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ORIGINAL ARTICLE

First molar eruption related to plaque acidogenicity in children of different socio-economic status

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Abstract

Objective. The aim of this study was to evaluate the association between the eruption stage of the lower first permanent molar and dental plaque acidogenicity. Socio-economic status (SES), gender, and oral hygiene condition were also variables considered. **Material and methods.** 230 children between 6 and 8 years of age were recruited from one public and one private primary school with different SES in Lima, Peru. Clinical examinations were performed to assess lower first permanent molar eruption stage, plaque acidogenicity, and oral hygiene condition. Bivariate associations were analyzed through chi-square tests and the variable interactions were analyzed through a hierarchical log-linear analysis with backward elimination. **Results.** 21.8% of the population had highly acidogenic plaque, 34.3% acidogenic plaque, and 43.9% non-acidogenic plaque. Of the lower first permanent molars, 46.1% were fully erupted, whereas 53.9% were partially erupted. According to the final log-linear model, children with fully erupted molars and non-acidogenic plaque are less frequent in low than in high SES. Also, the frequency of children with partially erupted molars and acidogenic to highly acidogenic plaque is higher in males than in females. Finally, fully erupted molars and non-acidogenic plaque are more frequent in children with good hygiene than in children with moderate to poor hygiene. **Conclusions.** Association between eruption stage of the lower first permanent molar and plaque acidogenicity was not significant in a bivariate context. However, in a multivariate context, socio-economic status, oral hygiene condition, and gender had an impact on the association between the two main variables.

Key Words: Log-lineal analysis, molar eruption, oral hygiene condition, plaque acidogenicity, socio-economic status

Introduction

Several studies have established that the cariogenic potential of the oral microorganisms depends partially on their capacity to release acids [1-4]. In this context, plaque acidogenicity is defined as the bacterial ability to produce lactic acid immediately after exposure to fermentable carbohydrates [1,5,6], and it is measured *in vivo* as its velocity to decrease plaque pH under the critical level of 5.5.

The aforementioned chemical changes are associated with differences in dental caries experience in both dentitions [3,5–8], due primarily to their demineralization effect over dental enamel. Furthermore, certain oral events have previously been cited as being associated with changes in dental plaque composition. Although the eruption of deciduous

and permanent teeth is one of the best known factors [9-13], the association between eruption stages and dental plaque acidogenicity has been overlooked in the literature [14,15].

The eruption of first permanent molars is an important milestone for the development of dental caries [12,14–17] because of its influence on plaque composition changes [12,13], the retentiveness of partially erupted surfaces [12,14], and the lower resistance to acid solubility of the recently erupted enamel [17–19]. For all these reasons, the eruption of first permanent molars has been considered as a second infection window in dental caries development in children [20].

Since a misbalance in oral conditions, such as increased plaque acidogenicity acting over newly erupted molars, may derive from the development

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and progress of dental caries, the association between dental eruption stages of first permanent molars and plaque acidogenicity may be important in the design of better preventive measures for this specific age group [12]. If the association between both variables is null, then the teeth are probably resistant enough to defend against acid attack; however, the existence of this association could lead to even more attention being given to recently erupted molars for decreasing the effect of the acid attack.

Only two studies have previously assessed the association between dental eruption and plaque acidogenicity [14,15]. Although these studies found that both variables were significantly associated, certain others factors were not considered, such as oral hygiene [4,21], gender [22–24], and socioeconomic status (SES) [25]. These factors have been demonstrated to be individually related to dental eruption or plaque acidogenicity. A multivariate analysis could permit evaluation of the association between dental eruption and plaque acidogenicity by controlling for possible confounders or effect modifiers.

The purpose of this study was to evaluate the association between eruption stages of the lower first permanent molar and plaque acidogenicity by controlling for socio-economic status, gender, and oral hygiene condition.

Material and methods

The study was authorized by the Ethics Board of the Universidad Peruana Cayetano Heredia.

Study population

The study population included 269 children between 6 and 8 years of age attending two primary schools located in two districts with different SES in Lima (Peru). A public school in Puente Piedra and a private school in Pueblo Libre were selected as representing low and high SES, respectively. Compared to the high SES, the low SES had a lack of basic services, e.g. potable water and drains and poor access to electricity, garbage collection services, public transportation, and dental/medical care services.

