

**STUDIES ON DETERMINATION OF SUSCEPTIBILITY TO DENTAL CARIES
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ABSTRACT

Dental caries is a multifactorial disease. The interactions of various factors like microorganisms, carbohydrate diet, hygiene habits and socioeconomic status are involved in causation of dental caries. For assessment of dental caries few tests demonstrating oral bacteria and their products have been accepted as these oral microflora and their products play an important role in causation and progression of dental caries. In the present study, susceptibility to dental caries was determined by Snyder test and Lactobacillus count. To study the risk factors associated with dental caries, a self designed questionnaire was distributed among the school children. About 84% children were found to be susceptible to dental caries. The Snyder test is more simple and rapid to perform as compared to Lactobacillus count in carrying out large surveys for assessment of dental caries. The study of other risk factors is also equally important in determination of risk of dental caries.

KEYWORDS

Dental caries, microflora, Snyder test, Lactobacillus count

INTRODUCTION

Dental caries is ubiquitous and is one of the most prevalent infectious diseases of human beings. It is a localized, progressive demineralization of hard tissue of the teeth.

Normally, many different types of bacteria live in the human mouth. They build up on the teeth in a sticky film called plaque. Human dental plaque harbors nearly 200-300 species of microorganisms¹. Dental plaque is a complex ecosystem with highly diverse acid tolerant & acid producing microorganisms. The fermentation of carbohydrates by acidogenic oral microorganisms is the key factor in the development of dental caries. Carious lesion results primarily from the dissolution of mineral in enamel and dentine due to acids released by these microbes². The primary acid tolerant bacteria associated with the dental plaque are *Streptococcus mutans*, *S.mitis*, *S.salivarius*, *S.sanguis*, *Lactobacillus acidophilus*, and *Actinomyces*³. Of these mutans, Streptococci are the most cariogenic pathogens as they are highly acidogenic producing short chain carboxylic acids which dissolve hard tissues like enamel and dentine⁴. In addition, they ferment sucrose and produce insoluble extra cellular polysaccharides which enhance their adherence to the tooth surface and encourage biofilm formation⁵.

Dental caries is the most important global oral health problem. Despite general advances in the overall health status of the people, the prevalence of dental caries in school children is up to 90%⁶. There is strong association between dental caries and systemic diseases like cardiovascular diseases, rheumatoid arthritis and osteoporosis⁷. The tooth loss due to poor periodontal health affects 20% of world's population and can lead to significant morbidity and premature deaths. The economic impact of oral diseases is an important consideration with up to 10% of public health expenditure in developed

countries related to curative dental care⁸. Early detection of individuals susceptible to dental caries is therefore important so as to institute special preventive programmes for them. This may help to reduce the prevalence of dental caries in school children.

OBJECTIVES

The present study was undertaken

- (1) To determine the susceptibility to dental caries among school children in Solapur city by Snyder test & Lactobacillus count.
- (2) To analyze various factors associated with susceptibility to dental caries which can help to plan the preventive strategies for population at greater risk for development of dental caries.
- (3) To compare the results of Snyder test with Lactobacillus count used for assessment of dental caries.
- (4) To assess the convenience of Snyder test over conventional method of Lactobacillus count for assessment of susceptibility to dental caries.

MATERIALS AND METHODS

In the present study school going children of Solapur in the age group 6-12 years from eight different schools were selected randomly. The schools were selected based on the socioeconomic status. About two hundred school children were included in the study.

For the assessment of various factors associated with susceptibility to dental caries, data were collected by self designed questionnaire. Questionnaire included 24 questions related to demographic profile, oral hygiene habits, dietary habits & health consciousness. School children were asked to fill up the questionnaire with the help of their

parents and to return back duly filled questionnaire.

To determine the susceptibility to dental caries, Snyder test⁹ and Lactobacillus counts were performed. For Snyder test, saliva sample was used. Consent for collecting saliva of school children was obtained from head master of respective schools. About 0.2 ml of saliva (collected in sterile tubes) was inoculated aseptically to a tube of liquefied Snyder agar (50⁰c). The tube was rotated between the palms of two hands to mix the contents. After the medium was solidified, the tube was incubated at 37⁰C for a period of 24-72 hrs. The results were categorized into three categories as 'marked,' 'moderate' 'slight' susceptibility to dental caries based on color change from green to yellow within a period of 24, 48, 72 hrs respectively [plate1] If color change was not observed after 72 hrs, test was reported as negative. [plate2]

Lactobacillus count was determined by spreading 0.1 ml saliva on selective Lactobacillus medium in duplicate (Lactobacillus selection Ox gall agar). The plates were incubated at 37⁰C in candle jar [plate3] for 48hrs. Following incubation colony

count of colonies exhibiting typical morphology of Lactobacillus was done [plate4]. Identification of Lactobacilli was confirmed by catalase test and gram staining. The count was expressed as number of colony forming units per milliliters [cfu/ ml] of saliva.

