Original Article



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Prevalence and Genetic Analysis of Bitter Taste Perception for Phenylthiocarbamide (PTC) Among Some Muslim Populations of Uttar Pradesh, India

Ruqaiya HUSSAIN, Ahsana SHAH, *Mohammad AFZAL

Human Genetics and Toxicology Lab, Section of Genetics, Department of Zoology, Aligarh Muslim University, Aligarh, Uttar Pradesh, India

*Corresponding Author: Email: afzal1235@rediffmail.com

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Abstract

Background: The ability to taste Phenylthiocarbamide (PTC), a bitter organic compound, described as a bimodal autosomal trait is widely used to know the heritable trait in both genetic and anthropological studies. The present study was carried out to analyze the prevalence of PTC taste sensitivity and to determine the gene frequencies among some Muslim populations of Uttar Pradesh, India. This study has some physiological relevance to highlight the adaptability of endogamous groups to behavioral traits in the same place.

Methods: Unrelated, healthy individuals of both sexes (Male-403, Female-418) belonging to different populations of Uttar Pradesh, India were randomly selected with the age range of 16-45 years observed for phenylthiocarbamide to taste sensitivity. PTC tasting ability was measured by using a serial dilution method of Harris and Kalmus.

Results: The phenotypic frequency of tasters was higher as compared to non-tasters, and the same is statistically significant ($\chi^2 = 11.92$, df = 5, P = 0.036). There were more females among tasters (67.94%) than males (64.76%). This observation was statistically significant ($\chi^2 = 14.79$, df = 5, P = 0.011).

Conclusion: The frequency of PTC tasters is greater than non-tasters and the females have lower non-taster phenotypes as compared to males. This type of study will provide background information about genetic structure of population and serves as useful interaction of genetics, food preferences and dietary patterns.

Keywords: Phenylthiocarbamide (PTC), Serial dilution method, Threshold distribution, Gene frequency, Indian Muslims

Introduction

Human population genetics aims to study the population in terms of genetic variation. This variation can be quantified by determining the gene frequencies of alleles at segregating loci which characterize one population and distinguish with another. Tasting ability to phenylthiocarbamide (PTC) by an individual is considered as a useful and important tool to study the genetic diversity in human populations. Taste and smell affects food preferences and dietary habits, thereby directly influencing the eating behavior of an individual. As taste threshold increases with age, abnormality in taste function may contribute to poor dietary intake in the elderly (1). Bitter taste perception is a conserved chemical sense against the ingestion of naturally toxic substances in mammals (2).

The experience of bitterness occurs after certain chemicals contact taste receptors located in cells on the surface of the tongue. Some investigators hypothesize that this sense provides information so that people do not ingest bitter-tasting toxic chemicals (3).

Studies of sensitivity to the bitter tasting anti thyroid compound, PTC have shown this to be an inherited trait and non-taster status has been linked to a variety of medical and health disorders (4). A high incidence of non-tasters has been reported among patients with nodular goiter (5-6), congenital athyreotic cretinism (7, 8), and dental caries (9).

Bitter perception generally occurs through bitter taste receptors located on the surface of taste cells of the tongue (10). These receptors are encoded by T2R genes that show 25–89% amino acid sequence identity between the 25 different members of this gene family. These differences presumably allow a wide variety of different chemical shapes, sizes, and functionalities to be bound by these receptors and perceived as bitter.

In humans, responses to some bitter compounds show a bimodal distribution that distinguishes two phenotypes, tasters and non-tasters. The beststudied example of these is the ability to taste PTC and other structurally related compounds (11).

The importance of the ability to taste bitter chemical compound PTC was realized by Fox (12). Thereafter, Synder (13) showed that the inheritance of the ability to taste PTC was dependent on a single autosomal dominant gene. PTC is a bitter tasting, harmless chemical compound (14), which is a member of a class of compounds known as "thioureas." These compounds are having the chemical group N-C=S, which is responsible for their characteristic bitter taste (15, 16).

Kim et al. (17) have identified a small region on Chromosome 7q, and harbours a gene that encodes a member of the TAS2R bitter taste receptor family. A major locus on chromosome 7q35q36 and a secondary locus on Chromosome 16p have been localized by genome scan for PTC taster gene (18).Tasters are those who taste the substance (PTC) while non-tasters cannot taste at all. Tasters have the genotype TT and the non-tasters have tt.

