Title: Exploring the relationships between taste phenotypes, genotypes, ethnicity, gender and taste perception using Chi-square and regression tree analysis

Qian Yang[a], Ann-Marie Williamson[b], Anne Hasted[c], Joanne Hort[ad]

[a]Sensory Science Centre, Division of Food, Nutrition and Dietetics, University of Nottingham, Sutton Bonington Campus, LE12 5RD, United Kingdom

[b]Unilever R&D, Colworth, United Kingdom

[c]Qi Statistics, Kent, United Kingdom

[d]Feast & Riddet Institute, Massey University, New Zealand

Abstract

It is well known that perceived taste intensity varies greatly among individuals, and that several factors including taste phenotypes (PROP Taster Status (PTS), Sweet Liking Status (SLS), Thermal Taster Status (TTS)), ethnicity and gender, contribute to variation in taste responsiveness, although such factors are usually investigated in isolation. This study aimed to investigate the association between different taste pheno/genotypes, explore whether these taste phenotypes associated with ethnicity (Caucasian vs Asian) and gender, and determine the relative effects of the different factors on perceived taste intensity. As analysis of this type of data with ANOVA can be difficult due to confounding factors, interactions, and small sample sizes in subcategories, the use of regression tree analysis as an alternative approach was investigated. To that end, two-hundred and twenty-three volunteers were phenotyped for their PTS, SLS and TTS and genotyped for TAS2R38 –rs713598 and gustin –rs2274333. They also rated their perceived intensity of five basic taste and metallic solutions on a gLMS scale. No significant association between the three taste phenotypes were found indicating PTS, SLS and TTS are independent taste phenotypes. However, the results indicated that Asians were not only more likely to be PROP supertasters, but also more likely to be thermal tasters or Low Sweet Likers, compared to Caucasians. Gender was also significantly associated with SLS, where males were more likely to be High Sweet Likers. For perceived taste intensity, traditional ANOVA analysis proved to be challenging. The alternative approach, using regression trees, was shown to be an effective tool to provide a visualised framework to demonstrate the multiple interactions in this dataset. For example, ethnicity was the most influencing factor for perceived sour and metallic taste intensity, where Asians had heightened response compared to Caucasians. The regression tree analysis also highlighted that the PTS effect was dependent on ethnicity for sour taste, and PTS and TTS effect was dependent on ethnicity for metallic taste. This study is the first study to use regression tree analysis to explore variation in taste intensity ratings, and demonstrated it can be an effective tool to handle and interpret complex sensory datasets.

1. Introduction

Taste perception occurs when certain compounds released from food dissolve in saliva and interact with taste receptor cells within taste buds located in the oral cavity. Most mammals are able to detect five different modalities of taste: sweet, bitter, sour, salty and umami (Chaudhari and Roper, 2010) whilst some other sensations have also been purported to be potential tastes including fatty acid, metallic, kokumi and calcium (Bachmanov *et al.*, 2014, Dipatrizio, 2014, Ohsu *et al.*, 2010, Bartoshuk, 1978).

Perceived intensity of taste and other oral sensations has been shown to vary greatly among individuals and may be one of the most important determinants of food preference and consumption affecting nutritional and health status (Stewart *et al.*, 2010, Tepper *et al.*, 2014, Ullrich *et al.*, 2004). Many factors have been shown to affect perceived taste intensity perception such as health status (Overberg *et al.*, 2012, Bertereteche *et al.*, 2004), age (Bilash *et al.*, 1959, Mojet *et al.*, 2001, Monteleone *et al.,* 2017, Vignini *et al*., 2019), gender (Hirokawa *et al.*, 2006, Michon *et al.*, 2009, Bartoshuk *et al.*, 1994, Monteleone *et al.*, 2017, Vignini *et al.*, 2019), ethnicity (Williams *et al.*, 2016, Bowser *et al.*, 2019), genetics (Kim *et al.*, 2003b, Chen *et al.*, 2009) and taste phenotypes (Yang *et al.*, 2014, Bajec and Pickering, 2008, Dinnella *et al*., 2018, Yang *et al.*, 2019). Findings, however, are often conflicting across different studies. For example, some studies have found that women tend to be more sensitive to taste compared to men (Hirokawa *et al.*, 2006), whilst other studies failed to reveal a significant effect (Chang *et al.*, 2006, Melis *et al.*, 2013). In general, some significant effects of ethnicity on perceived taste intensity have been observed (Williams *et al.*, 2016, Bowser *et al.*, 2019), but there is a lack of research comparing Asian and Caucasian populations. It is worth noting that some of the earlier studies used a relatively small sample size, which could contribute to conflicting findings.

Naes *et al*., (2018) outlined some key areas as to why individual differences are important in sensory and consumer science, highlighting the various geno and phenotypes impacting sensitivity to sensory perception. So far, many studies have investigated the effect of different taste phenotypes such as PROP Taster Status (PTS) (Bell and Song, 2005, Yang *et al.*, 2014), Sweet Liking Status (SLS) (Kim *et al.*, 2014, Yeomans *et al.*, 2007) and Thermal Taster Status (TTS) (Bajec and Pickering, 2008, Skinner *et al.*, 2018) on perceived taste intensity. PROP taster status (PTS) has been studied extensively since it was first reported by Fox (1932) and individuals can be classified into three groups: PROP supertasters (pST) – perceiving PROP as extremely bitter, medium-tasters (pMT) who perceive PROP as moderately bitter, and nontasters (pNT) who perceive PROP as tasteless (Lim *et al.*, 2008). TAS2R38, a bitter taste receptor genotype, identified by Kim *et al.* (2003a), is known to influence PROP tasting ability (Drayna *et al.*, 2003). Polymorphisms in TAS2R38 have been shown to account for up to 85% of variation in PROP tasting ability (Wooding *et al.*, 2004), with PAV/PAV genotypes perceiving PROP as most bitter. Melis *et al.* (2013) further suggested that gustin (rs2274333) also contributed to perceived PROP intensity ratings but a later study failed to find such findings (Feeney and Hayes, 2014).

A number of studies have found PROP bitterness perception is positively correlated with perceived intensity of other tastants and trigeminal stimuli including bitter, sweet, salty, sour, fat, astringent, metallic, and temperature (Yang *et al.*, 2014, Bajec and Pickering, 2008, Bartoshuk *et al.*, 1998, Tepper and Nurse, 1997, Dinnella *et al*., 2018). The distribution of pNT is known to vary greatly across ethnicity and was summarised by Guo and Reed (2001) who showed that the percentage of pNT was between 2 to 37% in Africans, 7 to 37% in Europeans, 2 to 67% in Indians, and 5 to 23% in Chinese. Robino et al. (2014) has also reported differences in ethnicity along the Silk Road. Some studies found that females perceived PROP intensity higher than males (Bartoshuk *et al.*, 1994, Shen *et al.*, 2016, Robino *et al.*, 2014, Monteleone *et al.*, 2017).

Sweet Liking Status (SLS) refers to individual variation in preferred sweetness in sugary solutions, which was first reported by Pangborn (1970b). Researchers have grouped individuals as sweet likers who increasingly prefer increasing levels of sweetness; and sweet dislikers who prefer lower levels of sweetness and show increasing dislike as sweetness increases (Methven *et al.*, 2016, Kim *et al.*, 2014). Different Sweet Liking Status classification methods have been used in different studies (Garneau *et al.*, 2018, Holt *et al.*, 2000, Kim *et al.*, 2014, Pangborn, 1970a, Yang *et al.*, 2019). A recent study has adopted cluster analysis followed with validation test (Pearson’s correlation analysis) as a way to standardise the classification method and re-named individuals as High Sweet Likers – prefer sweeter solutions, Medium Sweet Likers – prefer mid sweet solutions, Low Sweet Likers - prefer low sweet solutions (Yang *et al.*, 2019). Some studies have found sweet dislikers perceive sweetness intensity as more intense than sweet likers (Drewnowski *et al.*, 1997, Looy *et al.*, 1992, Peterson *et al.*, 1999), which was suggested to be one of the reasons why sweet dislikers’ overall liking declined as sweetness increased. However, other studies failed to find such differences (Kim *et al.*, 2014) or found the effect is subjective to sweetness levels (Methven *et al.*, 2016, Yang *et al.*, 2019). Interestingly, Yeomans *et al.* (2009) found that PROP supertasters were more likely to be sweet dislikers, indicating these two taste phenotypes are associated with each other. However, a recent study failed to find such association (Yang *et al.*, 2019).

