# Screening For Phenylthiocarbamide Taste Sensitivity Response Status On Apparently Healthy Subjects: A Pilot Study

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ABSTRACT

The dynamics of studying genetic variation in human population using phenylthiocarbamide has contributed immensely in influencing our knowledge of variations and diversity globally. The pilot study aims to screen for the taste sensitivity response status using different concentrations of a stock solution of 0.10% phenylthiocarbamide. 232 subjects were randomly recruited using a drawn questionnaire. 84.9% were tasters while 15.1 % were non-tasters. Of the 133 males, 52.3% were Tasters while 85.7% were non-Tasters. The 99 females, constituted 47.7% Tasters and 14.3% non-tasters. The Chi-Square analysis on gender was significant p-<0.000. The result of the different age range showed a decrease in the population of tasters with increase in age and a reverse among non-tasters. The phenotypic status of the age range of subjects was not significant p> 0.433. The combined allelic frequency of tasters and non - tasters was 0.61 and 0.39. Similarly, the individual allelic frequencies of male and female tasters were 0.52 and 0.78 while male and female non-tasters were 0.48 and 0.22, respectively. The pooled homozygous and heterozygous outcome for males was the same (0.50), but females were 0.66 and 0.34. The homozygous and heterozygous outcome of the combined population was 0.52 and 0.48. The highest cases of tasters were recorded at a threshold of 0.025 and 0.50 with female subjects responding most. The study provides insight to the distribution of the Phenylthiocarbamide phenotype, the heterozygous, homozygous and allelic frequencies of the population, which may serve as an index for directing dietary choice or diagnoses by health care professionals.

Key words: Phenylthiocarbamide, response status, taste sensitivity, phenotype

## Introduction

The use of genetic characteristics is essential in understanding the dynamics of variations in human population (My 46 Trait Profile, 2019). Human genetic variation denotes the genetic differences in and among populations (Hussain *et al.*, 2014). Today, many polymorphic genetic variants are used for better understanding of human diversity (Padmavathi, 2013). These multiple markers of any given gene in the human population (alleles), are referred to as polymorphism (Fareed *et al.*, 2012; Emerson, 2016). These

variations can be quantified by determining the gene frequencies of alleles at segregating loci that characterize one population and distinguish from another (Alimba *et al*., 2010).

Phenylthiocarbamide (PTC) otherwise called Phenylthiourea, is one of the most commonly used genetic taste sensitivity conducted on human populations over the years (Fareed *et al.,* 2012; Genetic Science Learning, 2016). It is an organic compound made up of phenyl ring with a molecular weight of 152.218g/mole (Daştan *et al*., 2015a, b). Analysis of the taste sensitivity of PTC as well as some related substances is genetically controlled and is governed by a pair of alleles, Dominant 'T" for tasting and recessive 't' for non-tasting. There are therefore two common forms of allele of the PTC gene and at least five rare forms (Robert *et al*., 2008). Individuals possessing genotypes "TT" and "Tt" are tasters and persons with genotypes "tt" are non-tasters respectively (Padmavathi, 2013).

The shape of the receptor protein determines how strongly it can bind to PTC, because all individuals possessing two copies of every gene combination of the bitter taste gene variants determines whether one will find PTC intensely bitter, somewhat bitter or without taste at all (Eriksson *et al*., 2012; Keller and Adise, 2016). The inability to taste certain compounds has long been associated with simple recessive Mendelian inheritance, and a large number of taste and odorant receptors have been cloned and sequenced in the last few decades to establish the molecular basis of these traits (My 46 Trait Profile, 2019; Rahim *et al.,* 2018; Harem, 2019).