The ability to taste PTC is a dominant genetic trait, and the test to determine PTC sensitivity is

one of the most commonly used genetic tests on humans. The strong genetic basis for sensitivity to PTC has been used as a tool to trace family lineages and population migration patterns (11, 19). It was previously used in paternity testing before the advent of DNA markers (20).

A review of the literature reveals that human populations show a tremendous variation in the frequency of tasters which ranges from 10% to 98% (21-23). Among population groups of India, the frequency of taster allele (T) is higher among population groups of islands, followed by people in North and South India and is low in the people of West and Central India, as well as among scheduled tribes (24).

The aim of the present study was to analyze the gene frequency, relative fitness value and threshold distribution for phenylthiocarbamide among some Muslim Populations of Uttar Pradesh, India to show their genetic heterogeneity. Males and females of different populations have also been compared for the taste sensitivity of this trait. We also tried to find out the status of PTC tasters and non-tasters in different Muslim Populations of North India.

Materials and Methods

Populations

Muslims of India comprise more than 12% of the population, yet their genetic structure has not been well investigated. Muslims belong to two major sects: Sunnis and Shias, while each sect has different biradaries, which are grouped under Ashraf and Ajlaf (25).

The former comprise higher rank Muslims like Syeds, Sheikhs, Pathans and Mughuls while the latter comprise Qureshis, Ansaris, and Saifis. A large number of the Ajlaf may also be converted from local indigenous population of other faiths (26).

The Aligarh city in Uttar Pradesh is situated between latitude 27.28° to 28.10° North, and 77.29° to 78.36°

east longitude and its total area is 34.05 km². Aligarh has almost a dry climate throughout the year. The

annual average rainfall in the district is 594.1mms and maximum temperature recorded is 44 °C.

The population of Aligarh is composed of Hindus, Muslims and Christians. Hindus comprise Brahmins, Jats, Banias, Thakurs and Balmikis while Muslims comprise Syed, Sheikh, Pathan, Shia, Sherwani, Ansari, Saifi and Qureshi populations.

Sample collection

A cross-sectional, analytical and randomized study was done to find out phenylthiocarbamide taste sensitivity and threshold distribution among different Muslim populations of Aligarh district, from March 2011 to October 2011. Out of a total number of about 1124 individuals, only 821 individuals consented to participate in this study. Among them 400 were males and 421 were females. A survey was conducted among healthy, normal and unrelated individuals with the age range of 16-45 years which were randomly selected from six populations viz.; Syed, Sheikh, Pathan, Shia, Sherwani and Ansari. Households were selected by door to door contact with the help of volunteers. Each subject was interviewed before screening. His or her general particulars (address, age, sex, ethnic group) were recorded. The prior informed written consent was taken from the individuals and their parents before their inclusion in the study. The questionnaire was designed to collect information about food preferences, socio economic factors, health status and medical history of family. The samples were collected from the Upper Court, Civil Lines, Hamdard Nagar, Friends Colony, AMU Campus, Sir Syed Nagar and Jamalpur areas of Aligarh.

The method to distinguish tasters from non-tasters was adopted as per the sorting technique with serial dilutions of Harris and Kalmus (27), because of its superiority in discerning the threshold of the individual with near perfection.

A solution of 0.13% of PTC was prepared by dissolving 130 mg of the PTC in 100ml of water (solution 14). The serial dilution from 1 through 14 was prepared taking 50ml of solution and adding 50ml of distilled water to it to make solution 13 which is diluted as half 14. The last solution was most dilute and designated as solution no. 1. The dilution is used for noting the threshold value. At first subjects were asked to taste two drops of PTC solution. The dilution number when tasted positive was recorded. If an individual did not taste even the solution 14 (strongest), then he was designated as non-taster. Taster and non taster status was also entered into the proforma and presented in the form of tables. After the test, the participant was asked to spit out the chemical and to rinse the mouth with water. Threshold levels for PTC were then recorded for males and females of each population. The distribution of the frequency of tasters and non-tasters is usually bimodal with antimode recording the lowest frequency separates the two distributions. The anitmodal point was taken to classify the subjects as tasters or non-tasters.

