Caries and Its Association with Infant Feeding and Oral Health-related Behavior in 18-month and 3-year-old Japanese Children

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Abstract

The primary aim of this study was to determine the prevalence of dental caries and its association with infant feeding and oral health-related behavior in Japanese children between the ages of 18 months and 3 years. A total of 387 infants were initially examined at the age of 18 months (or 19 months in some cases) and then again at 3 years (or at 3 years 1 month in some cases). The primary objective was to identify factors contributing to the prevalence of caries in children aged 18 months. The secondary objective was to follow up children with no caries at the age of 18 months to seek potential correlations between background factors and the increment of caries by the age of 3 years. In an adjusted multivariate logistic regression model, children who continued breastfeeding were approximately 7 times more likely to have dental caries at 18 months of age than those who did not. Infants brought in for an oral examination and consultation at around 12 months of age were less likely to develop dental caries at 18 months of age than those who were not. Multivariate logistic regression analysis revealed that infants who did not receive a dental check-up at 12 months of age showed a significantly higher incidence of dental caries at 3 years of age. The present results suggest that prolonged breastfeeding is a risk factor for early childhood caries at 18 months of age, and that infants in whom a regular oral care program is implemented from the age of 12 months are less likely to develop early childhood caries at 18 months or 3 years.

Key words: Early childhood caries — Risk factor — Japanese children — Infant feeding — Oral health behavior
Introduction

National data have revealed a trend towards a decrease in the prevalence of dental caries in the primary dentition over the last decade in Japan.\(^{15}\) Yonezu \textit{et al.}\(^{41,42}\) also reported a decline in dental caries in Japanese children aged 18 months, 2 years, or 3 years. This improvement may be attributable to the preventive dental intervention program promoted among children aged between 18 months and 3 years under the Maternal and Child Health Act. The prevalence of caries in Japanese children is still significantly higher than that seen in developed countries in the west, however.\(^{13,25,32}\)

Caries in early childhood is a multi-factorial disease. These factors include a susceptible host, a fermentable carbohydrate intake, dietary practice, the presence of dental plaque, and a high number of cariogenic microorganisms such as \textit{Mutans streptococci} (MS).\(^{23}\) Early childhood caries (ECC) has also been associated with demographic characteristics such as oral hygiene practices and early childhood feeding practices.\(^{22,24,26–38}\) Yonezu \textit{et al.}\(^{43}\) found that children who were breast-fed at 18 months of age were several times more likely to have dental caries at 2 years of age than those who were bottle-fed. However, their study did not answer the question of why prolonged breast-feeding was correlated with more dental caries. In addition, scientific evidence regarding the association between breastfeeding and ECC is somewhat mixed. Although some studies suggest that prolonged breastfeeding indicates an increased risk of dental caries in the primary dentition,\(^{2,5,7,10,14,20,28,34,35,43}\) several others\(^{4,8,11,16,18,21}\) have observed no association between breastfeeding and ECC.

The World Health Organization\(^{39}\) and the Japanese Pediatric Society\(^{1}\) have recommended exclusively breastfeeding for at least 6 months and complementary breastfeeding up to 24 months of age due to various maternal and child health benefits. Thereafter, a trend towards longer breastfeeding has been reported in many western countries,\(^{3}\) and also in Japan. It is therefore important to identify which factors are associated with the development of dental caries in breast-fed children as soon as possible if effective preventive programs are to be developed. Further research with careful control of persistent confounding factors, including cariogenic food/drinks in the diet, inadequate oral hygiene practices, and occlusal characteristics is needed to elucidate this issue and better inform infant feeding guidelines. Therefore, it is of great importance to begin implementing preventive measures at a very young age. Information on individual risk and specific factors to be addressed in developing preventive programs for the very young is scarce, however.

The primary objective of this study was to identify which factors contribute to the prevalence of caries in children aged 18 months. The secondary objective was to follow up children with no caries at the age of 18 months to seek potential correlations between background factors and the increment of caries by the age of 3 years.

Materials and Methods

1. Participants

This was a prospective study starting with infants aged 18 months (including a few aged 19 months). This cohort was born between 2010 and 2011. All were brought to a public health center in “K” city in Tokyo for regular dental examinations.

There is no water fluoridation in this region. A total of 414 children receiving oral examinations were followed up until the age of 3 years (including some aged 3 years and 1 month). All the children were Japanese and in good general health, and all showed age-appropriate cognitive development.