Thermal taster status (TTS) was first reported by Cruz and Green (2000), whereby part of the population perceive a taste when their tongue is warmed or cooled (Cruz and Green 2000), and are named thermal tasters (TT). Sweetness, bitterness, metallic and sourness are most often reported by TT during warming or cooling (Yang *et al.*, 2014, Skinner *et al.*, 2018, Bajec and Pickering, 2008, Cruz and Green, 2000). Those not perceiving a taste sensation from temperature stimulation are called thermal non-tasters (TnT) (Green and George, 2004). Between 20 to 50% of the tested populations have been found to be TT (Green and George, 2004b, Bajec and Pickering, 2008, Yang *et al.*, 2014, Skinner *et al.*, 2018). Several studies have also found that TT have heightened intensity responsiveness to some taste and trigeminal stimuli (Bajec and Pickering, 2008, Green and George, 2004b). However, Yang *et al.* (2014) and Skinner (2017) failed to find a significant TTS impact on individual perceived taste responsiveness.

Looking at literature, the findings remain unclear, as different factors have been found to contribute to perceived taste intensity, whereas others failed to find an effect. Some positive findings could be due to the small sample sizes used in earlier studies (e.g. in Bartoshuk *et al.*, (1994), 10 NT and 9 ST were evaluated). Most studies have investigated the effect of a taste phenotype in isolation (Bartoshuk *et al.*, 1988, Methven *et al.*, 2016, Green and George, 2004a). Similarly, the impact of ethnicity and gender on taste perception have also been investigated (Williams *et al.*, 2016, Bartoshuk *et al.*, 1994, Michon *et al.*, 2009) but again mainly as individual factors. However, individuals are combinations of different phenotypes and genotypes, ethnicity and gender, and there is a need to understand the impact of these combinations. Unbalanced numbers in factors can contribute to inconsistency in results, however, due to the nature of this type of study and population-based convenience samples, it is difficult to balance all the factors of interest across participants.

Furthermore, these factors interplay with each other, for example, Yang *et al.* (2014) found that the effect of TTS is more apparent in MT than the other two PROP phenotypes, and Dinnella *et al.*, (2018) reported a much more pronounced age effect on fungiform papilla density in males than females. This indicates a complex interplay among different factors.

Due to the complex nature of these type of studies, data is always associated with unbalanced groups. Analysis of Variance (ANOVA) is not usually an appropriate technique for understanding the separate effects in survey data (where there is limited option to control the characteristics of subjects in the test), ANOVA requires data with a structure design to allow estimation of the separate effects on a response and so is of limited applicability in this context (Sheskin, 2011). Other statistical techniques thus need to be explored to better understand this type of results.

Regression trees are a type of decision tree, and a data mining approach (Breiman *et al*., 1984, Bozkir and Sezer, 2011). There are many different types of decision trees, including ‘classification trees’ which predict outcomes for categorical data and regression trees which predict outcomes in numerical data. Regression trees use algorithms to produce predictive graphical models to identify subgroups in the data with differing levels of response between groups and homogeneity of response within groups. They have not been widely used in the field of sensory and consumer science. However, they have been commonly used in medical and health care applications over the past three decades (Podgorelec *et al.*, 2002a, Tsien *et al.*, 1998) with applications to support early diagnosis (Jabbar *et al.*, 2014, Tsien *et al.*, 1998) and to predict risk factors in some diseases (Dimopoulos *et al.*, 2018). Recently, regression trees were shown to be a useful technique to understand factors contributing to wine consumption using 21 variables for analysis from a survey (e.g. demographic information, price importance, location etc.) (Jovanović Miomir, 2017). The main advantage of a regression tree is that it shows hierarchical and graphical representations of interactions between variables (Loh, 2011, Machuca et al., 2017), which helps researchers to visualise the structure of interdependence of the data in a graphical tree format. As survey data is often an unstructured dataset, regression trees can be useful to understand individual characterisation in physiological and demographic measures. However, regression trees can suffer the risk of overfitting in very large datasets as small splits will be statistically significant. However, CHAID attempts to stop overfitting from the beginning, as the tree only splits when there is significant association (Shanthi, 2019). In some cases, analysts control the size of the splits by specifying in the CHAID algorithm the smallest acceptable size of a subgroup (Haughton & Oulabi, 1997). The CHAID technique is appropriate for datasets of varying sizes (n=41 to 2000) (Arroyo *et al.*, 2018, Álvarez-Álvarez *et al.*, 2011, Díaz-Pérez and Bethencourt-Cejas, 2016, Antipov and Pokryshevskaya, 2009).

The overall aim of this study was to investigate the interrelationships between taste phenotypes, genotypes, ethnicity, gender and taste perception, through the application of regression tree analysis. The objectives were to i) determine the association between three different taste phenotypes (PTS, TTS, SLS), and explore whether these taste phenotypes were associated with gender and ethnicity (specifically Caucasian and Asian); and ii) determine the relative impact of these factors on taste perception.

2. Materials and Methods

This study was approved by the University of Nottingham Medical School Research Ethics Committee. All participants signed and gave informed consent before taking part in the study. All data were collected using Compusense Cloud (Compusense, Canada) at the Sensory Science Centre, University of Nottingham. Participants were invited to attend two sessions over two separate testing days.

2.1. Subjects and sessions

Two hundred and twenty-three volunteers (160F, 63M; age range 18 – 65 years old; 156 Caucasians, 67 Asians) participated in this study. Subject characteristics were shown in Table 1. In this study, participants were divided into Asian and Caucasian based on understanding that there is more phenotypic homogeneity among Asian subgroups in comparison to Caucasian (Leow, 2017). Participants who were smokers or self-reported took medication known to affect taste and aroma perception were excluded from this study.

The first session involved gLMS scale training, phenotyping for PROP Taster Status and Sweet Liking Status together with taste intensity perception measurements. The second session ran on a one to one basis to facilitate ease of Thermal Taster Status phenotyping and buccal swab collection for genotyping.

2.2. gLMS training

In order to ensure participants understood the nature of the gLMS scale and facilitate its correct use for intensity rating, a gLMS scale reference sheet was given to each participant. To emphasise the general nature of the top of the scale, participants were asked to think of the strongest sensation of any kind they had experienced previously, or the strongest sensation they could imagine happening to them and write these down at the top of the gLMS scale. Following Bartoshuk *et al.* (2002), they were also asked to rate the intensities of 15 remembered sensations relative to their own strongest sensation. Reference sheets were always provided in subsequent experiments, and participants were encouraged to refer to their reference sheet for guidance when using the gLMS scale.

2.3. Taste Phenotyping methods

2.3.1. PROP Taster Status determination

A 0.32mM 6-n-Propylthiouracil (PROP) (Sigma Aldrich, UK) solution was prepared by dissolving PROP in water on a low heat stirring plate. Each subject was instructed to roll a saturated cotton bud, which had previously been dipped in the PROP solution (22 ± 2 ºC), across the tip of the tongue for approximately 3s. Participants were informed that the bitterness may take a few seconds to reach its maximum and were instructed to rate its maximum intensity using the gLMS scale. After a two min break, the procedure was repeated to collect duplicate ratings (Lim *et al.*, 2008).

Participants who rated bitterness intensity below barely detectable on the gLMS were classified as pNT. Those who rated the bitterness intensity above barely detectable, but below moderate were classified as pMT. Those who rated the intensity above moderate were classified as pST (Lim, Urban et al. 2008).

2.3.2. Sweet Liking Status determination

Four sucrose solutions (17g/L, 78 g/L, 168/L and 397g/L) and a water sample were served monadically in ascending sweetness concentration to each participant (Yeomans *et al.*, 2007). All samples were prepared the day before tasting, stored in a fridge (5±2°C) and brought out from the fridge at least an hour before testing to serve at ambient temperature (20 ± 2°C). 10 ml of each sample labelled with a random 3-digit code was served. A two-minute break was given between samples and participants were instructed to rinse their mouth with water (Evian, Danone, France) to cleanse their palate. Participants were asked to rate how much they liked each sample on a Labelled Affective Magnitude (LAM) Scale (Schutz and Cardello, 2001).

Sweet Liking Status classification was based on Yang *et al.* (2019). This used agglomerative hierarchical clustering (AHC) analysis of overall liking of the four sucrose solutions, followed by Pearson correlation tests between each individual's results and cluster means to validate cluster grouping. Regrouping was applied until the correlation coefficient reached above 0.6 within each cluster group, generating categories of High Sweet Likers, Medium Sweet Likers and Low Sweet Likers. Those participants whose correlation coefficients were lower than 0.6 in any of the three groups were categorised into an Unclassified group

2.3.3. Thermal Taster Status determination

A circular advanced thermal stimulator peltier thermode (Medoc, Israel) was used to heat and cool the anterior tip of the tongue. Two temperature trials (warming and cooling) were used. For the warming trial, the thermode temperature began at 35ºC, was cooled to 15ºC and then re-warmed to 40ºC and held there for 1s. For the cooling trial, the thermode temperature began at 35ºC, was cooled to 5ºC and held there for 10s (Bajec and Pickering, 2008). Two replicates of each temperature trial were conducted. The temperature ramp for all trials was 1ºC/s. Warming trials always preceded cooling trials to avoid possible adaptation from the intense, sustained cold stimulation. A break of two minutes was given before proceeding to the next trial to allow the tongue temperature/sensation to return to normal.