Statistical analysis

The phenotypes were recorded for PTC taste sensitivity for each individual, and the allele frequencies were calculated according to Hardy-Weinberg law (28) using a gene counting method. The level of heterozygosity was calculated using the formula,

 $Hetrozygosity_{=} 1- \sum H_{o}$

Where Ho is the homozygosity of the allele, $H_0 = \sum Pi^2$

Chi-square test: It is used for the measurement of the size of the discrepancy between the observed and expected values at particular degrees of freedom.

 $\chi^2 = \sum (\underline{\text{Observed - Expected}})^2$ Expected

Results

PTC thresholds

A well-defined bimodal distribution of the taste sensitivity was observed in all the communities investigated as shown in (Fig. 1). Fig. 2 presents the threshold values among six populations which ranged from 8.29 to 9.59 in males, 7.76 to 9.03 in females and 8.09 to 8.95 as combined. The means and standard deviations of the thresholds for males, females and combined population groups were calculated as 9.08 ± 0.195 , 7.99 ± 0.163 , 8.51 ± 0.13 respectively.



Fig. 1: Graph showing threshold distribution of PTC trait among combined population groups of North India



Fig. 2: Graph showing of mean threshold values of males, females and combined populations for PTC trait of North India

The Pathan shows the highest threshold value (9.59 in males and 8.35 in females) while the Syed the lowest (8.47 in males and 7.76 in females) and the Sheikh, Sherwani, Shia and Ansari showed the intermediate threshold values. In overall population, it is interesting to note that the males show a higher mean threshold value than females.

Phenotypic frequency

Out of 821 subjects studied 545 (66.38%) were tasters and 276 (33.62%) were non-tasters to PTC. Among 400 males, 140 (35.00%) were non-tasters and among 421 females, 136 (32.30%) were nontasters, and this shows more males were observed to be non-tasters of PTC as compared to females. Again females were having more tasters (285, 67.70%) than males (260, 65.00%). Table 1 and Fig. 3 presents the phenotypic frequencies of tasters and non-tasters for the male, female and total combined populations. Table 2 shows the χ^2 values for PTC tasting ability among different populations. The phenotype frequency of taster showed that the percentage of taster was higher as compared to nontasters, and is statistically significant ($\chi^{2=}$ 11.92, df = 5, P = 0.036). It is observed that the highest phenotypic frequencies for PTC tasters were found among Syed (73.75% in males and 72.83% in females) while least among Ansari (52.33% in males and 61.04% in females). The highest non-tasters frequencies were found among Syed (26.25% in males and 27.17% in females) while least among Ansari (47.67% in males and 38.96% in females). We also observed that more females were PTC tasters than males which is statistically significant ($_{\gamma}2=31.20$, df= 5, P < 0.00001).

Allele frequency

Table 3 shows the allelic frequencies for PTC tasters and non-tasters among male, female and combined populations. Overall, allelic frequency for the nontaster (t) is 0.5798. Allele frequency for the nontaster (t) varies in different populations. The allelic frequency for the non-taster (t) for males and females are found to be 0.5916 and 0.5683 respectively. The highest allelic frequency for, non-taster (t) was found among Ansaris for being males and females 0.6904 and 0.6242 respectively. In the Syed population the t-allele frequencies for males and females were found to be least i.e. 0.5123 and 0.5212 respectively. The χ^2 difference in allelic frequencies for different population was non-significant because there is no rigid caste system in Muslims, only biradaries are there.

Populations	Male		Female		Combined	
_	Taster	Non-taster	Taster	Non-taster	Taster	Non-taster
Syed	73.75	26.25	72.83	27.17	73.25	26.74
Sheikh	68.92	31.08	66.27	33.73	67.51	32.48
Pathan	63.79	36.21	64.52	35.48	64.16	35.83
Sherwani	67.39	32.61	72.55	27.45	70.10	29.90
Shia	66.07	33.93	69.64	30.36	67.86	32.14
Ansari	52.33	47.67	61.04	38.96	56.44	43.55
$M \pm SE$	65.00 ± 1.66	35.00 ± 1.66	67.70 ± 1.63	32.30 ± 1.63	66.38 ± 1.64	33.62 ± 1.64

Table 1: Phenotype frequency for PTC tasting ability in North Indian Muslim Populations

M = mean and SE = standard error.

