

1 **TAS2R38 predisposition to bitter taste associated with differential**
2 **changes in vegetable intake in response to a community-based**
3 **dietary intervention**

4

5 *Association of TAS2R38 variation and the responsiveness to lifestyle interventions*

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7 Larissa Calancie¹, Thomas C. Keyserling^{2,3}, Lindsey Smith-Taillie⁴, Kimberly Robasky⁵, Cam
8 Patterson⁶, Alice S. Ammerman^{2,4}, and *Jonathan C. Schisler⁷

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10 ¹Center for Health Equity Research, School of Medicine at The University of North Carolina at
11 Chapel Hill, Chapel Hill, NC 27599, USA; ²Center for Health Promotion and Disease Prevention;
12 ³Division of General Medicine and Clinical Epidemiology at The University of North Carolina at
13 Chapel Hill, Chapel Hill, NC 27599, USA; ⁴Department of Nutrition, Gillings School of Global
14 Public Health; ⁵Q2 Solutions | EA Genomics, Morrisville, North Carolina. 27560, USA;
15 ⁶Presbyterian Hospital/Weill-Cornell Medical Center, New York, NY 10065, USA; ⁷McAllister
16 Heart Institute, Department of Pharmacology, and Department of Pathology and Lab Medicine at
17 The University of North Carolina at Chapel Hill, Chapel Hill, NC 27599, USA.

18

19 *Address correspondence and inquiries to:

20 Jonathan C. Schisler, MS, PhD

21 UNC McAllister Heart Institute

22 2340C Medical Biomolecular Research Bldg

23 111 Mason Farm Rd CB# 7126

24 Chapel Hill, NC 27599-7126 USA

25 Telephone: 919-843-8708

26 schisler@unc.edu

27

28 **Abstract**

29 **Background:** Although vegetable consumption is associated with decreased risk for a variety of
30 chronic diseases, few Americans meet the CDC recommendations for vegetable intake. The
31 *TAS2R38* gene encodes a taste receptor that confers bitter taste sensing from chemicals found in
32 some vegetables. Common polymorphisms in *TAS2R38*, including rs713598, rs1726866, and
33 rs10246939, lead to coding substitutions that alter receptor function and result in the loss of bitter
34 taste perception.

35 **Objective:** Our study examines whether bitter taste perception *TAS2R38* diplotypes were
36 associated with vegetable consumption in participants enrolled in either an enhanced or a
37 minimal nutrition counseling intervention within a community-based dietary intervention.

38 **Methods:** DNA was isolated from the peripheral blood cells of study participants (N = 497) and
39 analyzed for polymorphisms using genotyping arrays. The Block Fruit and Vegetable screener
40 was used to determine frequency of vegetable consumption. Mixed effects models were used to
41 test differences in frequency of vegetable consumption between intervention and genotype
42 groups over time.

43 **Results:** There was no association between baseline vegetable consumption frequency and the
44 bitter taste diplotype ($p = 0.937$), however after six months of the intervention, we observed an
45 interaction between bitter taste diplotypes and time ($p = 0.046$). Participants in the enhanced
46 intervention increased their vegetable consumption frequency ($p = 0.020$) and within this
47 intervention group, the non-bitter and intermediate-bitter tasting participants had the largest
48 increase in vegetable consumption. In contrast, in the minimal intervention group, the bitter
49 tasting participants reported a decrease in vegetable consumption.

50 **Conclusions:** Non- and intermediate-bitter taste blind participants increased vegetable
51 consumption in either intervention group more than those who perceive bitterness. Future
52 applications of precision medicine could consider genetic variation in bitter taste perception
53 genes when designing dietary interventions.

54 **Author summary**

55 Most Americans under consume vegetables, despite clear associations between vegetable
56 consumption and health benefits. Vegetables, such as broccoli, kale, and Brussels sprouts,
57 contain bitter-tasting compounds, leading to taste aversion. Common polymorphisms on the
58 *TAS2R38* taste receptor gene (rs713598, rs1726866, and rs10246939) influence the perception of
59 bitter taste. We tested whether genetic predisposition to bitter taste influenced vegetable intake in
60 a dietary intervention and found that *TAS2R38* diplotypes were related to vegetable consumption.
61 Combining precision medicine approaches that identify taste profiles and personalizing dietary
62 advice could help engage intervention participants and improve the impact of dietary
63 interventions.

64 **Introduction**

65 Few Americans consume the recommended amount of dark green and orange vegetables, despite
66 the association between vegetable consumption and reduced risk of chronic diseases [1]. Public
67 health practitioners and researchers aim to increase vegetable consumption through dietary
68 interventions, but the impact of interventions on fruit and vegetable intake yields mixed results.
69 For example, some interventions resulted in increased vegetable consumption by participants [2–
70 4], whereas others did not significantly affect vegetable consumption [5]. In instances where
71 interventions increase vegetable intake, the effects are generally small and participants often do
72 not reach recommended intake levels [6,7].

73
74 One possible explanation for the mixed results of dietary intervention studies is heterogeneity of
75 participants regarding characteristics that strongly influence vegetable intake, such as taste
76 preferences. Taste is an important determinant of fruit and vegetable intake in adults and children
77 in the United States (US) [8,9]. While phytonutrients in vegetables, such as phenols, flavonoids,
78 isoflavones, terpenes, and glucosinolates, seem to be protective against certain cancers, their
79 bitter taste can be a deterrent to consumption [10]. Vegetable sweetness and bitterness were
80 found to be independent predictors of more or less preference for sampled vegetables and
81 vegetable intake, respectively, and the ability to detect a bitter tasting compound called
82 propylthiouricil (PROP) was related to vegetable taste preferences [11].

83
84 Identified in 2003 [12], the *TAS2R38* gene encodes a G protein coupled receptor that functions as
85 a taste receptor, mediated by ligands such as PROP and phenylthiocarbamide that bind to the
86 receptor and initiate signaling that can confers various degrees of taste perception [13].

87 Vegetables in the brassica family, such as collard greens, kale, broccoli, cabbage, and Brussels
88 sprouts, contain glucosinolates and isothiocyanates, which resemble PROP, and therefore much
89 of the perceived “bitterness” of these vegetables is mediated through *TAS2R38* [14]. Bitter taste
90 receptors in the TS2R family are also found in gut mucosal and pancreatic cells in humans and