2. Study methods

1) Behavioral and dietary survey

Prior to the examinations, the parents or guardians of the children received a letter containing information about the survey in
which they were asked to give consent for an oral examination of their children; they were also requested to complete a 22-item questionnaire addressing infant feeding practices, dietary history, snack food frequency, oral hygiene practices, and demographic information. Each of these factors was included based on previous studies by this group and similar research indicating their association with dental caries. The main items covered were as follows:

1. Sex (male/female)
2. Breast- or bottle-feeding at the time of oral examination (Yes or No)
3. Birth weight (g) and height (cm)
4. Finger and/or pacifier habit (Yes or No)
5. Frequency of sweet snack intake (times per week)
6. Frequency of sweet beverage intake (times per week)
7. Frequency of snacks between meals (times per day)
8. Dental check-up at 12 months of age (Yes or No)
9. Frequency of tooth brushing by parents or guardians (times per day)
10. Application of topical fluoride (Yes or No)
11. Presence of sibling in home (Yes or No)
12. Presence of grandfather and/or grandmother in home or cohabitation with grandparents (Yes or No)
13. Attending nursery school (Yes or No)
14. Wake-up time and bedtime
15. Time of night-time meal

3. Clinical oral examinations
1) Assessment of dental caries

Caries examinations and assessments of visible plaque were conducted by a single examiner trained in pediatric dentistry. The examinations were performed with the patient in a supine position on the dental chair. A dental light, mirror, and explorer were used, with the examinations being primarily visual. Cotton roll was used for moisture control and a cotton swab for removal of plaque on tooth surfaces when necessary.Decayed and filled surfaces were recorded. The criteria for cavitated lesions were either demonstrable loss of enamel structure upon visual examination or softness at the base of lesion in response to the explorer. An area of distinct chalky white enamel on a smooth surface or adjacent to a pit or fissure site, with no loss of enamel, was required for determination of a non-cavitated lesion.

The tooth was considered present if any part of the clinical crown was exposed in the oral cavity. The dental surfaces were also examined to verify the presence of structural defects in the enamel. Hypoplasia of the enamel was defined as a break in its continuity due to a reduction in its thickness. Opacity was defined as a change in the translucence of the enamel, or a qualitative defect that could vary in color from white to yellow and brown.

2) Caries activity test

The same examiner carried out a caries activity test in children aged 18 months using the CAT 21 test kit (Cariostat method, Willdent, Co., Osaka, Japan) to predict their risk for future development of caries. The Cariostat test is widely used in many clinical, epidemiological, and fieldwork-related studies in Japan.

A dental plaque sample collected from the labial surface of the maxillary incisors with a sterile cotton swab was then inoculated into test medium and incubated at 37°C for 48 hr. Cariostat scores indicate acid production by oral bacteria in plaque through comparison against a 4-level color scale (0, 1.0, 2.0, and 3.0) that corresponds to a pH value of 6.1 ± 0.3, 5.4 ± 0.3, 4.9 ± 0.3, and 4.7 ± 0.3, respectively.

3) Assessment of occlusal characteristics

Visual examination of occlusal characteristics was carried out by the same examiner, who recorded the presence of open bite, excessive over jet, deep bite, anterior cross bite, posterior cross bite, or crowding of incisors.

The diagnostic criteria selected were adopted from an earlier study conducted by Yonezu et al.40.
4. Study outcome

To understand why there is a large difference in the incidence of dental caries between 18 months and 3 years is an important public health issue. The primary objective of this study was to identify which factors contribute to the incidence of caries in children aged 18 months. The secondary objective was to follow up children with no caries at the age of 18 months to determine potential correlations between background factors and the increment of caries at the age of 3 years.

5. Statistical analysis

Statistical analysis was performed using the JMP software (version 10.0.2; SAS Institute Inc., NC, USA). Bivariate analyses of the association between dental caries and covariates were performed using the Chi-square and Fisher’s exact test. A value of p<0.05 was considered as statistically significant. Stepwise backward logistic regression served for the multivariable assessment of factors related to dental caries. The corresponding odds ratios (OR) and their 95% confidence intervals (95%CI) were determined.

6. Ethical considerations

The protocol of this study was approved by the Ethics Committee of Tokyo Dental College (Approval No.651). The mothers or guardian were asked to provide informed consent for participation in the study. The data on the identity of each infant/child were entered into the database as a numerical code only.

Results

1. Risk factors contributing to prevalence of caries in children aged 18 months

Twenty-seven of the 414 infants examined were excluded from analysis as the principal investigation procedures were not completed in this group. Therefore, the data used for the analysis were restricted to those on the remaining 387 children.

The overall prevalence of caries in these 387 infants was 7.49% at 18 months of age. Dental caries was more common in boys (9.5%) than in girls (5.35%), although this difference was not statistically significant.

Among all the variables listed and analyzed in Table 1–4, only the following factors were found to be associated with the prevalence of caries (p<0.05): (1) visible plaque at oral examination; (2) Cariostat score, ≥v2; (3)
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breastfeeding continued at 18 months of age; (4) breast and/or bottle feeding at bed time; (5) sweets intake, \( \geq 4 \) times/week; (6) frequency of snacks between meals, \( \geq \) twice/day; (7) frequency of tooth brushing by parents, \(< \) twice/day; and (8) no experience of dental check-up at 12 months of age.

Among the 114 infants who continued breastfeeding, 20 (14.9%) presented with dental caries, whereas among those who ceased breastfeeding, this was only 3.6%. In addition, the prevalence of caries was significantly higher (14.6%) in children who were breast- and/or bottle-fed at night than in those who were not (3.6%).

In the adjusted multivariate logistic regression model (Table 5), it was observed that dental caries was more likely to occur in children who brushed less than once a day; those who ate snacks more than twice a day; those with visible dental plaque on oral examination; and those with a high Cariostat score. In addition, infants who continued breastfeeding were about 7 times more likely to have dental caries than those who did not (OR = 7.1; 95%CI = 2.86–19.46). Infants brought in for an oral examination and consultation at around 12 months of age were less likely to develop dental caries than those who were not.

