INFLUENCE OF HOST-RELATED RISK INDICATORS ON DENTAL CARIES IN PERMANENT DENTITION

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ABSTRACT

The aim of this study was to assess the individual and grouped influence of host-related factors on dental caries experience in permanent dentition of 7-9 year-old children. One hundred and twenty one children were recruited applying a stratified cluster sampling without replacement. Clinical examinations, Colorimetric Test and non-stimulated and stimulated saliva collection were performed; the samples obtained were later incubated using Snyder culture medium. Simple and multiple binary logistic regression analyses were performed to assess the individual and grouped influence of the host-related factors and establish a model for predicting dental caries in permanent dentition. 78.5% of the evaluated children presented dental caries in permanent dentition. Binary logistic regression analyses revealed that only dental caries in deciduous dentition and stimulated salivary flow were significantly associated to dental caries in permanent dentition. The final multivariate model showed that the adjusted OR was 1.76 for an increase of 1 tooth in dmft and 2.29 for a decrease of 1 ml in stimulated salivary flow. Dental caries in deciduous dentition is a risk indicator for dental caries in permanent dentition, and stimulated salivary flow is a protective factor. Variables like enamel resistance to acid dissolution and buffer capacity are not related to dental caries in permanent dentition, making them inappropriate for risk studies.

Key Words: *dental caries experience, non-stimulated salivary flow, stimulated salivary flow, salivary buffer capacity, enamel resistance to acid dissolution*

INFLUENCIA DE FACTORES RELACIONADOS AL HOSPEDADOR SOBRE LA CARIES DENTAL EN DENTICION PERMANENTE

RESUMEN

El propósito de este estudio fue establecer la influencia individual y agrupada de los factores relacionados al hospedador sobre la experiencia de caries dental en dentición permanente de niños de 7 a 9 años de edad. Se reclutaron 121 niños aplicando un muestreo estratificado simple. Se realizaron exámenes clínicos, la Prueba Colorimétrica y la recolección de saliva estimulada y no estimulada; las muestras obtenidas fueron incubadas posteriormente empleando el medio de Snyder. Para establecer la influencia individual y agrupada se llevaron a cabo análisis de regresión logística simple y múltiple, a fin de establecer un modelo de predicción de riesgo de caries dental en dentición permanente.78.5% de los niños evaluados presentaron caries dental en dentición permanente. El análisis de regresión logística sólo encontró asociación significativa entre caries dental en dentición permanente con la experiencia de caries dental en dentición decidua y el flujo salival estimulado. El modelo multivariado final mostró que el OR ajustado para el incremento del ceod en 1 pieza era de 1.76, y para la disminución de 1 ml del flujo salival estimulado fue de 2.29.

Se encontró que la experiencia de caries dental en dentición decidua es un indicador de riesgo para caries dental en dentición permanente, y el flujo salival estimulado un factor de protección. Las variables como resistencia del esmalte a la desmineralización y la capacidad buffer de la saliva no mostraron relación con la caries dental en dentición permanente, lo que los hace inapropiados para estudios de riesgo.

Palabras Clave: *experiencia de caries dental, flujo salival no estimulado, flujo salival estimulado, capacidad buffer de la saliva, resistencia del esmalte a la desmineralización*

INTRODUCTION

Dental caries, defined as a chronic multifactorial disease, has been broadly studied in the last decades and a great amount of risk factors have been reviewed (1, 2). However, those variables referred to the host have not received the same attention as others, at least in children (3). A possible explanation of the lack of studies regarding influence of host-related factors on dental caries could be the assessment difficulties in field or in vivo settings.

The term "host-related factors" refers to all the individual's intra-oral characteristics –tooth and saliva– which play an important role in the development and progression of dental caries (4). Dental caries prevalence and experience in the deciduous dentition are associated to their counterparts in the permanent dentition (1). In fact, past caries experience compared to other variables had a relatively strong association with future caries increment, being the best predictor of caries increment in children (5-7).

Although saliva influences the cariogenic potential in several ways, there are two characteristics of saliva which might be useful for detecting high risk of caries development: flow rate and buffer capacity. One of the most important caries-preventive functions of saliva is to dilute bacterial substrates, a process that is usually referred to as oral clearance (1, 8-11). Despite this evidence, salivary flow rate has been scarcely studied among children. The function of buffering agents in saliva, in particular the carbonic acid/bicarbonate system, is to return the pH to the normal range as soon as possible following exposure to components whose pH differ from normal salivary pH (9). Buffering capability is related to salivary flow rate and has been measured by the amount of acid needed to lower salivary pH during a fixed time interval (1).

