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PROP taster status not related to reported cruciferous vegetable intake among ethnically diverse children

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Abstract

Sensitivity to the taste of 6-n-propylthiouracil (PROP) (a bitter tasting chemical related to the phenylthiocarbamide found in cruciferous vegetables) has been related to dietary intake or preferences of cruciferous vegetables among adults and young children, but not middle aged children or adolescents. We hypothesized that PROP taste sensitivity is related to lower reported dietary intake of cruciferous vegetables, primarily among younger children (i.e. a moderating effect of child age). This study examined the relationship of PROP sensitivity to reported dietary intake across three days in two age groups of youth (9–10 years and 17–18 year), while statistically controlling for physical activity, social desirability and reporting bias. Cross sectional design was employed with a multi-ethnic (White, African American, Hispanic, and Other) sample of 843 males and females. Children were recruited from and data were collected in local elementary and high schools that had at least 30% ethnic minority enrollment. Children providing nonplausible reports of dietary intake were deleted from the analyses. BMI was calculated and expressed in z-scores. Energy intake and physical activity were measured by three telephone conducted 24-hour dietary recalls with the Nutrient Data System for Research (NDSR) and 5 days of Actigraph activity monitor. The primary analyses included 347 students. PROP sensitivity was not related to intake of cruciferous vegetables. Intakes of the cruciferous vegetables were low, which may explain the lack of relationship.

Keywords

6-n-propylthiouracil; PROP; taste sensitivity; dietary intake; vegetables; food groups; children

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1. INTRODUCTION

6-n-propylthiouracil (PROP) is chemically related to the bitter substance (PTC-phenylthiocarbamide) in cruciferous vegetables; and whether a person can taste PROP is genetically determined. Taste sensitivity to PROP has been proposed to account for individual differences in food preferences and eating habits. Three studies have been published on the relationship of PROP taste sensitivity to reported dietary intake using food frequency questionnaires (FFQs) among children, all showing no relationship. FFQs, however, have limitations. Other limitations of these studies included small sample sizes (range: 30–81); limited age ranges (range: 3–6), no report of ethnic background of the sample; and none controlled for energy intake (i.e. children who eat more calories likely consume more of most foods), physical activity (i.e. children who are very active likely eat more of many foods), or social desirability of response, all of which have been related to diet, and thereby could confound the findings. Since some late adolescents develop a preference for some bitter foods (e.g. coffee, alcohol), PROP taste sensitivity may diminish with age.

Our hypothesis was: PROP taste sensitivity is related to lower dietary intake of cruciferous vegetables, primarily among the younger children (i.e. a moderating effect of child age). This paper examined whether PROP taste sensitivity was related to dietary intake using multiple 24-hour dietary recalls (24hdr) among children ages 9–10 years (elementary school age) and 17–18 years (high school age), while statistically controlling for energy intake, physical activity levels, social desirability, and over- or under-reporting response bias.

2. METHODS and MATERIALS

2.1. Design

A cross sectional design was used with stratification on ethnicity (White, African American, Hispanic, and Other), gender (male/female), age (9–10 and 17–18), BMI status (<85%tile and ≥85%tile) and PROP taster status (non-taster, medium taster and super-taster). The study was approved by the Baylor College of Medicine Institutional Review Board. The parents of all children completed informed consent and all children provided assent.

2.2. Study Sample

A total sample of 843 children was recruited. All 9 and 10 year old children were recruited from five elementary schools and 17 and 18 year olds from three high schools, each with greater than 30% ethnic minority representation from the Houston Independent School District in Houston, Texas. Children were excluded from participating for: 1) medical conditions or medications that interfered with taste, diet, or physical activity; or 2) developmental limitations that affected the child's ability to understand or provide age appropriate responses to the questions posed during testing. Recruitment was conducted in three annual waves to efficiently use staff.

Posterior power analysis was based on a three-way analysis of variance balanced on PROP sensitivity (non-tasters, medium tasters, and super tasters), race/ethnicity (White, African American, Hispanic, and Other), and annual household income (<$30,000, $30,000–$59,000, >$60,000) as factors. Given an alpha level significance of 0.05, an intra-class correlation (ICC) of 0.01, an average cluster size (m) of 83, and 347 participants after clustering [=unclustered size* (1+(m−1)*ICC)], there was adequate power (≥ 80%) to detect significance for moderate taster status and socioeconomic main effects (d=0.55) and large race-ethnicity main effects (d=0.81) and large interactions (d=1.31) with PROP sensitivity effects.