2. Risk factors contributing to prevalence of caries in children aged 3 years

Seventeen of the 358 children who were caries-free at 18 months of age were excluded from the analysis as all the investigation procedures were not completed in this group. Among the 341 children who remained in the study, 43 (12.6%) developed dental caries at 3 years of age. Dental caries was more common in boys (13.7%) than in girls (11.6%), although this difference was not statistically

| Life style variables                  | Prevalence of dental caries | Total | p-value |
|--------------------------------------|-----------------------------|-------|---------|
|                                      | No n (%)                    | Yes n (%) |       |         |
| Brother and/or sister at home        | 151 (89.9)                  | 17 (10.1) | 168   | NS      |
| Yes                                  | 151 (89.9)                  | 17 (10.1) | 168   | NS      |
| No                                   | 207 (94.5)                  | 12 (5.5)  | 219   |         |
| Grandfather and/or grandmother at home | 29 (90.6)                | 3 (9.4)   | 32    | NS      |
| Yes                                  | 29 (90.6)                   | 3 (9.4)   | 32    | NS      |
| No                                   | 329 (92.7)                  | 26 (7.3)  | 355   |         |
| Attending nursery                    | 113 (93.4)                  | 8 (6.6)   | 121   | NS      |
| Yes                                  | 113 (93.4)                  | 8 (6.6)   | 121   | NS      |
| No                                   | 245 (92.1)                  | 21 (7.9)  | 266   |         |
| Wake-up time                         |                            |          |       |         |
| After 8 a.m.                         | 83 (95.4)                   | 4 (4.6)   | 87    | NS      |
| Before 8 a.m.                        | 275 (91.7)                  | 25 (8.3)  | 300   |         |
| Bedtime                              |                            |          |       |         |
| After 9 p.m.                         | 97 (91.5)                   | 9 (8.5)   | 106   | NS      |
| Before 9 p.m.                        | 261 (92.9)                  | 20 (7.1)  | 281   |         |
| Evening meal                         |                            |          |       |         |
| After 7 p.m.                         | 145 (93.6)                  | 10 (6.5)  | 155   | NS      |
| Before 7 p.m.                        | 213 (91.8)                  | 19 (8.2)  | 232   |         |

Chi-square or Fisher’s exact test used to determine differences among groups. \( p<0.05 \) indicates statistical significance. NS = not significant
Among all the variables listed and analyzed in Table 6–9, a total of 7 factors identified at the age of 18 months were found to be associated with the incidence of caries (p<0.05):

1. less than 50 cm height at birth;
2. Cario-stat score, $\geq 2$;
3. visible plaque on oral examination;
4. wake-up after 8 o’clock;
5. brothers in their family;
6. frequency of tooth brushing by parents, $< 2$ times/day; and
7. frequency of tooth brushing by parents, $\geq 2$ times/day.

### Table 3 Association between oral health behaviors/feeding practices and dental caries at 18 months of age

| Oral health behaviors, feeding practices | Prevalence of dental caries | Total | p-value |
|-----------------------------------------|----------------------------|-------|---------|
|                                         | No n (%) | Yes n (%) |       |
| Sweet snack intake                      |           |           |       |
| $\geq 4$ times/week                     | 93 (86.9) | 14 (13.1) | 107   | 0.01   |
| $< 4$ times/week                        | 265 (94.6)| 15 (5.4)  | 280   |        |
| Sweet beverage intake                   |           |           |       |
| $\geq 4$ times/week                     | 106 (91.4)| 10 (8.6)  | 116   | NS     |
| $< 4$ times/week                        | 251 (95.0)| 19 (5.0)  | 270   |        |
| Frequency of snacks between meals       |           |           |       |
| $\geq$ twice/day                        | 204 (89.5)| 24 (10.5) | 228   | 0.007  |
| $< $ twice/day                          | 154 (96.9)| 5 (3.1)   | 159   |        |
| Breastfeeding at 18 months of age       |           |           |       |
| Yes                                     | 114 (85.1)| 20 (14.6) | 134   | <.0001 |
| No                                      | 244 (96.4)| 9 (3.6)   | 253   |        |
| Bottlefeeding at 18 months of age       |           |           |       |
| Yes                                     | 36 (92.3) | 3 (7.7)   | 39    | NS     |
| No                                      | 322 (92.5)| 26 (7.5)  | 348   |        |
| Breast and/or bottlefeeding in bed      |           |           |       |
| Yes                                     | 117 (85.4)| 20 (14.6) | 137   | <.0001 |
| No                                      | 241 (96.4)| 9 (3.6)   | 250   |        |
| Visible plaque at examination           |           |           |       |
| Yes                                     | 41 (78.9) | 11 (21.2) | 52    | <.0001 |
| No                                      | 317 (94.6)| 18 (5.4)  | 335   |        |
| CAT score                               |           |           |       |
| $\geq 2$                                | 113 (87.6)| 16 (12.4) | 129   | 0.002  |
| $< 2$                                   | 245 (95.0)| 13 (5.0)  | 258   |        |
| Dental check-up at 12 months of age     |           |           |       |
| Yes                                     | 101 (98.1)| 2 (1.9)   | 103   | 0.009  |
| No                                      | 255 (90.4)| 27 (9.6)  | 282   |        |
| Application of topical fluoride         |           |           |       |
| Yes                                     | 61 (95.3) | 3 (4.7)   | 64    | NS     |
| No                                      | 297 (92.0)| 26 (8.1)  | 323   |        |
| Frequency of tooth brushing by parents  |           |           |       |
| $\geq 2$ times/day                      | 101 (98.1)| 1 (1.0)   | 102   | 0.002  |
| $< 2$ times/day                         | 257 (90.2)| 28 (9.8)  | 185   |        |