Participants were asked to record the taste quality and intensity perceived during temperature stimulation, only if they perceived any. In order to ensure accuracy regarding taste quality reported by TT, a set of taste solutions, the same sample set used for the intensity ratings (Section 2.5) were given to participants in Session 1 to ensure participants were familiar with different taste qualities. TT were defined as those who perceived a taste sensation from both replicates at either warming or cooling trials, whereas TnT were defined as those who did not perceive any 'tastes' throughout the temperature trials (Green & George, 2004). This also left a group of individuals with inconsistent responses (e.g. only perceive taste from a single temperature trial), who were deemed the Uncategorised group.

2.4. Taste Genotyping determination

Isohelix Buccal swab kits (Cell Projects, Kent, UK) were used for collecting buccal cells from the inside of the cheek for DNA extraction. Buccal swab samples were collected by instructing participants to rub the sterile swab firmly against the inside of their cheek for one minute. The swab head was placed inside the associated tube together with a 'Dri-capsule' on top and stored at ambient temperature (20± 2°C). A label with the participant's study number was placed on the tube for identification and anonymity. Samples were sent to LGC Genomics (Herts, UK) for genotyping. Single nucleotide polymorphisms for TAS2R38 – rs713598 and gustin – rs2274333 were genotyped using the Kompetitive allele specific PCR (KASP) method (He *et al.*, 2014).

2.5. Perceived taste intensity rating measurement

Suprathreshold taste solutions were prepared at the following concentrations: sweet - glucose (117.32g/L), sour - citric acid (1.5g/L), salt- sodium chloride (10g/L), bitter - quinine (0.017g/L), umami/savoury - monosodium glutamate (20g/L) and metallic - ferrous sulphate (0.83g/L) (Sigma Aldrich, UK). All taste samples were prepared using deionised water the day before tasting and stored in a fridge (5±2°C). All samples were brought out from the fridge at least an hour before tasting, served at ambient temperature (20 ±2°C). 10 ml of each sample labelled with random three-digit codes was presented, in duplicate, randomised across participants. Participants were given a one-minute break between stimuli, and water (Evian, Danone, France) as a palate cleanser. Participants were asked to rate the perceived intensity of each tastant on the gLMS scale. The reference gLMS scale was provided during this session and participants were encouraged to refer back to their own reference scale during rating.

2.6. Data Analysis

All statistical analyses were performed using XLSTAT version 2018. 07 (Addinsoft, Paris, France) at an α-risk of 0.05.

Chi-square analysis was used to examine associations among taste phenotypes, genotypes, ethnicity and gender. One-factor ANOVA was applied on PROP intensity across PROP taster status, for sweet liking status, two-factor ANOVA (sweet liking status and sucrose concentration) was applied on overall liking of the four sucrose solutions to confirm robustness of phenotype classifications.

For perceived taste intensities, the average between the two replicates was used for statistical analyses.

Initially it was planned to run a five factor ANOVA in PTS, TTS, SLS, Ethnicity and Gender to determine if the factors studied impacted on taste perception. However, significant associations were found between ethnicity and taste phenotypes, which created confound effects, thus five-factor ANOVA is inappropriate. Consequently, the data was split into the two separate ethnicity groups, and four-factor ANOVA models were fitted for each ethnicity group (Caucasian vs Asian) separately. However due to low sample size in some subcategories (e.g. only 1 subject in Asian HSL and PROP NT group), including interactions in ANOVA model became problematic, so ANOVA could not be used to investigate possible interactions.

Due to the complex nature of the data and interactions in the data, a regression tree analysis was explored to investigate whether it would be a useful tool to highlight the relative impact of the factors studied. Regression tree can break down a dataset into smaller subsets and use a simple algorithm to build a tree structure to demonstrate the subsets in branches. In this case, subsets of taste perception behaviour. It therefore provides a framework to quantify the values of outcomes and the probabilities of achieving them. Stopping rules in the regression tree can be set by user to allow the tree to grow to an optimum size. The interpretation of the results is generally summarised in a tree, which is easier to understand and interpret (Bozkir and Sezer, 2011). Consequently, a regression tree was generated from the intensity ratings for each taste modality, with the five factors as independent variables. The CHAID option was applied with a minimum 5% of participants as the node size. The CHAID technique depends on a significance test at each branch, allowing the branching to stop when splits are not significant.

3. Results

3.1. Phenotype distribution

3.1.1. PROP Taster Status

In this study, seventy four participants (33%) were classified as pST, 102 participants (46%) were classified as pMT, and 47 participants (21%) were classified as pNT. As expected, pST rated PROP bitterness significantly higher than pMT and pNT; and pMT rated significantly higher than pNT ($p<0.0001$) (Figure 1).

3.1.2. Sweet Liking Status

Ninety-four participants (42%) were classified as High Sweet Likers (HSL), and 38 participants (17%) were classified as Medium Sweet Likers (MSL), 59 participants (26%) were classified as Low Sweet Likers (LSL) and 32 participants (14%) were classified as Unclassified due to their inconsistent responses.

The ANOVA data showed there is a significant Sweet Liking Status group difference ($p<0.0001$), where in general, HSL rated liking of sucrose solutions significantly higher than the other three groups, and MSL and UN gave significant higher liking scores than LSL, indicating LSL did not like the sweet solutions as much as the other three SLS groups. As expected, A significant SLS*Concentration interaction was also observed ($p<0.0001$), where HSL significantly liked the sweeter sucrose solutions (78 to 397 g/L) over the low sweetened sucrose solution (0 to 17g/L) (Figure 2A). MSL liked most the medium sweetened samples (78g/L and 168g/L) compared to low sweet (0-17g/l) and high sweet (397g/l) samples (Figure 2B). LSL significantly disliked the high sweetened solutions (168g/L and 397 g/L) than less sweetened solutions (0 to 78g/L) ($p<0.05$) (Figure 2C). No clear trend was observed for UN group (Figure 2D).

3.1.3. Thermal Taster Status

In this study, 86 participants (39%) were classified as TT, and 109 participants (49%) were classified as TnT, the remaining 28 participants (12%) were classified as Uncategorised due to their inconsistent responses. The most reported taste sensations during the warming trial were sweet (35%), metallic (16%) and bitter (12%). For the cooling trial, the most reported taste sensations were bitter (28%), metallic (19%), sweet (16%), sour (15%) and minty (11%). Other sensations such as salty and spicy were also reported during both warming and cooling trials (Table 2).

3.2. Associations between taste phenotypes, genotypes, ethnicity and gender

3.2.1. Associations between taste phenotypes and genotypes

As expected, a significant association was observed from Chi-square analysis between PTS phenotype and TAS2R38 genotype ($p<0.0001$). Participants who carried the C:C genotype were more likely to be pST (67%) and less likely to be pNT

(6%); whereas participants who carried the G:G genotype were more likely to be pNT (51%) and less likely to be pST (3%) (Table 3).

Interestingly, the association between PTS phenotype and gustin genotype (rs2274333) approached significance (p=0.09), where a higher proportion of pST (53%) carried the G:G genotype compared to pNT (14%) (Table 4).

3.2.2. Associations between taste phenotypes/genotypes and ethnicity

Not unexpectedly, a significant association between PTS phenotype and ethnicity was observed (p<0.0001), where 55% Asians were pST and 9% were pNT, compared to 24% of Caucasians being pST and 26% pNT (Table 5). This data was supported with the significant association between TAS2R38 (rs713598) and ethnic group (p<0.0001), where a higher proportion of Asians (43%) carried PROP tasting genotype (C:C) than Caucasians (17%). In addition, a higher proportion of Caucasians (17%) carried the non-PROP tasting G:G genotype compared to Asians (6%) (Table 6).

Furthermore a significant ethnicity^gustin association was observed (p<0.0001). Asians (38%) were more likely to carry G:G genotype compared to Caucasians (7%), as shown in Table 7.

The association between SLS and ethnicity was very close to significance (p=0.07), with 31% of Asians classified as HSL, compared to a higher proportion of Caucasians (47%). In addition, 24% and 33% of Asians were classified as MSL and LSL respectively, whereas lower proportions were observed in Caucasians (14% MSL and 24% LSL) (Table 8).

