Reduced taste responsiveness and increased food neophobia characterize obese adults
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Abstract
The aim of the present study was to investigate the relationship between two well-established markers of taste perception, 6-n-propylthiouracil (PROP) responsiveness and fungiform papillae number, in obese and healthy-weight subjects. The association between taste responsiveness and food neophobia attitude was evaluated to understand if these variables are linked to nutritional status of subjects.

Forty healthy-weight (Body Mass Index: 22.67 ± 0.43 kg/m²) and forty-five obese (Body Mass Index: 37.57 ± 0.77 kg/m²) subjects were involved. PROP responsiveness and fungiform papillae number were positively correlated to each other in both groups of subjects (healthy-weight: r= 0.67, p<0.001; obese: r=0.83, p<0.001). PROP responsiveness ratings and fungiform papillae number were significantly negatively correlated with food neophobia scores in both group of subjects (p<0.01). Subjects characterized as significantly less sensitive and more neophobics had a higher Body Mass Index. Especially, obese men showed significant lower taste responsiveness (p<0.05) and higher food neophobia scores (p<0.05) compared to obese women and healthy-weight subjects, both sexes.

The nutritional status of the subjects seems to be linked to taste responsiveness and food neophobic attitude. These data suggest that, between several factors which could play a role in the control of
body weight, understand how sensory perception affects eating behavior could give important
information to study variables which may determine food habits.

**Keywords:** PROP; fungiform papillae; BMI; overweight; sensory perception; eating behavior.

### 1. Introduction

Sensory perception varies widely across individuals but the link to actual eating behaviour, nutrition
and health is not that clear (Tepper, 2008). Possible explanations for this great individual variability
are environmental factors (Köster, 2009) as well as genetic background (Bajec & Pickering, 2008).
One of the most studied genetic sources of individual variation is the ability to taste the bitter
compound 6-n-propylthiouracil (PROP) (e.g. Yackinous & Guinard, 2001; Duffy, 2007; Tepper,
2008; Tepper et al., 2009). Previous studies reported that PROP responsiveness is associated with
sensitivity to a variety of oro-sensory stimuli. Super-tasters (i.e., subjects highly responsive to
PROP) perceive saltiness, sweetness, and sour more intensely than medium and non-tasters (i.e.,
subjects less responsive to PROP) (Duffy et al., 2003; Hayes & Duffy, 2007; Prescott et al., 2004).
These differences in taste responsiveness have a remarkable effect on food acceptance, with for
example, non-tasters more likely to be sweet likers while super-tasters more likely to be sweet
dislikers (Yeomans et al., 2007).

PROP responsiveness is also related to anthropometric, physiological and behavioral measurements
but literature data are controversial. Different studies showed an inverse association between PROP
responsiveness and Body Mass Index (BMI) (Tepper & Ullrich, 2002; Goldstein et al., 2005; Burd
et al., 2012) whereas others did not (Bajec & Pickering, 2010; Villarino et al., 2009; Borazon et al.,
2012). Moreover, a wide range of literature suggests that PROP responsiveness is positively related
to density of lingual fungiform papillae which are structures containing taste buds. Subjects with a
higher number of fungiform papillae are more sensitive to tastes (Bartoshuk, 2000; Delwiche et al.,
2001; Hayes et al., 2008; Masi et al., 2015). However, there are also recent findings not supporting
the association between PROP responsiveness and fungiform papillae (Fisher et al., 2013; Garneau et al., 2014; Webb et al., 2015).

Previous research led by our group (Bertoli et al., 2014; Proserpio et al., 2016) showed that overweight and obese subjects have a reduced taste sensitivity that might increase food desire, thus leading to excessive energy intake and weight gain. A recent neuroimaging study seems to support this hypothesis showing that gustatory stimulation induced differential fMRI brain activation patterns in obese compared to healthy subjects (Szalay et al., 2012).

