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Exploring the effects of genotypical and phenotypical variations in bitter taste sensitivity on perception, liking and intake of brassica vegetables in the UK



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ABSTRACT

Brassicaceous vegetables (BV) have chemoprotective effects and yet consumption of BV in the UK is low. Previous studies suggest perception, liking and intake of BV are influenced by bitter taste sensitivity which this study further explores. Phenotypical taste sensitivity of 136 subjects was classified using propythiouracil (PROP) and sodium chloride and fungiform papillae density (FPD) was measured from tongue images. Polymorphisms of TAS2R38 and gustin (CA6) genes were analysed. Liking and bitterness of four raw vegetables (two BV (broccoli and white cabbage) and two non-BV (spinach and courgette)), as well as habitual consumption, were evaluated.

There was a significant association between TAS2R38 genotype and PROP taster status (p < 0.0001) and between FPD and PROP taster status (p = 0.029). Individuals with greater sensitivity for PROP predominantly had TAS2R38 PAV/PAV genotype and greater FPD. BV were perceived as more bitter than non-BV (p < 0.0001) with PAV/PAV subjects perceiving significantly stronger bitter intensity. There was a significant difference in liking for the four vegetables (p = 0.002), and between consumers of different TAS2R38 genotype (p = 0.0024). Individuals with TAS2R38 AVI/AVI genotype liked BV more. Regarding intake, both PAV/PAV and AVI/AVI individuals consumed more total vegetables and BV than PAV/AVI. Although PROP nontasters tended to consume more vegetables and BV than the other two phenotype groups, liking and vegetable intake were not significantly affected by taste phenotype. Although there was not a significant effect of CA6 genotype on bitterness ratings, there was a significant interaction between CA6 and TAS2R38, and in addition CA6 genotype was significantly associated with BV intake. However, these effects require validation as the proportions of the population with the CA6 G/G genotype was extremely small (7%).

Our results confirmed that bitter taste perception in vegetables was influenced by both genotype and phenotype of bitter taste sensitivity. Moreover, our findings demonstrated that neither genotype nor phenotype of taste sensitivity alone accurately predict vegetable liking and intake as demographic factors were found to have a substantial influence.

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1. Introduction

Obesity has become a major public health concern in the last two decades (OECD, 2014). Improving diet by increasing the intake of vegetable and fruit instead of energy-rich food has been highlighted as an important way to control obesity, and may also reduce the risk of cancer and cardiovascular diseases (Nasser, 2001; Rolls, 2007). It is well known that food preference and choice

are determined by various factors (Mela, 2007; Shepherd, 2007). Taste has been reported as one important factor in food perception, liking and choice (Galindo, Schneider, Stahler, Tole, & Meyerhof, 2012; Grimm & Steinle, 2011). It is also reported as one of the main reasons for rejection of vegetables, particularly brassicaceous vegetables (BV) (Drewnowski & Gomez-Carneros, 2000; Gorovic et al., 2011). Of potential health benefit in brassicaceous vegetables are glucosinolate compounds and their hydrolysis products which are thought to reduce the risk of cancer (Jahangir, Kim, Choi, & Verpoorte, 2009; Verkerk et al., 2009). However, the thiourea group (N—C=S) within these compounds is predominantly responsible for their bitter taste, which may, therefore, contribute to their

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low consumption (Dinehart, Hayes, Bartoshuk, Lanier, & Duffy, 2006: Drewnowski, 2000).

The difference between humans in their phenotypical response to the thiourea group was discovered by Fox (1932) in the early 20th century. According to their phenotypical responses to PROP, individuals can be classified into three groups, PROP supertasters, medium-tasters, and nontasters (Bartoshuk, 1993; Zhao, Kirkmeyer, & Tepper, 2003). Further research led to the conclusion that approximately one third of the population could be categorised as "taste-blind" (PROP nontaster) to the thiourea group (Drayna, 2005). A genetic difference in a bitter taste receptor has since been found to be the major cause of bitter "blindness" to the thiourea group (Bakke & Vickers, 2011; Kim, Breslin, Reed, & Drayna, 2004; Prodi et al., 2004). The TAS2R38 receptor specifically detects compounds containing the bitter thiourea group in PTC and PROP. Three functional single nucleotide polymorphisms (SNPs) within this gene have been found to explain up to 85% of the observed variance in PTC and PROP taste sensitivity (Drayna, 2005; Kim et al., 2003). These polymorphisms encode amino acid substitutions at position 49 (alanine/proline, A49P), 262 (valine/ alanine, V262A), and 296 (isoleucine/valine, I296V) (Drayna, 2005; Kim et al., 2003). Generally, the TAS2R38 gene has two common haplotypes: Pro-Ala-Val (PAV), the dominant (sensitive) variant, and AVI (Ala-Val-Ile), the recessive (insensitive) one (Kim et al., 2004). Individuals with the homozygous AVI haplotype (AVI/AVI) are unable to taste bitter from PROP or other compounds with thiourea groups. In contrast, individuals who carry homozygous of the PAV haplotype (PAV/PAV) have a low detection threshold for PROP and perceive a strong bitter intensity from PROP. Others that are heterozygous with the PAV/AVI haplotype perceive a medium level intensity from PROP solutions (Calo et al., 2011; Hayes, Bartoshuk, Kidd, & Duffy, 2008).