Only 230 schoolchildren from the study population fulfilled the selection criteria: a voluntary consent letter signed for their parents; no systemic conditions or use of medication; and lower permanent first molars both erupting.

Clinical examination

All dental examinations were performed in the morning by one previously trained examiner (J.P.)

following standardized criteria [26]. Previously, replicate examinations of plaque acidogenicity and oral hygiene condition had been performed by the only examiner and then contrasted with those performed by a fully trained assessor (E.D.) on a random sample of 10 children selected from a similar population. Intra- and inter-examiner reliability for the plaque acidogenicity test was 1.00 in both cases (weighted kappa, p < 0.001) and for oral hygiene 0.94 and 0.91, respectively (weighted kappa, p < 0.001 in both cases).

Each lower first permanent molar was classified according to eruption stage as *partially erupted* if the occlusal surface was not in occlusion or *fully erupted* if the tooth was in full occlusion [12]. Oral hygiene condition was registered through the Simplified Oral Hygiene Index [27] and classified as *good* if the index score ranged from 0 to 1.2, or *moderate to poor* if ranging from 1.3 to 6.0 [28].

Acidogenicity was measured as the velocity with which the pH of plaque drops below 5.5. The sampling/scraping method was used [29–31]. However, a modification was made in relation to the instrument used to read the pH levels. Strips of pH indicator paper (pH indicator strip 4.0–7.0 Merck®) were used because of the impossibility of accessing one pH-meter. The use of pH indicator paper has been cited as an easy, rapid, and reliable method for assessing pH levels [32].

Plaque samples from lingual surfaces of both lower first permanent molars were removed using sterile dental scalers, disregarding quantity, and immediately transferred to a sterile recipient and mixed with 1 cc of 10% glucose (weight/volume). The pH level was then measured using strips of indicator paper at 1-min and 2-min intervals: if pH was lower than 5.5 at the first interval, then plaque was considered as *highly acidogenic*; if pH was lower than 5.5 at the second interval, plaque was considered as *acidogenic*, and if it was constantly equal or higher than 5.5, plaque was considered as *non-acidogenic* [33]. No repeat measurements of the pH level were done.

Statistical analysis

Because eruption stages were not statistically different between right and left lower first permanent molars (chi-square test, p = 0.062), only first molars from the left side were considered in the statistical analysis. Descriptions of the variables and the codes used in the statistical analysis are exhibited in Table I. Bivariate associations between plaque acidogenicity and eruption stage, as well as with each covariable (oral hygiene condition, gender, and SES), were analyzed using chi-square tests.

Table I. Variables used in the multivariate analysis (log-linear model)

Variable	Description and codes			
Plaque acidogenicity (PLAQUE)	0 = non-acidogenic 1 = acidogenic 2 = highly acidogenic			
Eruption stage (ERUPTION)	0 = partially erupted 1 = fully erupted			
Oral hygiene condition (HY-GIENE)	0 = good			
	1 = moderate to poor			
Socio-economic status (SES)	0 = low			
	1 = high			
Gender (GENDER)	0 = male			
	1 = female			

The interaction between plaque acidogenicity, eruption stage, oral hygiene, SES, and gender was analyzed fitting a log-linear model to a multidimensional contingency table $(3 \times 2 \times 2 \times 2 \times 2)$. Fitting of the final log-linear model was done in two stages: first, using hierarchical log-linear analysis to locate significant interactions, and then using general log-linear analysis in order to build a final model. Model fitting was done in the present study, with the frequency of cases in each cell taken as the dependent variable. Finally, odds ratios (OR) were calculated on the basis of model parameters.

Results

The evaluated schoolchildren presented a mean age of 7.3 years (SD = 0.75), with the majority of them being 8 years of age (47.8%). Distribution according to SES, gender, oral hygiene condition, plaque acidogenicity, and eruption stage is presented in Table II.