2.3. Measures

Parents completed a demographic questionnaire, including child's medical history (in regard to exclusionary criteria), household membership, and household socioeconomic status (SES).

2.3.1. PROP measurement—PROP taster status was determined using the impregnated paper screening test. This method used 2 paper discs, one impregnated with NaCl (1.0 mol/l) and the other with the PROP solution (50 mmol/1). The students were first asked to rinse their mouth with bottled water. They were then instructed to place the NaCl disk on the tip of their tongue for 30 seconds or until the disk was completely wet with their saliva and then spit it into a trash can. They were asked to rate the intensity of the taste using a Labeled Magnitude Scale (LMS) with ratings from 0 – 100, with descriptors of “barely detectable” to “weak” to “moderate” to “strong” to “very strong” and “strongest imaginable”. After they finished this first taste test and rating, they rinsed their mouth with bottled water. After 60 seconds they were asked to taste a second disk (PROP) and rate its taste using the same procedure and scale. Staff measured each child's markings on the LMS using a metric ruler and recorded the number for each disk. If the child rated the PROP disk ≤16.5 mm they were classified as a “non-taster”. Those who rated the PROP disk at ≥ 67 mm were classified as “super tasters”. Medium tasters fell in between. If their PROP rating was borderline at ~15 mm and they rated the NaCl disk much higher (at least a 30 mm difference on the LMS), they were classified as non-tasters. And if they rated the PROP at ~67 mm and gave a much lower rating to the NaCl, they were classified as super tasters. Test-retest PROP assessment was performed on 56 participants. The test-retest correlations were 0.79 for NaCl and 0.85 for PROP. The Kappa statistic measuring agreement between the time 1 and time 2 PROP categories was good (K=0.52). Most (70%) participants were classified into the same category at time 2. The remaining participants' (30%) PROP assessment differed by one category only.

2.3.2. Dietary assessment—Dietary assessment was conducted by registered dietitians trained and certified in 24-hour dietary recall procedures using the NDSR, a computerized dietary assessment program (Nutrient Data System for Research, #2005, 7/2005, University of Minnesota, Minneapolis, USA), over three non-consecutive days (2 weekdays and 1 weekend day). All recalls were collected via telephone interview and the target period was the 24-hours prior to the phone call. A 3 day intensive classroom training of the use of the NDSR software, the scripted interview, the two-dimensional food models used as aids for portion size estimation, the multiple pass data collection method, and probing methods per NDSR guidelines was conducted. Interviewers were eligible to conduct dietary intake interviews when they completed their certification interview and practice project with the trainer. All recalls conducted by the NDSR-certified interviewers were reviewed by a supervisor for accuracy and completeness before the interviews were considered complete and ready to be archived. The supervisor was ultimately responsible for managing missing foods, creating recipes for multi-ingredient foods or brand name items that were not in the NDSR database and ensuring that all recalls were collected following study-specific objectives. Participants completed dietary recalls independently, but supplemental information was gathered from the parent regarding the types of milk, fats, etc. purchased for the home. Students were given a Food Amounts Booklet with 2-dimensional food models at the school data collection. They were instructed to take it home and keep it next to their telephone to use during the telephone conducted recalls reporting their food eaten amounts to the dietitian. An attempt was made to overlap the days of collection of dietary intake data with accelerometry data collection. However, to minimize correlated errors, dietary data were collected on non-consecutive days; and to minimize the effect of the expectation of 24-hour recall on dietary intake, the phone calls were unannounced. When

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calls were made, families were sometimes not home. Cruciferous vegetable intake was defined as any mention of arugula, bok choy, broccoli, brussel sprouts, cabbage, cauliflower, kale, radish, spinach, swiss chard, turnip or watercress in the intake or ingredient files, and extracted and quantified from the NDSR output and averaged across days for each participant. Intake was adjusted for total EI using Willet's residual method to remove the effect of variation in EI.

