

Dental caries, salivary parameters and plaque scores as caries risk predictors among 12 year old school children – A follow up study

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ABSTRACT

Introduction: Dental caries is a disease with multifactorial etiology and many other factors influence indirectly. The important factors are Streptococci mutans, Lactobacilli counts, saliva flow rate, buffering capacity and past caries experience.

Objective: To find the association between caries increment and various risk factors: Caries experience, dental plaque, salivary flow rate, buffering capacity, Streptococcus mutans and Lactobacilli counts.

Method: Dental caries and plaque scores were assessed using Modified Dentition Status and Treatment Needs and Silness and Loe index respectively. Stimulated saliva was collected to estimate salivary flow rate, buffering capacity, Streptococcus mutans and lactobacilli colony forming units. Bivariate analysis was carried out using caries increment (dichotomous for DMFT and DMFS) and each variable dichotomized at baseline (Pearson's χ^2 test with continuity correction as required).

Result: WPDMMFT and WPDMMFS were associated with caries increment (DMFT and DMFS) after 8 months ($p=0.01$, $p=0.04$ respectively). Salivary Streptococcus mutans counts alone showed a statistically significant association for caries increment (WPDMMFT and WPDMMFS).

Conclusion: The results of the present study suggest initial caries to be the strongest predictor of caries occurrence in future.

Keywords: Dental Caries, plaque, Streptococcus mutans and Lactobacilli colony forming uni

Introduction

Dental caries is defined as a progressive, irreversible microbial disease of multifactorial nature affecting the calcified tissues of the teeth characterized by demineralization of the inorganic portion and destruction of the organic portion of the tooth. It is a disease of civilization¹. It ranks amongst the most common of human diseases mainly because of its frequency of occurrence. Among the numerous factors causing dental caries, the important ones are *Streptococcus mutans*, *Lactobacilli* counts, saliva flow rate, buffering capacity and past caries experience². It is also modified by factors like type of diet taken, oral hygiene practices, use of fluoride and other preventive measures and dental visits which are dependent on socioeconomic status^{3, 4}.

Although the severity of the dental disease in terms of its life threatening potential is limited except in rare instances, certain important consequences must be stressed. Dental caries and its sequel often involve pain and affect esthetics. Treatment of dental caries is costly both in terms of time and money⁵.

It is prudent to prevent dental caries by applying suitable preventive procedures to avoid complications. In developing countries where there is scarcity of resources the high risk people should be carefully selected. Although few models are available which have better predictive power in caries risk assessment they have not been validated among Indian population⁶⁻⁸.

Hence the present study was conducted with an objective to find the association between caries increment and various risk factors: Caries experience, dental plaque, salivary flow rate, buffering capacity, *Streptococcus mutans* and *Lactobacilli* counts. An additional objective was to assess the caries increment using modified DMFT.

Material and Method

The present study was conducted among 100, 12 years old school children of Belgaum city, which is located in the southern part of India. This is a part of the longitudinal study which is being conducted to assess the effect of preventive measures on caries risk. A part of the study has been published⁹.

There are a total of 285 schools in Belgaum city. A total of four schools were randomly selected for the study. Permission to conduct the study was obtained from Deputy Director of Public Instructions (DDPI) and school authorities. Ethical clearance was also obtained from Institutional Review Board (IRB, Ref no: KLEU /07-08/ D-9141). Informed consent and assent was obtained from parents and children respectively. The following parameters were assessed-

1. Clinical examination: Children were examined for plaque and dental caries.
 - i. Silness and Loe plaque index was used to assess the amount of plaque¹⁰.
 - ii. Dental caries was assessed using modified WHO Dentition Status and Treatment Needs. The initial lesions (WP) were taken into account and all the surfaces were also considered in order to make the index more sensitive. DMFT and DMFS were denoted as WPDMFT and WPDMFS respectively indicating the inclusion of initial lesions¹¹. A single trained and calibrated examiner recorded both the indices. The intra examiner reliability was found to be 0.78 and 0.86 respectively for both the indices respectively.

2. Saliva collection: Simplified techniques of salivary assessments were used to make them cost effective and applicable for the field study. Children were asked to chew a modeling wax made into a form of pellet (0.5 x0.5 centimeters) for 3 minutes to obtain stimulated saliva.