When bivariate associations between variables were evaluated, statistically significant associations were found between PLAQUE and SES (p < 0.001), PLAQUE and GENDER (p = 0.001), and between PLAQUE and HYGIENE (p < 0.001), but not

Table II. Distribution of evaluated students according to the study variables

Variable	n	%	
Socio-economic status (SES)			
Low	166	72.2	
High	64	27.8	
Gender (GENDER)			
Male	130	56.5	
Female	100	43.5	
Oral hygiene condition (HYGIENE)			
Moderate to poor	169	73.5	
Good	61	26.5	
Plaque acidogenicity (PLAQUE)			
Highly acidogenic	50	21.8	
Acidogenic	79	34.3	
Non-acidogenic	101	43.9	
Eruption stage (ERUPTION)			
Partially	124	53.9	
Fully	106	46.1	

between PLAQUE and ERUPTION (p = 0.072). Cross-tabulation between these variables can be seen in Table III.

Hierarchical log-linear analysis with backward elimination was conducted in order to find the significant interactions and main effects. Interactions between five and four variables were not appropriated to propose a model (K-way tests, p = 0.975 and 0.925, respectively); however, at least one third-order interaction resulted significantly (K-way test, p = 0.008). Therefore partial association tests were conducted in order to find out which three of the five variables interacted significantly (Table IV). Although at least one two-order interaction and main effects were also significant (K-way tests, p < 0.001 in both cases), they were not taken into account because they were included in higher-order interactions.

Thereafter, through the general log-linear analysis, a model with the minimum number of non-zero parameters was sought so that the model could give almost the same cell counts as those observed in the five-way contingency table (Table V). Log-likelihood ratio was calculated to check the goodness-of-fit of the final model (p = 0.904). This indicated that the model including three third-order interactions permits the cell counts to be predicted similarly to the saturated hierarchical model (which includes all possible interactions and main effects).

Table V gives the parameters and significant ORs for the final model, relating ERUPTION, GENDER, SES, and HYGIENE to PLAQUE. According to the final model, interaction between ERUPTION, PLAQUE, and SES indicated that the presence of children with fully erupted permanent lower first molars and non-acidogenic plaque is less frequent in the low than in high SES. In addition, the interaction between ERUPTION, PLAQUE, and GENDER demonstrated that the frequency of partially erupted lower first permanent molars and acidogenic or highly acidogenic plaque is higher in males than in females.

Similarly, interaction between ERUPTION, PLAQUE, and HYGIENE indicated that children with good oral hygiene condition more frequently presented fully erupted lower first permanent molars and non-acidogenic plaque than those with moderate to poor oral hygiene. Finally, interaction between HYGIENE, GENDER, and SES indicated that male and female children with moderate to poor oral hygiene were more common in low SES than in high SES. ORs for each described interaction are presented in Table V.

Discussion

The study population included children attending one public and one private primary school located in two districts with different SES. Because both schools were selected for convenience, based on the

Table III. Plaque acidogenicity related to eruption stage, socio-economic status, gender, and oral hygiene condition of the evaluated students

Variables	Highly acidogenic plaque		Acidogenic plaque		Non-acidogenic plaque		p-value
	\overline{n}	(%)	n	(%)	n	(%)	
Eruption stage (ERUPTION)							0.072
Partially	21	(16.9)	41	(33.1)	62	(50.0)	
Fully	29	(27.4)	38	(35.8)	39	(36.8)	
Socio-economic status (SES)							< 0.001
Low	46	(27.7)	68	(41.0)	52	(31.3)	
High	4	(6.3)	11	(17.2)	49	(76.6)	
Gender (GENDER)							< 0.001
Male	20	(15.4)	49	(37.7)	61	(46.9)	
Female	30	(30.0)	30	(30.0)	40	(40.0)	
Oral hygiene condition (HYGIENE)		, ,		. ,			< 0.001
Moderate to poor	50	(29.6)	73	(43.2)	46	(27.2)	
Good	0	(0.0)	6	(9.8)	55	(90.2)	

Chi-square test was used.

institutional association with the schools' principals, the students recruited are not representative of the Peruvian population. However, owing to the lack of similar studies in Peru, the present results could be generalized, under the proper considerations, to the population living in both districts of Lima who fulfil the selection criteria.