Many studies have been conducted to obtain data related to PTC taste perceptions at population levels (Risso *et al*., 2016; Beauchamp and Jul, 2017; Roper *et al* 2017) and compared to disease cases and other various personal characteristics related to taste perception (Melis *et al.,* 2019). It has been established that differences exist between tasters and non-tasters of PTC in terms of their life styles and susceptibility to certain diseases such as goiter, diabetes, peptidic ulcer, depression, alcoholism and schizophrenia (Cardullo, and Holt, 1951; Bachmanov *et al.,* 2014; Dastan *et al*., 2015a). Other studies have also linked PTC taste sensitivity to dietary choices (Bachmanov *et al.,* 2014; Igbeneghu *et al*., 2014; Igbeneghu *et al., 2016;* Keller and Adise; 2016). Strong genetic basis for sensitivity to PTC has also been used as a tool to trace family lineages, population migration patterns and in paternity testing before the advent of DNA markers (Hussain et *al*., 2014 ; Robert *et al*., 2008). These studies have indeed revealed that variations exist in all human populations ranging from 10% to 98% (Hussain *et al*., 2014; Risso *et al*., 2016).

In our perspective therefore, there are paucity of reports on PTC sensitivity taste in Nigerian population. The few studies acknowledged, reported for the south west geo political zone of the country (Bakare *et al*., 2009; Alimba *et al.,* 2010; Igbenehu *et al*., 2016). The aim of this study therefore was to screen for the taste sensitivity response status of PTC in the multi-ethnic society that makes up the Middle Belt geo pollical zone of Nigeria. This study provided an insight to the distribution of the PTC phenotype and allelic frequencies in the sampled population and highlighted the implication on medical thrust of the people.

## Materials and Methods

### Study design

The participants were adequately briefed on the aim of the study and their expectations as subjects before they were recruited. A total of 232 apparently healthy individuals comprising of male and female, aged 16 to 30 years, who were not on any viral or bacterial drugs at the time of test were recruited for the study using a drawn questionnaire. The subjects recruited met the enrollment criteria and consented to participate in the study either personally or through their parents or relations

**Study area**

This study was carried out in a cohort in Nasarawa State, Nigeria located on coordinate- latitude 8.8471$^{o}$N and longitude 7.8776$^{o}$E on the Northern and Eastern hemisphere (Akwa *et al* ., 2007). Nasarawa State has a population of about 1.8 million people. It is an extremely diverse multiethnic groups comprising of about 29 languages, some of which include Mada, Aguta, Alago, Basa, Ebira, Eggon, Gbagyi, Gwandara, Kanuri and Tiv .

**Ethical clearance**

The ethical clearance for the study was obtained from the ethics and grievances committee of the Federal Medical Center, Keffi while individual subjects provided written informed consent either personally or by their parents or relations to participate in the study. Ethical clearance approval: NHREC/21/12/2012, FMC/KF/0040/21.

**Phenylthiocarbamide Test**

A stock solution containing 0.10% phenylthiocarbamide was prepared in distilled water and serial dilutions were made to obtain five concentrations in mg/ml (0.005, 0.05, 0.25, 0.50, and 1.0). Distilled water was used as control. Taste sensitivity to PTC was ascertained using a filter paper impregnated with the different dilutions of PTC. The impregnated paper was administered starting with the weakest PTC concentration in the order of increasing concentrations for between 20 to 30 seconds.

The serially diluted concentrations of PTC were used as points to determine the genotypes of PTC sensitivity taste intuitively (Stern, 1943; Asaad et al., 2014). Individuals whose sensitivity taste score was 0.005 and 0.05, or 0.05 or 0.50 and 1.0 were regarded as 'TT', or 'Tt', 'tt' respectively. Participants were provided with water after each successive step of PTC application to rinse their mouths. Subjects who could not taste the impregnated paper at the lowest dilution concentration, were designated as non-tasters. The threshold levels of the respondents were then recorded for the sampled population (Genetic Science Learning, 2016).

**Gene frequency:**

The Gene frequencies of the population were determined using intuitive approach and calculated by Hardy–Weinberg method to determine their heterozygosity (Stern, 1943; Rushell, 1998).