In our studies taste sensitivity was measured through the 3-Alternative Forced Choice (3AFC; ASTM E 679-04 (2011), a robust and reliable procedure, which is, however, difficult to apply in an ambulatory context involving obese subjects undergoing a weight-loss therapy. Faster and easier approaches, such as the count of the fungiform papillae and PROP responsiveness, would be more appropriate in this context, due to their simpler, but reliable, procedures (Zhao et al., 2003; Rankin et al., 2004). Indeed, taste response to PROP, as well as the density of fungiform papillae, are well-studied markers of genetic variation in taste and oral sensation perception (e.g. Bartoshuk et al., 1994; Miller & Reedy, 1990; Zuniga et al., 1997; Bajec & Pickering, 2010; Tepper 2008; Duffy et al., 2010; Tepper et al., 2014; Feeney & Hayes, 2014). Moreover, the fungiform papillae number, which is not a reported measure, could be helpful in order to avoid biased report ratings.

Besides individual variation in taste responsiveness, food neophobia (literally the fear of novel food) is another aspect to be considered as it plays an important role in shaping food preference and rejection (Pliner & Hobden, 1992). This behavior has been largely studied in omnivores, including humans but its association with taste perception and nutritional status is under debate. Knaapila and colleagues (2011) reported a weak correlation between food neophobia scores and BMI in young women but not in men. Other authors observed that BMI is higher in food neophobic than in food neophilics (Finistrella et al., 2012; Knaapila et al., 2015).

In a previous study, we hypothesized that obese adults may have a higher neophobic attitude than healthy controls but, unexpectedly, we did not find significant differences (Proserpio et al., 2016).
This maybe was due to the deliberately or unwittingly biased report ratings that obese subjects gave about their eating behaviour (Klesges et al., 1988). It is well recognized that obese subjects have the tendency, either intentional or as a form of self-deception, to answer to dietary and eating behaviour questions as expected by the interviewer (Heitmann, 1996).

In this context, among all the several factors which could play a role in the control of body weight, the relation between taste perception and food neophobia is still under investigation.

The aim of the present study was to compare taste perception in obese and healthy-weight subjects using two well-established markers of taste responsiveness, i.e. PROP responsiveness and fungiform papillae number. The relationship between these two markers was also investigated, since we hypothesized that if these two measurements are related, one of these methods could be preferred to investigate taste responsiveness when the 3AFC or similar procedures are not easy to be performed (i.e. ambulatory context). Finally, due to the lack of agreement in the literature, the association between taste responsiveness and food neophobia attitude was evaluated in order to understand if these variables are linked to the nutritional status of the subjects. Gender has been also considered due to its role on BMI and food neophobia attitude (Monteleone et al., 2017).

2. Materials and Methods

2.1 Subjects

Eighty-five adults completed the study. Forty-five obese subjects were recruited among patients admitted to the Department of Medical Sciences and Rehabilitation before starting their weight loss treatment (IRCCS Istituto Auxologico Italiano). Forty healthy-weight subjects were recruited among employees of the Faculty of Agriculture and Food Sciences of the University of Milan.

Sample size was chosen assuming a standardized effect size around 0.70, $\alpha = 0.05$ and $\beta = 0.20$, which gives approximately 35 subjects for each BMI group. All subjects were invited to a screening session, around 9:00 am, to assess the anthropometric measurements by collecting body weight (to the nearest 0.1 kg) and standing height (to the nearest 0.1 cm) using the same calibrated scale on a
telescopic vertical steel stadiometer (SECA 220; Germany), with subjects dressed only in underwear. BMI was calculated accordingly \([\text{weight (kg)}/\text{height (m}^2\text{)}]\). Subjects with BMI higher than 30 were classified as obese, while subjects with BMI between 18 and 25 were classified as healthy-weight. Participants’ characteristics are presented in Table 1.