The age group was selected because it represents the time period when lower first permanent molars erupt. This event has been shown to modify the oral microflora [12,13] and has been cited as a second window for caries infectivity [20]. The study contemplates three of the four health dimensions described on the Health-Field Model [34]. The field

Table IV. Interactions and mean effects between plaque acidogenicity, eruption stage socio-economic status, gender, and oral hygiene condition of the evaluated students

Interaction effect	Partial chi-square	df	p-value
ERUPTION*PLAQUE*SES	6.45	2	0.038
ERUPTION*PLAQUE*HYGIENE	8.72	2	0.021
ERUPTION*PLAQUE*GENDER	6.25	2	0.049
ERUPTION*HYGIENE*SES	0.06	1	0.805
ERUPTION*HYGIENE*GENDER	0.06	1	0.811
ERUPTION*SES*GENDER	3.19	1	0.074
PLAQUE*HYGIENE*SES	1.54	2	0.463
PLAQUE*HYGIENE*GENDER	1.14	2	0.566
PLAQUE*SES*GENDER	1.15	2	0.564
HYGIENE*SES*GENDER	5.17	1	0.037
ERUPTION*PLAQUE	4.81	2	0.078
ERUPTION*HYGIENE	0.05	1	0.820
ERUPTION*SES	0.20	1	0.651
ERUPTION*GENDER	2.41	1	0.120
PLAQUE*HYGIENE	54.66	2	< 0.001
PLAQUE*SES	11.05	2	0.004
PLAQUE*GENDER	8.01	2	0.016
HYGIENE*SES	12.30	1	< 0.001
HYGIENE*GENDER	1.70	1	0.192
SES*GENDER	0.30	1	0.584

df = degrees of freedom.

Asterisks represent interaction between variables.

related to health care organization was not considered in the actual design because few if any of the subjects referred to dental visits in the 6 months prior to the study.

In relation to the methodology followed in the present study, one important issue was the relatively uncommon method used to determine plaque acidogenicity [33]. Although three different methods have been established in the literature, i.e. the sampling/scraping method, the electrode system, and the indwelling electrode system [35–38], there is no ideal method for measuring plaque pH [7,36]. Since these methods measure pH at different dental locations and at different dental plaque depths, they could yield differing results with respect to both pH drop and recovery [35,36].

The advantages and disadvantages of these methods have been discussed in the literature [7,35,36]. The sampling/scraping method requires neither sophisticated equipment nor stricter recollection methods, thus diminishing time and costs of data collection [29–31]. A modification of the original sampling/scraping method was used in this study. The pH level was measured using pH indicator paper more than a pH meter [33]. The use of pH indicator paper for measuring acid concentration in the oral cavity has been compared with more sophisticated techniques [32]. It was found that the results obtained using indicator paper were reliable compared to the other methods, the conclusion being that this technique may have applications in clinical practice [32].

Quantification of the plaque present was not sought in the present study. The amount of plaque collected from each child was on average similar, although it was dependent on the amount of plaque available at selected teeth. This situation was not expected to be a problem because small differences in plaque wet weight have not previously been reported as a bias factor for acidogenicity measure-

Table V. Results of the log-linear model for evaluated students

Effect	Parameter value	SE	OR	95% CI		
					Upper bound	
(ERUPTION = 1)*(PLAQUE = 0)*(SES = 0)	-2.72	1.10	0.07	0.01	0.56	
(ERUPTION = 0)*(PLAQUE = 1)*(GENDER = 0)	1.34	0.53	3.82	1.36	10.71	
(ERUPTION = 0)*(PLAQUE = 2)*(GENDER = 0)	1.29	0.61	3.64	1.10	11.99	
(ERUPTION = 1)*(PLAQUE = 0)*(HYGIENE = 0)	1.63	0.82	5.10	1.02	25.46	
$(HYGIENE = 1)^*(GENDER = 0)^*(SES = 0)$	2.30	1.09	9.92	1.17	84.53	
(HYGIENE =1)*(GENDER =1)*(SES =0)	3.25	1.02	25.77	3.47	191.22	

SE = Standard error.

95% CI = 95% confidence interval.

Only significant interactions are exhibited (p < 0.05).