2.3.3. Anthropometrics—Anthropometry was conducted after breakfast. Trained and certified staff collected all measurements using standardized protocols. Weight (wt) was measured in kilograms (kg) using a SECA Alpha 882 scale (SECA Corporation, Hamburg, Germany). Height (ht) was measured in centimeters (cm) using a PE-AIM-101 Stadiometer (Perspective Enterprises, Olney, Maryland). Waist circumference (WC) was measured in cm just above the iliac crest after marking those points on both sides of the body using a Gulick measuring tape with tensioning device (Fitness Wholesale, Park Twinsburg, OH). Triceps skinfold (SF) was measured in mm using a Lange caliper from Cambridge Scientific Industries (Cambridge, MD, USA). The location for the triceps measurement was determined by measuring the distance between the lateral projection of the acromion process and the inferior margin of the olecranon process of the ulna. The tape measure was used to measure the distance between these two points when the elbow was flexed at a 90-degree angle and the subject was standing. Wt, ht, WC and SF were each measured twice by one individual and averaged. When the weight, height, WC, or SF measurement difference between the first and second reading were much different (>1cm for height, >.2 kg for weight, >2 cm for WC, or >10% for skinfolds), a third measurement was obtained by the same individual, and the two closest values averaged. BMI z-scores were calculated using the CDC growth charts. To enhance inter-rater reliability and provide quality control of anthropometric measurement a standardization and certification training was conducted annually for each technician and continuous monitoring was conducted throughout data collection. The standardization and certification training involved comparing trainee with an accomplished senior technician's measurements of the same individuals. Trainees were certified and approved to conduct data collection when nine out of 10 measurement values corresponded closely (within 1 cm for height,.2 kg for weight, 2 cm for WC, and 10% for skinfolds) with the senior technician's values. In addition, throughout data collection every 10^{th} participant was measured by the senior technician in addition to the staff technician. Staff technicians who obtained measurements discrepant from the senior technician's measurements were required to complete further training.

2.3.4. Physical Activity—PA was recorded over a five-day period including weekdays and weekend days using accelerometers (Actigraph, Shalimar, Florida). Staff placed the monitor in the appropriate body location on the children and adolescents, who were instructed to wear it at waist level above the right hip in accordance with manufacturer guidelines for five complete days. The monitor began recording the next midnight. Accelerometers were programmed to record intensity of activity in one-minute epochs. Time not worn (or sleep time) was determined by 20 or more minutes of consecutive zeros. Participants were asked to maintain a diary of activities performed during times not worn and a review of these diaries revealed that participants were usually sleeping or showering when not wearing their accelerometers. A valid day was determined as a minimum of 800 minutes of wear time from 6AM to midnight and was calculated as the total daily minutes recorded less the time not worn. Minutes when the accelerometer counts were above the threshold (>2999) were summed daily and then averaged across all valid days for moderate to vigorous physical activity (MVPA). MVPA was assessed for participants who provided at least four valid days. Children providing less than five full days were asked to rewear the accelerometer to obtain complete days.

2.3.5. Social desirability of response—Social desirability of response reflects giving answers that are socially acceptable or expected, not necessarily what was done, and has been observed in regards to self-report of diet in both adults and children. It was assessed using the 9-item “Lie Scale” from the Revised Children's Manifest Anxiety Scale, which is a nine-item scale with a yes/no response format, and “lie” score determined by summing the “yes” responses. The instrument has shown good reliability and validity in children across a variety of ethnic groups.

2.4. Procedures

All child anthropometric and questionnaire measures were conducted at school at times arranged with school administrators (during non-academic class time for elementary school age students and before school hours for the high school students). Dietary and activity monitor measures were completed at home within two weeks after school data collection.

2.5. Statistical Analyses

2.5.1. Reporting status—Because underreporting is common in dietary assessment, dietary data were classified into reporting bias groups: plausible and non-plausible (under- or over-) reporters. Although Goldberg's equation has been widely used to establish reporting bias cut-off values, it has several limitations: (1) the individual's physical activity level is assumed, (2) the error assigned to the PA level is not accounted for, and (3) only the most extreme inaccurate reporting is identified. The method was improved through the use of predicted energy requirements (pER) published by the Institute of Medicine. Steps to classify individuals by reporting bias included: each individual's predicted energy requirement (pER) was calculated using 2002 Dietary Reference Intake; age and gender specific mean coefficients of variation for reported energy intakes (CV_{rEI}) were calculated where each individual's CV_{rEI} was equal to the standard deviation of the individual's recalls divided by mean of the individual's recalls; age-gender specific coefficients of variation for predicted energy report (CV_{pER}) and coefficients of variation for total energy expenditure (CV_{mTEE}) were obtained from the literature; one standard deviation (+1SD) was computed and used as a cut-off for rEI as a percentage of pER for age-gender specific groups where, $1SD = \sqrt{(CV^2_{rEI}/\# \text{ of recalls} + CV^2_{pER} + CV^2_{mTEE})}$; each individual's reported energy intake as a percentage of their predicted energy requirement (%rEI/pER) was computed; and each individual was classified as an underreporter (%rEI/pER < 100%−1.4 SD), plausible reporter (100%−1.4 SD<%rEI/pER < 100%+1.4 SD), or overreporter (%rEI/pER> 100% +1.4 SD). Through post hoc analyses, Huang et al found that a larger cut-off (from 1SD to 1.4 SD) offered acceptable plausibility while retaining a larger portion of the data, therefore, 1.4SD was used.