Table 1. Participants’ characteristics (data are reported as mean values ± SEM)

<table>
<thead>
<tr>
<th></th>
<th>Healthy-weight (n=40)</th>
<th>Obese (n=45)</th>
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<tbody>
<tr>
<td></td>
<td>Women (n=21)</td>
<td>Men (n=19)</td>
</tr>
<tr>
<td>Age (years)</td>
<td>40.38 ± 1.37</td>
<td>41.84 ± 2.74</td>
</tr>
<tr>
<td>BMI (kg/m(^2))</td>
<td>21.59 ± 0.53</td>
<td>22.86 ± 0.60</td>
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The exclusion criteria were: aged > 65 years, experienced ageusia, pharmacological therapy that could modify taste perception, smokers and diabetics. All subjects were invited to take part to one session before lunch from 12.00 to 13.00, and were assessed for their taste responsiveness in pre-prandial condition. Subjects were also asked to complete a questionnaire concerning food neophobia. This study was approved by the Ethic Committee of the IRCCS Istituto Auxologico Italiano and written informed consent was obtained from all subjects after full explanation of the study. This study was conducted according to the guidelines laid down in the Declaration of Helsinki.

2.2 Taste responsiveness assessment

2.2.1 PROP responsiveness

PROP responsiveness was established using PROP-impregnated filter paper according to the procedure described by Bartoshuk and colleagues (2003). 3 cm\(^2\) filter papers (Whatman) were soaked in a saturated aqueous PROP (6-n-propyl-2-thiouracil, Sigma-Aldrich, Spa, Milano) solution heated to near boiling temperature. Papers were air dried and stored at room temperature in small glassine envelopes for a maximum of 24 hours. Each paper contained around 1.6 mg PROP. PROP
crystallizes into the filter paper making it a convenient vehicle to deliver a measured amount of material into the mouth. Comparing the average perceived bitterness of PROP papers with those of PROP solutions, PROP paper falls between the perceived bitterness of 0.001 and 0.0032 M PROP (Bartoshuk et al., 2003). Using paper filter has the advantage of being easy to administer to subjects in ambulatory conditions and it has been used rather than solutions since it is equally valid and shows high test-retest reliability (Zhao et al., 2003; Rankin et al., 2004).

Prior to the test, subjects practiced the general version of the Labeled Magnitude Scale (gLMS; Green et al. 1993, 1996) by rating a list of remembered or imagined oral sensations (e.g., the sweetness of cotton candy and the burn of cinnamon gum) to give them experience using the scale in the broad context of normal oral perception (Green et al., 2012). Subjects were instructed that PROP intensity should be rated in the context of other sensations and that the endpoint of the scale equalled the most intense sensations imaginable, such as hearing a jet plane take off overhead or looking into the sun. After these explanations about the use of the scale, each respondent was instructed to put a filter paper on the tongue, wait 10 seconds and rate the perceived bitter intensity using the gLMS.

**2.2.1 Fungiform papillae number evaluation**

Fungiform papillae number was calculated using a method previously described by our group (Proserpio et al., 2016) with a procedure proposed by Nachtsheim and Schlich (2013) and Bakke and Vickers (2011). Subjects’ tongues were painted with a blue food dye (F.Ili Rebecchi, Color Dolci, Spa, Milano, Italy). A circle of filter paper (7 mm diameter) was positioned on the centre of their tongue at 1–2 cm from the tip. In a bright room several photos of the tongue were taken using a 12-megapixel digital camera (FUJIFILM USA, Inc.) in macro mode with no flash. After the selection of the best photograph, Adobe Photoshop was used to draw three outlined circles of the same size as the filter paper circle that had been placed on the tongue. Those papillae that were in contact with the outlined circle were counted if more than 50% of individual papillae were within
the boundary. The fungiform papillae were counted independently by three researchers and the mean of the counts was calculated.

2.3 Food neophobia assessment

Food neophobia was measured using the Italian translation of the Food Neophobia Scale (FNS) (Pliner & Hobden, 1992), as described by Proserpio and colleagues (2016). The FNS consists of ten statements, such as “I do not trust new foods” with a seven-category response scale ranging from “strongly disagree” (score 1) to “strongly agree” (score 7). Half of the statements are worded in reverse relative to food neophobia (e.g. “I like foods from different countries”), so responses to these statements were reversed when calculating the score. The FNS score was computed as a sum of the responses, yielding a range of 10–70, with higher scores indicating higher food neophobia. Internal consistency, as measured by Cronbach’s alpha, was 0.87.

