



# Influence of the PROP bitter taste phenotype and eating attitudes on energy intake and weight status in pre-adolescents: A 6-year follow-up study



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## HIGHLIGHTS

- We examined PROP& eating attitudes as predictors of weight in preadolescents during a 6 yr follow up.
- PROP was not related to current BMI or change in BMI from baseline.
- Baseline BMI and physical activity were the strongest predictors of weight.
- Other predictors were: child restraint,PROP\*gender interaction & maternal factors.
- PROP and eating attitudes are modest predictors of weight in preadolescents.

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## ABSTRACT

The PROP bitter-taste phenotype is a marker for food preferences and eating behavior, and may associate with differences in body weight in children. Previous work has shown that PROP status in combination with eating attitudes are better predictors of weight status in preadolescents, than either factor alone. However, no studies have examined the role of PROP phenotypes in body weight change in children over time. The primary objective of this study was to investigate current weight status and change in weight status in children from preschool (baseline) to preadolescence as a function of eating attitudes and PROP phenotype. Other measures included self-reported food intakes and physical activity by activity monitor. Seventy-three lean (BMI percentile =  $57.7 \pm 3.2\%$ ) children with mean age =  $10.3 \pm 0.5$  yrs, participated in the follow up. There were no group differences in energy intake, current BMI-percentile or change in BMI percentile from baseline by PROP phenotype in either boys or girls. However, there was a trend for non-taster girls to show a downward shift in BMI-percentile at follow up. Hierarchical regression analysis revealed that baseline BMI percentile and physical activity energy expenditure were the strongest predictors of current weight (28.5% variance), followed by child restraint, the taster  $\times$  gender interaction, and the maternal BMI  $\times$  maternal emotional eating interaction, accounting for 7.1%, 6.0% and 4.8% of variance in the model, respectively. These findings suggest that PROP status and eating attitudes are modest predictors of weight status in preadolescent children.

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## 1. Introduction

The prevalence of pediatric obesity has increased three-fold during the past thirty years [34]. Obesity in children is of extreme concern because weight status tends to track over time, and overweight children are 2–6 times more likely to become obese adults when compared to normal-weight children [16,28]. Eating patterns are also established early in the lifecycle, and once entrenched are difficult to change [2].

Thus, it is important to identify the critical determinants of eating behavior in children order to instill more healthful dietary habits early in life as well develop better primary prevention strategies for childhood obesity.

Food preferences are mediated, in part, by genetic predispositions [23,43]. Our laboratory has been examining the role of the 6-n-propylthiouracil (PROP) taste phenotype as a general index of food preferences and eating behaviors in children and adults [39,41]. Individuals who are taste blind to PROP (i.e., non-tasters) perceive less intensity from a range of oral sensations including sweetness, bitterness, oral pungency and creaminess. In contrast, those who are moderately or extremely responsive to PROP bitterness (medium or super-tasters, respectively) perceive more intensity from these same oral sensations. These differences are apparent at an early age and are shown to influence

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food selection. Studies in young children have shown that in comparison to taster children, non-tasters are more likely to accept bitter-tasting vegetables and fruit juices [4,40,45] and soy foods [44]. In addition, non-taster children gave higher acceptability ratings to full-fat milk [20] and they reportedly consumed more added fats in the diet than tasters [21]. The relative contribution of these dietary patterns to energy intake in PROP-classified children is unknown.

The involvement of the PROP bitter taste phenotype in body weight status in children is controversial. Two studies in preschool children reported that male non-tasters were heavier than male tasters, but the opposite (non-significant) trend was seen in girls [19,21]. Other studies reported no differences in body weight as a function of PROP status in this age group [27]. A large, population-based study in older children (7–18 yrs of age) of different ethnic and socioeconomic groups also found no differences in weight related to PROP status [3].

Parental characteristics such as weight status, eating attitudes and child feeding practices are known to play a critical role in the development of children's food patterns and body weight [12,17,28]. Goldstein et al. [14] investigated the contribution of PROP taster phenotype and several maternal characteristics to these outcomes in a convenience sample of 9 year old children. Results showed that non-tasters consumed more energy than tasters, but no differences in body weight were found among the groups. Rather, maternal body mass index (BMI; kg/m<sup>2</sup>), restriction of child's food intake and concern about child weight were positive predictors of children's BMI percentile, whereas pressure to eat was a negative predictor. Maternal disinhibition (i.e., loss of eating control) was also associated with higher BMI percentile in girls but not boys [14]. These data imply that maternal characteristics can potentially override the influence of PROP status on body weight in older children.