91 rodents. These receptors influence release of hormones involved in appetite regulation, such as
92 peptide YY and glucagon-like peptide-1, and therefore may influence caloric intake and the
93 development of obesity [15]. Thus, bitter taste perception may affect dietary behaviors by
94 influencing both taste preferences and metabolic hormonal regulation.
95
96 Three variants in the *TAS2R38* gene – rs713598, rs1726866, and rs10246939 – are in high
97 linkage disequilibrium in most populations and result in amino acid coding changes that lead to a
98 range of bitter taste perception phenotypes [16,17]. The PAV haplotype is dominant; therefore,
99 individuals with at least one copy of the PAV allele perceive molecules in vegetables that
100 resemble PROP as tasting bitter, and consequently may develop an aversion to bitter vegetables.
101 In contrast, individuals with two AVI haplotypes are non-bitter tasters. PAV and AVI haplotypes
102 are the most common, though other haplotypes exist that confer intermediate bitter taste
103 sensitivity (AAI, AAV, AVV, and PVI) [18]. This taste aversion may apply to vegetables in
104 general [19]. Therefore, dietary interventions aiming to increase vegetable intake may have
105 different outcomes depending on individuals' perceptions of the taste.
106
107 While many studies have examined whether certain participant and intervention characteristics
108 influence differential response to dietary interventions, such as age, sex, race, education, disease
109 state, and intervention delivery methods [20,21], we are not aware of studies examining whether
110 genes associated with bitter taste perception moderate participants' responses to dietary
111 interventions. The Heart Healthy Lenoir (HHL) Project offers a unique opportunity to test a
112 concept that the genetic predisposition to bitter taste perception may associate with a differential
113 response to a dietary intervention among a diverse, community-based study population [22,23].
114 In this paper we tested the following two hypotheses:
115
116 1. Participants with the *TAS2R38* non-bitter taste diplotype will consume more servings of
117 vegetables per day at baseline than participants with intermediate or bitter taster diplotypes.

118
119 2. The *TAS2R38* diplotype will moderate the effect of the HHL intervention on vegetable
120 consumption such that participants with a bitter taste diplotype will have a lower increase in
121 reported vegetables intake than the non-bitter taste participants after 6 months of the
122 intervention.

123

124 **Results**

125 STUDY POPULATION

126 *Demographics.* Participant characteristics at baseline and after 6-months are shown in **Table 1**.
127 There were several differences between participants in the minimal versus the enhanced
128 intervention groups. More women, Caucasians, highly educated, and non-smokers participated in
129 the enhanced intervention compared to the minimal intervention at baseline. Despite attrition,
130 there were no significant differences in participant characteristics within each intervention
131 group at baseline and after 6-months.

132

133 *TAS2R38 genetic characterization.* All three alleles located in the *TAS2R38* gene are common
134 variants in both African and Caucasian American populations [24] similar to our sample enrolled
135 in HHL (**Supplemental Table 3**). In our CAU participants the three alleles had similar
136 frequencies and were in high linkage disequilibrium (**Table 2**). The linkage disequilibrium was
137 not as high across the pairwise allele comparisons in the AA participants (R^2 range 0.46 – 0.95,
138 $D' > 0.98$) in part due to the difference in allele frequency of rs1726866 (**Table 2**). Therefore, we
139 used the phased genotypes to determine the haplotypes found in our population. In our AA
140 population, PAV was the most frequent haplotype, followed by AVI, haplotypes that encode the
141 bitter and non-bitter polymorphisms, respectively (**Table 2**). This distribution was reversed in
142 our CAU population. Demonstrating the genetic diversity between AA and CAU populations,
143 nearly one-third the AA haplotypes were AAI (intermediate-taster phenotype) whereas the CAU
144 haplotypes were almost exclusively PAV (bitter tasters) or AVI (non-bitter tasters) (96%).

145

146 The PAV is a dominant allele, therefore instead of relying on an index SNP or haplotypes, we
147 used a dominant model to derive a bitter taste phenotype score based on the diplotype (**Table 3**).
148 Contingency analysis of the bitter taste phenotype revealed that the percentage of bitter-tasting
149 participants was similar between AA and CAU (**Figure 1**). However, among those not falling
150 into the bitter tasting category, we observed a higher proportion of non-bitter tasters in CAUs
151 (29%) versus AAs (12%) and three times as many intermediate tasters in AAs versus CAUs
152 (**Figure 1**), likely due to the prevalence of the AAI (intermediate-taster) haplotype in our AA
153 population (**Table 2**).

154

155 ASSOCIATIONS BETWEEN VEGETABLE CONSUMPTION AND GENETIC PREDISPOSITION TO BITTER
156 TASTE

157 *Bitter taste diplotypes did not associate with differences in baseline vegetable intake.* We first
158 measured associations between baseline vegetable intake and *TAS2R38* phenotypes using model
159 1. Sex, education, and household income were positively associated with reported vegetable
160 consumption frequency scores, as expected (**Table 4**). Participants reported similar vegetable
161 consumption frequency independent of their genetic predisposition toward bitter taste sensitivity,
162 $p = 0.937$ (**Figure 2, Table 4**). Thus, we rejected our first hypothesis that participants would
163 report different vegetable consumption frequency scores at baseline according to their *TAS2R38*
164 diplotype. These data suggest that within our HHL population, the *TAS2R38* polymorphisms
165 were not associated with vegetable intake. This finding is consistent with another study
166 examining the association between self-reported vegetable intake and PROP sensitivity in a
167 community-based population [25].

168

169 *Participants with non-bitter or intermediate-bitter taste diplotypes increased vegetable intake*
170 *after the intervention.* Using model 2, we incorporated variables to measure the impact of the
171 different interventions over time and to measure interactions between *TAS2R38* diplotypes,

172 intervention intensity, and time (**Table 4**). We observed the same associations between reported
173 vegetable consumption frequency scores and sex, education, and household income. Consistent
174 with our second hypothesis, we observed an interaction between phenotype and time (**Figure 2**).
175 Non-bitter taste participants reported 0.65 higher vegetable intake frequency scores, or about
176 0.20 servings of green salads or other vegetables per day, at the end of the intervention.
177 Vegetable intake frequency scores also increased by 0.55 among intermediate bitter tasters.
178 Intake scores only increased 0.04 among bitter tasters at the end of the intervention. Importantly,
179 we did not see differences in participant demographics (**Table 1**) or allele frequencies, linkage
180 disequilibrium, or haplotype distributions (**Supplemental Tables 3, 4, 5**) due to intervention
181 attrition at the 6-month time point.

182
183 *Vegetable intake increased in the enhanced dietary intervention.* Given the enhanced
184 intervention included tailored dietary goals and behavior change strategies, we hypothesized that
185 participants in the enhanced intervention would have a greater increase in vegetable intake. As
186 expected, the change in vegetable intake frequency scores was higher in the enhanced
187 intervention group compared to the minimal group over time (**Figure 3**). In fact, participants in
188 the minimal intervention group reported a decrease of 0.19 in vegetable intake frequency scores,
189 whereas participants in the enhanced intervention group increased their reported scores by 0.58,
190 suggesting that the enhanced intervention contributed to dietary changes regarding vegetable
191 intake.

192
193 *Bitter taste perception and the intensity of the dietary intervention may influence vegetable*
194 *intake.* Although the enhanced intervention associated with increased reported vegetable intake
195 (**Figure 3**), could this response be modified by the *TAS2R38* phenotype? Despite significant
196 main effects, the three-way interaction between intervention group, phenotype, and time was not
197 statistically significant, $p = 0.392$. Still, the 3-way interaction analysis trends similar to those
198 seen in the 2-way interactions (**Figure 4**). Non-bitter and intermediate-bitter tasting participants

199 in the enhanced intervention increased their vegetable intake frequency score the most (delta =
200 0.71 and 0.89, respectively). Consistent with our hypothesis, bitter tasting participants in the
201 minimal intervention were the only group that decreased their vegetable intake (delta = -0.44),
202 however there was an increase among bitter tasting participants in the enhanced intervention
203 (delta = 0.50). Our data suggest that these *TAS2R38* alleles and resulting phenotypes may impact
204 a person's response to dietary interventions regarding vegetable intake.