Chi-square or Fisher’s exact test used to determine differences among groups. p<0.05 indicates statistical significance. NS = not significant.
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### Table 4
Association between oral characteristics and dental caries at 18 months of age

| Oral characteristics variables                          | Prevalence of dental caries | Total | p-value |
|-----------------------------------------------------------|----------------------------|-------|---------|
|                                                           | No n (%) | Yes n (%) |       |         |
| Crowding in maxillary incisors                            |           |           |       |         |
| Yes                                                       | 27 (100) | 0         | 27    | NS      |
| No                                                        | 331 (91.9) | 29 (8.1) | 360   |         |
| Crowding in mandibular incisors                            |           |           |       |         |
| Yes                                                       | 19 (90.5) | 2 (9.5)   | 21    | NS      |
| No                                                        | 339 (92.6) | 27 (7.4) | 366   |         |
| Deep bite                                                 |           |           |       |         |
| Yes                                                       | 30 (96.8) | 1 (3.2)   | 31    | NS      |
| No                                                        | 328 (92.1) | 28 (7.9) | 356   |         |
| Open bite                                                 |           |           |       |         |
| Yes                                                       | 8 (100)   | 0         | 8     | NS      |
| No                                                        | 350 (92.4) | 29 (7.7) | 379   |         |
| Excessive over jet                                        |           |           |       |         |
| Yes                                                       | 20 (100)  | 0         | 20    | NS      |
| No                                                        | 338 (92.1) | 29 (7.9) | 377   |         |
| Anterior cross bite                                       |           |           |       |         |
| Yes                                                       | 45 (88.2) | 6 (11.8)  | 51    | NS      |
| No                                                        | 313 (93.2) | 23 (6.4) | 336   |         |
| Inlocking of incisors                                     |           |           |       |         |
| Yes                                                       | 14 (100)  | 0         | 14    | NS      |
| No                                                        | 344 (92.2) | 29 (7.8) | 373   |         |
| Abnormal position of upper labial frenum                  |           |           |       |         |
| Yes                                                       | 203 (94.4) | 12 (5.6) | 215   | NS      |
| No                                                        | 155 (90.1) | 17 (9.9) | 172   |         |
| Teeth with structural defects in enamel                   |           |           |       |         |
| Yes                                                       | 5 (100)   | 0         | 5     | NS      |
| No                                                        | 353 (92.4) | 29 (7.6) | 382   |         |
| Number of erupted teeth                                   |           |           |       |         |
| <16                                                       | 138 (95.8) | 6 (4.2)  | 144   | NS      |
| ≥16                                                       | 220 (90.5) | 23 (9.5) | 243   |         |
| Digital sucking habit                                     |           |           |       |         |
| Yes                                                       | 72 (97.3) | 2 (2.7)   | 74    | NS      |
| No                                                        | 286 (91.4) | 27 (8.6) | 313   |         |
| Use of pacifier                                           |           |           |       |         |
| Yes                                                       | 14 (100)  | 0         | 14    | NS      |
| No                                                        | 344 (92.2) | 29 (7.8) | 373   |         |

Chi-square or Fisher’s exact test used to determine differences among groups. \( p < 0.05 \) indicates statistical significance. NS = not significant.
Table 5 Multivariable logistic regression model for dental caries risk factors at 18 months of age

|                                    | Adjusted OR | 95% CI         | p-value |
|------------------------------------|-------------|----------------|---------|
| Number of erupted teeth            |             |                |         |
| <16                                | 1           |                |         |
| ≥16                                | 3.41        | 1.264–10.771   | 0.014   |
| Visible plaque at examination      |             |                |         |
| Yes                                | 3.824       | 1.4–10.411     | 0.009   |
| No                                 | 1           |                |         |
| CAT score                          |             |                |         |
| ≥2                                 | 2.862       | 1.133–7.42     | 0.026   |
| <2                                 | 1           |                |         |
| Breastfeeding at 18 months of age  |             |                |         |
| Yes                                | 7.106       | 2.857–19.455   | <.0001  |
| No                                 | 1           |                |         |
| Frequency of meals                 |             |                |         |
| ≥twice/day                         | 3.989       | 1.45–13.248    | 0.006   |
| <twice/day                         | 1           |                |         |
| Frequency of tooth brushing by parents |         |                |         |
| ≥twice/day                         | 1           |                |         |
| <2 twice/day                       | 9.476       | 1.805–175.895  | 0.004   |
| Dental check-up at 12 months of age|             |                |         |
| Yes                                | 1           |                |         |
| No                                 | 3.8         | 1.011–24.888   | 0.048   |

OR = odds ratio; CI = confidence interval. p<0.05 indicates statistical significance.