Log-likelihood ratio = 12.33; degrees of freedom = 20 (p = 0.904).

ments [39]. The plaque samples were immediately exposed to glucose [8,40,41] to determine how quickly they produce acids (acidogenicity) and concurrently reduce the pH to values at which enamel loses its integrity [42].

Another important issue could be that the plaque sampled to determine acidogenicity may not have been the plaque primarily responsible for the development of dental caries in the evaluated subjects. Controversy still exists in this regard, but it has been shown that acids released from plaque affect every tooth surface [7]. This fact may support the use of different surfaces for plaque collection [1,2,4,7,39–41], although lingual surfaces provide an excellent site for plaque accumulation [37].

According to the bivariate analyses, plaque acidogenicity was associated with SES, gender, and oral hygiene condition but not with eruption stage of lower first permanent molars. Nevertheless, it has previously been established that the extent of plaque accumulation, as well as its composition, is related to tooth eruption stage, tooth functional use, and specific anatomy [12]; in fact, teeth that are in full occlusion and fully functional retain less plaque than those that are not in occlusion and not fully erupted [12].

In order to assess the association between eruption stage of the lower first permanent molar and plaque acidogenicity within a multivariate context, a log-linear model was fitted which demonstrated that SES, oral hygiene condition, and gender, in that order according to OR values, seemed to influence the association between both variables.

Hence, it was found that the presence of fully erupted lower first permanent molars and non-acidogenic plaque increased 15-fold and 5-fold in children from high SES and with good oral hygiene, respectively. The present results indicate that the association between lower first permanent molar eruption stage and plaque acidogenicity is favorably modulated by better living conditions and adequate hygiene habits. Tooth eruption seemed to be slowed down in low SES groups probably related to

deprived nutritional status [43,44]. It is also known that oral hygiene is frequently neglected in low SES groups [45], which may expose them to an increased variety of dental plaque microorganisms changing the environment and modulating acidogenicity [46].

On the other hand, partially erupted lower first permanent molars with acidogenic and highly acidogenic plaque were 3.1-fold and 2.3-fold more frequent among boys, which concurs with the fact that eruption is delayed between boys compared with girls [22–24]. Nevertheless, no biological explanation for differences in plaque acidogenicity between boys and in girls has been reported.

Current evidence indicates that increasing caries activity is associated with an enrichment of plaque with organisms having a relatively high capacity for acidogenesis and acid tolerance. Such organisms include the lactobacilli, mutans streptococci (MS), and the so-called "low-pH" non-mutans streptococci (non-MS), as well as other types of "low-pH" organisms [8]. So far, only a few studies have reported on the flora of erupting molar teeth [14,15].

Since the microflora is an essential component of the caries process [12], it is important to completely understand the association between tooth eruption stages and dental caries induction. In relation to plaque acidogenicity, the authors recognize that in the present study an attempt was made to control some of the several factors influencing the oral environment by using some selection criteria (general health status and medication). Nevertheless, variables such as diet habits, previous exposure to fluoride, and use of oral health services were not analyzed. Further studies are therefore required to evaluate the impact of those factors among the present results. The use of alternative ways of assessing plaque acidogenicity as well as inclusion of the frequency of acidogenic episodes [7] in the statistical modeling could be the following steps. Similarly, the evaluation of other permanent and deciduous teeth and the use of more complete scales to classify dental eruption not only by simple

dichotomization (partially or fully erupted) in future studies will also bring about a better understanding of this phenomenon.

Conclusions

Association between eruption stage of the lower first permanent molar and plaque acidogenicity was not significant when assessed in a bivariate context. However, within a multivariate context, SES, oral hygiene condition, and gender seemed to influence the association between both variables.

