infancy may be maintained as continued feeding habits, and these later habits serve as the proximate causes of tooth decay in childhood. Likewise, in vitro studies report that the potential cariogenicity of breast milk is enhanced with the addition of outside sugars<sup>33</sup>, suggesting the possibility that in children, interaction with dietary sugars may influence whether breastfeeding will be caries promoting. Regardless of the mechanistic pathway, early-life behaviours appear to be important indicators of children at high caries risk.

Among the limitations of this study, a sizeable proportion of the sample was ineligible for analysis as a result of incomplete dietary recalls or loss to followup, which is common in longitudinal studies in communities of lower socio-economic status. However, the analytic sample and the originally enrolled cohort were similar in the characteristics measured, and the results were largely unchanged in a sensitivity analysis that included additional participants with only a single 24-hour recall, which together suggest a low probability of selection bias. Imperfect measurement is unavoidable in collecting dietary information from participants, but all fieldworkers were extensively trained and calibrated to follow a standard protocol for ensuring data accuracy. The prospective birthcohort design was a significant study strength, along with the ability to record detailed daily feeding histories through 24-hour recalls. The use of repeated recalls helps to account for daily intraperson variability. However, as a limitation, no more than two recalls were completed with each participant. This protocol is used in the US National Health and Nutrition Examination Survey<sup>34</sup>. Completing a larger number of 24-hour recalls or using multiple-day dietary food records might have yielded greater accuracy, but both of these alternatives would have increased participant burden.

A key future direction for dental professionals and other health practitioners will be how to incorporate the possible cariogenicity of frequent infant feeding into sound recommendations for parents and families. To meet children's nutritional needs, Brazilian guidelines recommend up to five meals a day for infants and young children who are no longer breastfeeding<sup>19</sup>. The period of transition from exclusive breastfeeding to a complementary diet that begins to incorporate other foods is a critical moment in infant development and a time of heightening risk for nutritional deficiencies<sup>27</sup>. As evidence of caries risk accumulates for frequent feeding, in particular, frequent or nocturnal breastfeeding beyond age 1–2 years<sup>25</sup>, it will be critical to take an evidence-based and balanced approach to integrating dental considerations into established best practices for child nutrition promotion. Caries prevention is far from the primary concern in developing infant-feeding guidelines, but, as the most common chronic childhood disease worldwide<sup>35</sup>, it is worthy of place among other health and nutritional considerations.

In this population, there was a prospective association between the total daily feeding frequency at 12 months of age, including high-frequency breastfeeding and bottle-use, and dental caries status at age 3 years. These findings suggest that dental caries prevention could be accomplished by emphasising that the breastfed or bottle-fed child be nursed at less frequent intervals once nutritionally sufficient complementary foods have been introduced. Limiting the frequency of other snacks or drinks may also result in caries prevention. Importantly, any feeding advice from dental professionals and other health workers must be aligned with the complete nutritional needs of the growing child and with caregivers' beliefs and values.

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#### **Conflicts of interest**

The authors declare that they have no conflicts of interest related to this research.

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