For determination of saliva flow rate, children were asked to chew paraffin cube for 5mins. Then the saliva was collected and measured. Data were analyzed by simple statistical methods like frequency, cross tabulation, mean, percentages, standard deviation and P value determination.

RESULTS

The susceptibility to dental caries among school children in the present study was highly alarming. About 84% children were found susceptible to dental caries. Females (86.84%) were found more susceptible than males (82.25%) as presented in table 1. The mean standard deviation of susceptibility was found 0.43 ± 0.23 and the P value was < 0.01 which is statistically significant.

plate 1



Tube 1: Uninoculated Snyder tube

Tube 2: No color change indicates little or no susceptibility to dental caries

Tube 3: Slight color change indicates mild susceptibility to dental caries

Tube 4: Significant color change indicates moderate susceptibility to dental caries

Tube 5: Complete color change indicates high susceptibility to forming dental caries

Plate - 1



Plate 2
showing color changes in Snyder agar inoculated with saliva samples after 72 hrs.



Plate 3
showing candle jar incubation.



Plate 4
showing colonies of Lactobacilli on Lactobacillus selection Oxgall agar medium

Table 1
Gender wise distribution of school children Susceptible to dental caries.

Gender	Susceptible		Total
	No.	%	
Male	102	82.25	124
Female	66	86.84	76
Total	168	84	200

The subject population studied was divided into two age groups viz. children having 5-10 years of age and 10-15 years of age. The children of 5-10 years of age group were found to be more susceptible to dental caries (Table 2)

Table 2
Age wise distribution of school children susceptible to dental caries.

Age group (Years)	Susceptible		Total
	No.	%	
5 – 10	55	85.93	64
10-15	113	83.08	136
Total	168	84	200

Based on Kuppaswami's socioeconomic status scale,¹⁰ school children were divided into five socioeconomic classes as upper I, upper middle II, lower middle III, Upper lower IV and Lower V. Children belonging to upper I Socioeconomic class were highly susceptible to dental caries followed by upper middle II, Upper lower IV, lower V and lower middle III class. (Table 3)

Table 3
Relationship between susceptibility to dental caries and Socioeconomic Status.

Socioeconomic Status	Susceptible		Total
	No.	%	
Upper I	31	96.87	32
Upper middle II	21	87	24
Lower middle III	15	65.21	23
Upper lower IV	96	83.47	115
Lower V	5	83.3	6
Total	168	84	200

The saliva flow rate of children was determined and found in between 0.01 – 2.0 ml / min. Saliva flow rate values were divided into four classes as shown in table 4. The school children with low saliva rate were found more susceptible to dental caries.

Table 4
Saliva flow rate wise distribution of Children susceptible to dental caries.

Saliva flow rate (ml / min)	Susceptible		Total
	No.	%	
0.0-0.05	120	85	141
0.05 – 1.0	43	80	54
1.0 – 1.5	01	-	01
1.5 – 2.0	04	-	04
Total	168	84	200

Questions regarding brushing frequency and dietary habits were asked. Children brushing once in a day were more susceptible than those who brush twice a day. Children having purely vegetarian diet were more susceptible to caries than children with mixed diet. The children consuming high carbohydrate diet (sugary diet) and having soft drink drinking habit were found to have more susceptibility to dental caries. (Table 5)

Table 5
Relation between oral hygiene habits, type of diet, candy eating habits (sugary diet), soft drink drinking habits & susceptibility to dental caries.

Habit		Susceptible		Total
		No.	%	
Brushing Frequency	Once	128	88.88	144
	Twice	40	71.42	56
Type of diet	Purely Vegetarian	94	86.23	109
	Mixed	74	81.31	91
Chocolate/ Candy eating habit	Frequent	130	84.96	153
	Occasional	38	80.85	47
Soft drink drinking habit	Frequent	30	85.71	35
	Occasional	138	83.28	165

Lactobacillus count was done and expressed as number of Lactobacillus colonies per millilitre of saliva [cfu/ml of saliva]. This count was found in between 0 to greater than 10,000. This Lactobacillus count was divided into five categories as shown in table 6

Table 6
Distribution of saliva samples showing Lactobacillus count and positive Snyder test.

Lactobacillus count (cfu/ml) of Saliva Sample	No. of Saliva Specimens of positive for Snyder test	Color change in Snyder agar medium after		
		24 hrs.	48 hrs.	72 hrs.
0	2	0	1	1
1-1000	4	0	2	2
1000-5000	10	0	4	6
5000-10000	22	2	10	10
> 10000	162	65	142	149
Total	200	67	159	168

As shown in table 6, at low lactobacillus count of saliva, number of positive Snyder tests for respective saliva samples were found to be very low. As the count of lactobacilli increased, number of positive Snyder tests for respective saliva sample was found to be increased.

Table 7
Correlation of Positive Snyder test and Lactobacilli Count / ml of Saliva.