Enamel resistance to acid dissolution seems to be the only previously reported tooth factor (12, 13). This parameter has been used to assess enamel cariostatic properties (1). It is a passive characteristic of enamel that delays mineral loss caused by bacterial acid attack. Furthermore, this property allows saliva to equilibrate oral conditions, promoting remineralization and avoiding dental caries progression. Some studies have mentioned it as an indicator for caries risk assessment (12-14), although it seems more suited for evaluating remineralization procedures (15, 16).

A recent consensus considers that any dental caries assessment should be based on several risk factors and should consider not only their cumulative effect but also their interactive effect on dental caries (1, 2, 6, 17). Although risk factors for dental caries have been extensively studied, some of these factors –like salivary characteristics– have been overlooked, especially among children, and disregarded in multivariate studies. This fact precludes the understanding of their cumulative and interactive effect on dental caries.

The aim of this study was to assess the individual and grouped influence of the host-related risk indicators (dental caries experience in deciduous dentition, non-stimulated and stimulated salivary flows, salivary buffer capacity and enamel resistance to acid dissolution) on dental caries in the permanent dentition of 7-9 year old children.

MATERIALS AND METHODS

Approval for the study was previously obtained from the Ethics Board at the Universidad Peruana Cayetano Heredia.

Study sample

121 children, 7 to 9 years old (28% of the total target population), were recruited applying a stratified cluster sampling without replacement within nine underserved communities from Lima, Peru. The sample largely represents the Peruvian infantile population living in low income urban communities, where the lack of basic services such as potable water and drains; poor access to electric energy, garbage recollection services, public transportation and dental or medical care services are the prevailing characteristics.

The evaluated age group was chosen because of the susceptibility to caries attack of the permanent teeth erupting during this period of life and because it is the recommended age to evaluate enamel resistance by the Colorimetric test because of the eruption of upper permanent incisors (13, 15, 16).

For the sample selection, every block was identified as a cluster and a blocks map was designed. Then, blocks were selected with a probability proportional to their size, i.e. the number of homes in each block. This procedure allowed for the selection of children with equal probability. Every home within each block chosen where a 7-to-9-year-old child resided was invited to participate in the study through interviews with the child's parents. Only those children whose parents returned a signed consent form were enrolled in the study. Non-response rate was only of 2.4%.

Clinical examination

All dental examinations were conducted between 9 AM and noon by one previously trained examiner (EKD) following standardized and widely accepted criteria, as recommended by the WHO report on oral health surveys (18), and based on the diagnostic criteria for caries prevalence surveys published by the British Association for the Study of Community Dentistry (BASCD) (19). Radiographic examinations were not included and dental caries was recorded at the level of enamel lesion.

Training and calibration exercises were conducted based on the guidelines recommended by BASCD (19). To establish the levels of intra- and inter-examiner reliability, replicate examinations were performed prior to the beginning of the study by the only examiner and a fully trained assessor on BASCD criteria on a random sample of children from the study population. The generalized Kappa coefficient was calculated at surface level for decayed, filled and sound surfaces in permanent dentition. Acceptable levels (20, 21) of intra- and inter-examiner reliability were obtained (0.93 and 0.85 respectively, $p<.001$).

Procedures of data recollection

During the initial interviews, parents were asked for the birth date of their children and the age of each child was calculated as the time interval (in years) between birth and examination dates.

The saliva collection was performed with the children seated. In order to assess non-stimulated salivary flow, children were asked to spit into plastic cups during 1 minute, measured using a chronometer. These samples were placed in graduated disposable syringes and the number of milliliters of saliva was read from the graduated syringe to the nearest 0.1 ml and recorded (22). To determine the stimulated salivary flow, each child was given a 1-gram piece of unflavored paraffin wax to chew for 1 minute. The saliva produced was spit into a plastic cup and collected into a graduated disposable syringe. Again the amount of saliva collected was read and recorded (22, 23). The stimulated saliva samples were stored on ice and transported to the laboratory.

200 µl from each saliva sample were transferred to a test tube containing 8 ml sterile Snyder's test agar maintained semisolid at 45ºC. Then the medium was mixed uniformly with the inoculums and allowed to solidify. The tubes were incubated in upright position at 37ºC (23, 24). Medium color change was observed every 24 hours for 3 days, and classified from green to yellow as "marked" –if the medium remained green all the time–, "moderate" –if the medium changed to yellow at 48-72 hours of incubation–, "slight" –if change was produced at 24 to 48 hours– and "negative" –if the medium color changed from green to yellow within the first 24 hours of incubation– as described by Snyder (24, 25). A non-inoculated medium served as negative control.