205
206 *Vegetable intake associated specifically with TAS2R38 variants and not other variants in related*
207 *TAS2R genes.* Other genes in *TAS2R* family are also implicated in taste perception,
208 neuroendocrine function, appetite, and satiety [26] as well as human aging [27]. We extracted the
209 genotypes of these related family members (**Supplemental Table 6**) and along with the
210 *TAS2R38* variants we used principal components analysis with the adjusted predicted vegetable
211 intake as a supplementary variable to determine if other *TAS2R* genes associate with the
212 responsiveness to our dietary interventions. In our AA and CAU groups we identified the two
213 components that corresponded to the highest loading for vegetable intake (**Figure 5A, 5B**). Not
214 surprisingly, this resulted in segregation of the *TAS2R38* bitter taste phenotypes and revealed that
215 the three *TAS2R38* alleles were highly correlated to the variance of PC4 or PC2 in the AA or
216 CAU groups, respectively (**Supplemental Table 7**). We also identified another associated locus
217 common to both AA and CAU populations that harbors *TAS2R20* and *TAS2R50* (**Table 5,**
218 **Supplemental Table 7**). However, when we used a mixed model approach to look at the
219 association of these individual SNP or the SNP : time interaction and reported vegetable intake,
220 we only observed an association with two *TAS2R38* alleles, rs713598 and rs10246939. Another
221 locus of interest included the *TAS2R3*, *TAS2R4*, and *TAS2R5* genes that had high correlation in
222 PC2 in the CAU group (**Figure 5B, Supplemental Table 7**). However, like the other loci we
223 analyzed, we did not find any association with vegetable intake either analyzed with both
224 populations or only within the CAU group (**Supplemental Table 8**). These data suggest that
225 *TAS2R38* is likely the largest genetic contributor to our association analysis. The other SNPs we

226 identified in this analysis, however, may play other roles that contribute to taste perception and
227 diet.

228

229 **Discussion**

230 The primary goal of HHL was to reduce CVD-related health disparities in a rural population in
231 North Carolina. In this report, we tested the concept that participants in a dietary intervention
232 designed to promote heart healthy eating patterns may respond differently according to their
233 genetic predisposition of bitter taste perception mediated by the *TAS2R38* gene and allelic
234 variants that can affect receptor signaling and hence, perception of bitter taste compounds found
235 in many vegetables. Our HHL sample was represented by two ancestral populations, African and
236 Caucasian Americans, and we were cognizant of the genetic population structure of our cohort.
237 When we analyzed the diplotypes and corresponding phenotypes of our cohort, we observed
238 similar proportion of bitter taste participants in the AA and CAU groups (**Figure 1**). There was a
239 striking difference, however, in the proportion of non-bitter and intermediate bitter tasters such
240 that the CAU group had nearly triple the frequency of non-bitter tasters (**Figure 1**), consistent
241 with a recent study on the natural selection of *TAS2R38* haplotypes [24]. Although we lacked the
242 power to stratify our HHL cohort for robust, focused analyses within each ancestry group, we
243 accounted for ancestry in our analyses and the variable accounting for ancestry in either of our
244 models did not approach our defined level of statistical significance (**Table 4**). Although these
245 data suggest that ancestry did not associate with changes in reported vegetable consumption in
246 our cohort, future studies should consider and seek to define differences in allele frequency and
247 interactions with other biological factors that contribute to taste perception in distinct ancestral
248 populations to determine the applicability of precision medicine to dietary interventions.

249

250 We found differences in vegetable consumption frequencies between intervention participants at
251 follow-up according to their bitter taste perception phenotype characterized by common coding
252 variants in the *TAS2R38* gene (**Figure 2**). Participants with *TAS2R38* diplotypes associated with

253 non-bitter tasting increased vegetable consumption more than participants whose genotypes were
254 associated with bitter taste perception (**Figure 2**). Our findings are consistent with other studies
255 that observed differential vegetable preferences according to the presence of bitter taste
256 perception SNPs [11,28]. However, other studies suggest that bitter taste sensitivity is not
257 associated with food selection due to other factors such as attitudes toward foods, cultural norms,
258 and one's food environment [29,30]. More research is needed to better understand how genetic
259 taste variation and other factors influence vegetable selection and consumption [30], and
260 importantly, how this information can help inform dietary interventions.

261
262 Not surprisingly, we also found that participants in the enhanced dietary intervention increased
263 their vegetable intake frequency scores more than those in the minimal intervention (**Figure 3**).
264 A review of behavioral interventions aiming to increase vegetable intake found that 17 of 22
265 studies reported small, but significant increases in vegetable intake [21]. Many dietary
266 intervention studies aim to change servings of total fruits and vegetables, while ours only
267 examined a subset of vegetable intake (green salads and other vegetables) and likely explains the
268 small changes we observed in daily servings of vegetables after the intervention. Moreover, the
269 study participants reported very low intake of vegetables as baseline; in retrospect, participants
270 may have benefitted from a more intensive vegetable consumption focus in the intervention than
271 they received. In some cases, participants in the minimal intervention group reported lower
272 vegetable intake frequency scores after 6 months than at baseline (**Figure 3**).

273
274 Participants who took part in the enhanced intervention increased their vegetable intake over the
275 course of the intervention, irrespective of the *TAS2R38* phenotype, whereas participants in the
276 minimal intervention showed mixed results based on *TAS2R38* phenotype (**Figure 4**). Non-bitter
277 taste participants in the minimal intervention group increased their vegetable intake while bitter
278 tasters in the same intervention group decreased their vegetable consumption (**Figure 4**). Our
279 findings demonstrate that all participants in the enhanced condition, even those who are likely to

280 perceive bitterness in some vegetables, increased vegetable consumption during the intervention.
281 Biological sensitivity to bitter taste is likely one of many factors contributing to participants'
282 decisions about vegetable consumption. Participants that perceive bitterness may choose to
283 consume vegetables that are less bitter, such as carrots or cooked vegetables [31] or food
284 preparation strategies that minimize the bitter taste. Participants may have also modified their
285 preferences toward vegetable consumption over the course of the enhanced intervention; studies
286 suggest that repeated exposure to foods and beverages can alter preferences for those foods and
287 beverages [32–34]. Since participants were receiving information about the benefits of a
288 vegetable-rich diet, they may have been more willing to overcome taste aversions and perhaps
289 even modify their taste preferences during the 6-month enhanced intervention.

290
291 There were several limitations in this study. Frequency of vegetable intake questions did not
292 specifically target vegetables that are high in bitter compounds [11,31]. Additionally, cooking
293 methods were not assessed, and cooking can affect consumers' vegetable preferences [35,36].
294 Moreover, we did not include self-reported vegetable juice and vegetable soup intake in our
295 outcome variable. These items were excluded because they are likely to have added salt or sugar,
296 which suppresses bitterness [36,37]. Also, there was 22% attrition at the 6-month follow up;
297 however, the haplotype frequencies were similar at baseline and follow-up (**Supplemental Table**
298 **5**, so the differences seen between baseline and 6 months are not likely due to differences in
299 genotypes. Additionally, our sample size limited our ability to detect a statistically significant
300 interaction between genotype and intervention group at two time points and, given multiple
301 comparisons, some significant findings may be due to chance. Despite these limitations, the
302 significant main effects suggest that both genotype and intervention group influenced
303 participants' vegetable consumption frequency (**Figure 4**). Future studies with larger sample
304 sizes and more participants per phenotype and intervention group at each time point should be
305 powered to identify additional three-way statistical interactions.