Table 2: The χ^2 values for PTC tasting ability in North Indian Muslim Populations

Populations	n	Male		Female		Combined	
-		Tasters	Non-tasters	Tasters	Non-tasters	Tasters	Non-tasters
Syed	172	59(0.3767)	21(2.37)	67(0.8900)	25(0.4275)	126(1.22)	46(2.42)
Sheikh	157	51(0.033)	23(0.5309)	55(4.59)	28(0.1523)	106(0.030)	51(0.060)
Pathan	120	37(0.0263)	21(0.0143)	40(0.066)	22(0.2261)	77(0.089)	43(0.175)
Sherwani	97	31(2.55)	15(0.1434)	37(0.3293)	14(0.2666)	68(0.202)	29(0.399)
Shia	112	37(0.066)	19(5.24)	39(3.70)	17(0.1295)	76(0.037)	36(0.072)
Ansari	163	45(0.849)	41(6.27)	47(1.62)	30(0.3333)	92(2.43)	71(4.79)
Total	821	260	140	285	136	545	276

Parentheses=chi-square value (χ^2) and n =number of individuals.

 $\chi^2 = 31.20$, df = 5, *P*<0.00001(statistically significant at *P*<0.05).

Table 3: Allele frequency for PTC tasting ability in North Indian Muslim Populations

Populations	Male		Female		Combined	
	Т	t	Т	t	Т	t
Syed	0.4877	0.5123	0.4788	0.5212	0.4829	0.5171
Sheikh	0.4425	0.5575	0.4192	0.5808	0.4301	0.5699
Pathan	0.3983	0.6017	0.4043	0.5957	0.4014	0.5986
Sherwani	0.4289	0.5711	0.4752	0.5248	0.4532	0.5468
Shia	0.4175	0.5825	0.4490	0.5509	0.4331	0.5669
Ansari	0.3096	0.6904	0.3758	0.6242	0.3401	0.6599
Total	0.4084	0.5916	0.4317	0.5683	0.4202	0.5798

T and t are dominant and recessive alleles respectively.

Heterozygosity

Table 4 and Fig. 4 present the homozygosity and heterozygosity among male, female and combined population groups. The pooled heterozygosity for males and females is found to be 0.4832 and 0.4907 respectively. The highest heterozygosity was found among Syed i.e. 0.4994 and the least among Ansari i.e. 0.4489. All the population groups showed the little variation in observed heterozygosities. Overall, heterozygosity for combined population group was found to be 0.4873.

Relative fitness value (w)

Fitness values of different traits from the total population are presented in (Table 5). The highest fit genotype is taken as having fitness value of one, and the fraction of fitness of each genotype with fitness of highest trait is taken as relative fitness of the other genotypes Ayala (29). The highest fitness value of T trait was found among Syed (1.0) followed by Sherwani (0.96), Shia (0.93), Sheikh (0.92), Pathan (0.88) and Ansari (0.77).







Fig. 4: Graph showing Heterozygosity and Homozygosity for PTC tasting ability among different Muslim Populations of North India

Populations	Male		Female		Combined	
_	H_t	Ho	H_t	Ho	H_t	Ho
Syed	0.4997	0.5003	0.4991	0.5009	0.4994	0.5006
Sheikh	0.4934	0.5066	0.4869	0.5131	0.4902	0.5098
Pathan	0.4793	0.5207	0.4817	0.5183	0.4806	0.5194
Sherwani	0.4899	0.5101	0.4988	0.5012	0.4956	0.5044
Shia	0.4864	0.5136	0.4947	0.5053	0.4910	0.5090
Ansari	0.4275	0.5725	0.4691	0.5309	0.4489	0.5511
Total	0.4832	0.5168	0.4907	0.5093	0.4873	0.5127

Table 4: Heterozygosity and Homozygosity for PTC tasting ability in North Indian Muslim Populations

 H_{t} and H_{O} represents heterozygosity and homozygosity respectively.