Enamel resistance to acid dissolution was measured using the Colorimetric Test (13, 15, 16), which uses rounded absorbent paper discs 2.8 mm in diameter. Every disc is impregnated with crystal violet as an indicator. At pH 2.0 the discs turn yellow and when pH increases to its maximum (3.0) they return to their initial purple color. The activator used is hydrochloric acid at pH 1.9 (12-16, 25). This technique is based on the fact that acids produce the demineralization of dental enamel, releasing calcium and phosphate ions which in contact with crystal violet disc neutralize its acidity, changing its color (12-16, 25). Thus, a colorimetric scale is built where the first color is yellow and the last is purple, with intermediate values that correspond to the colors of the paper disc at pH values between 2.0 and 3.0 –yellow-green-purple– (12, 14, 25).

A paper disc exposed to hydrochloric acid was placed on the palatal surface of a permanent superior incisor –representative of all other teeth (12, 13, 15, 16)– previously cleaned and dried using cotton tips. After a one-minute period –measured using a chronometer– the paper disc was removed and compared to the colorimetric scale. According to this scale, enamel was considered resistant to acid dissolution when the result of the Colorimetric Test was between 1 and 5; or susceptible to acid dissolution if its values were equal to or higher than 6.

Statistical analysis

Simple binary logistic regression analyses were performed to assess the individual influence of dental caries experience in deciduous dentition, non-stimulated and stimulated salivary flows, salivary buffer capacity and enamel resistance to acid dissolution and each covariable (age and sex) on the probability of having clinically detectable dental caries (DMFT > 1 vs. DMFT = 0).

Thereafter, a multiple binary logistic regression analysis was conducted to evaluate the grouped influence of the host-related risk indicators and establish a model for predicting dental caries in permanent dentition. The multivariate analysis was undertaken including those variables with moderate significance ($P < .25$) in the univariate models previously developed. The presence of interactions between risk indicators or covariables was also

TABLE I: Description of the sample according to the host-related risk indicators		
Risk Indicators	**n**	**%**
Salivary buffer capacity		
Marked	32	26.4
Moderate	16	13.2
Slight	21	17.4
Negative	52	43.0
Enamel resistance to acid dissolution		
Resistant	15	12.4
Non-resistant	106	87.6
Dental caries experience in deciduous dentition (dmft)		
Mean + S.D.	5.54 ± 2.65	
Non-stimulated salivary flow (ml)		
Mean + S.D.	1.18 ± 0.64	
Stimulated salivary flow (ml)		
Mean + S.D.	2.54 ± 1.17	

S.D. = Standard Deviation

explored (26). The G values and final significance of each model were considered in the selection of the appropriate final model (26).

Finally, the coefficient of determination (R^2) was calculated as a measure of the percentage of variance on dental caries in permanent dentition that is explained for those variables included in the final model (26). Accuracy, sensitivity and specificity of the final model were also estimated.

Every statistical analysis were performed using SPSS® 11.0 software.

RESULTS

The sample selected was conformed for 60 (49.6%) female and 61 (50.4%) male children with a mean age of 8.01 + 0.78 years. In addition, 78.5% [$IC_{95\%}$(71.2; 85.8)] of the evaluated children presented dental caries in permanent dentition and a mean DMFT of 2.17 [$IC_{95\%}$(1.89; 2.45)]. Distribution of the sample according to each host-related risk indicator is shown in Table I.

Simple binary logistic regression analyses were conducted to assess the bivariate association between each host-related risk indicator or covariable and the presence of dental caries in permanent dentition. Only dental caries experience in deciduous dentition ($P < .001$) and stimulated salivary flow ($P = .036$) were significantly associated to the presence of dental caries in permanent dentition (Table II).