No. of Lactobacilli / ml (CFU / ml)	No. of Saliva Samples studied	Snyder test positive after		
		24 hours	48 hours	72 hours
0 – 10000	38	2(5.2%)	17(44%)	19(50%)
> 10000	162	65(40%)	142(87.6%)	149(91.9%)

Thus, from table 7 it is observed that 5.2% saliva samples containing < 10,000 Lactobacilli / ml of Saliva induced positive color change in 24 hrs as contrasted to 40 % of saliva samples with counts > 10,000 Lactobacilli / ml. Thus, positivity of Snyder test increases with increase in Lactobacilli Count.

DISCUSSION

Dental caries is a multifactorial disease influenced by many factors including age, gender, micro-organisms, saliva flow rate, diet and oral hygiene. In the present study, females were found more susceptible to dental caries than males. This finding is consistent with study of Rehman et al., (2008)¹¹. High susceptibility to dental caries in female children can be attributed to number of facts including early teeth eruption in females in comparison to males, differences in

hormonal levels & financial dependence. Susceptibility to dental caries is more in age group 5-10 years than 10-15 years. This finding correlates with the studies of Wyne et al., (2002)¹². In the present study the finding of an inverse relationship between socioeconomic status & susceptibility to dental caries in children correlates with the study of Rehman et al., (2008)¹¹. This can be attributed to high consumption of sugary diet and faulty food habits in upper socioeconomic status. Oral care undoubtedly begins with good oral hygiene. Frequent brushing helps to remove cariogenic bacteria and fermentable substances. The continuous flow of saliva also reduces the cariogenic flora on the teeth. Saliva also acts as a buffering agent during the continuous acid production in oral cavity. In present study more number of children who brush once a day (88.88%) was found to be susceptible to dental caries than those who

brush twice a day (71.42%). Also, less salivation (0.5 ml / min) is found to be associated with more susceptibility to dental caries compared to more salivation (1.0 ml – 2.0 ml / min) as is observed with study done by Daniels et al., (1975)¹³.

Among the children studied with different dietary habits, the susceptibility to dental caries was high with vegetarian dietary habit (86.23%) than mixed diet (81.31%). This finding is consistent with the study by Shah et al., (2004)¹⁴. The protein rich non vegetarian diet maintains alkaline conditions. This might be the reason for less number of cases among non vegetarians.

The studies done by Rehman et al., (2008)¹¹ indicate a linear correlation of sugar consumption with susceptibility to dental caries in children. This observation supports the results of our present investigation. Sowers et al., (2006)¹⁵ found that consumption of carbonated drinks especially by children is a predominant cause of dental caries and

should be strictly discouraged. Findings in the present study are consistent with Sowers et al., (2006)¹⁵.

The present investigation shows that there exists close correlation between positivity of Snyder test and Lactobacillus count. Counts between 0-10000 Lactobacilli / ml of Saliva showed only 5.2% positive Snyder test in 24 hours for respective saliva samples while counts greater than 10000 Lactobacilli/ml of saliva showed 40% positive Snyder test. This is a ratio of 1:32 indicating that as Lactobacilli count increased, rate of positivity of Snyder test also increased. The positivity of Snyder test increased to 87.6% and 91.9% with Lactobacilli count greater than 10000 / ml of saliva after 48 hrs. and 72 hrs. respectively. These findings are similar to the findings revealed during earlier studies by Snyder et al., (1941)¹⁶. On the basis of present findings and previous studies by Marshall et al., (1950)¹⁷ Snyder test proves more effective in predicting caries activity as follows:

Positive Color change in 24 hrs	: marked caries susceptibility
No Color change in 72 hrs	: Negative Susceptibility
Positive Color change in 48 hrs	: moderate caries susceptibility
Positive Color change in 72 hrs	: Slight caries Susceptibility

The periodic testing of children by Snyder test would give the useful information to dentist as to whether individual is less caries active or is entirely in a state of caries activity. Accordingly the prophylactic measures also can be applied. Also, dentist would get an idea whether individual responds to the prophylactic measures or not.

The disadvantage of Snyder test is that positive color change of medium is due to acid production by not only Lactobacilli but also those organisms introduced into the medium by the inoculums of saliva. But if we consider dental caries as the result of demineralization by acid, then one can use Snyder test as a qualitative test to assess the possibility to dental caries. Though Lactobacillus count is accepted as standard method for assessing possibility of dental caries, it is laborious compared to Snyder test. Thus, the simplicity and rapidity of Snyder test proves its

usefulness in assessing possibility of caries activity during large Surveys.

CONCLUSION

Snyder test used in the present study for determination susceptibility to dental caries is very simple, economic and rapid test as compared to Lactobacillus count.

In the present study, 84% school children were found to be susceptible to dental caries. The study revealed an inverse relationship between socioeconomic status & susceptibility to dental caries among school children. Female children were found to be more susceptible to dental caries. Higher susceptibility was found in children of 5-10 years age group. Susceptibility to dental caries was higher in children with upper (I) & upper middle (II) class of socioeconomic status. From present study it is concluded that

oral hygiene, dietary habits and saliva flow rate play an important role in dental caries

development.

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