



ASSESSMENT OF 2D:4D RATIO AMONG 14-16 YEAR OLD SCHOOL STUDENTS WITH DIFFERENT GENETIC SENSITIVITY TO BITTER TASTE OF 6-N PROPYLTHIOURACIL – A CROSS SECTIONAL SURVEY

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ABSTRACT

Aims: To compare the caries experience and 2D:4D ratio of 14-16 year old school going students in Davangere city with different genetic sensitivity to bitter taste of 6-n propylthiouracil.

Methods and Material: A cross sectional survey was conducted among 350 school children aged 14-16 years. Based on PROP test children were categorized into tasters and non-tasters. Caries experience and 2D:4D ratio of left hand were recorded using Decayed Filled Missing teeth (DMFT) index and Vernier Calipers respectively. The categorical data was compared using Chi square test.

Results: The percentage of caries free children (DMFT=0) was significantly higher ($p=0.017$) among tasters (58.8%) compared to non-tasters (44.6%). There was no significant association between taste sensitivity and 2D:4D ratio ($p=0.98$) and caries experience and 2D:4D ratio ($p=0.84$).

Conclusions: Majority of tasters were caries free compared to non-tasters. There was no significant association between taste sensitivity and 2D:4D ratio and caries experience and 2D:4D ratio.

KEYWORDS : Dental caries, PROP test, Taste perception, 2D:4D ratio, 6-n propylthiouracil

INTRODUCTION:

Dental caries is one of the most prevalent infectious diseases to afflict mankind.¹ The development of dental caries is dependent on interrelationships among susceptible host/ tooth surfaces, specific oral bacteria and dietary carbohydrates.² Several studies have indicated that individuals with high sugar intake have higher caries rate which is positively related to sweet score and total sugar exposure.³ Various risk markers like salivary counts of *S. mutans* and lactobacilli, socioeconomic factors, past caries experience, frequency and amount of sugar intake have been evaluated to identify individuals at high risk for dental caries.

Inherited behavior and taste thresholds may play an important role in the frequency of carbohydrate intake.⁴ Genetic taste sensitivity dictates the fondness or refusal of food by children, who are accordingly categorized into tasters or non-tasters. The ability to taste bitter thiourea compound such as 6-n-propylthiouracil (PROP) is inherited. PROP tasters are more sensitive to many oral sensations, including bitter and sweet tastes and the sensation of fats. PROP status influences food selection and dietary habits in children.⁵ In clinical practice, PROP is a medication used in the treatment of Grave's disease (Hyperthyroidism).⁶ It is extremely bitter and can be tasted at a very low concentration and this property of PROP proved to be a useful tool in determining the genetic sensitivity levels to bitter and sweet taste⁷ which in turn appeared to be influenced by TAS2R38 gene.⁸

Recently, literature revealed a new biological marker- hormonal fingerprint to predict caries risk at a very early stage of life and found to be more stable, reproducible and consistent for each individual. The second to fourth digit ratio (2D:4D) is sexually differentiated in humans and men have lower 2D:4D than women due to men's greater relative length of the ring finger, in comparison with their index finger. It might be useful as a retrospective indirect marker for assessing taste perception. The probable mechanism is that the common genes HOX A and HOX D underlie the development of both digits and gonads, which in turn are known to influence the taste behavior in the human brain.⁹

In Dental literature, there is a lack of research that focuses on the link between taste perceptions, 2D:4D ratio and dental caries experience. So, a study is planned to compare the caries experience and 2D:4D ratio of tasters and non-tasters.

MATERIALS AND METHODS:

The present study was a cross sectional survey conducted among 350 school children of both sexes aged 14-16 years in Davangere city. The sample size was calculated scientifically using the formula,

$$n_A = \left(\frac{z_{1-\alpha/2} + z_{1-\beta}}{P_A - P_B} \right)^2$$

Type I (α) error was fixed at 0.05 and
Type II (β) error at 0.2

- $p_A = 0.3$ (proportion of tasters based on study done by Verma et al)⁹
- $p_B = 0.4$ (proportion of non-tasters based on study done by Verma et al)⁹

Overall sample size was estimated to be 350.

The study was conducted for a time period of three months. Permission to conduct the study was obtained from the Principals of schools in Davangere city. Ethical approval was obtained from the Institutional Review Board, Bapuji Dental College and Hospital, Davangere. Voluntary informed consent was obtained from the parents of study participants prior to the start of the study after informing about the research details through a participant information form. Subjects suffering from systemic diseases or/ on long term/recent/current regimen of medication that can affect taste perception; with known allergy/history of adverse reactions to propylthiouracil; with acute dental distress, requiring emergency dental treatment and subjects undergoing orthodontic treatment were excluded from the study.

Description of the proforma used for data collection:

A self-designed format was used to record all relevant data pertaining to study. It included provision for recording demographic details and categorization of students into tasters and non-tasters using Green's Labeled magnitude scale,¹⁰ dental caries experience and 2D:4D ratio.