Table 5: Relative fitness value (w) for PTC tasting ability in North Indian Muslim Populations

Populations	Male (w)		Fen	Female (w)		Combined (w)	
	Tasters	Non-tasters	Tasters	Non-tasters	Tasters	Non-tasters	
Syed	1.0	0.55	1.0	0.69	1.0	0.61	
Sheikh	0.94	0.65	0.91	0.87	0.92	0.75	
Pathan	0.87	0.75	0.89	0.91	0.88	0.82	
Sherwani	0.91	0.68	0.99	0.71	0.96	0.69	
Shia	0.89	0.71	0.96	0.78	0.93	0.74	
Ansari	0.71	1.0	0.84	1.0	0.77	1.0	
Total	0.88	0.73	0.93	0.83	0.91	0.77	

(w) Represents relative fitness value.

Discussion

The sense of taste is a powerful predictor of food selection. Human infants show an innate pleasure response to sweet taste, but dislike bitterness and reject bitter-tasting foods (30). Whereas sweetness serves as a sensory cue for energy rich foods, bitterness often predicts toxicity (30).

The human sense of taste can be categorized into five basic tastes sweet, bitter, sour, salty and umami, that are critical for nutrition and survival. Bitter perception has a particularly significant role, as it protects us from ingesting naturally toxic substances which are typically bitter in taste (3).

The best known example of variation in sensitivity to a bitter compound is that of phenylthiocarbamide (PTC). In 1931 Fox (12) observed that to some individuals the simple chemical compound phenylthiocarbamide (PTC), has an intensely bitter taste, while to others it is tasteless. Harris and Kalmus (27) found that the distribution of PTC tasting thresholds was bimodally distributed, but there were some intermediate individuals. The major gene TAS2R38 on chromosome 7 responsible for this trait was identified as a member of the TAS2R bitter taste receptor gene family consisting of a single coding exon 1002 bp long, encoding a 333 amino acid, 7-transmembrane domain G protein- coupled receptor (17), that responds to bitter stimuli (31-35) and the milestones of this discovery have been summarized (36). Bufe et al. (31) demonstrated that alleles of hTAS2R38 codes for functionally different receptor types that directly affect perception of bitterness containing compounds. Two major forms of this bitter receptor gene were identified in most of the world's populations, designated as the 'major taster' form and the 'major non-taster' form. These two forms differ in 3 amino acid positions, numbers 49, 262, and 296 (37).

The ability or inability to taste the PTC is a classic inherited Mendelian trait that has long been known to vary in human population. This trait is of genetic, epidemiologic and evolutionary interest and has been shown to correlate with a number of dietary preferences and thus have important implications for human health (31, 38, 39).

The main objective of this study was to investigate the threshold distribution and determine the gene frequencies of dominant and recessive alleles for PTC taste sensitivity among different Muslim populations of Uttar Pradesh, North India. The present study provides brief information on the distribution of PTC tasters and non-tasters in different populations of North Indian Muslims. Some studies on Muslim populations have been attempted earlier in Uttar Pradesh (40). These are described here for comparison. The present work shows some differences from earlier studies on PTC taste blindness among Muslim populations of North India.

Data on PTC taste ability is vast and a great deal of work has been done over the world. Many studies have reported that in world population, approximately 30% of them are PTC non tasters and 70% are tasters (11, 41). The prevalence of taste blindness or an inability to taste bitter chemicals ranges from 3% in West Africa, to 6-23% in China, 40% in India and 30% in US population (11, 42). The average frequency of t allele among Indian populations is 53.4 percent (varies from 8.8% among scheduled caste of Andhra Pradesh to 89.2% in Munda of Ranchi, Bihar) while in European populations it varies from 25 to 57% which is little higher but similar to that of South west Asian (43). For PTC taste ability the overall frequency of allele t for Muslims is 51.3% (44), which is 57.98% in our case. For the Ansari from Bihar, the frequency of the t allele is 71% (26), which is 65.99% in the present study. Aarzoo and Afzal (45), reported that the overall frequency of allele t is 58% for Muslims which is similar to the present data.

In our study we found the percentage frequency of 33.62% for non-tasters and the allele frequency t, as 0.5798. Singh and Singh (46), observed percentage frequency of 31.3% and allele frequency t as 0.56 among the Muslims of Ambedkar Nagar District of Eastern Uttar Pradesh, North India. Our data are almost similar to this study. The value of non-taster allele *t* varies from 0.44-0.56 among Muslims of Eastern Uttar Pradesh (46). In India the frequency of t-allele shows variation in different ethnic groups and castes (45, 24). In the present study the overall frequency of tasters was 66.38% and it varies in different populations of India (47).