Notably, an original and interesting finding in this study was that a significant TTS^ethnicity association was observed (p=0.001), where 51% Asians were classified as TT, reporting a taste sensation during either warming or cooling trials and only 30% of Asians were TnT. Instead, a much lower proportion of TT (33%) and a much higher proportion of TnT (57%) were observed in Caucasians (Table 9).

3.2.3. Associations between taste phenotypes/genotypes and gender

No significant association between PTS and gender (p=0.8) was observed in this study. A significant SLS^gender association was found (p=0.03) in this study, where 52% of males were classified as HSL, compared to a lower proportion in females (38%); and 31% of females were classified as LSL, compared to 14% of males (Table 10).

3.2.4. Associations between different taste phenotypes

Importantly, no significant associations were found among any of the three taste phenotypes ($p>0.05$), indicating these three taste phenotypes are likely to be independent phenotypes.

3.3. Perceived Taste Intensity Rating

This study investigated the effect of the different taste phenotypes (PTS, SLS and TTS), ethnicity and gender on perceived intensity of taste solutions. Four-factor ANOVA within the Caucasian group showed that limited effects were observed for all the factors examined ($p>0.05$), apart from a significant gender effect on bitterness ($p=0.04$), as shown in Figure 3. Females perceived the bitter intensity significantly higher than males (Figure 3d). Within the Asian group, interestingly, a significant PTS effect was found for salty and metallic taste ($p<0.05$), and an approaching significant effect was found for sour taste ($p=0.09$). As shown in Figure 4a, pST rated saltiness and sourness significantly higher than pNT, where pMT was not significantly different to pST or pNT. However, for metallic taste, it was for both pST and pNT that rated it significantly higher than pMT. An additional TTS effect on metallic taste also approached significance ($p=0.07$), where TT rated metallic taste as significantly higher than Unclassified group.

No significant SLS effect was found for any of the tastes ($p>0.05$) measured here. In addition, no significant gender effect was found for any of the individual tastes ($p>0.05$).

3.4. Regression Tree Analysis

ANOVA demonstrated that it is difficult to decouple the complexity of the data, especially investigating interactions in a complex and relatively small dataset. As an alternative approach, regression tree analysis was explored to interpret the data. For each individual taste intensity, a regression tree was generated with the five factors as independent variables, with a minimum 5% of participants in the node size. No regression tree was generated for sweet, salty, bitter and umami taste.

For sour taste, regression tree analysis resulted in seven terminal nodes (see Figure 5). The first split, and therefore most influencing factor, was for ethnicity: It shows that Asians had significantly higher predicted sour intensity ratings (1.56 on gLMS scale, 30% of total participants) than Caucasians (1.5 on gLMS scale, 70% total participants).

Among the Asian group, PROP Taster Status comes into play where pST (1.59 on gLMS scale, 16.6% of total participants) had significantly higher predicted sour taste intensity rating than pMT & pNT group (1.51 on gLMS scale, 13.5% of the total participants). Whereas among the Caucasian group, PROP Taster Status also comes into play, however, pST and pMT were grouped together (1.51 on gLMS scale, 51.6% of total participants), and had a significant higher predicted sourness intensity than pNT (1.45 on gLMS scale, 18.4% of total participants).

For metallic taste, a regression tree was also generated, resulting in 7 terminal nodes, as shown in Figure 6. Interestingly, ethnicity again was the first split, where Asians (1.3 on gLMS scale, 30% of total participants) had significantly higher predicted metallic intensity ratings than Caucasians (1.15 on gLMS scale, 70% of the total participants). No further split was observed under the Caucasian group. However, for the Asian group, a further split was observed for PROP Taster Status, where pMT (1.09 on gLMS scale, 10.8% of total participants) had significant lower predicted metallic intensity than pST & pNT group (1.41 on gLMS scale, 19.3% of total participants). No further split was observed for Asian pMT group, but Asian pST/pNT was further differentiated by Thermal Taster Status phenotype. TT (1.48 on gLMS scale, 10.8% of total participants) perceived the metallic taste as significantly higher than TnT/Unclassified group (1.32 on gLMS scale, 8.5% of total participants).

4. Discussion

4.1. Phenotype distribution and associations

PTS distribution (33% pST, 16% pMT and 21% pNT) in the current study is in general agreement with distributions observed in previous studies (Lim et al., 2008, Guo and Reed, 2001). Although a slightly higher pST proportion was observed, this was because higher proportion of Asians (55%) are pST compared to Caucasians (24%). This finding agrees with previous studies (Guo and Reed, 2001, Risso et al., 2016, Rankin et al., 2004) that approximately 25% of the population in Western countries were pST and pNT respectively. In this study, 24% pST and 26% pNT in Caucasians were observed. As expected, a much lower proportion of pNT (9%) was found in Asian participants, which agrees with Guo and Reed (2001)'s finding that only 13.7% of Chinese were pNT.

As expected, TAS2R38 genotype is significantly associated with PTS phenotype ($p<0.0001$), which supported previous findings (Kim et al., 2003b, Prodi et al., 2004) that bitter receptor gene TAS2R38 play a key role in PROP tasting ability. Interestingly, an approaching significant association between PTS phenotype and gustin genotype (rs2274333) was found, where pST carried more G:G genotype. This finding disagrees with previous findings (Barbarossa, et al., 2015, Melis et al., 2013), where an opposite trend was found that pST were more likely to carry the A:A genotype of gustin (rs2274333). Further investigation on ethnicityˆgustin in the current study revealed that Asians were more likely to carry G:G genotype. The approaching significant PTS^Gustin association is undoubtedly due to the fact that Asians are more likely to be pST and carry G:G genotypes. By analysing the association between PTS and gustin genotype within each ethnic group, no significant associations ($p>0.05$) were found. This is in agreement with another investigation conducted by Feeney and Hayes (2014) that no significant association between PTS and gustin genotype was found. The findings here highlight the importance of characterising recruited participants to better understand the relationship between taste phenotype and genotype, due to international immigration over the last 20 years, recruited participants in Western countries are becoming more ethnically diverse (Abel and Sander, 2014).

In addition, this study did not find any significant association between PTS and gender ($p=0.8$). However, a range of previous research have reported that females were more likely to be supertasters (Bartoshuk et al., 1994, Shen et al., 2016, Robino et al., 2014), but there are also some studies like ours who failed to find such association (Chang et al., 2006, Garneau et al., 2014).

SLS distribution (42% HSL, 17% MSL, and 26% LSL) supported recent published research where a slightly lower proportion (34%) of the participants were classified as HSL, and slightly higher proportion (35%) of the participants were classified as LSL (Yang et al., 2019). This could be due to the fact that previous studies used 5 sucrose solutions (6g/L to 360g/L) following Methven et al. (2016)'s approach, whereas the current study used the four sucrose solutions (17g/L to 397g/L) following Yeomans et al. (2007)'s approach. Interestingly, an approaching significant SLSˆethnicity ($p=0.07$) was found, where higher proportion of Caucasians are HSL, and higher proportion of Asians are LSL. This finding builds on the same trend observed in a recent published study by the same research group (Yang et al., 2019). An older study supported the current finding, where US students of European descent gave much higher pleasantness ratings for sweeter cookies compared to Taiwanese students (Bertino et al., 1983). However, it is worth noting that in the current small Asian cohort (n=67), mainly as international students studying at University, 81% were Chinese (n=54), 9% were Indian (n=6), 4% were Pakistan (n=3) and 7% were other Asian background (n=5). Although variation within Asian ethnic groups on food preference and intake has been previously reported (Abdullah et al., 2016), one of the limitations of this study is that ethnic group within Asian participants were not further analysis due to small sample size in subcategories. A larger sample size would be needed to further investigate Asian subethnic groups.

Interestingly, this study found that males were more likely to be HSL than females. This finding supports previous work by Turner-Mcgrievy et al. (2013) where a higher proportion of males were found to be sweet likers. A recent study failed to find a significant association between SLS and gender, but a similar trend was observed (Yang et al., 2019). Several studies have reported that women are associated with greater sweet cravings, especially for chocolate (Zellner et al., 1999, Roininen et al., 2001). In general, women are reported to have experienced more frequent craving in everyday life (Lafay et al., 2001, Hallam et al., 2016), which is believed to relate to hormonal changes (Dye et al., 1995, Asarian and Geary, 2013). However, gender has been found to significantly affect food choices, where females have a higher general health interest than males (Roininen et al., 2001), indicating although females had greater cravings for sweets, they are more conscious with sweet consumption due to health concerns. In this study, HSL referred to participants who prefer high sweetened solutions, whereas LSL prefer low sweetened solution. The fact that higher proportion of females were LSL could be related to females' general health interest. The relationship between Sweet Liking Status phenotype and sweet craving is yet to be investigated. One of the limitations for this study is that gender and ethnicity were not balanced, further studies with more balanced numbers of gender and ethnic groups to further explore these relationships are needed.