A number of studies have suggested that fungiform papillae density (FPD) on the anterior tongue also plays an important role in the bitter taste intensity perceived from PROP (Bakke & Vickers, 2011; Delwiche, Buletic, & Breslin, 2001). Bartoshuk, Duffy, and Miller (1994) suggested that the PROP supertaster group had the highest FPD. Recent studies have investigated a further genetic explanation for PROP taste sensitivity through understanding the genetic cause of FPD variation, where more papillae may result in greater receptor cells concentration. Evidence suggests that gustin (CA6), a trophic factor for taste bud development, is strongly associated with PROP taste sensitivity and FPD (Barbarossa et al., 2015; Calo et al., 2011; Melis, Atzori et al., 2013; Padiglia et al., 2010). Individuals who present higher PROP sensitivity are more likely to carry the A/A genotype of CA6 on SNP rs2274333, and those with lower PROP sensitivity tend to have G/G genotypes (Calo et al., 2011; Melis, Atzori et al., 2013). In addition, individuals with the G/G genotype had lower FPD and showed more gross morphological changes in fungiform papillae (Melis, Atzori et al., 2013). However, results from Feeney and Hayes (2014) failed to find an association between 12 SNPs within CA6 gustin genotype and FPD.

The relationship between bitter taste sensitivity and food preference has become a prevalent research topic over the last twenty years. The study of Drewnowski, Henderson, Levine, and Hann (1999) showed that PROP phenotype supertaster young women tended to have lower preference for the bitter taste perceived from brassica vegetables such as broccoli and Brussel sprouts compared with nontasters. Moreover, results from Bell and Tepper (2006) also suggested that PROP nontaster children consumed significantly more bitter vegetables used in their study (broccoli, olives and cucumber) than medium tasters and supertasters. As the underlying cause of PROP taste phenotype is the genetic difference in bitter receptor TAS2R38, it is likely that TAS2R38 genotype will play an important role in bitter perception of brassica vegetables.

Sandell and Breslin (2006) reported that PAV/PAV individuals' rated bitter intensity of various brassica (but not non-brassica) vegetables on average 60% higher than the AVI/AVI group. However, only a few studies have investigated the relationship between TAS2R38 and brassica vegetable intake. Sacerdote et al. (2007) analysed TAS2R38 genotype in 634 healthy subjects randomly sampled from the Italian EPIC cohort and concluded that individuals with AVI/AVI genotype had significantly higher brassica vegetable intake. Duffy et al. (2010) also found AVI/AVI subjects had higher overall vegetable consumption in 59 college-aged adults in the US. However, not all studies supported this relationship. Catanzaro, Chesbro, and Velkey (2013) did not find any significant difference in preference for brassica vegetables between PROP taster status groups within 139 college-age participants in US. Other studies (Baranowski et al., 2011; Keller & Tepper, 2004; Lumeng, Cardinal, Sitto, & Kannan, 2008) have also found no significant association between phenotypes of PROP sensitivity and actual vegetable intake in children. Furthermore, Timpson et al. (2005) did not find any association between TAS2R38 genotype and brassica vegetable intake in 4286 British women aged 69-79 years old.

In summary, the effects of taste sensitivity on vegetable preference and intake remain controversial from previous studies. Most studies only consider intensity or preference or intake and do not investigate these factors together. Very few studies have utilised actual tasting of vegetables in their study. No studies to date have combined the genotypes for both TAS2R38 and gustin, as well as phenotypical measures of taste sensitivity, into a study of vegetable perception, liking and intake. Therefore, the aim of this study was to investigate both phenotype (PROP taste status and FPD) and genotype (TAS2R38 and CA6) measurements of taste sensitivity, and their effects on brassica vegetable perception, liking and intake.

2. Materials and methods

2.1. Subjects

Healthy adults age 18–55 years, were recruited by advertisement within and around the University of Reading, UK. Exclusion criteria included smoking, pregnancy, relevant food allergies, major medical conditions and medication used that could impact on taste perception. The study had been given a favourable opinion to proceed by the University of Reading Ethics committee (study number 12/04).

2.2. Suprathreshold test

A suprathreshold test was used to classify PROP taster status of the participants. They were classified into supertaster, medium taster and nontaster groups using the method described by Tepper, Christensen, and Cao (2001). In order to determine the perceived intensity of bitterness from PROP and saltiness from sodium chloride, three levels of PROP (0.000032 M, 0.00032 M, 0.0032 M) and salt (0.01 M, 0.1 M, 1 M) were used, and participants were asked to rate the intensity on a Labelled Magnitude Scale (LMS). To ensure that participants were comfortable and able to make proper use for this scale, a practice scale was used where participants were asked to rate their memory of food that had the strongest intensity of sweet, bitter, sour, and salt from their own food experience.

Using the classification method from Tepper et al. (2001), if participants rated the NaCl solutions higher than PROP, they were classified as nontasters, if intensity rating of PROP solution higher than NaCl, they were supertasters. Medium-tasters were classified as those who gave similar perceived intensity ratings for PROP and NaCl

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