- i. Salivary flow rate: Saliva from oral cavity was sucked using a sterile disposable syringe and amount of saliva secreted per minute was calculated.
- ii. Salivary buffering capacity: 0.5ml of saliva was added to 1.5ml of 0.005 molarity of hydrochloric acid (HCL). Buffering capacity of saliva was determined by assessing the change in pH using commercially available Indikrom paper, which have a predetermined pH range and categorized accordingly.
- iii. Microbial assessment: By means of a sterile disposable syringe 0.5 ml aliquot of saliva collected directly from the oral cavity was injected in a previously labeled sterile bottle containing 2ml of transport medium. The samples were processed on the same day in the Department of Microbiology, Jawaharlal Nehru Medical College.

Laboratory procedure: The samples were vortexed to uniformly mix the saliva and the media using a cyclomixer. Using an inoculation loop (standard loop with 4mm inner diameter) 10 ml of the vortexed sample was streaked on Mitis salivarius agar selective for *Streptococcus mutans* and on Rogosa SL agar for *Lactobacillus*. The Mitis salivarius agar plates were incubated in an anaerobic jar for 48 hours at 37°C in an incubator and similar procedure were followed for Rogosa SL agar plates, which were incubated for 96 hours.

All the parameters were reassessed after 8 months. The subjects were reexamined by the same examiner in order to avoid inter examiner variation.

Statistical methods: The data was analysed using SPSS statistical package (version 17.0 SPSS Inc. Chicago III, USA). Descriptive statistics including the means and standard deviations were calculated for the continuous variables. Paired t test was used to find the difference between the mean values at baseline and follow-up. Bivariate analysis was carried out using caries increment (dichotomous for DMFT and DMFS) and each variable dichotomized at baseline (Pearson's χ^2 test with continuity correction as required). Risk ratio was also calculated to estimate risk. For assessment of caries increment using initial lesions (WPDMFT and WPDMS) the data was trichotomized and analyzed. P-value <0.05 was considered statistically significant.

Results

The present study was conducted to assess the effect of various risk factors on caries increment. There were totally 100 children at the baseline, during follow-up 4 children dropped out from the study. Of the 96 children who remained in the study, 49 were boys and 47 were girls. Almost all of them used tooth paste and tooth brush to clean their teeth and most of them brushed once daily. At baseline 23 were free caries when DMFT and DMFS were considered and only 4 were free from caries when initial lesions were considered. During follow-up examination the number reduced to 20 and 2 respectively for DMFT and initial lesions.

It was observed that there was increase in follow-up mean values compared to baseline values for all the variables except for plaque scores, salivary *Streptococcus mutans* and *Lactobacilli* counts. Statistically significant difference was observed for dental caries with initial lesions and

bacterial counts whereas statistically significant difference was observed for dental caries, salivary secretion rate and buffering capacity between baseline and follow-up values (Table 1).

All the variables were dichotomized (0- acceptable value for each variable, 1- higher than acceptable value). When DMFT increment was considered, it was found that 7 children developed new carious lesions during 8 months. WPDmFT and WPDmFS were associated with caries increment (DMFT) after 8 months ($p=0.01$), (Table 2). When DMFS increment was considered, it was found that 9 children developed new carious lesions during 8 months. WPDmFT and WPDmFS were associated with caries increment (DMFS) after 8 months ($p=0.04$), (Table 3).

Table 4 and 5 represent analysis of various risk factors and caries increment (WPDmFT). Since many initial carious lesions remineralized during followup, three categories were made to assess caries status during follow-up for WPDmFT and WPDmFS. Salivary *Streptococcus mutans* counts alone showed a statistically significant association for caries increment (WPDmFT and WPDmFS).

Crude risk ratios with 95% confidence interval for salivary secretion rate, buffering capacity and streptococcus mutans count showed a possible association with new caries lesions (both DMFT and DMFS)- Table 6.

Discussion

The present study was conducted among 12 year old Belgaum children to find the association between caries increment and various risk factors. Out of 100 children 4 children could not be followed as they changed the school, thus the final sample was 96. There was no difference in the socio demographic characteristics among the study population (the data has not been presented in this article) 9. The age group of 12 year old was chosen as this is a WHO global monitoring age for dental caries. Only children with permanent dentition were selected in order to avoid discrepancies between mixed and permanent dentition with regard to microbial counts as stated by Schlagenhauf U et al 12. Children in the present study had relatively low dental caries expressed as mean DMFT although the prevalence was 76%. This is in accordance with study conducted by Mascarenhas AK who found that 22% of children were free of dental caries and mean DMFT and DMFS were 2.78 and 4.20 respectively 13.