TTS distribution (39% TT and 49% TnT) agrees with previous literature, where between 20 to 50% of the population have been classified as TT (Bajec and Pickering, 2008, Yang et al., 2014, Skinner et al., 2018, Green and George, 2004a). The most reported taste sensations reported in this study agrees with published evidence

(Skinner et al., 2018, Yang et al., 2014), and again highlighted sweetness was most popular reported taste sensation during warming, and bitter and sour tastes were the most popular perceived tastes during cooling. Interestingly, metallic taste was once again reported as one of the most frequent reported taste responses perceived from both warming and cooling trial since its first reported by Yang et al. (2014), which warrants further investigation. The reasons why a wider range of TT proportion were reported in previous studies (between 20 to 50%) (Bajec and Pickering, 2008, Yang et al., 2014, Skinner et al., 2018, Green and George, 2004a), were believed to be the variation in classification methods used across different studies. However, this study has suggested that apart from the classification methods, ethnically diverse tested population also contribute to such variation. It is well known that ethnic diversity in terms of food preference and eating habit exist (Green et al., 2003, De Castro, 2007), and it would be interesting to investigate the role of Thermal Taster Status in shaping the diversity of ethnically diverse diets. Yang (2015) has already reported that TT significantly disliked a strawberry flavoured drink served at extreme temperatures (e.g. warm, cold and frozen) more than TnT, but no studies to date have looked at whether differences in food choices across ethnicities could be linked to Thermal Taster Status. In addition, no significant association between TTS and gender was observed (p=0.6), supporting Yang et al. (2014)'s previous finding.

Although this study has revealed that Asians were more likely to be pST, TT and LSL, it also revealed these three taste phenotypes are likely to be independent phenotypes. Consequently, if an Asian participant is a pST, it does not necessarily mean that he/she is also a TT or LSL. More research as to whether these latter phenotypes have genetic drivers is warranted. The association between taste geno and phenotypes with ethnicity is very interesting, however, the dataset is small in this study (only 67 in Asian population), and research with larger datasets is required to confirm such findings.

4.2. Perceived Intensity rating

Due to confounding effect between ethnicity and taste phenotypes, including ethnicity in ANOVA is inappropriate. Thus a four-factor ANOVA within each ethnic group was conducted. ANOVA revealed limited effects across different taste phenotypes and gender, apart from a significant gender effect on bitterness in Caucasians, and a significant PTS effect for salty, sour and metallic tastes ($p<0.05$) in Asians. PTS effect for salty taste follows previous findings, where pST and pMT have a general heightened taste responsiveness (Yang et al., 2014, Bajec and Pickering, 2008, Bartoshuk et al., 1998). An additional TTS effect on metallic taste approached significance (p=0.07) in Asians. In the current study and previous studies (Yang et al., 2014, Skinner et al., 2018), TT perceived metallic taste as one of the most commonly reported taste sensation during temperature trials, whether the heightened metallic response is linked to their ability to perceive a metallic taste from temperature is currently unknown and warrants further investigation. However, it is worth noting that this effect is only apparent in the Asian group not in the Caucasian group. Conflicting results have been observed in the literature for the effect of Thermal Taster Status in taste sensitivity, where some studies reported that TT have heightened taste response (Bajec and Pickering, 2008, Pickering et al., 2010), whereas others did not (Yang et al., 2014, Bajec and Pickering, 2010).

In general, no significant SLS effect was found for any of the taste qualities (p>0.05) measured in this study. Previous research has found that the impact of SLS is dependent on sweetness concentration, where an effect was found at lower sugar solutions (3% and 6% sucrose solutions), but such an effect was diminished at intense solutions (above 12% sucrose solution) (Yang et al., 2019). The sweet sample used in this study is glucose (117.32g/L) which is quite intense, which might supress SLS effect.

In general, the effects observed across Asians and Caucasians were not the same, especially for PROP Taster Status. For example, a significant PROP Taster Status effect was found for salty, sour and metallic in Asians, but no such effect was found for Caucasians. This indicates interactions exist between Ethnicity and PROP Taster Status. Previous studies have found significant PTS*gender (Bajec and Pickering, 2008), PTS*TTS (Yang et al., 2014) and ethnicity*gender (Williams et al., 2016) interaction in taste sensitivity. However, the data here is rather complex due to confounded effects and small sample sizes in subcategories, which makes investigating interactions using ANOVA models impossible.

4.3. Regression tree

Conventional ANOVA demonstrated it can be difficult to investigate the complex and relatively small dataset with interactions, thus the alternative approach – regression tree analysis was used to explore the datasets. Interestingly, regression tree was only generated for sour and metallic tastes, but not for sweet, salty, bitter and umami tastes, indicating the effects were not powerful enough to explain the variation in these taste intensities. For sour taste, the first split was ethnicity, with Asians had higher predicted intensity than Caucasians. Within each ethnic group, PTS comes into play, where pST had higher predicted rating than pMT and pNT in Asians, but both pST and pMT combined had higher predicted rating than pNT (Figure 5). From ANOVA output, only significant PTS effect in Asian group was found but not in Caucasian group, indicating regression tree could provide additional information than just traditional ANOVA. Regression tree analysis also provides a visualised relationship tree that makes it easier to understand and interpret the data, and suggested there is a significant interaction between ethnicity and PROP Taster Status, where ANOVA could not deliver due to the complex nature of the data.

For metallic taste, ethnicity again was the first split, with Asians had higher predicted intensity ratings than Caucasians. Interestingly, no further split was found under Caucasians. But a further PTS and TTS splits were observed under Asians, where pMT had significantly lower predicted metallic ratings than both pST and pNT. This agrees with ANOVA output that pMT rated lowest (Figure 4a). A further TTS split was observed under Asian pST/pNT group, where TT had a significantly higher predicted intensity rating than TnT/Unclassified. This data was supported with the ANOVA output that TT perceived metallic as strongest (Figure 4b). Looking at the regression tree output, three-way interactions (ethnicity*PROP Taster Status*Thermal Taster Status) are likely to occur, however, ANOVA was not able to demonstrate such complex relationship due to confounded effects and small sample size.

One of the limitations in this study is that the subject number of Caucasians and Asians was not equal, as well as the subject numbers in different taste phenotypes, however, it is not realistic to have a balanced design for this type of study. This study found that regression tree could be a useful statistical tool to analyse complicated datasets that have a slightly unbalanced design. However, larger sample size in the Asian group would still be needed to further validate the finding in this study, especially the differences between Asians and Caucasians.

The impact of PTS, SLS, TTS, ethnicity and gender were not fully examined in any previous research, the data here suggests that the relationship is rather complex, and multiple interactions are likely to occur. Regression tree analysis provided additional information and can be used as a different approach to look at the data compared to traditional ANOVA analysis. There are multiple statistical techniques available for every data analysis, other techniques such as Partial Least squares discriminant analysis could also be used to analyse this kind of datasets, however one of the advantages of regression tree is that it can visualise the relationship between the variables and related categorical predictors within a tree structure (Gandomi et al., 2013, Miller et al., 2014). We speculate that regression tree analysis provides a useful output of the dataset structure to aid interpretation. Thus, regression trees may therefore be a useful tool to use in the field of sensory and consumer science to better understand and visualise complex datasets.

5. Conclusion

Although this study was performed on a relatively small sample size, the findings continue to support previous observations that pST were more likely to carry the PROP tasting genes (TAS2R38) and that males were more likely to be High Sweet Likers than females. However, it also brought new insights concerning ethnic differences across different taste pheno and genotypes, where a significant association was observed between ethnicity and taste genotypes. This highlighted the importance of characterising recruited participants. For the first time it is shown that Asians were not only more likely to be PROP supertasters, but also more likely to be thermal tasters or Low Sweet Likers. However, interestingly, no significant association across these three taste phenotypes was found, indicating these three taste phenotypes are independent.