306

307 The T2R gene family represents a collection of 28 genes found on chromosomes 5, 7, and 12
308 [26,38] that are expressed in taste bud cells. Given the ability of people to distinguish more
309 distinct bitter tasting compounds than the number of receptors suggests T2R receptors likely
310 respond to more than one bitter ligand [39]. We expanded our SNP-level analysis to cover 20
311 T2R genes to look for other taste receptors that may provide some insight into the phenotype of
312 our HHL participants. Although our results at the individual SNP level in other T2R genes did
313 not identify associations to changes in vegetable intake within our intervention (**Table 5**), our
314 multivariate analysis (**Figure 5**) did identify other loci other than *TAS2R38* that should be
315 considered in future studies, including *TAS2R50* that recognizes the naturally occurring bitter
316 compounds amarogentin and andrographolide [40], and *TAS2R20*, a receptor with no known
317 natural ligand [41]. Within the CAU group our analysis identified SNPs from an additional locus
318 containing three genes in chromosome 7, recently identified as having long-range haplotype
319 structure with *TAS2R38* [42] that contains two receptors with undefined natural ligands, *TAS2R3*
320 and *TAS2R5* [41], and *TAS2R4*, a known receptor for quinine [43].

321
322 Given the American Heart Association recommends individual focused interventions for
323 increasing fruit and vegetable intake [44], our findings raise several important issues regarding
324 how we can develop precision medicine approaches in the context of taste perception to inform
325 dietary interventions for heart health. Measuring consumption of specific vegetables that contain
326 glucosinolates and isothiocyanates (e.g., collard greens, broccoli, Brussels sprouts, kale), as well
327 as vegetable preparation methods (e.g., cooked, fresh), could yield more robust associations
328 between bitter taste perception alleles and consumption of bitter vegetables. Conducting a
329 qualitative study among bitter tasters who consume vegetables to learn how and why they have
330 overcome a genetic predisposition to perceive compounds in vegetables as bitter may yield
331 strategies for interventions aiming to increase vegetable consumption. Future research could test
332 whether personalizing diets to specific genetic-based taste profiles increases consumption of
333 specific healthy foods more than generalized dietary advice. Supportive of this concept, a meta-

334 analysis of behavioral interventions found that tailored nutrition interventions aiming to increase
335 fruit and vegetable consumption were more successful than untailored interventions [45,46].

336
337 Nutrigenomics and other approaches to tailor nutrition advice and interventions based on genetic
338 and metabolic profiles are increasing as scientists overcome technological and data challenges
339 [47]. In one study, genes associated with energy metabolism were used to personalize a low
340 glycemic index weight management program informed by the Mediterranean diet for participants
341 [48]. The authors observed greater diet adherence to the genetically tailored diets, as well as
342 longer-term reductions in BMI and improved blood glucose levels compared to participants who
343 received a low glycemic index weight management program informed by the Mediterranean diet
344 that was not genetically-tailored [48]. A recent review of nutrigenomic studies did not report any
345 studies that used genes associated with taste perception to inform dietary intervention strategies
346 [47]. Recognizing the important influence that taste perception has on diet and tailoring dietary
347 interventions using this information may be a strategy for engaging participants and improving
348 dietary intervention outcomes.

349
350 Reducing heart health disparities requires attention to the many factors driving the disparities.
351 Despite high prevalence of cardiovascular disease among African Americans, this population is
352 under-represented in GWAS studies [49]. Likely explanations include mistrust between African
353 American community members and researchers due to the legacy of unethical medical and
354 genetic studies [50], and imbalances in information and power [51], as well as persistent biases
355 that influence research participation [52]. A strength of the HHL study was our community-
356 based participatory research (CBPR) approach where we worked with a community advisory
357 board, held focus groups with community members, and hired and trained community members
358 as study staff [53,54]. We believe these activities helped build trust between researchers and
359 community-based participants, and helped the research team better understand and meet the
360 expectations that community members had regarding their participation in the genomics portion

361 of this study. Moreover, these activities likely contributed to the high enrollment of African
362 Americans in the genomics arm of the HHL study. In addition to the genomics and lifestyle
363 counseling components of the study, HHL sought to address heart health disparities by
364 increasing access to healthy foods, promoting knowledge of heart healthy choices through a
365 collaboration with local restaurants that included information on healthful menu items and a
366 coordinated monthly newspaper column with information on healthy eating [55], and enhancing
367 clinical care for hypertension in the Lenoir community [23,53]. These strategies were designed
368 to address behavioral and environmental factors that drive heart health disparities in a rural NC
369 population. Combining precision medicine insights to engage participants with CBPR principles
370 and public health strategies that shape the context in which individuals live, work, and play may
371 be a promising approach for reducing cardiovascular health disparities in the US.

372
373 This study demonstrates a concept that genes associated with bitter taste perception can influence
374 frequency of vegetable intake in the context of a dietary intervention in a diverse, community-
375 based study sample. The variability in frequency of intake according to participants' bitter taste
376 perception phenotype could help explain why dietary change interventions report mixed results.
377 Taste has a strong influence over individuals' dietary habits and should be considered when
378 designing dietary change interventions and in developing novel precision medicine approaches to
379 lifestyle interventions.

380

381 **Methods**

382 *The Heart Healthy Lenoir (HHL) Project Overview.* The overall goal of the HHL Project was to
383 reduce Cardiovascular Disease (CVD) risk and disparities in CVD risk among Lenoir County,
384 North Carolina residents, as previously described [53,56]. It was conducted in Lenoir County
385 because of its location in the “stroke belt” [57] of eastern North Carolina, where rates of CVD
386 are higher than state and national averages [58] and because it has a large minority population
387 (40% African American) that experiences disproportionately higher rates of CVD [59]. The

388 overall Project included three coordinated studies: a lifestyle intervention study focusing on diet
389 and physical activity [22] a study to improve high blood pressure management at local clinical
390 practices [23] and a study examining associations between genetic markers and change in CVD
391 risk factors. The Project was designed and conducted with input from a local Community
392 Advisory Committee and approved and monitored by the University of North Carolina at Chapel
393 Hill's Institutional Review Board, with data collected from September 20, 2011 to November 7,
394 2014 and analyzed in 2017. This trial is registered as # NCT01433484 at clinicaltrials.gov.