Non cavitated incipient lesions (WP) were included in diagnosis of dental caries to avoid the possible underestimation of the disease 11. Studies by Manji and Tikwomwi; et al. demonstrated as high prevalence as 76% and mean DMFT of 2.97, and 60.4% (1.88), respectively, in 12 year old children when incipient lesions were included. When incipient lesions were excluded, the respective figures were 26.5% (0.55) and 23.1% (0.45) 14. Despite inclusion of incipient dental caries in the diagnosis, good intra examiner reliability was observed due to meticulous training and calibration of the examiner before conducting the examination. When DMFT+WP and DMFS+WP were compared no difference was observed. This could be attributed to underestimation of proximal lesions that might have occurred due to lack of, visibility and radiographic confirmation of these lesions. Inclusion of incipient lesion also made it feasible to observe changes in caries increment over short time period unlike cavitated lesions which require longer time.

Most of the children brushed once daily using tooth brush and tooth paste which could be attributed to educational messages through mass media and Indian culture of cleaning teeth in

the morning. Frequency of brushing was found to be positively associated with dental caries since brushing before going to bed is more crucial¹⁵.

There was increase in follow-up mean caries values compared to baseline values. This could be due to the fact that dental caries is a disease with a progressive character, the prevalence of which increases with age in any population independent of sex, urbanization and social status probably due to longer exposure time of the dentition to the etiologic factors of caries.

There were improvements in the salivary parameters and plaque scores. This may be due to more positive attitude of children towards saliva collection. As the children were already exposed to saliva collection procedures during baseline, they could chew wax comfortably without hesitation during follow up. This might have resulted in increased saliva secretion and buffering capacity and decrease in bacterial counts due to dilution. However the contribution of Hawthorne effect cannot be ruled out. (Hawthorne effect is a form of reactivity whereby subjects improve an aspect of their behavior being experimentally measured simply in response to the fact that they are being studied, not in response to any particular experimental manipulation.)

All the variables were dichotomized as 0- acceptable value for each variable and 1-higher than acceptable value. It was observed that atleast one third of the study subjects were in higher than acceptable value for all the variables. Bivariate analysis showed that WPDmFT and WPDmFS were associated with caries increment (DMFT and DMFS) after 8 months. Salivary *Streptococcus mutans* was other variable associated with caries increment (WPDmFT and WPDmFS). Various studies have proved initial caries to be the most valuable tool to identify children at high caries risk¹⁶⁻¹⁸.

Batchelor and Sheiham pointed out the limitations of the 'high-risk' approach for the prevention of dental caries. Nevertheless, in most developing nations caries is still a growing public health problem and in industrialized nations, where caries is no longer pandemic, groups of children remain at risk¹⁹.

Risk based preventive strategies, especially in developing countries may prove to be beneficial in optimum utilization of the scarce resources available for caries prevention. Though there is high prevalence of dental caries among Indian population there is a skewed distribution and hence risk based preventive strategies would yield promising results.

The results of the present study suggest initial caries to be the strongest predictor of future caries occurrence. However the lack of association with other variables could be due to small sample size and short follow-up period.

Conclusion

Initial caries was found to be the strongest predictor of caries occurrence in future. This study provides the basis to develop an affordable system for caries risk assessment where the clinicians can be encouraged to record initial lesion and provide appropriate preventive care.

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Table 1: Mean values of various risk factors at baseline and 8 months follow up

S.no	Variables	Baseline Values		After 8 Months Values		P values
		Mean	Std. Deviation	Mean	Std. Deviation	
1.	plaque scores	0.63	0.37	0.61	0.42	0.47
2.	DMFT	2.56	2.19	2.70	2.18	0.01
3.	WPDMFT	5.03	2.58	6.00	3.04	<0.001
4.	DMFS	3.26	3.49	3.44	3.49	0.01
5.	WPDMFS	5.86	3.63	6.98	4.43	<0.001
6.	Salivary secretion rate	0.78	0.63	0.97	0.77	0.01
7.	Salivary buffering capacity	2.97	0.61	3.20	0.80	0.01
8.	Salivary <i>streptococcus mutans</i> counts (represented in \log_{10})	5.25	0.28	4.92	0.23	<0.001
9.	Salivary <i>lactobacilli</i> counts (represented in \log_{10})	4.97	0.37	4.69	0.32	<0.001

Table 2: Bivariate analysis of various risk factors and caries increment (DMFT)