3. Statistical analysis

Statistical analysis was performed using STATGRAPHICS PLUS v.16 software (Manugest KS Inc.). The data were normally distributed according to Shapiro-Wilk test. An ANCOVA with BMI (healthy-weight vs obese), gender and their two-way interaction as independent variables and PROP responsiveness, fungiform papillae number, and food neophobia scores as dependent variables was performed. Age was included as covariate. When a significant effect (p<0.05) was found, Tukey’s post-hoc test was used.

Correlations between all measurements (PROP responsiveness, fungiform papillae and food neophobia scores) were examined using Pearson's correlation coefficient with a minimum significance level defined as p<0.05. The relationship between food neophobia score and taste responsiveness (PROP responsiveness vs food neophobia scores; fungiform papillae vs food neophobia scores) in both healthy-weight and obese subjects, was also explored comparing slopes and intercepts of the regression lines through analysis of variance using the Comparison Regression
4. Results

4.1. Taste responsiveness and food neophobia assessment

Mean values for PROP responsiveness, fungiform papillae number and food neophobia according to BMI and gender are reported in Figure 1a-c. According to ANCOVA results, the interaction between BMI and gender was significant for all the variables investigated (PROP responsiveness: $F_{(1,80)}=4.19 \ p<0.05$; fungiform papillae: $F_{(1,80)}= 8.14 \ p<0.01$; food neophobia $F_{(1,80)}= 5.53 \ p<0.05$).

PROP responsiveness (Figure 1a) was significantly lower in obese men ($14.44 \pm 4.88$) than all other subjects (obese women: $32.80 \pm 4.05$; healthy-weight women: $40.01 \pm 4.51$; healthy-weight men: $40.04 \pm 4.67$), who were in turn comparable to each other.

Looking at Figure 1b, obese men showed significant lower fungiform papillae number ($9.40 \pm 0.80$) than other subjects (obese women: $13.47 \pm 0.66$; healthy-weight women: $15.21 \pm 0.74$; healthy-weight men: $15.36 \pm 0.77$).

Considering food neophobia scores (Figure 1c), obese men ($42.25 \pm 2.34$) were significantly more neophobic than obese women ($33.85 \pm 1.94$). Both obese women and men were significantly ($F_{(1,80)}=19.86; \ p<0.001$) more neophobic than healthy-weight subjects (men: $27.26 \pm 2.24$; women: $28.81\pm2.16$). The covariate age was not significant for all the measured variables.

4.2. Relationship between PROP responsiveness and fungiform papillae number

The correlation between fungiform papillae number and PROP responsiveness according to BMI and gender is reported in Figure 2a-b.

A significant positive correlation between PROP responsiveness ratings and fungiform papillae number was found in both groups of subjects (healthy-weight: $r= 0.67, \ p<0.001$; obese: $r=0.83$,
p<0.001). Healthy-weight subjects, both women and men, were distributed quite homogenously in
the space, while generally obese men tended to be positioned in the left part of the graph, indicating
lower taste responsiveness compared to obese women and confirming the reduced taste
responsiveness of obese men.

4.3. Relationship between taste responsiveness and food neophobia

Results showed that both models (PROP responsiveness vs food neophobia scores; fungiform
papillae number vs food neophobia scores) were significant \( \text{F}(3,81)=42.32, \, p<0.001; \, \text{F}(3,81)=25.91, \, p<0.001, \) respectively. The comparisons of regression lines, according to BMI and gender, are
shown in Figure 3a-b.

Considering PROP responsiveness vs food neophobia score (Figure 3a) a significant negative
correlation was found in both group of subjects (healthy-weight: r=-0.73 p<0.01; obese: r=-0.72
p<0.01). Similarly, looking at Figure 3b, a significant negative correlation between fungiform
papillae number and food neophobia scores was found in both BMI group (healthy-weight: r=-0.46
p<0.01; obese: r=-0.67 p<0.01).