TABLE II: Dental caries experience in permanent dentition (DMFT ≥ 1 vs. DMFT = 0) according to each host-related risk indicator

Risk Indicators	DMFT≥1 (n = 95)		DMFT=0 (n = 26)		Unadjusted OR	95% IC	p value
	n	%	N	%			
Sex							
Female	46	76.7	14	23.3	1.00		
Male	49	80.3	12	19.7	1.24	(0.52; 2.97)	0.624
Age (per 1-year increment)							
Mean ± S.D.	8.21 ± 0.77		7.88 ± 0.76		1.72	(0.98; 3.04)	0.061
Salivary buffer capacity							
Marked	23	71.9	9	28.1	1.00		
Moderate	12	75.0	4	25.0	1.17	(0.30; 4.62)	0.818
Slight	18	85.7	3	14.3	2.35	(0.55; 9.95)	0.247
Negative	42	80.8	10	19.2	1.64	(0.58; 4.62)	0.346
Enamel resistance to acid dissolution							
Resistant	11	73.3	4	26.7	1.00		
Non-resistant	84	79.2	22	20.8	1.39	(0.40; 4.78)	0.408
Dental caries experience in deciduous dentition (per 1-tooth increment)							
Mean + S.D.	6.22 ± 2.40		3.03 ± 1.93		1.99	(1.48; 2.68)	<0.001
Non-stimulated salivary flow (per 0.1-ml increment)							
Mean + S.D.	1.18 ± 0.65		1.17 ± 0.60		1.02	(0.51; 2.04)	0.947
Stimulated salivary flow (per 0.1-ml increment)							
Mean + S.D.	2.42 ± 1.12		2.97 ± 1.28		0.66	(0.45; 0.97)	0.036

S.D. = Standard Deviation

TABLE III: Results of the multiple binary logistic regression analysis					
Risk indicators	**Beta**	**S.E.**	**Adjusted OR**	**95% IC**	**p value**
Dental caries experience in deciduous dentition (per 1-tooth increment)	0.56	0.11	1.76	(1.42; 2.18)	<0.001
Stimulated salivary flow (per 1-ml increment)	-0.44	0.15	0.65	(0.48; 0.87)	0.004
Constant	-1.81				<0.001

S.E. = Standard Error

A multivariate model was then built taking into account main effects and interactions. The first multivariate model evaluated the risk indicators without taking into account the interactions (main effects only model) with the purpose of assessing the independent effect of each one on the presence of dental caries in permanent dentition while controlling for confounding. The next step was to explore the inclusion of possible two-factor and three-factor interactions in the main effects only model. However, no interaction term could be added to the model ($P > .10$ in all cases).

Based on the present findings, the final multivariate model was conformed by dental caries experience in deciduous dentition and stimulated salivary flow (Table III). The adjusted OR for an increase of 1 tooth in dental caries experience in deciduous dentition (dmft) was 1.76, which means that for every increase of 1 tooth in dmft, the probability of having clinically detectable dental caries in permanent teeth increases 1.76 times, controlling the stimulated salivary flow. Conversely, the adjusted OR for a decrease of 1 ml in stimulated salivary flow was 2.29 (inverse value for -0.44). This indicates that for every decrease of 1 ml in stimulated salivary flow, the probability of having clinically detectable dental caries in permanent teeth increases 2.29 times, controlling the dental caries experience in deciduous dentition.

Finally, the multivariate model, with a R^2 of 0.632, exhibited values of accuracy, sensitivity and specificity of 83.5%, 95.8% and 38.5% respectively. Individual contribution on the R^2 was of 0.576 and 0.056 for dmft and stimulated salivary flow respectively.

DISCUSSION

The present results indicate that approximately three-quarters of the examined children had dental caries in permanent dentition and, on average, two teeth with dental caries experience. Although the studied population is local, the findings reported here could be of broader interest. The use of a strict sampling technique added to the lack of previous reports assessing dental caries and associated factors in Peruvian children could allow us to generalize these results not only to the studied population but also to other Peruvian communities with similar socio-economic characteristics. However, further studies are encouraged in order to corroborate these findings.

According to the multivariate analysis conducted, only dental caries experience in deciduous dentition and stimulated salivary flow, in that order, were significantly associated to the presence of dental caries in permanent teeth. It has been previously demonstrated that dental caries in deciduous teeth is strongly associated with future caries increments (5-7, 27), and that past caries experience is used as a risk indicator to predict caries in permanent teeth (2, 7, 28). The present findings reinforce these statements. In fact, this unique factor explained almost sixty percent of the variability in the presence of dental caries in permanent dentition of the evaluated children. This finding supports the general idea that dental caries in deciduous teeth is the most valuable predictor for caries development in permanent teeth.

Stimulated salivary flow was the second factor associated to dental caries in permanent dentition. However, the relative contribution of this risk indicator in comparison with the dental caries experience in deciduous teeth was minor (about 6%). The physical removal of undesirable products by saliva is a relatively simple protective mechanism that is often overlooked. Nevertheless, it is an important factor. However, the wide variation in individual rates could limit the significance of this finding (1, 22).