2.5.2. Model testing—The distributional properties of the variables were assessed with descriptive statistics, frequencies, histograms and boxplots to determine possible violation of model assumptions. Initial investigation involved testing for differences in participant characteristics between students included and excluded from analyses.

Because of reporting bias, only plausible reporters were included in the final analyses. Models testing the relationship of PROP taster status to intake included variables known to influence type and amount of food, including gender, age, race/ethnicity, family income (socio-economic status), BMI z-score, MVPA min, and social desirability. Interaction terms between taster status and gender, age, race/ethnic group, and income were introduced since an earlier study showed that PROP taster status influenced BMI only in the top income group. Mixed model analyses of variance examined differences in diet among the dietary reporting bias groups. The mixed model adjusted for the school-related clustering effect for the nine schools where data were collected. Mixed model analyses of variance among only the plausible reporters determined the relationship between PROP taster status (3 groups) and diet with additional gender, race/ethnicity, and income factors. The model utilized BMI z-score, MVPA min, and social desirability as covariates.

3. RESULTS

Although 843 students were recruited for this stage of the study, 665 students (342 9–10 year olds and 323 17–18 year olds) provided complete data for the analyses; the remaining 178 students were missing data for one or more variables and were excluded. There were no significant differences in any characteristics between students' included and excluded from the analyses. Among the 665 (52.8% female and 47.2% male) elementary (342, 51.4%) and high school students (323, 48.6%), 347 (52.2%) were plausible reporters, and 318 (47.8%) nonplausible reporters. The demographic characteristics of the whole sample and by reporting bias are shown in Table 1. Females were more likely to be plausible reporters, while males were slightly more likely to be nonplausible reporters. No differences were detected across reporting groups by age (school), race/ethnicity, or annual household income.

No statistically significant differences were detected between plausible and nonplausible reporters. None of the interaction nor main effect terms were related to cruciferous vegetable intake, suggesting there was no relationship to consumption of foods containing the bitter tasting chemical related to PROP.

4. DISCUSSION

The current investigation showed no evidence that PROP taster status influenced cruciferous vegetable consumption. Thus, we rejected our hypothesis. The low intake of vegetables observed in the cohort is consistent with norms in the U.S. population where roughly 95% of children from 9–18 years of age do not consume the recommended amount of vegetables.

Since 6-n-propylthiouracil is related to the bitter tasting chemical found in cruciferous vegetables, one would expect sensitivity to the taste of PROP to be related to lower consumption of cruciferous vegetables, especially among younger children, before they learned to like bitter tastes. However the few published findings in children are conflicting with some studies showing that PROP tasters gave lower acceptance ratings to broccoli (a cruciferous vegetable), but other studies showing no difference in broccoli acceptability between taster groups. Interestingly, taster children gave lower acceptance ratings than non-taster children to other bitter vegetables and fruits, such as spinach and grapefruit-orange juice. Nevertheless, vegetable consumption did not differ between groups in studies where mothers reported children's intakes. Only one study offering a variety of vegetables as a snack to preschoolers showed that tasters consumed half as much bitter vegetables as non-tasters. It is possible that laboratory observations of taster-group differences in intake do not translate well into everyday diet behaviors, particularly across different economic, ethnic and cultural groups. A key feature of the latter study was that the children had been repeatedly exposed to vegetables in the classroom, and were very familiar with these foods. Thus, two major barriers to vegetable acceptance in children (i.e., familiarity and accessibility) were minimized in that study sample.