References

- [1] Margolis HC, Moreno EC. Composition of pooled plaque fluid from caries-free and caries-positive individuals following sucrose exposure. J Dent Res 1992;71:1776-84.
- [2] Gao XJ, Fan Y, Kent RL Jr, van Houte J, Margolis HC. Association of caries activity with the composition of dental plaque fluid. J Dent Res 2001;80:1834–9.
- [3] van Houte J. Role of micro-organisms in caries etiology. J Dent Res 1994;73:672–81.
- [4] Aranibar Quiroz EM, Lingström P, Birkhed D. Influence of short-term sucrose exposure on plaque acidogenicity and cariogenic microflora in individuals with different levels of mutans streptococci. Caries Res 2003;37:51-7.
- [5] Margolis HC, Moreno EC. Composition and cariogenic potential of dental plaque fluid. Crit Rev Oral Bio Med 1994;5:1–25.
- [6] FDI. Review of methods of identification of high caries risk groups and individuals. Federation Dentaire Internationale Technical Report No. 31. Int Dent J 1988;38: 177–89.
- [7] Dong YM, Pearce EI, Yue L, Larsen MJ, Gao XJ, Wang JD. Plaque pH and associated parameters in relation to caries. Caries Res 1999;33:428-36.
- [8] Lingström P, van Ruyven FO, van Houte J, Kent R. The pH of dental plaque in its relation to early enamel caries and dental plaque flora in humans. J Dent Res 2000;79:770-7.
- [9] Wan AK, Seow WK, Purdie DM, Bird PS, Walsh LJ, Tudehope DI. A longitudinal study of Streptococcus mutans colonization in infants after tooth eruption. J Dent Res 2003; 82:504–8.
- [10] Caufield PW, Dasanayake AP, Li Y, Pan Y, Hsu J, Hardin JM. Natural history of Streptococcus sanguinis in the oral cavity of infants: evidence for a discrete window of infectivity. Infect Immun 2000;68:4018–23.
- [11] Caufield PW, Cutter GR, Dasanayake AP. Initial acquisition of mutans streptococci by infants: evidence for a discrete window of infectivity. J Dent Res 1993;72:37–45.
- [12] Brailsford SR, Sheehy EC, Gilbert SC, Clark DT, Kidd EA, Zoitopoulos L, et al. The microflora of the erupting first permanent molar. Caries Res 2005;39:78-84.
- [13] Loesche WJ, Eklund S, Earnest R, Burt B. Longitudinal investigation of bacteriology of human fissure decay: epidemiological studies in molars shortly after eruption. Infect Immun 1984;46:765-72.
- [14] Carvalho JC, Ekstrand KR, Thylstrup A. Dental plaque and caries on occlusal surfaces of first permanent molars in relation to stage of eruption. J Dent Res 1989;68:773–9.
- [15] Maltz M, Barbachan e Silva B, Carvalho DQ, Volkweis A. Results after two years of nonoperative treatment of occlusal surface in children with high caries prevalence. Braz Dent J 2003;14:48-54.