In terms of perceived taste intensity, due to confounding effect between factors, five-factor ANOVA was inappropriate. Although 4-factor ANOVA was conducted under different ethnicity groups, interactions could not be included in the ANOVA model due to confounding effect and small sample size, which makes investigating interactions impossible using ANOVA. An alternative approach, regression tree was hence explored and was shown to highlight additional information beyond traditional ANOVA analysis. For example, regression tree has demonstrated two-way interaction in sour taste, and three-way interaction in metallic taste in a visualised framework, which makes data interpretation much easier and efficient. It also enabled clear visualisation at the impact of different factors studied, for example, it is clear that Asian pST TT perceive metallic taste at more intense level than pMT Caucasians in general. Additional and larger studies are needed to further validate the technique for this type of data, but this study has highlighted that regression tree analysis is a promising technique to handle complicated and multifactorial dataset for the sensory and consumer science.

Funding: This research was funded by Unilever R&D, Colworth, United Kingdom

Reference:

ABDULLAH, N.-F., TEO, P.S. and FOO, L.H. 2016. Ethnic Differences in the Food Intake Patterns and Its Associated Factors of Adolescents in Kelantan, Malaysia. *Nutrients* **8**, 551.

ABEL, G.J. and SANDER, N. 2014. Quantifying Global International Migration Flows. *Science* **343**, 1520-1522.

ÁLVAREZ-ÁLVAREZ, P., KHOURI, E., CÁMARA-OBREGÓN, A., CASTEDO-DORADO, F. and ANTA, M. 2011. Effects of foliar nutrients and environmental factors on site productivity in Pinus pinaster Ait. stands in Asturias (NW Spain). *Annals of Forest Science* **68**, 497-509.

ANTIPOV, E. and POKRYSHEVSKAYA, E. 2009. Applying CHAID for logistic regression diagnostics and classification accuracy improvement. *University Library of Munich, Germany, MPRA Paper* **18**.

ARROYO, L., MARÍN, D., FRANKEN, K., OTTENHOFF, T. and BARRERA, L. 2018. Potential of DosR and Rpf antigens from Mycobacterium tuberculosis to discriminate between latent and active tuberculosis in a tuberculosis endemic population of Medellin Colombia. *BMC Infectious Diseases* **18**.

ASARIAN, L. and GEARY, N. 2013. Sex differences in the physiology of eating. *American journal of physiology. Regulatory, integrative and comparative physiology* **305**, R1215-R1267.

BACHMANOV, A.A., BOSAK, N.P., LIN, C., MATSUMOTO, I., OHMOTO, M., REED, D.R. and NELSON, T.M. 2014. Genetics of Taste Receptors. *Current pharmaceutical design* **20**, 2669-2683.

BAJEC, M.R. and PICKERING, G.J. 2008. Thermal taste, PROP responsiveness, and perception of oral sensations. *Physiol. Behav.* **95**, 581-590.

BAJEC, M.R. and PICKERING, G.J. 2010. Association of thermal taste and PROP responsiveness with food liking, neophobia, body mass index, and waist circumference. *Food. Qual. Prefer.* **21**, 589-601.

BARBAROSSA, I. T., MELIS, M., MATTES, M. Z., CALO, C., MURONI, P., CRNJAR, R., & TEPPER, B. J. (2015). The gustin (CA6) gene polymorphism, rs2274333 (A/G), is associated with fungiform papilla density, whereas PROP bitterness is mostly due to TAS2R38 in an ethnically-mixed population. *Physiology & Behavior, 138*, 6-12.

BARTOSHUK, L.M. 1978. The psychophysics of taste. *Am J Clin Nutr* **31**, 1068-1077.

BARTOSHUK, L.M., DUFFY, V.B., FAST, K., GREEN, B.G., PRUTKIN, J. and SNYDER, D.J. 2002. Labeled scales (e.g., category, Likert, VAS) and invalid across-groupcomparisons: what we have learned from genetic variation in taste. *Food. Qual. Prefer.* **14**, 125-138.

BARTOSHUK, L.M., DUFFY, V.B., LUCCHINA, L.A., PRUTKIN, J. and FAST, K. 1998. PROP (6-n-propylthiouracil) supertasters and the saltiness of NaCl. *Ann Ny Acad Sci* **855**, 793-796.

BARTOSHUK, L.M., DUFFY, V.B. and MILLER, I.J. 1994. Ptc/Prop Tasting - Anatomy, Psychophysics, and Sex Effects. *Physiol. Behav.* **56**, 1165-1171.

BARTOSHUK, L.M., RIFKIN, B., MARKS, L.E. and HOOPER, J.E. 1988. Bitterness of Kcl and Benzoate - Related to Genetic Status for Sensitivity to Ptc/Prop. *Chem. Senses* **13**, 517-528.

BELL, G. and SONG, H. 2005. Genetic basis for 6-n-Propylthiouracil Taster and Supertaster Status determined across cultures. In *Genetic variation in taste sensitivity* (J. Prescott & B.J. Tepper, eds.), Marcel Dekker, New York.

BERTERETCHE, M.V., DALIX, A.M., D'ORNANO, A.M., BELLISLE, F., KHAYAT, D. and FAURION, A. 2004. Decreased taste sensitivity in cancer patients under chemotherapy. *Supportive care in cancer : official journal of the Multinational Association of Supportive Care in Cancer* **12**, 571-576.

BERTINO, M., BEAUCHAMP, G.K. and JEN, K.-L.C. 1983. Rated taste perception in two cultural groups. *Chem. Senses* **8**, 3-15.

BILASH, I., COOPER, R.M. and ZUBEK, P. 1959. The Effect Of Age On Taste Sensitivity. *Journal of Gerontology* **14**, 56-58.

BOWSER, S., FARNSWORTH, N., RUSSELL, K., SCHLECHTER, H., BERNSTEIN, S., COURVILLE, A.B., ZAMBELL, K., SKARULIS, M. and MUNIYAPPA, R. 2019. Sweet taste perception is greater in non-Hispanic black than in non-Hispanic white adults. *Nutrition* **59**, 103-107.

BREIMAN, L., FRIEDMAN, J., OLSHEN, R. AND STONE, C.J.. 1984. *Classification and Regression Trees*. Chapman and Hall, Wadsworth, New York

BOZKIR, A.S. and SEZER, E.A. 2011. Predicting food demand in food courts by decision tree approaches. *Procedia Computer Science* **3**, 759-763.

CHANG, W.-I., CHUNG, J.-W., KIM, Y.-K., CHUNG, S.-C. and KHO, H.-S. 2006. The relationship between phenylthiocarbamide (PTC) and 6-n-propylthiouracil (PROP) taster status and taste thresholds for sucrose and quinine. *Arch Oral Biol* **51**, 427-432.

CHAUDHARI, N. and ROPER, S.D. 2010. The cell biology of taste. *J Cell Biol* **190**, 285-296.

CHEN, Q.-Y., ALARCON, S., THARP, A., AHMED, O.M., ESTRELLA, N.L., GREENE, T.A., RUCKER, J. and BRESLIN, P.A.S. 2009. Perceptual variation in umami taste and polymorphisms in TAS1R taste receptor genes. *The American Journal of Clinical Nutrition* **90**, 770S-779S.

CRUZ, A. and GREEN, B.G. 2000. Thermal stimulation of taste. *Nature* **403**.

DE CASTRO, J.M. 2007. Socio-cultural determinants of meal size and frequency. *Brit J Nutr* **77**, S39-S55.

DÍAZ-PÉREZ, F.M. and BETHENCOURT-CEJAS, M. 2016. CHAID algorithm as an appropriate analytical method for tourism market segmentation. *Journal of Destination Marketing & Management* **5**, 275-282.

DIMOPOULOS, A.C., NIKOLAIDOU, M., CABALLERO, F.F., ENGCHUAN, W., SANCHEZ-NIUBO, A., ARNDT, H., AYUSO-MATEOS, J.L., HARO, J.M., CHATTERJI, S., GEORGOUSOPOULOU, E.N., PITSAVOS, C. and PANAGIOTAKOS, D.B. 2018. Machine learning methodologies versus cardiovascular risk scores, in predicting disease risk. *BMC Medical Research Methodology* **18**, 179.

DINNELLA, C., MONTELEONE, E., PIOCHI, M., SPINELLI, S., PRESCOTT, J., PIERGUIDI, L., GASPERI, F., LAUREATI, M., PAGLIARINI, E., PREDIERI, S., TORRI, L., BARBIERI, S., VALLI, E., BIANCHI, P., BRAGHIERI, A., CARO, A.D., DI MONACO, R., FAVOTTO, S. and MONETA, E. 2018. Individual Variation in PROP Status, Fungiform Papillae Density, and Responsiveness to Taste Stimuli in a Large Population Sample. *Chem Senses* **43**, 697-710.

DIPATRIZIO, N.V. 2014. Is fat taste ready for primetime? *Physiol. Behav.* **136**, 145-154.