Table 6 Association between sex/physical birth variables and dental caries at 36 months of age

| Sex and physical variables | Prevalence of dental caries | Total | p-value |
|----------------------------|-----------------------------|-------|---------|
|                            | No n (%) | Yes n (%) |       |
| Sex                        |           |           |       |
| male                       | 145 (86.3)| 23 (13.7) | 168    | NS      |
| female                     | 153 (88.4)| 20 (11.6) | 173    |         |
| Birth weight               |           |           |       |
| <2,500 g                   | 134 (89.9)| 15 (10.1) | 149    | NS      |
| ≥2,500 g                   | 164 (85.4)| 28 (14.6) | 292    |         |
| Birth height               |           |           |       |
| <50 cm                     | 173 (90.6)| 18 (9.4)  | 191    | 0.046   |
| ≥50 cm                     | 125 (83.3)| 25 (16.7) | 150    |         |

Chi-square or Fisher’s exact test used to determine differences among groups. p<0.05 indicates statistical significance. NS = not significant.
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All risk indicators were included in the multivariate logistic model. From these, 6 influenced the incidence of dental caries at 3 years of age (Table 10): less than 50 cm in height at birth (OR = 2.99; 95%CI, 1.42–6.54); frequency of tooth brushing by parents, < twice/day at 18 months of age (OR = 3.02; 95%CI, 1.18–9.06); no dental check-up at 12 months of age (OR = 5.02; 95%CI, 1.65–21.96); broth- ers in their family (OR = 2.66; 95%CI, 1.18–6.36); wake-up after 8 o’clock (OR = 5.34; 95%CI, 2.16–13.77); and visible plaque on oral examination (OR = 2.74; 95%CI, 1.06–6.86). It was observed that dental caries was more likely to occur in infants who continued breast- and/or bottle feeding during sleep at 18 months of age. However, this association was not statistically significant.

Discussion

1. Risk factors contributing to prevalence of caries in children aged 18 months

The figure of 7.49% for those with caries among 18 month-old participants found in the present study was higher than that seen in other similar studies in Japan. In a study published in 2014, Watanabe et al. reported that only 622 children (1.84%) exhibited dental caries among 33,655 children in Kobe city. Nakayama et al. investigated 1,675 children aged 18- to 23-months and reported that the prevalence of dental caries was 3.3%. In these studies, however, the oral examinations were carried out by general dentists in the ‘knee-to-knee’ position by visual inspection under natural light. It is sometimes difficult for general dentists to identify ECC. Therefore, it is possible that the prevalence of caries was underestimated. On the other hand, in this

| Life style variables                              | Prevalence of dental caries | Total | p-value |
|---------------------------------------------------|-----------------------------|-------|---------|
|                                                   | No n (%)                    | Yes n (%) |       |         |
| Brother and/or sister at home                     | 119 (82.6)                  | 25 (17.4) | 244 | 0.024 |
|                                                   | 179 (90.9)                  | 18 (9.1)  | 197 |       |
| Grandfather and/or grandmother at home           | 22 (78.6)                   | 6 (21.4)  | 28  | NS    |
|                                                   | 276 (88.2)                  | 37 (11.8) | 313 |       |
| Attending nursery                                 | 95 (88.8)                   | 12 (11.2) | 107 | NS    |
|                                                   | 203 (86.8)                  | 31 (13.3) | 234 |       |
| Wake-up time                                      | After 8 a.m.                | 59 (77.6) | 76  | 0.004 |
|                                                   | Before 8 a.m.               | 239 (90.2) | 265 |       |
| Bedtime                                           | After 9 p.m.                | 80 (88.9) | 90  | NS    |
|                                                   | Before 9 p.m.               | 218 (86.9) | 251 |       |
| Evening meal                                      | After 7 p.m.                | 118 (86.1) | 137 | NS    |
|                                                   | Before 7 p.m.               | 180 (88.2) | 204 |       |

Chi-square or Fisher’s exact test used to determine differences among groups. p<0.05 indicates statistical significance. NS = not significant.
study, all the examinations were performed by a pediatric dentist using a dental chair, dental light, mirror, and explorer, which allowed a more accurate diagnosis of initial lesions associated with caries.