**Statistical analysis**

The Sofastats 1.5 software (https://www.sofastatistics.com) was used to determine the frequencies and percentages of the population while the correlation analysis was performed between individual characteristics and PTC status using SPSS version 23.0 software package (www.ibm.com).

**Results**

Table 1: shows the phenotypic frequency distribution of the population according to the ability of subjects to taste phenylthiocarbamide. The study population was 232 subjects, 197 (84.9%) were tasters while 35 (15.1 %) were nontasters. The population comprised of 133 males out of which 103 (77.44%) were Tasters while 30 (22.56%) were non-Tasters, while among the 99 females, 94 (94.9%) were Tasters and 05(5.01%) non-tasters. The Chi Square value of this analysis on gender was 13.577 and a significant p-Value of $p<0.000$ ($p<0.005$).

Table 1: Phenotypic Frequency of the population sampled

| **Parameters** | **Phenylthiocarbamide Phenotype** | | **Total** | **Chi Square** | **p-Value** |
|---|---|---|---|---|---|
| | Tasters | Non-Tasters | | | |
| Population | 197 (84.91%) | 35 (15.08%) | 232 | | |
| Male | 103 (77.44%) | 30 (22.56%) | 133 | 13.577 | 0.000* |
| Female | 94 (94.9%) | 05 (5.01%) | 99 | | |

The result in table 2 shows the distribution of the sampled population based on the different age range of subject. The age range 16-20 years recorded (86.7%) of the population as tasters and (13.3%) as nontasters. This was followed by the age 21-25 years with (81.1%) tasters and (189%) nontasters. The last age group, 26-30 years had a population of 76.9% tasters and (23.1%) non tasters respectively. Whereas there was a decrease in the population of tasters with increase in age an inverse outcome was experienced among the nontasters where a corresponding increase was recorded. On the whole the result of the phenotypic status of the population showed a chi square value of 1.675 and a non-significant value of $p> 0.433$ ($p<. 0.005$)

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Table 2: Frequency distribution of subjects according to age range

| PTC Status | Age Group of Participants | Pearson Chi- | P- Value |
|---|---|---|---|

| | | | | Square | |
|---|---|---|---|---|---|
| | 16-20 | 21-25 | 26-30 | | |
| Tasters | 86.7%$_a$ | 81.1%$_a$ | 76.9%$_a$ | 1.675[a] | 0.433 |
| non-Tasters | 13.3%$_a$ | 18.9%$_a$ | 23.1%$_a$ | | |

Each subscript letter denotes a subset of age group _sub categories whose column proportions do not differ significantly from each other at the .05 level.

**Figure 1** shows the allelic frequency of the PTC tasting ability amongst the sampled population. The combined allelic frequency for non - tasters was 0.39 while tasters was 0.61. The allelic frequencies of male and female non-tasters were found to be 0.48 and 0.22, while for male and female tasters was 0.52 and 0.78 respectively.