DRAYNA, D., COON, H., KIM, U.K., ELSNER, T., CROMER, K., OTTERUD, B., BAIRD, L., PEIFFER, A.P. and LEPPERT, M. 2003. Genetic analysis of a complex trait in the Utah Genetic Reference Project: a major locus for PTC taste ability on chromosome 7q and a secondary locus on chromosome 16p. *Human genetics* **112**, 567-572.

DREWNOWSKI, A., HENDERSON, S.A., SHORE, A.B. and BARRATTFORNELL, A. 1997. Nontasters, tasters, and supertasters of 6-n-propylthiouracil (PROP) and hedonic response to sweet. *Physiol. Behav.* **62**, 649-655.

DYE, L., WARNER, P. and BANCROFT, J. 1995. Food craving during the menstrual cycle and its relationship to stress, happiness of relationship and depression; a preliminary enquiry. *Journal of affective disorders* **34**, 157-164.

FEENEY, E.L. and HAYES, J.E. 2014. Exploring associations between taste perception, oral anatomy and polymorphisms in the carbonic anhydrase (gustin) gene CA6. *Physiol. Behav.* **128**, 148-154.

FOX, A.L. 1932. The relationship between chemical constitution and taste. *P Natl Acad Sci USA* **18**, 115-120.

GANDOMI, A.H., FRIDLINE, M.M. and ROKE, D.A. 2013. Decision tree approach for soil liquefaction assessment. *ScientificWorldJournal* **2013**, 346285-346285.

GARNEAU, N.L., NUESSLE, T.M., MENDELSBERG, B.J., SHEPARD, S. and TUCKER, R.M. 2018. Sweet liker status in children and adults: Consequences for beverage intake in adults. *Food. Qual. Prefer.* **65**, 175-180.

GARNEAU, N.L., NUESSLE, T.M., SLOAN, M.M., SANTORICO, S.A., COUGHLIN, B.C. and HAYES, J.E. 2014. Crowdsourcing taste research: genetic and phenotypic predictors of bitter taste perception as a model. *Frontiers in Integrative Neuroscience* **8**, 33.

GREEN, B.G. and GEORGE, P. 2004a. 'Thermal taste' predicts higher responsiveness to chemical taste and flavor. *Chem. Senses* **29**, 617-628.

GREEN, B.G. and GEORGE, P. 2004b. Thermal taste predicts higher responsiveness to chemical taste and flavour. *Chem Senses* **29**.

GREEN, J., WATERS, E., HAIKERWAL, A., O'NEILL, C., RAMAN, S., BOOTH, M.L. and GIBBONS, K. 2003. Social, cultural and environmental influences on child activity and eating in Australian migrant communities. *Child: Care, Health and Development* **29**, 441-448.

GUO, S.-W. and REED, D.R. 2001. The genetics of phenylthiocarbamide perception. *Annals of Human Biology* **28**, 111-142.

HALLAM, J., BOSWELL, R.G., DEVITO, E.E. and KOBER, H. 2016. Gender-related Differences in Food Craving and Obesity. *The Yale journal of biology and medicine* **89**, 161-173.

HAUGHTON, D. and OULABI, S. 1997. Direct marketing modeling with CART and CHAID. *Journal of Direct Marketing* **11**, 42-52.

HE, C., HOLME, J. and ANTHONY, J. 2014. SNP Genotyping: The KASP Assay. In *Crop Breeding: Methods and Protocols* (D. Fleury & R. Whitford, eds.) pp. 75-86, Springer New York, New York, NY.

HIROKAWA, K., YAMAZAWA, K. and SHIMIZU, H. 2006. An examination of sex and masculinity/femininity as related to the taste sensitivity of Japanese students. *Sex Roles* **55**, 429-433.

HOLT, S.H.A., COBIAC, L., BEAUMONT-SMITH, N.E., EASTON, K. and BEST, D.J. 2000. Dietary habits and the perception and liking of sweetness among Australian and Malaysian students: A cross-cultural study. *Food. Qual. Prefer.* **11**, 299-312.

JABBAR, M.A., DEEKSHATULU, B.L. and CHNDRA, P. 2014. Alternating decision trees for early diagnosis of heart disease. In *International Conference on Circuits, Communication, Control and Computing* pp. 322-328.

JOVANOVIĆ MIOMIR, M. 2017. Decision tree analysis of wine consumers' preferences: evidence from an emerging market. *British Food Journal* **119**, 1349-1361.

KIM, J.Y., PRESCOTT, J. and KIM, K.-O. 2014. Patterns of sweet liking in sucrose solutions and beverages. *Food. Qual. Prefer.* **36**, 96-103.

KIM, U., JORGENSON, E., COON, H., LEPPERT, M., RISCH, N. and DRAYNA, D. 2003a. Positional cloning of the human quantitative trait locus underlying taste sensitivity to phenylthiocarbamide. *Science* **299**.

KIM, U.K., JORGENSON, E., COON, H., LEPPERT, M., RISCH, N. and DRAYNA, D. 2003b. Positional cloning of the human quantitative trait locus underlying taste sensitivity to phenylthiocarbamide. *Science* **299**, 1221-1225.

LAFAY, L., THOMAS, F., MENNEN, L., CHARLES, M.A., ESCHWEGE, E. and BORYS, J.-M. 2001. Gender differences in the relation between food cravings and mood in an adult community: Results from the Fleurbaix Laventie Ville Santé Study. *International Journal of Eating Disorders* **29**, 195-204.

LEOW, M.K. 2017. Characterization of the Asian Phenotype - An Emerging Paradigm with Clinicopathological and Human Research Implications. *International journal of medical sciences* **14**, 639-647.

LIM, J.Y., URBAN, L. and GREEN, B.G. 2008. Measures of individual differences in taste and creaminess perception. *Chem. Senses* **33**, 493-501.

LOH, W.-Y. 2011. Classification and regression trees. *Wiley Interdisciplinary Reviews: Data Mining and Knowledge Discovery* **1**, 14-23.

LOOY, H., CALLAGHAN, S. and WEINGARTEN, H.P. 1992. Hedonic response of sucrose likers and dislikers to other gustatory stimuli. *Physiol Behav* **52**.

MACHUCA, C., VETTORE, M.V., KRASUSKA, M., BAKER, S.R. and ROBINSON, P.G. 2017. Using classification and regression tree modelling to investigate response shift patterns in dentine hypersensitivity. *BMC Med Res Methodol* **17**, 120.

MILLER, B., FRIDLINE, M., LIU, P.-Y. and MARINO, D. 2014. Use of CHAID decision trees to formulate pathways for the early detection of metabolic syndrome in young adults. *Comput Math Methods Med* **2014**, 242717-242717.

MELIS, M., ATZORI, E., CABRAS, S., ZONZA, A., CALO, C., MURONI, P., NIEDDU, M., PADIGLIA, A., SOGOS, V., TEPPER, B.J. and BARBAROSSA, I.T. 2013. The gustin (CA6) gene polymorphism, rs2274333 (A/G), as a mechanistic link between PROP tasting and fungiform taste papilla density and maintenance. *Plos One* **8**, 1-7.

METHVEN, L., XIAO, C., CAI, M. and PRESCOTT, J. 2016. Rejection thresholds (RjT) of sweet likers and dislikers. *Food. Qual. Prefer.* **52**, 74-80.

MICHON, C., O'SULLIVAN, M.G., DELAHUNTY, C.M. and KERRY, J.P. 2009. The investigation of gender-related sensisitivity differences in food preception *J. Sens. Stud.* **24**, 922-937.

MOJET, J., CHRIST-HAZELHOF, E. and HEIDEMA, J. 2001. Taste perception with age: Generic or specific losses in threshold sensitivity to the five basic tastes? *Chem. Senses* **26**, 845-860.

MONTELEONE, E., SPINELLI, S., DINNELLA, C., ENDRIZZI, I., LAUREATI, M., PAGLIARINI, E., SINESIO, F., GASPERI, F., TORRI, L., APREA, E., BAILETTI, L.I., BENDINI, A., BRAGHIERI, A., CATTANEO, C., CLICERI, D., CONDELLI, N., CRAVERO, M.C., DEL CARO, A., DI MONACO, R., DRAGO, S., FAVOTTO, S., FUSI, R., GALASSI, L., GALLINA TOSCHI, T., GARAVALDI, A., GASPARINI, P., GATTI, E., MASI, C., MAZZAGLIA, A., MONETA, E., PIASENTIER, E., PIOCHI, M., PIRASTU, N., PREDIERI, S., ROBINO, A., RUSSO, F. and TESINI, F. 2017. Exploring influences on food choice in a large population sample: The Italian Taste project. *Food. Qual. Prefer.* **59**, 123-140.