Pre-adolescence (age range = 9–14 yr) is a period of rapid growth and development as well as greater personal independence in food selection and lifestyle behaviors. These factors can impact the growth curve trajectories for BMI of children. Indeed, Berkey and co-workers [5] reported that even modest changes in energy intake and physical activity modified the pattern of 1-yr weight change in adolescents in a national cohort of U.S. children. Individual differences in eating attitudes such as dietary restraint (i.e., conscious control of eating) or emotional eating which may have their origins in early childhood [8] also play a role in weight status and body satisfaction during preadolescence [26,51] especially among girls [35]. Since all the studies on PROP status and adiposity in children have been cross-sectional, the influence of this phenotype on changes in body weight during childhood is unknown.

The objective of this study was to address this gap in knowledge. Children who participated in our studies as preschoolers and their mothers were invited to participate in a follow up study. Self-reported food intakes were collected, and current weight status and 6-yr change in weight from baseline (4.5 yrs of age) were also determined. Other data collected included maternal weight, and eating attitudes. The primary hypothesis was that PROP status would be inversely related to current energy intake and weight status of the children in the follow up cohort.

## 2. Methods

### 2.1. Subjects and general procedures

The subject pool for this follow up study consisted of children who attended the Rutgers University Preschool between 1999 and 2003, and had participated in one of three studies investigating the relationship among PROP taster status and food preferences and/or body weight when they were  $4.2 \pm 0.3$  yrs of age [4,20,21]. The total number of children who previously participated in the preschool studies was 154, and 148 of those children and their families were still living in the Central New Jersey area. Families were contacted by mail with an invitation for each former student and his/her mother to participate in the follow-up

study. Mothers gave written, informed consent for themselves, and their child's participation. Oral assent was also obtained from each child. The research protocols were approved by the Rutgers University Institutional Review Board. Participants were screened with a general health questionnaire to ensure that they had no medical conditions or recent illness, or that they were not taking medications that might interfere with taste perception.

### 2.2. Classification by PROP taste phenotype

As preschoolers, the children were classified as PROP tasters or non-tasters using an age-appropriate method [20,21]. In the current study, children were screened for PROP status using the paper disk method, previously tested for validity and reliability in both preadolescents and adults [13,14,52]. Briefly, subjects place a filter paper disk impregnated with 1.0 mol/L NaCl on the tip of the tongue until it is thoroughly wet. They rate the taste intensity of the disk using the labeled magnitude scale (LMS), a 100-mm scale anchored with the phrases "barely detectable" to "strongest imaginable" [15]. This procedure is repeated with a second paper disk impregnated with 50 mmol/L PROP (6-propyl-2-thiouracil, P3755, Sigma-Aldrich). Subjects are instructed to rinse with spring water at room temperature before and in between tasting each paper disk. Subjects are categorized as non-tasters if they rate the PROP disk  $\leq 13$  mm on the LMS; they are categorized as super-tasters if they rate the PROP disk  $> 67$  on the LMS. All others are classified as medium tasters [52]. NaCl ratings do not vary with PROP status in this method [13,14,52]. Therefore, NaCl ratings are used as a reference standard to clarify the taster status of subjects who give borderline ratings to PROP, although this situation is rare (~4% occurrence rate). This strategy is based on the rationale that non-tasters give higher ratings to NaCl than to PROP, medium tasters give equivalent ratings to both stimuli and super-tasters give higher ratings to PROP than NaCl. The PROP taste test was conducted twice. The correlation between the two PROP ratings was high ( $r = 0.83$ ;  $p < 0.001$ ) and the mean of the two ratings was used to classify the subjects.

### 2.3. TAS2R38 genotype analysis

TAS2R38 is the major gene controlling PROP taste sensitivity [24]. Thus, children were also characterized by TAS2R38 genotypes as a check on the reliability of the PROP phenotyping. Cells were obtained by gently brushing the inside of the cheek with a swab (Epicentre, Madison WI), and genomic DNA was extracted using the extraction solution provided by the manufacturer (Epicentre). Alleles of the gene TAS2R38 (Accession # AF494231; rs713598 and rs172866) were genotyped for a variant site using allele-specific probes and primers purchased from Applied Biosystems (Foster City, CA). Samples were compared with a sequenced reference standard, and alleles that failed to cluster into one of three groups were re-genotyped as needed.

Although there are three variant sites in the gene associated with bitter sensitivity (A49P, V262A, and I296V), the last two are in perfect linkage disequilibrium in all human populations tested thus far, and therefore the third site (I296V) was not assayed but imputed [29]. Subjects were grouped by the first and second variant sites, A49P and V262A, respectively. Since these sites are not in perfect linkage disequilibrium, the haplotypes for the first two sites were imputed based on observed allele frequencies and knowledge gained through genotyping thousands of similar samples [29,30].

Subjects who were homozygous for the bitter-insensitive allele are referred to as AV/AV, those who are heterozygous for the bitter-insensitive allele are referred to as PA/AV, and those who are homozygous for the bitter-sensitive allele are referred to as PA/PA. Those with the rare form (AA) display moderate sensitivity to PROP and are included in the PA/PA group.

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