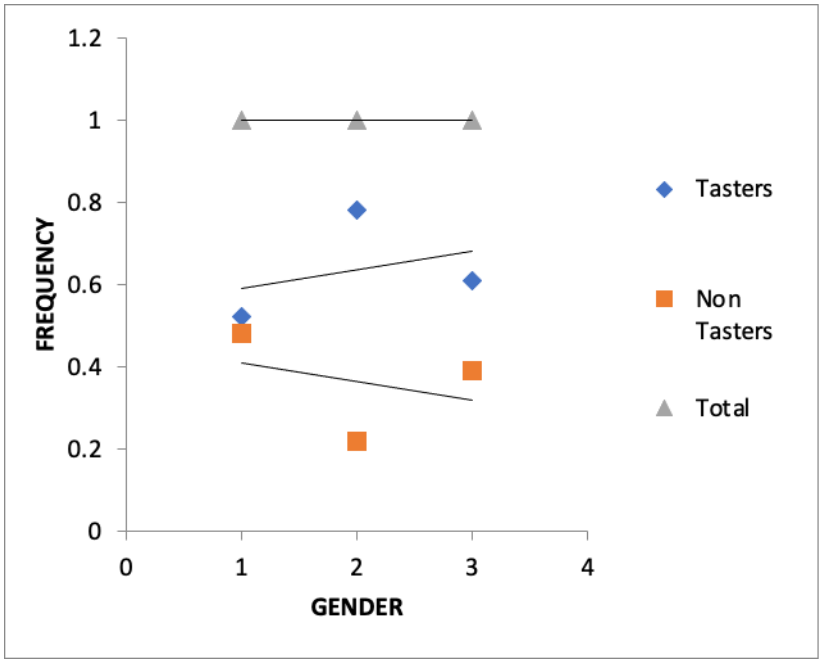



Figure 1: Allelic frequency for PTC tasting ability in sampled population.

Table 3 revealed the homozygous and heterozygous outcome of PTC taste sensitivity of the subjects. The pooled homozygous and heterozygous outcome for

males was the same (0.50), while that of the females was 0.66 and 0.34 respectively. On the whole, however the homozygous and heterozygous outcome for the combined population was found to be 0.52 and 0.48 respectively.

Table 3: Homozygosity and Heterozygosity for PTC tasting ability of sampled population

| **Phenotype** | **Males** | **Females** | **Total** |
|---|---|---|---|
| Homozygosity | 0.50 | 0.66 | 0.52 |
| Heterozygosity | 0.50 | 0.34 | 0.48 |
| Total | 1.00 | 1.00 | 1.00 |

**Figure 2** shows the percentage distribution of PTC taste sensitivity at different concentrations (mg/ml) amongst the subjects. The highest cases of tasters were recorded at concentrations (0.025 and 0.50) which were the same (100%) for females and 78.9 and 96.2% respectively for males.

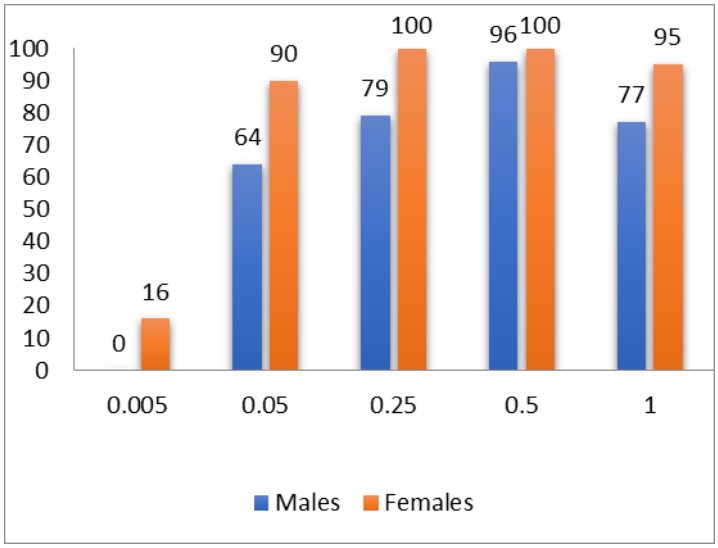



Figure 2: The frequency distribution of PTC taste sensitivity at different concentrations (mg/ml).

**Discussion**

Taste recognition is mediated by specialized taste cells that communicate with several brain regions through direct connections to sensory neurons. Taste perception is a two-step process. First, a taste molecule binds to a specific receptor on the surface of a taste cell. Then, the taste cell generates a nervous impulse, which is interpreted by the brain. For example, stimulation of "sweet cells" generates a perception of sweetness in the brain. Recent research has shown that taste sensation ultimately is determined by the wiring of a taste cell to the cortex, rather than the type of molecule bound by a receptor (Roper and Nirupa, 2017). As a result, if a bitter taste receptor is expressed on the surface of a "sweet cell," a bitter molecule is perceived as tasting sweet (Bachmanov *et al.,* 2014).