395
396 *Heart Healthy Lenoir (HHL) Interventions*. Participants in the HHL Project (N = 664 in total)
397 could take part in the lifestyle study (N = 339), the high blood pressure study (N = 525) or both
398 (N = 200). All participants were invited to take part in the genomics study. We utilized the data
399 collected at baseline and at the 6-month follow-up that included participants with complete data
400 for the variables of interest in this study, including bitter taste perception phenotype
401 characterized by three SNPs on the *TAS2R38* gene, vegetable intake frequency, and model
402 covariates (N = 497). Twelve participants of the 509 genotyped (2%) were missing data (other
403 than household income) and therefore removed from the analysis. The lifestyle intervention is
404 described in detail elsewhere [22]. Briefly, during the first 6 months, the dietary component of
405 this intervention included four counseling sessions that focused on improving dietary fat and
406 carbohydrate quality, consistent with a Mediterranean dietary pattern. The primary focus of the
407 second counseling session was on increasing fruit and vegetable consumption with a goal of
408 seven total servings per day. The high blood pressure intervention is also described in detail
409 elsewhere [23,53]. Participants in the high blood pressure study received limited dietary
410 counseling by phone, with only 13 receiving a counseling phone call before the 6-month follow-
411 up measurement visit. Accordingly, in this paper, the dietary intervention given to lifestyle study
412 participants is considered the “enhanced” intervention, while the intervention given to those who
413 only participated in the high blood pressure study is considered the “minimal” intervention.

414

415 *Genotyping procedure.* SNP status was obtained from 505 HHL participants at baseline via DNA
416 isolated from peripheral blood cells using the Infinium Human Omni Express Exome+ BeadChip
417 (Illumina). Genotypes were generated from genomic DNA using the Infinium workflow
418 essentially as described by the manufacturer. DNA was amplified, fragmented, precipitated with
419 isopropanol, and resuspended prior to hybridization onto BeadChips containing 50mer probes.
420 After hybridization, enzymatic single base extension with fluorescently labeled nucleotides was
421 conducted to distinguish alleles. Hybridized BeadChips were imaged using an Illumina iScan to
422 determine intensities for each probe. Corresponding genotypes were extracted from intensity data
423 and called using a standard cluster file within Illumina Genome Studio software. A MAIME-
424 compliant dataset of the microarray data generated is available at the NCBI database of
425 Genotypes and Phenotypes (dbGaP, study ID phs001471).

426
427 *Imputing SNPs.* All DNA samples identified as either African American (AA, N = 304) or
428 Caucasian American (CAU, N = 201) were imputed for a total of 505 samples. The array data
429 were exported into plink format converted into chromosome-specific variant call format,
430 applying the following filters: merge replicate probes, switch the alternate (ALT) or reference
431 (REF) sequence if deemed necessary by reference, exclude markers where neither REF nor ALT
432 matches the reference, exclude markers where REF is not AGCT. Additionally, in preparation
433 for imputing the following filters were further applied: remove markers not in the reference, fill
434 ALT values in from reference where genotype is entirely homozygous for reference. Samples
435 were imputed twice, once with the Michigan imputation server [60] and once with Beagle (v4.1)
436 [61]. All 505 samples imputed with Beagle were run against the 2504 sample reference panel
437 from 1000 genomes. The Haplotype Reference Consortium (HRC, 65k haplotypes) reference
438 panel was used to run the CAU samples on the Michigan imputation server, and the Consortium
439 on Asthma among African-ancestry Populations in the Americas (CAAPA) reference panel was
440 used to run the AA samples on the imputation server. A brief summary of coverage regarding the
441 panels and how they performed with the target marker set (the markers from the genotyping

442 array) is provided (**Supplemental Table 1**). However, the Illumina genotyping arrays are sparse
443 compared to the reference panels. We filtered our array data for conformity and the markers
444 remaining used for the variant call formatted files (VCF) are indicated (**Supplemental Table 2**).

445
446 *Phased genotype, haplotype, and diplotype analysis.* The phased genotyping data on
447 chromosome 7 for the three *TAS2R38* SNPs (rs713598, rs10246939, and rs1726866) were used
448 to extract the haplotypes of each study subject using the public server at usegalaxy.org [62] to
449 analyze the data with the VCFgenotype-to-haplotype tool (v1.0.0). VCFtools (v0.1.15) was used
450 to generate all genotype and haplotype frequencies as well as the linkage disequilibrium analyses
451 [63]. The resulting diplotype consisting of the three substitution mutations was used to determine
452 the bitter taste sensitivity phenotype using previously published PROP taste responsiveness with
453 a single PAV haplotype conferring bitter taste [18].

454
455 *Outcome variable.* We used the Block Fruit and Vegetable Screener [64] to assess vegetable
456 consumption in two mutually exclusive categories: green salads and other types of vegetables.
457 The Block F&V screener is valid for assessing high and low vegetable intake and has been used
458 in African American and White populations [64,65]. Frequency scores were calculated by adding
459 the frequency categories (0 = less than once/week; 1 = once/week; 2 = 2-3 times/week; 3 = 4-6
460 times/week; 4 = once/day; 5 = 2 or more/day) for the two questions. Frequency scores ranged
461 from 1-10. A score of four is equivalent to about one serving of vegetables per day and a score of
462 five is equivalent to two or more servings per day.

463
464 *Covariates.* The following covariates were included in the models: sex, age, household income,
465 education, and current smoking status. Taste perception diminishes with age [66] and females are
466 typically more taste sensitive than males [67]. Smoking reduces taste perception [68]. Race,
467 income, smoking status, and education levels are associated with vegetable consumption [69–
468 71]. Sex, smoking status (currently smoking, non-smoker), race (African American or

469 Caucasian), household income (reported in \$5,000 incremental categories), and highest year of
470 education achieved, were included as categorical variables. Income was defined as total
471 combined income of participants' household in the past year, including income from all sources
472 such as wages, salaries, Social Security or retirement benefits, and help from relatives. The mean
473 household income was imputed when data were missing (**Table 1**). Age was used as a
474 continuous variable.

475
476 *Statistical analysis.* We used mixed effects models with repeated measures and STATA's
477 margins command to estimate the adjusted predicted vegetable consumption score for
478 participants within each intervention group and phenotype group at baseline and 6-months follow
479 up. We tested two-way interactions (phenotype group : intervention group and phenotype group :
480 time) and a three-way interaction (phenotype group : intervention : time). Adjusted predicted
481 margins estimate the means for each group of interest, adjusting for the covariates in the mixed
482 effects models [72]. Predicted margins for vegetable consumption scores were contrasted to test
483 whether there were significant differences between participants by intervention group and
484 phenotype group over time. Statistical significance was defined as $p \leq 0.05$. Statistical analyses
485 were conducted in STATA 15.0 [73]. Principal components analysis and the p value of
486 individual SNPs or the SNP : time interaction using mixed effects models with repeated
487 measures was conducted in JMP Pro (v13.2.0, SAS).