The early detection of dental caries is very important in preventing its development into a more severe condition. Yonezu et al.\(^1\) reported that children who developed caries at 18 months of age exhibited high caries pro-

Table 8  Association between oral health behaviors/feeding practices and dental caries at 36 months of age

| Oral health behaviors, feeding practices variables | Prevalence of dental caries | Total | p-value |
|-------------------------------------------------|-----------------------------|-------|---------|
|                                                 | No n (%) | Yes n (%) |       |         |
| Sweet snack intake                              |           |           |       |         |
| ≥4 times/week                                   | 75 (86.2) | 12 (13.8) | 87   | NS      |
| <4 times/week                                   | 223 (87.8) | 31 (12.2) | 254  |         |
| Sweet beverage intake                           |           |           |       |         |
| ≥4 times/week                                   | 84 (82.4) | 18 (17.7) | 102  | NS      |
| <4 times/week                                   | 213 (89.5) | 25 (10.5) | 238  |         |
| Frequency of snacks between meals               |           |           |       |         |
| ≥twice/day                                      | 169 (86.7) | 26 (13.3) | 195  | NS      |
| <twice/day                                      | 129 (88.4) | 17 (11.6) | 146  |         |
| Breastfeeding at 18 months of age               |           |           |       |         |
| Yes                                            | 94 (85.5) | 16 (14.6) | 110  | NS      |
| No                                             | 204 (88.3) | 27 (11.7) | 231  |         |
| Bottlefeeding at 18 months of age               |           |           |       |         |
| Yes                                            | 30 (85.7) | 5 (14.3)  | 35   | NS      |
| No                                             | 268 (87.6) | 38 (12.4) | 306  |         |
| Breast and/or bottlefeeding in bed              |           |           |       |         |
| Yes                                            | 93 (83.0) | 19 (17.0) | 112  | NS      |
| No                                             | 205 (89.5) | 24 (10.5) | 229  |         |
| Visible plaque at examination                   |           |           |       |         |
| Yes                                            | 26 (72.2) | 10 (27.8) | 36   | 0.004   |
| No                                             | 272 (89.2) | 33 (10.8) | 305  |         |
| CAT score                                       |           |           |       |         |
| ≥2                                             | 85 (81.7) | 19 (18.3) | 104  | 0.037   |
| <2                                             | 213 (89.9) | 24 (10.1) | 237  |         |
| Dental check-up at 12 months of age             |           |           |       |         |
| Yes                                            | 93 (96.9) | 3 (3.1)   | 96   | 0.0005  |
| No                                             | 204 (83.6) | 40 (16.4) | 244  |         |
| Application of topical fluoride                 |           |           |       |         |
| Yes                                            | 55 (91.7) | 5 (8.3)   | 60   | NS      |
| No                                             | 243 (86.5) | 38 (13.5) | 281  |         |
| Frequency of tooth brushing by parents          |           |           |       |         |
| ≥twice/day                                      | 90 (93.8) | 6 (6.3)   | 96   | 0.027   |
| <twice/day                                      | 208 (84.9) | 37 (15.1) | 245  |         |

Chi-square or Fisher’s exact test used to determine differences among groups. \(p<0.05\) indicates statistical significance. NS = not significant.
Caries and Feeding Behavior in Children

Table 9  Association between oral characteristics and dental caries at 36 months of age

| Oral characteristics variables | Prevalence of dental caries | Total | p-value |
|--------------------------------|-----------------------------|-------|---------|
|                                | No n (%)                    | Yes n (%) |       |
| Crowding in maxillary incisors |                             |        |        |
| Yes                            | 18 (78.3)                   | 5 (21.7) | 23     | NS     |
| No                             | 280 (88.3)                  | 37 (11.7)| 317    |        |
| Crowding in mandibular incisors|                             |        |        |
| Yes                            | 15 (79.0)                   | 4 (21.2) | 19     | NS     |
| No                             | 283 (87.9)                  | 39 (12.1)| 322    |        |
| Deep bite                      |                             |        |        |
| Yes                            | 26 (86.7)                   | 4 (13.3) | 30     | NS     |
| No                             | 272 (87.5)                  | 39 (12.5)| 311    |        |
| Open bite                      |                             |        |        |
| Yes                            | 6 (75.0)                    | 2 (25.0) | 8      | NS     |
| No                             | 292 (87.7)                  | 41 (12.3)| 333    |        |
| Excessive over jet             |                             |        |        |
| Yes                            | 19 (95.0)                   | 1 (5.0)  | 20     | NS     |
| No                             | 279 (86.9)                  | 42 (13.1)| 321    |        |
| Anterior cross bite            |                             |        |        |
| Yes                            | 36 (85.7)                   | 6 (14.3) | 42     | NS     |
| No                             | 262 (87.6)                  | 37 (12.4)| 299    |        |
| Inlocking of incisors          |                             |        |        |
| Yes                            | 12 (92.3)                   | 1 (20.0) | 13     | NS     |
| No                             | 286 (87.2)                  | 42 (12.8)| 328    |        |
| Abnormal position of upper labial frenum |                 |        |        |
| Yes                            | 176 (89.8)                  | 20 (10.2)| 196    | NS     |
| No                             | 122 (84.1)                  | 23 (15.9)| 145    |        |
| Teeth with structural defects in enamel |                       |        |        |
| Yes                            | 4 (80.0)                    | 1 (20.0) | 5      | NS     |
| No                             | 294 (87.5)                  | 42 (12.5)| 336    |        |
| Number of erupted teeth        |                             |        |        |
| <16                            | 116 (87.9)                  | 16 (12.2)| 132    | NS     |
| ≥16                            | 182 (87.1)                  | 27 (12.9)| 209    |        |
| Digital sucking habit          |                             |        |        |
| Yes                            | 65 (91.9)                   | 6 (8.5)  | 71     | NS     |
| No                             | 233 (86.3)                  | 37 (13.7)| 270    |        |
| Use of pacifier                |                             |        |        |
| Yes                            | 10 (76.9)                   | 3 (23.1) | 13     | NS     |
| No                             | 288 (87.8)                  | 40 (12.2)| 328    |        |