NÆS, T., VARELA, P. and BERGET, I. 2018. *Individual Differences in Sensory and Consumer Science* (T. Næs, P. Varela & I. Berget, eds.) pp. 1-14, Woodhead Publishing.

OHSU, T., AMINO, Y., NAGASAKI, H., YAMANAKA, T., TAKESHITA, S., HATANAKA, T., MARUYAMA, Y., MIYAMURA, N. and ETO, Y. 2010. Involvement of the Calcium-sensing Receptor in Human Taste Perception. *The Journal of Biological Chemistry* **285**, 1016-1022.

OVERBERG, J., HUMMEL, T., KRUDE, H. and WIEGAND, S. 2012. Differences in taste sensitivity between obese and non-obese children and adolescents. *Arch Dis Child* **97**, 1048-1052.

PANGBORN, R.M. 1970a. Individual variation in affective responses to taste stimuli. *Psychon Sci* **21**.

PANGBORN, R.M. 1970b. Individual variation in affective responses to taste stimuli. *Psychonomic Science* **21**, 125-126.

PETERSON, J.M., BARTOSHUK, L.M. and DUFFY, V.B. 1999. Intensity and Preference for Sweetness is Influenced by Genetic Taste Variation. *J Am Diet Assoc* **99**, A28.

PICKERING, G.J., MOYES, A., BAJEC, M.R. and DECOURVILLE, N. 2010. Thermal taster status associates with oral sensations elicited by wine. *Aust. J. Grape Wine Res.* **16**, 361-367.

PODGORELEC, V., KOKOL, P., STIGLIC, B. and ROZMAN, I. 2002a. *Decision Trees: An Overview and Their Use in Medicine*.

PODGORELEC, V., KOKOL, P., STIGLIC, B. and ROZMAN, I. 2002b. Decision trees: an overview and their use in medicine. *Journal of medical systems* **26**, 445-463.

PRODI, D.A., DRAYNA, D., FORABOSCO, P., PALMAS, M.A., MAESTRALE, G.B., PIRAS, D., PIRASTU, M. and ANGIUS, A. 2004. Bitter taste study in a sardinian genetic isolate supports the association of phenylthiocarbamide sensitivity to the TAS2R38 bitter receptor gene. *Chem. Senses* **29**, 697-702.

RANKIN, K.M., GODINOT, N., M., C.C., TEPPER, B.J. and KIRKMEYER, S.V. 2004. Assessment of different methods for 6-n-propylthiouracil status classification. In *Genetic variation in taste sensitivity* (J. Prescott & B.J. Tepper, eds.), MARCEL dEKKER, New York.

RISSO, D.S., MEZZAVILLA, M., PAGANI, L., ROBINO, A., MORINI, G., TOFANELLI, S., CARRAI, M., CAMPA, D., BARALE, R., CARADONNA, F., GASPARINI, P., LUISELLI, D., WOODING, S. and DRAYNA, D. 2016. Global diversity in the TAS2R38 bitter taste receptor: revisiting a classic evolutionary PROPosal. *Sci Rep-Uk* **6**, 25506.

ROBINO, A., MEZZAVILLA, M., PIRASTU, N., DOGNINI, M., TEPPER, B.J. and GASPARINI, P. 2014. A Population-Based Approach to Study the Impact of PROP Perception on Food Liking in Populations along the Silk Road. *Plos One* **9**, e91716.

ROININEN, K., TUORILA, H., ZANDSTRA, E.H., DE GRAAF, C., VEHKALAHTI, K., STUBENITSKY, K. and MELA, D.J. 2001. Differences in health and taste attitudes and reported behaviour among Finnish, Dutch and British consumers: a cross-national validation of the Health and Taste Attitude Scales (HTAS). *Appetite* **37**, 33-45.

SCHUTZ, H.G. and CARDELLO, A.V. 2001. A labeled affective magnitude (LAM) scale for assessing food liking/disliking. *J. Sens. Stud.* **16**, 117-159.

SHEN, Y., KENNEDY, O.B. and METHVEN, L. 2016. Exploring the effects of genotypical and phenotypical variations in bitter taste sensitivity on perception, liking and intake of brassica vegetables in the UK. *Food. Qual. Prefer.* **50**, 71-81.

SHANTHI, R. 2019. *Multivariate Data Analysis using SPSS and AMOS*. MJP, Chennai, India.

SHESKIN, D. J. 2011. Handbook of parametric and nonparametric statistical procedures. Chapman and Hall/CRC.SKINNER, M. 2017. Exploring individual variation in oral perception. In *Food Science*, Vol PhD in Sensory Sciences, University of Nottingham, University of Nottingham.

SKINNER, M., ELDEGHAIDY, S., FORD, R., GIESBRECHT, T., THOMAS, A., FRANCIS, S. and HORT, J. 2018. Variation in thermally induced taste response across thermal tasters. *Physiol. Behav.* **188**, 67-78.

STEWART, J.E., FEINLE-BISSET, C., GOLDING, M., DELAHUNTY, C., CLIFTON, P.M. and KEAST, R.S.J. 2010. Oral sensitivity to fatty acids, food consumption and BMI in human subjects. *Brit J Nutr* **104**, 145-152.

TEPPER, B.J., BANNI, S., MELIS, M., CRNJAR, R. and TOMASSINI BARBAROSSA, I. 2014. Genetic Sensitivity to the Bitter Taste of 6-n-Propylthiouracil (PROP) and Its Association with Physiological Mechanisms Controlling Body Mass Index (BMI). *Nutrients* **6**, 3363-3381.

TEPPER, B.J. and NURSE, R.J. 1997. Fat perception is related to PROP taster status. *Physiol. Behav.* **61**, 949-954.

TSIEN, C.L., FRASER, H.S., LONG, W.J. and KENNEDY, R.L. 1998. Using classification tree and logistic regression methods to diagnose myocardial infarction. *Studies in health technology and informatics* **52 Pt 1**, 493-497.

TURNER-MCGRIEVY, G., TATE, D.F., MOORE, D. and POPKIN, B. 2013. Taking the Bitter with the Sweet: Relationship of Supertasting and Sweet Preference with Metabolic Syndrome and Dietary Intake. *J Food Sci* **78**, S336-S342.

ULLRICH, N.V., TOUGER-DECKER, R., O'SULLIVAN-MAILLET, J. and TEPPER, B.J. 2004. PROP taster status and self-perceived food adventurousness influence food preferences. *J Am Diet Assoc* **104**, 543-549.

VIGNINI, A., BORRONI, F., SABBATINELLI, J., PUGNALONI, S., ALIA, S., TAUS, M., FERRANTE, L., MAZZANTI, L. and FABRI, M. 2019. General Decrease of Taste Sensitivity Is Related to Increase of BMI: A Simple Method to Monitor Eating Behavior. *Disease Markers* **2019**, 8.

WILLIAMS, J.A., BARTOSHUK, L.M., FILLINGIM, R.B. and DOTSON, C.D. 2016. Exploring Ethnic Differences in Taste Perception. *Chem. Senses* **41**, 449-456.

WOODING, S., KIM, U.K., BAMSHAD, M.J., LARSEN, J., JORDE, L.B. and DRAYNA, D. 2004. Natural selection and molecular evolution in PTC, a bitter-taste receptor gene. *Am J Hum Genet* **74**, 637-646.

YANG, Q. 2015. Individual variation across PROP and Thermal taste phenotypes. In *Food Science*, Vol PhD, University of Nottingham.

YANG, Q., HOLLOWOOD, T. and HORT, J. 2014. Phenotypic variation in oronasal perception and the relative effects of PROP and Thermal Taster Status. *Food. Qual. Prefer.* **38**, 83-91.

YANG, Q., KRAFT, M., SHEN, Y., MACFIE, H. and FORD, R. 2019. Sweet Liking Status and PROP Taster Status impact emotional response to sweetened beverage. *Food. Qual. Prefer.*

YEOMANS, M.R., PRESCOTT, J. and GOULD, N.J. 2009. Acquired hedonic and sensory characteristics of odours: influence of sweet liker and propylthiouracil taster status. *Quarterly journal of experimental psychology (2006)* **62**, 1648-1664.

YEOMANS, M.R., TEPPER, B.J., RIETZSCHEL, J. and PRESCOTT, J. 2007. Human hedonic responses to sweetness: Role of taste genetics and anatomy. *Physiol. Behav.* **91**, 264-273.

ZELLNER, D.A., GARRIGA-TRILLO, A., ROHM, E., CENTENO, S. and PARKER, S. 1999. Food liking and craving: A cross-cultural approach. *Appetite* **33**.