The frequency of allele T among Indian population is 45.7% (48). In general frequency of allele T is higher among population groups with Mongoloid affinities from the Himalayan region but lower from the Mongoloid populations from the East and Southeast Asia and lowest among Scheduled tribes (24). In the present study the frequency of allele T was found to be 42.02%. The frequency of taster (T) is about 0.50 among European populations (5).

In the present investigation the frequency of T allele ranged from 0.34-0.48 and the non-taster allele t ranged from 0.52-0.65 among Muslims of Uttar Pradesh, North India. Our observations on PTC taste perception revealed that there is significant higher percentage frequency of tasters as compared to non-tasters among different populations in the present study which supports other studies (11, 41, 34). In the present study, we found the ratio of non-tasters among males was more than females as reported by others (24, 49). Previous studies suggest that the frequency values for non-taster of PTC may be unique to the specific populations studied (50). We found that females are more PTC tasters as compared to males which is in total conformity with other studies (50, 51). PTC taste thresholds vary in different populations and females are found to taste PTC at lower concentrations of thresholds than males, a small number of specific differences in taste ability have long been known and well-studied (11).

The present study revealed genetic affinities among different populations. It also provides a clear cut distribution of PTC taster and non-taster phenotypes among different Muslim populations of North India.

Acceptance and rejection of bitter fruits and vegetables, as well as sweet foods, added fats, spicy foods and alcoholic beverages had an association with PTC/PROP taste sensitivity (52-54). People who can taste PTC (taster) are more sensitive to salt, sweet foods, sharp tasting foods and spicy foods (55). Anatomical studies reported that tasters actually have more taste buds than non-tasters (56). Keller et al. (57) suggested that non tasters prefer high fat diet more, than low fat diet, while tasters show this lack of preference for food.

The PROP/PTC tasters also have other oral sensations for example, the PROP tasters perceive more fattiness in foods whereas non-tasters cannot discriminate, between high and low fat containing foods and consequently be more obese (more weight gain/higher body mass). Hence, the variation in genetic taste sensitivity may also play important role in dietary patterns and food habits. Aside its importance in genetic and anthropological studies, PTC taste sensitivity has been shown to be important in food selection, which may affect individual metabolism and physiology (58). The effect of genetic factors on food selection has acquired a new importance in cancer research. If PTC and PROP taster status helps to identify food preferences and food consumption, it might turn out to be a genetic marker for some of the major diet related chronic diseases (16).

On a larger scale, the PTC gene may be illustrative of ancient genetic variation that has been proposed to underlie common disease in modern populations (59). In addition, the mapping of the PTC genes will provide a powerful tool to examine the genetic basis for food preferences and the relationship between taste status and health outcomes (11). Finally, PTC presents a unique opportunity in the field of bitter taste transduction. Having a known gene with a strong effect on phenotype in vivo provides many opportunities for studies of taste physiology, biochemical function, and the molecular structure elucidation in the human sense of taste. The present paper reports the threshold distribution and gene frequencies of PTC taste sensitivity among different Muslim populations of North India. We also tried to find out the status of PTC tasters and non-tasters among these populations. Our observations reveal that the percentage of PTC tasters is greater than that of non-tasters and the, females have lower non-taster phenotypes than males. Although PTC itself has not been found in nature, the ability to taste PTC is strongly correlated with the ability to

taste other naturally occurring bitter substances, many of which are toxic (60, 55). Such studies have a great significance in understanding the adaptability of the populations to the same region which results in their varying response to threshold of sensitivity of the same genetic trait (61-63).

Conclusion

Females have higher non-taster phenotypes than males. Further, the adaptive fitness of tasters is higher than that of non-tasters for all phenotypes, which speaks of adaptive value of taster phenotype. Therefore, understanding the nature of the variation in bitter taste perception and its relationship to diet and other behavior aspects may have important implications for human health.

Ethical considerations

Ethical issues (Including plagiarism, Informed Consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

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