The intercepts of the healthy-weight versus obese subjects were significantly different from each
other in both cases (PROP responsiveness vs food neophobia score: \( \text{F}(1,3)=9.55 \, p<0.01 \); fungiform
papillae number vs food neophobia scores: \( \text{F}(1,3)=4.06 \, p<0.05 \). Similarly, the slopes of the healthy-
weight versus obese subjects were significantly different from each other in both cases (PROP
responsiveness vs food neophobia score: \( \text{F}(1,3)=14.41 \, p<0.001 \); fungiform papillae number vs food
neophobia scores: \( \text{F}(1,3)=8.24 \, p<0.001 \). The distribution of responses highlights that participants
classed as obese tended to have lower tastes sensitivity score and higher food neophobia than
healthy-weight participants.

5. Discussion
The aim of the present study was to investigate the relationship between PROP responsiveness and fungiform papillae number in healthy-weight and obese subjects. The association between taste responsiveness and food neophobia was also evaluated to understand if these variables are linked with subjects’ nutritional status. Our data showed lower taste responsiveness and higher food neophobia in subjects with higher BMI, especially men. Both measurements used to evaluate taste responsiveness were positively correlated to each other. Moreover, taste responsiveness was negative correlated with food neophobia scores in both BMI groups.

The present results support previous findings reporting that subjects who differ in their response to PROP, as well as other taste stimuli, have anatomical differences in the tongue, i.e. number of fungiform papillae (Yackinous & Guinard, 2002; Essick et al., 2003; Shahbake et al., 2005; Bajec & Pickering, 2008). This association has not been confirmed in more recent larger studies (Fisher et al., 2013; Garneau et al., 2014) which, however, did not include severe obese subjects. It is difficult to implement large population studies including obese subjects due to the recruitment phase and their low willingness to participate in laboratory tests. Nevertheless, it is crucial to consider this specific target population for their taste perception which may varies considerably according to diet and vice versa.

Although in the present study food preferences and dietary intake have not been considered, we can speculate that subjects with higher BMI could prefer high energy dense foods, rich in fat and sugar, to compensate their altered/reduced sensitivity (Bertoli et al., 2014; Proserpio et al., 2016), influencing their body weight. Moreover, recent evidence has shown that, in a small cohort of obese subjects, reduction in dietary fat content for 6 weeks improved taste sensitivity to fat (Newman et., 2016), highlighting the likely association between taste perception, food preference and consumption. Similarly, Wise and colleagues (2016) showed that the reduction of simple sugars in a group of obese subjects increased the perceived sweet taste intensity without decreasing the pleasantness.
Since obese subjects are often under weight-loss treatment in ambulatory context, it is essential to flash out easy but reliable methods to assess their taste responsiveness. In this context, fungiform papillae count, as well as PROP responsiveness, could be helpful to monitor taste responsiveness in particular group of subjects and settings. One advantage of fungiform papillae counting, rather than PROP evaluation, is that it is not a reported measure thus it could limit bias associated to the use of the scale.

The present results showed that, obese subjects, both women and men, were significantly more neophobic compared to the control group while we did not previously find differences in food neophobia accordingly to body weight (Proserpio et al., 2016). One possible explanation for the contrasting results is the deliberately or unwittingly biased report ratings that obese subjects give about their eating behaviours (Klesges et al., 1988). Indeed, it is well known that there is often discrepancy between self-reported and actual eating behavior in obese subjects, with for example dietary under-reporting (Gemming et al., 2015). In the present study, in order to limit this problem, a researcher explained to the obese subjects that the responses would not have been used by the dieticians to evaluate their food consumption or their adherence to the diet. It could be interesting and helpful in future studies to adopt implicit methods, such as behavioural tasks, physiological correlates (Finlayson et al., 2008) to evaluate eating behaviour in particular groups of subjects (e.g. overweight and obese subjects, restrained eaters) in order to avoid biased report ratings.