In this study, we did considered variation in taste sensitivity response to the bitter compound phenylthiocarbamide (PTC) to determine the existing Mendelian trait in the sampled population. Of the population sampled, our findings revealed a greater number of tasters to non-tasters. This outcome is similar to a common position revealed by other scholars (Fareed *et al*., 2012; Shivaprasad *et al.,* 2012; Daştan, *et al.,* 2015a; My 46 Trait Profile, 2019; Mairiga *et al.,* 2021). In their studies, Hussaini *et al.,* (2014) had 545 (66.38%) subjects out of 821 who were tasters while 276 (33.62%) were non-tasters. In like manner, out of a population of 2,500 who volunteered to participate in their studies, Dastan *et al*., (2015a) reported that the ratio of tasters to PTC were 2,045 (81.8%) while 455 (18.2%) were non-tasters. Shivaprasad *et al.,* (2012) also obtained a high frequency of tasters (61.41%) to non-tasters (38.58%) in their study.

Several other studies on Nigerian subjects have reported high frequencies of tasters to non-tasters in their reports (Bakare et al., 2009; Alimba et al., 2010; Igbeneghu et al., 2014). In their previous studies involving general population, Igbeneghu et al., (2016) recorded 76 (71.0%) and 31 (29.0%) of the 107 control subjects as tasters and non-tasters respectively. Similarly, they found also that 17 (32.7%) of the 52 males and 14 (25.5%) of the 55 females in the control group were non-tasters. They also revealed that the percentage of non-tasters in the test group (54.0%) was significantly higher than that of the non-tasters in the control group (29.0%). This implies that non-tasters of PTC were significantly more associated with tuberculosis patients than control subjects.

Even at this, the PTC perception frequencies obtained from the subjects in our study are slightly higher than the results obtained by them. It therefore beholds that there is a significant higher incidence of PTC tasters than non-tasters among general population, but with peculiarities.

In comparing PTC sensitivity response status based on gender, more of male and female subjects were tasters than non-tasters in our findings. This is similar to the report of Shivaprasad *et al.,* (2012), Hussaini *et al.,* (2014); and Daştan, *et al.,* (2015a). It was also observed that within the gender, the frequency of PTC sensitivity status in their respective studies recorded higher frequency of tasters in female than in male subjects (Shivaprasad *et al.,* 2012; Hussaini *et al.,* 2014; Daştan, *et al.,* 2015a; Mairiga *et al*., 2021). It has been reported that females are generally more of tasters than non-tasters, whereas this may be in line with the reports of anatomical studies which revealed that tasters have more taste buds than non-tasters, it still calls for further research since tasting ability may also be influenced by some other sensory and proprioceptive pathways (Shivaprasad *et al.,* 2012; Mairiga *et al*., 2021).

The outcome of the PTC phenotype status across the age groups in our sampled population recorded a decrease in the population of tasters with increase in age and an inverse result among non-tasters. There were decreases in the frequencies of tasters in the age group 16 – 20 years (86.7%) through to 26 – 30 years (76.9%) and a corresponding increase in the frequency of non-tasters with increase in age i.e., decrease in non-tasters in the age group 16 – 20 years (13.3%) through to the age group 26 – 30 years (23.1%). Though this outcome was not-significant, it however collaborates the position of some investigators that age modifies the genotype – phenotype relationship in taste sensitivity where younger subjects are more sensitive than older subjects to the bitterness of PTC (Hussaini *et al.,* 2014). There is however the need to investigate further on this finding since variation in the frequencies of allele may have little or nothing to do with age variations in PTC response.