In the oral cavity, there is a minimum volume of saliva after swallowing: the resting or residual volume (8, 11), which seems to increase when salivary flow rate is normally stimulated by the taste of sucrose together with optional flavoring agents (8, 11, 22). This increase in flow rate augments the clearance of sugars and acids after food consumption (11). In this study, stimulated salivary flow and dental caries in permanent dentition are negatively related, similarly to the findings of previous studies (10, 22).

It has been suggested that any caries prediction model should produce a sensitivity of 75% or higher and a specificity level of at least 85% (29). The model obtained had a sensitivity of 95.8%, which reflects its capability to reduce the number of false negatives, a desirable characteristic that avoids the need of more advanced costly and painful therapy in the long term due to the presence or progression of the disease (17).

There is a permanent need to identify the subjects with a greater predisposition to future dental caries in order to target the most effective preventive interventions at these individuals and motivate them to maintain appropriate self-care behaviors (28). Any model, regardless of its ultimate accuracy, would be based on a data collection system that is relatively quick, inexpensive, and requires a limited armamentarium, and be acceptable to those to whom it is applied (29). Further studies are encouraged to validate the proposed model as well as to compare it with alternative models.

The other two salivary properties evaluated (buffer capacity and non-stimulated flow) were not associated to dental caries in permanent dentition. The salivary buffering capacity has been thought to vary with caries activity (1), but it may exhibit a narrow range of values for most children, thus limiting its utility in risk discrimination (6, 28). This is why the buffer capacity tests have a low prediction value (8, 9) –in concordance to the present findings– because the decisive events in a caries attack take place in the plaque and below the enamel surface, where the buffering mechanisms are different from those in saliva (8).

Stimulated salivary flow could be a protective factor against dental caries because sucrose induces it, as opposed to non-stimulated salivary flow that remains unaffected. Besides this fact, the pH of non-stimulated saliva can be as low as 5.6, which in comparison to the pH of 7.8 in stimulated saliva can hardly have a protective effect against dental caries (8).

The present results show that enamel solubility in acid is not directly related to caries experience in the permanent dentition. However, it has been used repeatedly as a measurement to assess cariostatic properties of teeth and therapeutic agents (12-16, 25). Caries resistance is only partially dependent on the quality of the enamel and is heavily dependent on the composition of the oral fluids in contact with the enamel (1).

According to the literature, identifying children with existing caries, as well as those at high risk for developing caries should include: improved accuracy of screening examinations conducted in non-clinical settings; inexpensive procedures and equipment; ease of performance, requiring little technical skills and achievement of fast results (1, 6). Based on these concepts, a different technique to those usually employed to evaluate enamel resistance to acid dissolution was used in the present study.

The Colorimetric Test derives from the Color Reaction Time used by the Swiss during the sixties (12, 13). It is a simple and economic way to assess in situ the enamel resistance to acid solubility instead of using intra-oral appliances which may require the subject's cooperation and would take more time to obtain results. Minerals from the enamel surface should not be released rapidly when oral pH decreases (30). This test exposes enamel to acid during a short period of 1 minute. The indicator paper should remain the same color, corresponding to its initial pH value of 2.0 if minerals are not released and should change color with an increase in pH up to a value of 3.0 if minerals are released from the enamel (12, 13). According to these changes, an 8-color scale is obtained where enamel is considered very resistant to acid dissolution if the paper color corresponds to values 1 to 3, resistant for values 4 to 5, susceptible to acid dissolution for values 6 to 8 (12, 13, 15, 16). It should be noted that for the present analysis, the categories "very resistant" and "resistant" were collapsed due to the very few cases classified as very resistant to acid dissolution.

Although all human enamel surfaces are susceptible to acid exposure if the acid is strong enough (1, 13, 30), previous studies have demonstrated that upper incisors could be considered the average of all teeth when evaluating enamel resistance to acid dissolution through the Colorimetric Test (12, 13, 15, 16). Their accessibility made these teeth also ideal to assess enamel resistance.

In conclusion, dental caries experience in the deciduous dentition and stimulated salivary flow are shown to be contributing factors to dental caries development in permanent dentition. Because of the multifactor origin of dental caries, other risk factors, not considered in the present study, have previously shown to have an important role. Therefore, new studies should be developed to address the role of other risk factors. It would be interesting to evaluate how these two factors also interact with the others. Although the findings of the present study could theoretically only be generalized to the evaluated population, the authors believed that they might also be applicable to Peruvian communities with similar socioeconomic characteristics.

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