Chi-square or Fisher’s exact test used to determine differences among groups. p<0.05 indicates statistical significance. NS = not significant

Regression up to 3 years of age, especially in the maxillary anterior teeth. O’Sullivan et al. reported that the anterior teeth of the primary dentition, being the first teeth to erupt,
were immediately susceptible to carious attack if a child had inappropriate nursing habits and was infected with cariogenic microorganisms. The same report also noted that infection with MS was strongly associated with the development of the maxillary anterior caries pattern. This early type of infection has been suggested to be the major determinant of disease progression to the other teeth.

In earlier studies, Yonezu et al. reported that, children who were breast-fed at 18 months of age were 6 times more likely to

| Data obtained at 18 months of age | Adjusted OR | 95%CI       | p-value |
|----------------------------------|-------------|-------------|---------|
| Birth height                     |             |             |         |
| <50 cm                           | 2.993       | 1.428–6.535 | 0.0035  |
| ≥50 cm                           | 1           |             |         |
| Abnormal position of upper labial frenum |         |             |         |
| Yes                              | 1.972       | 0.937–4.218 | NS      |
| No                               | 1           |             |         |
| Use pacifier                     |             |             |         |
| Yes                              | 1           |             |         |
| No                               | 3.394       | 0.634–15.022| NS      |
| Visible plaque at examination    |             |             |         |
| Yes                              | 2.737       | 1.056–6.864 | 0.0387  |
| No                               | 1           |             |         |
| Wake-up time                     |             |             |         |
| After 8 a.m                      | 5.339       | 2.159–13.772| 0.0003  |
| Before 8 a.m                     | 1           |             |         |
| Bed time                         |             |             |         |
| After 9 p.m                      | 1           |             |         |
| Before 9 p.m                     | 2.043       | 0.814–5.572 | NS      |
| Breast and/or bottlefeeding in bed |         |             |         |
| Yes                              | 2.072       | 0.986–4.368 | NS      |
| No                               | 1           |             |         |
| Sweet beverage intake            |             |             |         |
| ≥4 times/week                    | 2.104       | 0.992–4.469 | NS      |
| <4 times/week                    | 1           |             |         |
| Brother and/or sister at home    |             |             |         |
| Yes                              | 2.657       | 1.170–6.364 | 0.0192  |
| No                               | 1           |             |         |
| Frequency of tooth brushing      |             |             |         |
| ≥twice/day                       | 1           |             |         |
| <twice/day                       | 3.019       | 1.184–9.055 | 0.0193  |
| Dental check-up at 12 months of age |         |             |         |
| Yes                              | 5.019       | 1.643–21.984| 0.0030  |
| No                               | 1           |             |         |

OR = odds ratio; CI = confidence interval; NS = not significant. p<0.05 indicates statistical significance.
have dental caries at 18 months of age than those who had been weaned before 18 months of age. The present results also indicate that children who continued breastfeeding were approximately 7 times more likely to have dental caries than those who did not (OR = 7.1; 95%CI = 2.86–19.46).

The correlation between prolonged breastfeeding and caries status has been investigated since the 1970s [2,7,10,14,20,28,34,35,43]. The results of these studies are inconclusive, however, and the cariogenic potential of human milk remains to be determined. Erickson et al. [6] reported that human milk did not cause a significant reduction in plaque pH and, consequently, enamel decalcification. On the other hand, Ripa [19] reported that while human milk was not considered to be a cariogenic agent under normal dietary conditions, repeated and prolonged exposure led to a larger decrease in plaque pH. More recently, it was shown that prolonged contact with human milk has a considerable acidulous potential and softens the enamel to a greater extent than bovine milk [12,31]. In addition, previous studies reported that at-will breastfeeding during sleep resulted in a higher MS count in the dental plaque and more dental caries among young children [2,9,28,30]. It should also be taken into account that factors contributing to dental caries in early childhood may vary between countries. Both high frequency and protracted breastfeeding were shown to be risk factors for dental caries in Japan [14,20,45]. No association was observed between duration of breastfeeding and dental caries in the USA, however [29].

Current WHO breastfeeding guidelines recommend exclusive feeding for the first 6 months of life and complementary breastfeeding for up to 2 years. In Japan, breastfeeding has also become more common over the last few decades, and more mothers breastfeed their children for longer periods of time than before. Yonezu et al. [45] found that 7.1% of children born in 1997–1999 were still being breast-fed at 18 months of age. Meanwhile, 31.8% of children born in 2010–2011 in the same city were being breast-fed at 18 months of age.

The evidence concerning infant feeding as a risk factor for dental caries is inconsistent. Consequently, no definitive optimal weaning times or breastfeeding practices have been determined to address the risk of dental caries. Tham et al. [29] conducted a systematic review and meta-analysis of accumulated evidence on the association between breastfeeding and dental caries with respect to specific windows of early childhood caries and provided quantitative evidence that is suggestive of the potential protective effects of breastfeeding up to 12 months, but high risk of dental caries in children beyond 12 months. The results of the present study revealed further evidence for the association between breastfeeding and dental caries.