It is possible that the inability to detect a relationship here was due to low reliability of PROP taster status assessment. Current test retest correlations within the measurement session were 0.79 for NaCl and 0.85 for self reported PROP values with a Kappa value of 0.52 between PROP categories. One study reported a somewhat higher test retest reliability (r=0.92), but this was with preschoolers using a categorical response (yes, no) with tastes of fluid from a cup as interpreted by the tester who also queried and interpreted the participants' verbal and facial responses. Another group had to substantially modify this first approach, apparently to deal with a lower socioeconomic group of preschoolers, but did not report a reliability coefficient. At third study reported a two step measurement process (a PROP threshold test followed by a suprathreshold rating test) with preschoolers, but did not report a test retest reliability. This latter procedure rejected data from 10% of participants, however, due to inconsistent responses. Ours is the first report of PROP testing with 9–10 year old children or 17–18 year old adolescents. A test-retest correlation of 0.80 or higher has been identified as desirable.

Boys and older children eating more of many nutrients and food groups has been reported. No effect was identified that reflected or might mediate a recently reported significant PROP taster status by income interaction term, wherein PROP taster status was related to BMI only in the higher socioeconomic status group in this sample. The current study was adequately powered to detect moderate effects among PROP taster status groups, the primary hypothesis. However, the study was not powered to detect moderate effects in the various sub groups, and larger samples (or a non clustered design) would be needed to examine differences by income, race/ethnicity, or gender.

Future research on PROP sensitivity and dietary intake should engage large samples determined from power calculations, control for possible confounding relationships and reporting bias, and measuring the specific gene or perhaps epigenetic influences on the gene. The limitations on this research were the high loss of sample due to nonplausible reporting (which could both lower power to detect differences, and in which some bias by sample loss could influence outcome), and the regional urban generally lower income ethnic minority nature of the sample. It is possible that relationships with PROP sensitivity emerge only among higher SES samples or with more reliable measures. Alternatively, limiting the sample to plausible reporters alone enhanced a previous report's ability to detect true relationships in this sample; and while ethnic minorities expanded the samples previously tested, race/ethnic group was not related to cruciferous vegetable, nor broccoli intake. Controlling for clustering by school limited power to detecting primarily large effects.

PROP sensitivity was not related to intake of cruciferous vegetables, which contain the bitter tasting chemical related to PROP. Generally low vegetable consumption may have limited the ability to detect this relationship.

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Abbreviations

BMI	body mass index
EI	energy intake
FFQ	food frequencies
ICC	intra-class correlation
LMS	Labeled Magnitude Scale

MVPA	moderate to vigorous physical activity
NDSR	Nutrient Data System for Research
PA	physical activity
PROP	6-n-propylthiouracil
SES	socioeconomic status
SF	skin fold
WC	waist circumference
24hdr	twenty-four hour dietary recall
85%tile	eighty-fifth percentile
cm	centimeter
ht	height
kg	kilogram
mm	millimeter

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Table 1

Frequencies and percentages for participant characteristics by reporting status

Characteristic	Plausible (n=347)	Nonplausible[a] (n=318)	Total (n=665)
Taster Status	n(%)	n(%)	n(%)
Non-taster	96(27.7)	96(30.2)	192(28.9)
Taster	137(39.5)	110(34.6)	247(37.1)
Super Taster	114(32.9)	112(35.2)	226(34.0)
Gender[b]			
Female	199(57.3)	152(47.8)	351(52.8)
Male	148(42.7)	166(52.2)	314(47.2)
Age			
9	125(36.0)	97(30.5)	222(33.4)
10	64(18.4)	56(17.6)	120(18.0)
17	122(35.2)	121(38.1)	243(36.5)
18	36(10.4)	44(13.8)	80(12.0)
Race/Ethnicity			
White	95(27.4)	93(29.2)	188(28.3)
African American	106(30.5)	99(31.1)	205(30.8)
Hispanic	97(28.0)	93(29.2)	190(28.6)
Other	49(14.1)	33(10.4)	82(12.3)
Obesity			
Underweight	118(34.0)	118(37.1)	236(35.5)
Healthy weight	86(24.8)	82(25.8)	168(25.3)
Overweight	143(41.2)	118(37.1)	261(39.2)
Obese			
Annual Household Income	8(2.3)	4(1.3)	12(1.8)
<$30,000	218(62.8)	159(50.0)	377(56.7)
$30,000–$59,000	74(21.3)	58(18.2)	132(19.8)
>$60,000	47(13.5)	97(30.5)	144(21.7)

[a]Nonplausile reports include 250 under-reporters and 68 over-reporters

[b]Significant association between gender and reporting status (X^2=6.07, df=1, p=0.014)