S.no	Variables		No caries increment	Caries increment (DMFT)	Chi square value	P value
	plaque	0	69	6	0.25	0.61
		1	20	1		
1.	DMFT	0	20	3	0.57	.44
		1	69	4		
2.	WPDMFT	0	2	2	5.6	.01
		1	87	5		
3.	DMFS	0	20	3	0.57	.44
		1	69	4		
4.	WPDMFS	0	2	2	5.6	.01
		1	87	5		
5.	Salivary secretion rate	0	30	2	0.07	0.78
		1	59	5		
6.	Salivary buffering capacity	0	4	0	--	1*
		1	85	7		
7.	Salivary <i>streptococcus mutans</i> counts	0	27	2	0.01	0.9
		1	62	5		
8.	Salivary <i>lactobacilli</i> counts	0	4	1	0.05	0.81
		1	85	6		

*- Fisher's Exact Test applied

Table 3: Bivariate analysis of various risk factors and caries increment (DMFS)

S.no	Variables		No caries increment	Caries increment (DMFS)	Chi square value	P value
1.	plaque	0	68	7	0.001	0.97
		1	19	2		
2.	DMFT	0	20	3	0.47	.48
		1	67	6		
3.	WPDMFT	0	2	2	3.88	0.04
		1	85	7		
4.	DMFS	0	20	3	0.47	.48
		1	67	6		
5.	WPDMFS	0	2	2	3.8	0.04
		1	85	7		
6.	Salivary secretion rate	0	30	2	0.13	.71
		1	57	7		
7.	Salivary buffering capacity	0	4	0	--	1*
		1	83	9		
8.	Salivary <i>streptococcus mutans</i> counts	0	27	2	0.3	.58
		1	60	7		
9.	Salivary <i>lactobacilli</i> counts	0	4	1	0.002	.96
		1	83	8		

*- Fisher's Exact Test applied

Table 4: Analysis of various risk factors and caries increment (WPDMFT)

S.no	Variables		Caries reversal	No caries increment	Caries increment (WPDMFT)	Chi square value	P value
1.	plaque	0	12	25	38	0.04	0.98
		1	3	7	11		
2.	DMFT	0	1	10	12	3.4	0.18
		1	14	22	37		
3.	WPDMFT	0	0	2	2	1.0	0.60
		1	15	30	47		
4.	DMFS	0	1	10	12	3.0	0.18
		1	14	22	37		
5.	WPDMFS	0	0	2	2	1.0	0.60
		1	15	30	47		
6.	Salivary secretion rate	0	4	12	16	0.56	0.75
		1	11	20	33		
7.	Salivary buffering capacity	0	1	0	3	2.0	0.35
		1	14	32	46		
8.	Salivary <i>streptococcus mutans</i> counts	0	3	18	8	15.5	<0.001
		1	12	14	41		
9.	Salivary <i>lactobacilli</i> counts	0	1	2	2	0.26	0.87
		1	14	32	49		

Table 5: Analysis of various risk factors and caries increment (WPDMFS)

S.no	Variables		Caries reversal	No caries increment	Caries increment (WPDMFS)	Chi square value	P value
1	plaque	0	13	28	34	0.92	0.63
		1	3	6	12		
2	DMFT	0	2	11	10	2.59	0.27
		1	14	23	36		
3	WPDMFT	0	0	2	2	0.95	0.62
		1	16	32	44		
4	DMFS	0	2	11	10	2.59	0.27
		1	14	23	36		
5	WPDMFS	0	0	2	2	0.95	0.62
		1	16	32	44		
6	Salivary secretion rate	0	5	14	13	1.5	0.47
		1	11	20	33		
7	Salivary buffering capacity	0	2	0	2	4.26	0.11
		1	14	34	44		
8	Salivary <i>streptococcus mutans</i> counts	0	3	18	8	12.9	0.002
		1	13	16	38		
9	Salivary <i>lactobacilli</i> counts	0	1	2	2	0.13	0.93
		1	15	32	44		

Table 6: Risk ratios for caries increment (DMFT and DMFS) during 8 months period.

S.no	Variables	DMFT Risk Ratio	95% confidence interval		DMFS Risk Ratio	95% confidence interval	
			Lower	Upper		lower	upper
1.	plaque	.57	.06	5.0	1.02	0.19	5.3
2.	DMFT	.38	.08	1.8	.59	.13	2.6
3.	WPDMFT	.05	.007	.49	.08	.01	.67
4.	DMFS	.38	.08	1.8	.59	.13	2.6
5.	WPDMFS	.05	.007	.49	.08	.01	.67
6.	Salivary secretion rate	1.2	.23	6.9	1.8	.36	9.4
7.	Salivary buffering capacity	1.08	1.0	1.1	1.1	1.0	1.1
8.	Salivary <i>streptococcus</i> <i>mutans</i> counts	1.08	.19	5.9	1.5	.3	8.0
9.	Salivary <i>lactobacilli</i> counts	.28	.02	2.9	.38	.03	3.8