The link between food neophobia and nutritional status is not that clear. On one hand, food neophobia might limit the variety of the diet overall, thus reducing energy intake; on the other hand, food neophobics could prefer to consume traditional foods with a higher energy density compared with healthier food versions, resulting in a higher BMI (Laureati et al., 2015). This second line of thought is in accordance with the present findings as well as earlier results (Knaapila et al., 2011, 2015). Knaapila and colleagues (2011) speculated that the two aforementioned opposite lines of though can compensate each other, thus explaining the lack of association found by other authors in children (Laureati et al., 2015).
The present results suggested also that food neophobia is negatively correlated to taste responsiveness in both healthy-weight and obese subjects. It means that some aspects linked to sensory perception may be involved in the refuse of novel foods affecting the variety of the diet. The obese subjects involved, particularly men, were characterized as being more neophobic and were also significant less sensitive compared to the healthy-weight subjects. Studies on large population samples identified that, independently from the nutritional status, gender as significant predictor of PROP bitterness intensity, with male showing lower ratings than females (Monteleone et al., 2017; Fischer et al., 2013; Garneau et al., 2014) and it is more generally reported that men are less sensitive to stimuli than females (Doty & Cameron, 2009). Moreover, men were found in large cohorts of subjects more neophobic compared to women (Monteleone et al., 2017; Siegrist et al., 2013). This has been explained by the greater involvement of women rather than men in food purchase and preparation leading women to be more familiar with a broader set of foods than men (Hartmann et al., 2013). Contrary to the present findings, no significant variation in taste sensitivity for participants scoring higher and lower in food neophobia were found in our previous study (Proserpio et al., 2016). This discrepancy could be due to the different methods used to evaluate taste responsiveness as well as food neophobia and perception categorization. According to the present results it has been proposed that the fear of trying new foods mediates the effect of PROP responsiveness on food liking (Ullrich et al., 2004), thus potentially influencing eating behavior and energy intake. Ullrich and colleagues (2004) showed that PROP tasters, who were more food adventurous, liked stronger tasting foods (e.g., chili peppers, pungent condiments, bitter fruits and vegetables) more than who were less food adventurous. Further researches are needed to understand the complex relation between food neophobia, taste perception and its implication in food choice and consumption.

Some limitations of the present study should be considered. First, the reduced number of subjects does not enable results generalization, thus further research in larger groups is recommended.
Second, it could be useful in future researches to consider also food preferences and dietary data which have not been evaluated in the present study.

**Conclusions**

The current study suggests that body weight could have an impact on the measured variables. However, the causality of this relationship cannot be inferred from association studies. Other studies are needed to understand whether is the nutritional status that affects taste responsiveness, thus influencing eating behaviour, or whether is the altered taste responsiveness that affects BMI. These variables, as well as other factors (i.e. genetic, metabolic, anthropometric and behavioral variables) further need to be examined since they could give important information to better understand the complex issue of overeating.

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The author contributions were as follows: CP, ML and EP designed the study. CP carried out the experiment, performed the statistical analysis and wrote the manuscript. CP, ML, EP, CI regularly discussed the experiment, analyzed the results, and provided useful suggestion during the writing. All authors read and approved the final manuscript.

The authors declare no conflict of interest.

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Figure 1a-c. Mean values of the studied variables (PROP responsiveness ratings, fungiform papillae number, and food neophobia scores) ± standard error by BMI and gender. Mean values marked with different superscript letters are significantly different (Tukey, p<0.05).
Figure 2a-b. Scatterplots representing the correlation between PROP responsiveness and fungiform papillae number by BMI and gender.
Figure 3a-b. Comparison of the slopes and the intercepts of regression lines between the ‘PROP responsiveness vs food neophobia scores’ and the ‘fungiform papillae number vs food neophobia scores’ in the two groups of subjects, according to BMI and gender (--- obese; __ healthy-weight).