Clinical Examination:

The subjects were made to sit comfortably on a chair with back rest and the oral cavity was examined under natural lighting condition. The entire study was conducted in school setup.

Prop Test:

Procurement and preparation of Prop impregnated filter paper: Pure samples of Prop was obtained from Sigma Aldrich Company, Dharwad. Sterile Prop strips containing 6-n propylthiouracil were prepared at Bapuji College of Pharmacy College, Davangere. Whatman filter paper was prepared in the required size of 2x2 cm and sterilized in an autoclave at 121°C for 15 minutes. These strips were pre-weighed and stored in desiccators. 10 mL of ethyl alcohol

was used to dissolve 10 mg/mL of PROP in a beaker. Ten autoclaved Whatman filter paper strips were soaked for a period of one hour to allow absolute absorption of PROP. These soaked strips were taken out of beaker and kept for drying at room temperature. The difference between the pre-weighed and the post-weighed strips provided the appropriate amount of drug infused on each strip, which was calculated at 1.6 mg of PROP for each strip.⁹

PROP test procedure:

PROP sensitivity was carried out by placing the filter paper containing approximately 1.6mg of 6-n-propylthiouracil on the dorsal surface of the anterior two-third region of the tongue for 30 seconds (Figure 1). Based on their ability to rate the intensity to bitter taste on a modified Green's Labeled magnitude scale (LMS) these children were subdivided into two groups as tasters (>15) and non-tasters (<15).¹¹

Assessment of Caries experience:

Caries experience was recorded using DMFT index based on WHO criteria.¹² For the purpose of analysis the sample was stratified into three categories based on DMFT values as done in previous study by PP Hegde et al.¹³ into following: DMFT- 0 (low risk); DMFT – 1-3 (medium risk) and DMFT>3 (High risk).

2D:4D Ratio assessment:

The ratio of the lengths of the index finger (2D) and ring finger (4D) of left hand was determined using Vernier calipers (Figure 2) and the study participants were categorized into high 2D:4D, low 2D:4D and equal 2D:4D digit ratio.⁹

Statistical Analyses:

Data obtained was compiled systematically in Microsoft excel spread sheet and a master table was prepared. Statistical analyses were performed using SPSS software version 20. Categorical data was compared using Chi square test. Significance level was kept at p≤0.05.

RESULTS:

In the study population of 350 school subjects, 54.6% were males and 45.4% were females (Table 1). The proportion of tasters was 60.3% and non-tasters was 39.7%.

A statistically significant difference was observed between gender and taste perception (p=0.005). Among tasters majority were male subjects and among non-tasters, majority were females (Table 2).

A statistically significant difference was observed between caries risk (categorized according to Decayed Missing Filled teeth scores) and genetic taste sensitivity (p=0.017). Percentage of caries free children were significantly high in taster group compared to non-taster group. However, no significant difference was observed between the proportion of medium caries risk and high caries risk groups between tasters and non-tasters (Table 3).

There was no significant difference (p=0.84) in the distribution pattern of 2D:4D ratio among tasters and non-tasters (Table 4). Also, there was no significant difference in the distribution of 2D:4D ratio among different caries groups (p=0.98) (Table 5).

TABLES:

TABLE 1: Distribution of study participants according to Gender

	Frequency	Percent (%)
Males	191	54.6
Females	159	45.4
Total	350	100

TABLE 2: Gender wise distribution of taster population

	Gender		Total n(%) of taster population	Chi square value	p value
	Male n(%)	Female n(%)			
Tasters	128 (60.67)	83 (30.33)	211 (60.3)	7.95	0.005*
Non Tasters	63 (45.32)	76 (54.67)	139 (39.7)		

*significant at p<0.05

TABLE 3. Distribution of caries risk among taster population

	Caries risk			Total n(%) of taster population	Chi square value	p value
	No caries risk n(%)	Medium caries risk n(%)	High caries risk n(%)			
Tasters	124 (58.7)	66 (31.2)	21 (9.9)	211 (60.3)	8.112	0.017
Non Tasters	62 (44.6)	64 (46.04)	13 (9.3)	139 (39.7)		*

Caries risk categorized as DMFT- 0 (low risk); DMFT – 1-3 (medium risk) and DMFT>3 (High risk)

*significant at p<0.05

TABLE 4. Distribution of 2D:4D ratio among taster population

	2D:4D ratio			Total n(% of taster population)	Chi square	p value
	Low 2D:4D ratio n(%)	Equal 2D:4D ratio n(%)	High 2D:4D ratio n(%)			
Tasters	131 (62.08)	33 (15.6)	47 (22.27)	211 (60.3)	0.348	0.84
Non Tasters	82 (58.99)	23 (16.5)	34 (24.4)	139 (39.7)		

TABLE 5. Distribution of 2D:4D ratio of study subjects based on caries experience.