It has been reported that children who are breast-fed for protracted periods of time also have more frequent cariogenic food intake. Multivariate logistic regression analysis in the present study (Table 5) revealed that dental caries was more likely to occur in children who brushed fewer than 1 time per day, those who ate snacks more than 2 times per day, those with visible dental plaque on oral examination, and those with high Cariostat scores.

On the other hand, children brought in for oral examination and consultation at around 12 months of age were less likely to develop dental caries than those who were not. Therefore, early establishment of good oral hygiene habits, reducing the frequency of high carbohydrate intake, reducing the frequency of breastfeeding, and being brought in for oral examination/consultation at 12 months of age are essential in preventing dental caries in early childhood.

2. Risk factors contributing to prevalence of caries in children aged 3 years

Among previous studies [22,24,26,36–38], the factors found to be associated with ECC have included high consumption of sugary snacks or beverages, irregular feeding practices, poor oral hygiene, and higher levels of MS. While the above-mentioned studies have identified a number of risk factors associated
with ECC, most of these studies were conducted in preschool children (3-years old or older). From a life-course perspective, the present longitudinal study highlighted the effect of early life circumstances and experiences on later health.

The present results showed that even if caries-free, children who continued nocturnal breast- and/or bottle-feeding in bed at 18 months of age were more likely to develop dental caries at 3 years of age. Although this disparity was not statistically significant, it does suggest that children who continue breast- and/or bottle-feeding in bed may be at risk of developing dental caries, and this cannot be ignored. Therefore, breast-fed children need to be monitored more closely, and aggressive methods of preventive care should be instituted.

In addition, the present results indicate that children whose oral hygiene condition is evaluated as “poor” at the 18-month oral examination are significantly more likely (OR = 2.74) to develop dental caries at 3 years of age than those in whom it is evaluated as “good”. The present results also showed a close relationship between the number of brushing times at 18 months of age and later caries development up to 3 years of age. Hallonsten et al.72 and Yonezu et al.44 reported that visible plaque on the incisors was highly correlated with ECC in 18-month-old children. The present results indicate an association between visible plaque on the incisors and ECC in children aged 18 months. In addition, other researchers have shown that the presence of visible plaque was not only associated with the presence of ECC but was also an indicator of the early stages of caries and the severity of disease41. Taken together with the present results, this emphasizes the importance of plaque control in very young children. Wendt & Birkhed30 reported that if the habit of daily brushing was established as early as the age of 1 year, children were more likely to be free of caries by the age of 3 years. This indicates that parents should be encouraged to brush their child’s teeth as soon as the primary teeth erupt. Yonezu et al.41, however, reported that there was no close relationship between the frequency of brushing times and caries development. This may indicate that parents have difficulty in achieving a good brushing technique, and that the method in use may be unpleasant to the child. Also, it should always be recognized that the quality of cleaning is more important than its frequency. Therefore, suitable training in brushing technique should be given to parents by the time their child is 18 months of age.

Many studies have investigated caries risk factors at specific ages. The present study has several advantages over such single outcome studies, however. This study provided a better mechanism for understanding strategies for caries prevention in Japan. It also had a noteworthy result, showing that children who were brought in for a dental check-up and consultation before 18 months of age had a significantly lower prevalence of caries at 18 months and 3 years of age. The question of whether children who are not brought in for a health examination before 18 months of age have poorer dental health remains to be answered. The present findings, however, support the proposition that special preventive dental care programs for infants and children should be started at around 12 months of age and continued every 6 months thereafter in Japan.

Finally, the present study collected a wide range of information regarding perinatal and early childhood conditions and adjusted for such confounding factors. Warren et al.35 reported that the absence of interdental spaces was weakly associated with greater decay experience in the primary dentition. Subramanian et al.27 also reported that children with no interdental spacing in the primary dentition were at higher risk for ECC. In present study, children who exhibited crowding in the maxillary incisors at 18 months of age were more likely to develop dental caries at 3 years of age than those who did not, although this association was not statistically significant. It should not be forgotten, however, that the highest prevalence of dental caries occurred in the proximal surfaces of the maxillary incisors.
This study had several strengths. Data on several key variables in relation to ECC were collected prospectively. These included infant feeding and oral hygiene practices, sweet consumption, fluoride use, and dental care utilization, which were used to control for confounding factors. The present study had several limitations, however. Caries outcomes were measured as "yes" or "no" at 18 months and 3 years of age, which was not suitable for maximizing statistical power. In addition, only a relatively small number of children living in the suburbs of Tokyo were investigated. This may limit the generalizability of the results in Japan as a whole. These issues need to be addressed in further investigation.

Conclusions

In conclusion, the present results showed that prolonged breastfeeding was a risk factor for ECC at 18 months of age after adjustment for a number of important confounding factors. A close association was also demonstrated between quality of oral hygiene and ECC at 18 months and 3 years of age. Children brought in for regular examination and consultation under the oral care program from 12 months of age were less likely to develop ECC at 18 months and 3 years of age. These results indicate that early life factors play an important role in the development of ECC in Japan.

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