The report obtained by Dastan *et al.*, (2015a) on age groups in their study however differs from our findings. To determine the PTC perception frequencies in their sampled population they adopted the Age Classification Index of Turkish Elder Sciences and Technologies Foundation (TUYEV) to categorize the participants into 'young' (14-29), 'middle' (30-49) and 'old' (50>) age groups. The ratios of PTC taste perception frequencies observed in the 'middle age' group was evidently higher than other groups with 93.8% value and lowest in the individuals of the 'young age' 80.6%. In justifying their findings, they were of the view that the exact taste perception had occurred due to the complete formation of taste buds on the tongues of the people in the middle age group which were not in the young and old group. They maintained that the nature of and functions of the taste buds would have been damaged or the taste buds weakened due to old age. In our study we could not use a standard matrix to classify the ages of our subjects, this may form the basis for the differences in our findings. While their age range extended beyond 50 years of participants, ours was within 30years, equivalent to their 'young age group'. Their claims on the occurrence of exact taste perception due to complete formation of taste buds on the tongues of the people in the 'middle age group' which were not in the young and old age groups and the damage and weakening of the taste buds due to old age requires further study as we could not substantiate these claims in our study.

The combined allelic frequency of the PTC tasting phenotype in our findings was in favour of tasters (0.61) against non-tasters (0.39). The allelic frequencies for male and female tasters obtained was 0.52 and 0.78 while for non-tasters was 0.48 and 0.22 respectively. A similar result was reported by Hussaini *et al.,* (2014). In their study, Igbeneghu et al., (2016) also reported the allelic frequencies of 0.46 and 0.54 in the control group of a Nigerian population while those of tasters and non-tasters in the test group were 0.27 and 0.73 respectively. Our findings on homozygosity and heterozygosity revealed a male homozygous status of 0.5168, and female 0.5093. While the heterozygous status was 0.4832 for male and 0.4907 for female respectively. The combined frequency values for homozygosity (Ho) and heterozygosity (Ht) of the population were Ho. 0.5127 and Ht. 0.4873 respectively.

Differences in allele frequencies is said to contribute to group differences in the incidence of some forms of monogenic and common diseases (Richard and Tatiana, 2016). For the monogenic diseases, the frequency of causative alleles usually correlates best with ancestry, whether

familial, ethnic, or geographical (Limborska *et al.*, 2002; . Risch, *et al.,* 2002; Rahim *et al*., 2018). Even with common diseases involving numerous genetic variants and environmental factors, investigators point to evidence that suggest the involvement of differentially distributed alleles with small to moderate effects which are taken into cognizance by health-care professionals in diagnoses (Lu *et al*., 2014).

For instance, when people take ill, complains of changes in their sense of taste are prominent as illness can make food or medicine unpalatable (Nala, 2015). Research has shown that the sensory shift may be due to a protein that triggers inflammation (Feng et al., 2015) In their study on mice, Feng et al., (2015) posited that mice that lack the ability to produce the protein called Tumour Necrosis Factor α (TNF α) are less sensitive to bitter flavour than normal. The higher incidences of PTC sensitivity taste among general population could be justified by the health condition of the people. Research has further shown that people with infections, autoimmune disease or other inflammatory conditions have higher levels of TNF α which is known to reduce food intake than in healthy people. Some other variations on the other hand are beneficial to human, as they prevent certain diseases and increase the chances to adapt to the environment (Risch, *et al.,* 2002; Lu *et al*., 2014)**.**

**Conclusion**

The study of human genetic variation has contributed greatly in influencing our knowledge of variations and diversity of human population globally. This pilot study has provided insight to the distribution of the PTC phenotype in the population that make up the Middle Belt zone of Nigeria. And further revealed the heterozygous, homozygous and allelic frequencies of the population which may be important indices for health care professionals to use in diagnoses.

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**Conflict of interest.**

There was no conflict of interest among the authors

**Author's contributions**

The research concept and methodology were conceived and agreed upon by the researchers. The sample collections were carried out by Dr Mairiga, the statistically analysis was done by Dr Yahaya and verified by Dr Iroanya. All authors contributed in the writing and editing of the manuscript. The manuscript was approved by all the authors. Dr Mairiga was appointed the corresponding author.

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