488

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500 **References**

- 501 1. Kimmons J, Gillespie C, Seymour J, Serdula M, Blanck HM. Fruit and Vegetable Intake
502 Among Adolescents and Adults in the United States: Percentage Meeting Individualized
503 Recommendations. *Medscape J Med*. 2009;11(1):26.
- 504 2. Bowen DJ, Beresford SAA, Christensen CL, Kuniyuki AA, McLerran D, Feng Z, et al.
505 Effects of a Multilevel Dietary Intervention in Religious Organizations. *Am J Heal*
506 *Promot*. 2009;24(1):15–22.
- 507 3. Emmons KM, Stoddard AM, Fletcher R, Gutheil C, Suarez EG, Lobb R, et al. Cancer
508 prevention among working class, multiethnic adults: results of the healthy directions-
509 health centers study. *Am J Public Health*. 2005; 5(7):1200–5.
- 510 4. Djuric Z, Ellsworth JS, Ren J, Sen A, Ruffin MT, IV. A randomized feasibility trial of
511 brief telephone counseling to increase fruit and vegetable intakes. *Prev Med*. 2010;50(5–
512 6):265–71.
- 513 5. Dzewaltowski DA, Estabrooks PA, Welk G, Hill J, Milliken G, Karteroliotis K, et al.
514 Healthy youth places: a randomized controlled trial to determine the effectiveness of
515 facilitating adult and youth leaders to promote physical activity and fruit and vegetable
516 consumption in middle schools. *Health Educ Behav*. 2009;36(3):583–600.
- 517 6. Pomerleau J, Lock K, Knai C, McKee M. Interventions designed to increase adult fruit
518 and vegetable intake can be effective: a systematic review of the literature. *J Nutr*.
519 2005;135(10):2486-2495.
- 520 7. Thomson CA, Ravia J. A Systematic Review of Behavioral Interventions to Promote
521 Intake of Fruit and Vegetables. *J Am Diet Assoc*. 2011;111(10):1523–35.
- 522 8. Guillaumie L, Godin G, Vézina-Im L-A. Psychosocial determinants of fruit and vegetable
523 intake in adult population: a systematic review. *Int J Behav Nutr Phys Act*. 2010;7(1):12.
- 524 9. Rasmussen M, Krølner R, Klepp K-I, Lytle L, Brug J, Bere E, et al. Determinants of fruit
525 and vegetable consumption among children and adolescents: a review of the literature.
526 Part I: quantitative studies. *Int J Behav Nutr Phys Act*. 2006;3(1):22.

- 527 10. Drewnowski A. Taste preferences and food intake. *Annu Rev Nutr.* 1997;17(1):237–253.
- 528 11. Dinehart ME, Hayes JE, Bartoshuk LM, Lanier SL, Duffy VB. Bitter taste markers
529 explain variability in vegetable sweetness, bitterness, and intake. *Physiol Behav.*
530 2006;87(2):304–13.
- 531 12. Kim UK, Jorgenson E, Coon H, Leppert M, Risch N, Drayna D. Positional Cloning of the
532 Human Quantitative Trait Locus Underlying Taste Sensitivity to Phenylthiocarbamide.
533 2003;299(5610):1221–5.
- 534 13. Kim UK, Drayna D. Genetics of individual differences in bitter taste perception: lessons
535 from the PTC gene. *Clin Genet.* 2005;67(4):275–80.
- 536 14. Bufe B, Breslin PAS, Kuhn C, Reed DR, Tharp CD, Slack JP, et al. The Molecular Basis
537 of Individual Differences in Phenylthiocarbamide and Propylthiouracil Bitterness
538 Perception. *Curr Biol.* 2005;15(4):322–7.
- 539 15. Rozengurt E. Taste Receptors in the Gastrointestinal Tract. I. Bitter taste receptors and α -
540 gustducin in the mammalian gut. *Am J Physiol - Gastrointest Liver Physiol.*
541 2006;291(2):G171–7.
- 542 16. Genick UK, Kotalik Z, Ledda M, Souza Destito MC, Souza MM, A. Cirillo C, et al.
543 Sensitivity of Genome-Wide-Association Signals to Phenotyping Strategy: The PROP-
544 TAS2R38 Taste Association as a Benchmark. Matsunami H, editor. *PLoS One.*
545 2011;6(11):e27745.
- 546 17. Kim UK, Breslin PAS, Reed D, Drayna D. Genetics of Human Taste Perception. *J Dent*
547 *Res.* 2004;83(6):448–53.
- 548 18. Boxer EE, Garneau NL. Rare haplotypes of the gene TAS2R38 confer bitter taste
549 sensitivity in humans. *SpringerPlus.* 2015;4(1):505–508.
- 550 19. Duffy VB, Hayes JE, Davidson AC, Kidd JR, Kidd KK, Bartoshuk LM. Vegetable Intake
551 in College-Aged Adults Is Explained by Oral Sensory Phenotypes and TAS2R38
552 Genotype. *Chemosens Percept.* 2010;3(3–4):137–148.
- 553 20. Carcaise-Edinboro P, McClish D, Kracen AC, Bowen D, Fries E. Fruit and Vegetable

- 554 Dietary Behavior in Response to a Low-Intensity Dietary Intervention: The Rural
555 Physician Cancer Prevention Project. *J Rural Heal*. 2008;24(3):299–305.
- 556 21. Ammerman AS, Lindquist CH, Lohr KN, Hersey J. The efficacy of behavioral
557 interventions to modify dietary fat and fruit and vegetable intake: a review of the
558 evidence. *Prev Med*. 2002;35(1):25–41.
- 559 22. Keyserling TC, Samuel-Hodge CD, Pitts SJ, Garcia BA, Johnston LF, Gizlice Z, et al. A
560 community-based lifestyle and weight loss intervention promoting a Mediterranean-style
561 diet pattern evaluated in the stroke belt of North Carolina: the Heart Healthy Lenoir
562 Project. *BMC Public Health*. 2016;16:732–754.
- 563 23. Cené CW, Halladay JR, Gizlice Z, Donahue KE, Cummings DM, Hinderliter A, et al. A
564 multicomponent quality improvement intervention to improve blood pressure and reduce
565 racial disparities in rural primary care practices. *J Clin Hypertens*. 2017;19(4):351–360.
- 566 24. Risso DS, Mezzavilla M, Pagani L, Robino A, Morini G, Tofanelli S, et al. Global
567 diversity in the TAS2R38 bitter taste receptor: revisiting a classic evolutionary PROPosal.
568 *Sci Rep*. 2016;6(1):25506.
- 569 25. Drewnowski A, Kristal A, Cohen J. Genetic taste responses to 6-n-propylthiouracil among
570 adults: a screening tool for epidemiological studies. *Chem Senses*. 2001;26(5):483–489.
- 571 26. Bachmanov AA, Beauchamp GK. Taste Receptor Genes. *Annu Rev Nutr*.
572 2007;27(1):389–414.
- 573 27. Campa D, De Rango F, Carrai M, Crocco P, Montesanto A, Canzian F, et al. Bitter taste
574 receptor polymorphisms and human aging. Glendinning JI, editor. *PLoS One*.
575 2012;7(11):e45232.
- 576 28. Bell KI, Tepper BJ. Short-term vegetable intake by young children classified by 6-n-
577 propylthiouracil bitter-taste phenotype. *Am J Clin Nutr*. 2006;84(1):245–251.
- 578 29. Tepper BJ, White EA, Koelliker Y, Lanzara C, D’Adamo P, Gasparini P. Genetic
579 Variation in Taste Sensitivity to 6-n-Propylthiouracil and Its Relationship to Taste
580 Perception and Food Selection. *Ann N Y Acad Sci*. 2009;1170(1):126–139.