- [16] Härkäne T, Larmas MA, Virtanen JI, Arjas E. Applying modern survival analysis methods to longitudinal dental caries studies. J Dent Res 2002;81:144–8.
- [17] Sánchez-Pérez TL, Sáenz-Martínez LP, Gómez-López ME, Pérez-Quiroz J. [Enamel resistance to acid dissolution and its correlation with dental caries]. Salud Publica Mex 1995;37: 224-31.
- [18] Rodríguez Miró MJ, Elías Avila L, Gispert Abreu E. [Effect of a mineralizing solution (Minersol) on acid solubility resistance of enamel]. Rev Cubana Estomatol 1988;25: 11–21.
- [19] Kotsanos N, Darling AI. Influence of posteruptive age of enamel on its susceptibility to artificial caries. Caries Res 1991;25:241–50.
- [20] Straetemans MM, van Loveren C, de Soet JJ, de Graaff J, ten Cate JM. Colonization with mutans streptococci and lactobacilli and the caries experience of children after the age of five. J Dent Res 1998;77:1851-5.
- [21] Borgström MK, Edwardsson S, Svensäter G, Twetman S. Acid formation in sucrose-exposed dental plaque in relation to caries incidence in schoolchildren. Clin Oral Investig 2000;4:9-12.
- [22] Moslemi M. An epidemiological survey of the time and sequence of eruption of permanent teeth in 4-15-year-olds in Tehran, Iran. Int J Paediatr Dent 2004;14:432-8.
- [23] Nizam A, Naing L, Mokhtar N. Age and sequence of eruption of permanent teeth in Kelantan, north-eastern Malaysia. Clin Oral Investig 2003;7:222-5. Epub 2003 Aug 28.
- [24] Wedl JS, Schoder V, Blake FA, Schmelzle R, Friedrich RE. Eruption times of permanent teeth in teenage boys and girls in Izmir (Turkey). J Clin Forensic Med 2004;11: 299-302.
- [25] Saleemi MA, Hägg U, Jalil F, Zaman S. Dental development, dental age and tooth counts. A prospective longitudinal study of Pakistani children. Swed Dent J 1996;20: 61-7.
- [26] WHO. Oral Health Surveys: basic methods, 4th edn. Ginebra: World Health Organization; 1997.
- [27] Greene JC, Vermillion JR. The simplified oral hygiene index. J Am Dent Assoc 1964;68:7–13.
- [28] Greene JC. The oral hygiene index development and uses. J Periodontol 1967;38:Suppl:625–37.
- [29] Frostell G. A method for evaluation of acid potentialities of foods. Acta Odontol Scand 1970;28:599–622.
- [30] Lingström P, Holm J, Birkhed D, Bjorck I. Effects of variously processed starch on pH of human dental plaque. Scand J Dent Res 1989;97:392–400.
- [31] Tahmassebi JF, Duggal MS. Comparison of the plaque pH response to an acidogenic challenge in children and adults. Caries Res 1996;30:342-6.
- [32] Galgut PN. The relevance of pH to gingivitis and periodontitis. J Int Acad Periodontol 2001;3:61-7.
- [33] Gómez-Herrera B. [Integral clinical examination in pedodontics: methods], 1st edn. Cartagena: Editorial Corporación CDI; 1997. p. 232–3.
- [34] Dever GEA. Epidemiology: focus on prevention. In: Dever GEA, editor. Epidemiology in health care management, 1st edn. Germantown: Aspen Publishers; 1984. p. 1–24.
- [35] Lingström P, Imfeld T, Birkhed D. Comparison of three different methods for measurement of plaque-pH in humans after consumption of soft bread and potato chips. J Dent Res 1993;72:865-70.
- [36] Schachtele CF, Jensen ME. Comparison of methods for monitoring changes in the pH of human dental plaque. J Dent Res 1982;61:1117-25.
- [37] Hall AF, Creanor SL, Strang R, Foye R. Determination of plaque pH changes within the trough of an in situ appliance used to study mineral changes in early carious lesions. Caries Res 1997;31:50-4.

- [38] Huang GF, Guo MK. Resting dental plaque pH values after repeated measurements at different sites in the oral cavity. Proc Natl Sci Counc Repub China B 2000;24:187–92.
- [39] Damen JJ, Buijs MJ, ten Cate JM. Acidogenicity of buccal plaque after a single rinse with amine fluoride stannous fluoride mouthrinse solution. Caries Res 2002; 36:53–7.
- [40] Sansone C, Van Houte J, Joshipura K, Kent R, Margolis HC. The association of mutans streptococci and non-mutans streptococci capable of acidogenesis at a low pH with dental caries on enamel and root surfaces. J Dent Res 1993;72:508–16.
- [41] Gerardu VA, Buijs MJ, ten Cate JM, van Loveren C. The effect of a single application of 40% chlorhexidine varnish on the numbers of salivary mutans streptococci and acidogenicity of dental plaque. Caries Res 2003;37:369–73.

- [42] Margolis HC, Zhang YP, Lee CY, Kent RL Jr, Moreno EC. Kinetics of enamel demineralization in vitro. J Dent Res 1999;78:1326–35.
- [43] Alvarez JO, Caceda J, Woolley TW, Carley KW, Baiocchi N, Caravedo L, et al. A longitudinal study of dental caries in the primary teeth of children who suffered from infant malnutrition. J Dent Res 1993;72:1573–6.
- [44] Alvarez JO. Nutrition, tooth development, and dental caries. Am J Clin Nutr 1995;61:410S-6S.
- [45] Chen MS. Children's preventive dental behavior in relation to their mothers' socioeconomic status, health beliefs and dental behaviors. ASDC J Dent Child 1986;53:105–9.
- [46] Bretz WA, Corby PM, Hart TC, Costa S, Coelho MQ, Weyant RJ, et al. Dental caries and microbial acid production in twins. Caries Res 2005;39:168-72.