	2D:4D ratio				Chi square	p value
	Low 2D:4D ratio n(%)	Equal 2D:4D ratio n(%)	High 2D:4D ratio n(%)	Total (%)		
Low caries risk	112 (60.2)	29 (15.5)	45 (24.1)	186 (100)	0.387	0.98
Medium caries risk	80 (61.5)	22 (16.9)	28 (21.5)	130 (100)		
High caries risk	21 (61.7)	5 (14.7)	8 (23.5)	34 (100)		

Caries risk categorized as DMFT-0 (low risk); DMFT – 1-3 (medium risk) and DMFT>3 (High risk)

DISCUSSION:

In the present study, the majority of the study subjects were tasters. This is in contrast with the study findings of Jyotirmai et al, where, majority of the study subjects were non tasters. This difference in the prevalence rates of the present study from previous studies may be attributed to the fact that exact comparisons with previous studies are not feasible due to: (a) different geographic distributions;(b) different methods of PROP strip preparation; (c) different concentrations of PROP strips used; (d) age span and sex discrepancies of the participants; (e) different cut-off scores for grouping taste status.¹⁴

Taste differences between females and males have been studied with conflicting results.^{15,16} As described for food intake, sex hormones could directly act in the central nervous system e.g. in the brainstem, while alternatively estrogens or progesterone may affect taste receptors or taste buds. In a study conducted by Jyotirmai et al, females were found to be more likely to be tasters than males.¹⁴ Perhaps the reason of this could be that women have more fungiform papillae and more taste buds than men as observed in a cross sectional study.⁷ In the present study, contrasting result was found where the proportion of tasters are more in males compared to females.

Students aged 14-16 years were included in the study as 15 years which is the mean age of 14-16 years is WHO index age group.¹² At this age, the permanent teeth are exposed to the oral environment for 3-9 years. Hence, assessment of caries experience in permanent dentition can be conveniently carried out in these age group children.

PROP has been documented in the literature in relation to the genetic sensitivity to expect the response of individuals through Green's labeled magnitude scale to bitter taste.¹⁰ Taste perception of humans vary according to their degree of taste sensitivity to phenylthiocarbamide (PTC) and its structurally similar drug referred to as PROP. Sensitivity of PROP test is known to be a reliable test in assessing this genetic sensitivity test to bitter taste, which is an inherent genetic feature.⁷ Hence PROP test was chosen to assess the taste thresholds of the subjects.

The 2D:4D ratio has been projected as a putative marker for prenatal hormone exposure as well as homeobox (HOX) and androgen receptor gene expression. The individual variability of 2D:4D is established in utero during the second trimester and appears stable during postnatal life. This evidence proposed that 2D:4D can be used as a useful indicator for assessing prenatal sex hormone action in the body, brain and behavior along with the taste perception.¹⁷

In the present study, the percentage of caries free children was significantly higher in tasters compared to non-tasters. The results of the present study are in accordance with similar studies^{18,19} where, there was increase in the overall caries experience in non-tasters compared to tasters, as the genetic ability to taste PROP decreased. The findings of this study can be attributed to the fact that taste perception plays a key role in determining individual food preferences and dietary habits.²⁰ Tasters prefer less cariogenic diet since they perceive taste at low concentration therefore, have decreased caries risk. Whereas in non-tasters, who perceive taste at high concentration are more inclined towards cariogenic diet and hence experience increased caries risk.

In the present study, no significant association was observed between taste sensitivity and 2D:4D ratio. This finding is in contrast with the study findings of Verma et al.⁹ CR Lakshmi et al,¹⁵ who reported 80% of non-tasters had low 2D:4D ratio whereas in the present study, only 59% of non-tasters had low 2D:4D ratio. The probable mechanism might be increased estrogen levels is associated with high 2D:4D ratio which is linked with increased caries risk as seen in non-tasters who are sweet likers and vice versa. Also, in females, increased estrogen/progesterone levels results in polyphagia (increased snacking between meals) and decreased sucrose threshold which increases the caries risk.²¹

In the present study, no significant difference was observed in the distribution pattern of 2D:4D ratio among different caries risk groups. There are no studies, which have tested the association between these two parameters, so comparison could not be made with other studies in this regard. High 2D:4D ratio is linked with individuals who are predominantly non tasters in the present study. Non tasters have increased estrogen levels which results in low sucrose threshold and hence increased caries risk.

In the present study, there was no significant relationship of genetic taste sensitivity and 2D:4D ratio and dental caries experience and 2D:4D ratio. So, future studies with larger sample size are needed to explore the association between these variables.

The present study has limitations as it used a cross sectional survey design, in which establishing association is difficult. Further studies with analytical study designs should be carried out to find the association between taste perception and dental caries risk; taste perception and 2D:4D ratio; dental caries risk and 2D:4D ratio. Also, the present study was conducted among 14-16 year old school students. The interventions based on PROP sensitivity might be more valid in children because studies have shown that in contrast

to children, adults' sweet predilection was not affected by genotyping of bitter receptors.²²

CONCLUSIONS:

The percentage of caries free children was significantly higher in tasters compared to non-tasters. There was no significant association between taste sensitivity and 2D:4D ratio and caries experience and 2D:4D ratio and future investigations on larger groups are needed.

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