- 581 30. Tepper BJ. Nutritional Implications of Genetic Taste Variation: The Role of PROP
582 Sensitivity and Other Taste Phenotypes. *Annu Rev Nutr.* 2008;28(1):367–388.
- 583 31. Mikołajczyk-Stecyna J, Malinowska AM, Chmurzynska A. TAS2R38 and CA6 genetic
584 polymorphisms, frequency of bitter food intake, and blood biomarkers among elderly
585 woman. *Appetite.* 2017;116:57–64.
- 586 32. Pliner P. The Effects of Mere Exposure on Liking for Edible Substances. *Appetite.*
587 1982;3(3):283–290.
- 588 33. Anzman-Frasca S, Savage JS, Marini ME, Fisher JO, Birch LL. Repeated exposure and
589 associative conditioning promote preschool children’s liking of vegetables. *Appetite.*
590 2012;58(2):543–553.
- 591 34. Stein LJ, Nagai H, Nakagawa M, Beauchamp GK. Effects of repeated exposure and
592 health-related information on hedonic evaluation and acceptance of a bitter beverage.
593 *Appetite.* 2003;40(2):119–129.
- 594 35. Bongoni R, Verkerk R, Steenbekkers B, Dekker M, Stieger M. Evaluation of Different
595 Cooking Conditions on Broccoli (*Brassica oleracea* var. *italica*) to Improve the Nutritional
596 Value and Consumer Acceptance. *Plant Foods Hum Nutr.* 2014;69(3):228–234.
- 597 36. Drewnowski A, Gomez-Carneros C. Bitter taste, phytonutrients, and the consumer: a
598 review. *Am J Clin Nutr.* 2000;72(6):1424–35.
- 599 37. Bresin P, Beauchamp G. Salt enhances flavour by suppressing bitterness. *Nature.*
600 1997;387:563.
- 601 38. Bachmanov AA, Bosak NP, Lin C, Matsumoto I, Ohmoto M, Reed DR, et al. Genetics of
602 taste receptors. *Curr Pharm Des.* 2014;20(16):2669–2683.
- 603 39. Behrens M, Meyerhof W. Signaling in the Chemosensory Systems. *Cell Mol Life Sci.*
604 2006;63(13):1501–1509.
- 605 40. Behrens M, Brockhoff A, Batram C, Kuhn C, Appendino G, Meyerhof W. The Human
606 Bitter Taste Receptor hTAS2R50 Is Activated by the Two Natural Bitter Terpenoids
607 Andrographolide and Amarogentin. *J Agric Food Chem.* 2009;57(21):9860–9866.

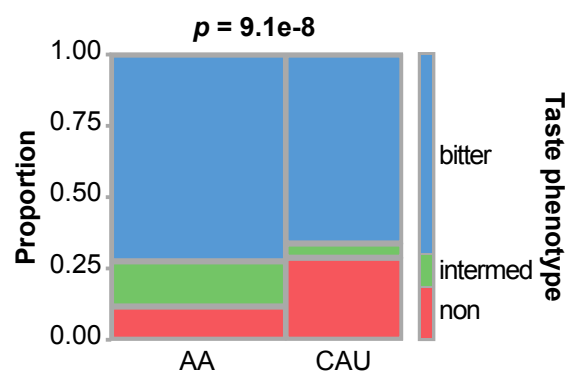
- 608 41. Meyerhof W, Batram C, Kuhn C, Brockhoff A, Chudoba E, Bufe B, et al. The Molecular
609 Receptive Ranges of Human TAS2R Bitter Taste Receptors. *Chem Senses*.
610 2010;35(2):157–170.
- 611 42. Roudnitzky N, Behrens M, Engel A, Kohl S, Thalmann S, Hübner S, et al. Receptor
612 Polymorphism and Genomic Structure Interact to Shape Bitter Taste Perception. Gojobori
613 T, editor. *PLOS Genet*. 2015;11(9):e1005530.
- 614 43. Upadhyaya JD, Chakraborty R, Shaik FA, Jaggupilli A, Bhullar RP, Chelikani P. The
615 Pharmacochaperone Activity of Quinine on Bitter Taste Receptors. Abe K, editor. *PLoS*
616 *One*. 2016;11(5):e0156347.
- 617 44. Artinian NT, Fletcher GF, Mozaffarian D, Kris-Etherton P, Van Horn L, Lichtenstein AH,
618 et al. Interventions to Promote Physical Activity and Dietary Lifestyle Changes for
619 Cardiovascular Risk Factor Reduction in Adults: A Scientific Statement From the
620 American Heart Association. *Circulation*. 2010;122(4):406–441.
- 621 45. Kroeze W, Werkman A, Brug J. A systematic review of randomized trials on the
622 effectiveness of computer-tailored education on physical activity and dietary behaviors.
623 *Ann Behav Med*. 2006;31(3):205–223.
- 624 45. Noar SM, Benac CN, Harris MS. Does tailoring matter? Meta-analytic review of tailored
625 print health behavior change interventions. *Psychol Bull*. 2007;133(4):673–693.
- 626 47. Wittwer J, Rubio-Aliaga I, Hoefft B, Bendik I, Weber P, Daniel H. Nutrigenomics in
627 human intervention studies: Current status, lessons learned and future perspectives. *Mol*
628 *Nutr Food Res*. 2011;55(3):341–358.
- 629 48. Arkadianos I, Valdes AM, Marinos E, Florou A, Gill RD, Grimaldi KA. Improved weight
630 management using genetic information to personalize a calorie controlled diet. *Nutr J*.
631 2007;6:29–37.
- 632 49. Lek M, Karczewski KJ, Minikel E V., Samocha KE, Banks E, Fennell T, et al. Analysis of
633 protein-coding genetic variation in 60,706 humans. *Nature*. 2016;536(7616):285–291.
- 634 50. Corbie-Smith G. The continuing legacy of the Tuskegee Syphilis Study: considerations for

- 635 clinical investigation. *Am J Med Sci.* 1999;317(1):5–8.
- 636 51. Corbie-Smith G, Thomas SB, Williams M V, Moody-Ayers S. Attitudes and beliefs of
637 African Americans toward participation in medical research. *J Gen Intern Med.*
638 1999;14(9):537–546.
- 639 52. Popejoy AB, Fullerton SM. Genomics is failing on diversity. *Nature.*
640 2016;538(7624):161–164.
- 641 53. Halladay JR, Donahue KE, Hinderliter AL, Cummings DM, Cene CW, Miller CL, et al.
642 The Heart Healthy Lenoir project--an intervention to reduce disparities in hypertension
643 control: study protocol. *BMC Health Serv Res.* 2013;13(1):441–452.
- 644 54. Skinner HG, Calancie L, Vu MB, Garcia B, DeMarco M, Patterson C, et al. Using
645 community-based participatory research principles to develop more understandable
646 recruitment and informed consent documents in genomic research. *PLoS One.* 2015;10(5):
647 e0125466.
- 648 55. Thayer LM, Pimentel DC, Smith JC, Garcia BA, Sylvester LL, Kelly T, Johnston LF,
649 Ammerman AS, Keyserling TC. Eating Well While Dining Out: Collaborating with Local
650 Restaurants to Promote Heart Healthy Menu Items. *Am J Health Educ.* 2017;48(1):11-21.
- 651 56. Pitts SBJ, Vu MB, Garcia BA, McGuirt JT, Braxton D, Hengel CE, et al. A community
652 assessment to inform a multilevel intervention to reduce cardiovascular disease risk and
653 risk disparities in a rural community. *Fam Community Health.* 2013;36(2):135–146.
- 654 57. Howard G, Labarthe DR, Hu J, Yoon S, Howard VJ. Regional Differences in African
655 Americans' High Risk for Stroke: The Remarkable Burden of Stroke for Southern African
656 Americans. *Ann Epidemiol.* 2007;17(9):689–696.
- 657 58. Centers for Disease Control and Prevention. Interactive atlas of heart disease and stroke.
658 Atlanta, Ga: CDC. 2014.
- 659 59. Mozaffarian D. Dietary and policy priorities for cardiovascular disease, diabetes, and
660 obesity. *Circulation.* 2016;133(2):187–225.
- 661 60. Das S, Forer L, Schönherr S, Sidore C, Locke AE, Kwong A, et al. Next-generation

- 662 genotype imputation service and methods. *Nat Genet.* 2016;48(10):1284–1287.
- 663 61. Browning BL, Browning SR. Genotype Imputation with Millions of Reference Samples.
664 *Am J Hum Genet.* 2016;98(1):116–126.
- 665 62. Afgan E, Baker D, van den Beek M, Blankenberg D, Bouvier D, Čech M, et al. The
666 Galaxy platform for accessible, reproducible and collaborative biomedical analyses: 2016
667 update. *Nucleic Acids Res.* 2016;44(W1):W3–10.
- 668 63. Danecek P, Auton A, Abecasis G, Albers CA, Banks E, DePristo MA, et al. The variant
669 call format and VCFtools. *Bioinformatics.* 2011;27(15):2156–2158.
- 670 64. Block G, Gillespie C, Rosenbaum EH, Jenson C. A rapid food screener to assess fat and
671 fruit and vegetable intake. *Am J Prev Med.* 2000;18(4):284–288.
- 672 65. Gary TL, Baptiste-Roberts K, Gregg EW, Williams DE, Beckles GLA, Miller EJ, et al.
673 Fruit, vegetable and fat intake in a population-based sample of African Americans. *J Natl*
674 *Med Assoc.* 2004;96(12):1599–605.
- 675 66. Mennella JA, Pepino MY, Reed DR. Genetic and Environmental Determinants of Bitter
676 Perception and Sweet Preferences. *Pediatrics.* 2005;115(2):e216–222.
- 677 67. Bartoshuk LM, Duffy VB, Miller IJ. PTC/PROP tasting: anatomy, psychophysics, and sex
678 effects. *Physiol Behav.* 1994;56(6):1165–1171.
- 679 68. Peterson DI, Lonergan LH, Hardinge MG. Smoking and taste perception. *Arch Environ*
680 *Heal An Int J.* 1968;16(2):219–222.
- 681 69. Drewnowski A. Obesity and the food environment: Dietary energy density and diet costs.
682 *Am J Prev Med.* 2004;27(3, Supplement):154–162.
- 683 70. Serdula MK, Gillespie C, Kettel-Khan L, Farris R, Seymour J, Denny C. Trends in fruit
684 and vegetable consumption among adults in the United States: behavioral risk factor
685 surveillance system, 1994-2000. *Am J Public Health.* 2004;94(6):1014–1018.
- 686 71. Grimm KA, Foltz JL, Blanck HM, Scanlon KS. Household Income Disparities in Fruit
687 and Vegetable Consumption by State and Territory: Results of the 2009 Behavioral Risk
688 Factor Surveillance System. *J Acad Nutr Diet.* 2012;112(12):2014–2021.

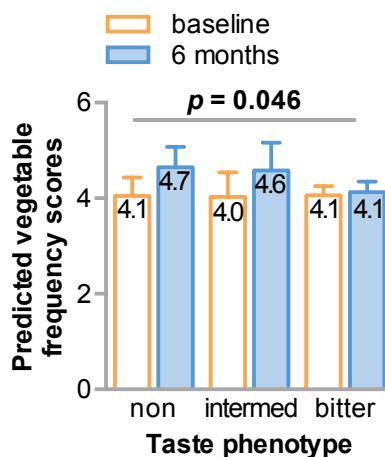
- 689 72. Williams R. Using the margins command to estimate and interpret adjusted predictions
690 and marginal effects. *Stata J.* 2012;12(2):308–331.
691 73. StataCorp. STATA 15.0. College Station, TX.

692 **Figures**



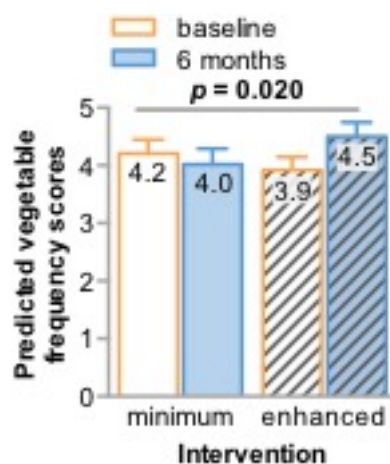
693 **Figure 1: *TAS2R38* bitter taste phenotype distribution in the HHL cohort.** Contingency plot
694 and p value of the Fisher's Exact Test in comparing the distribution (proportion) of taste
695 phenotypes in the AA and CAU group.

696



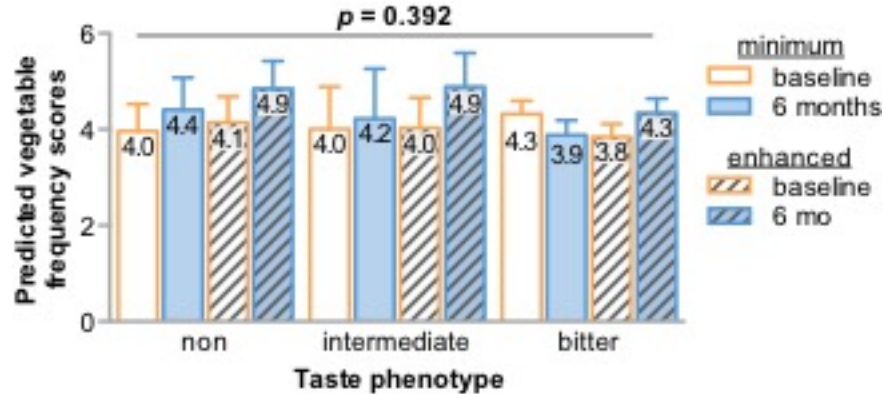
697 **Figure 2: Vegetable intake at baseline and after 6-months categorized by *TAS2R38* bitter**
698 **taste phenotype.** Bar plots of the predicted vegetable intake adjusted for sex, ancestry, age,
699 education, income, and smoking status represented by the mean \pm 95% confidence intervals at
700 either the onset of the study (baseline) or at the 6-month follow up, grouped by non-bitter (non),
701 intermediate-bitter (inter), or bitter tasting phenotype. The p value of the interaction between
702 taste phenotype and time is indicated.

703



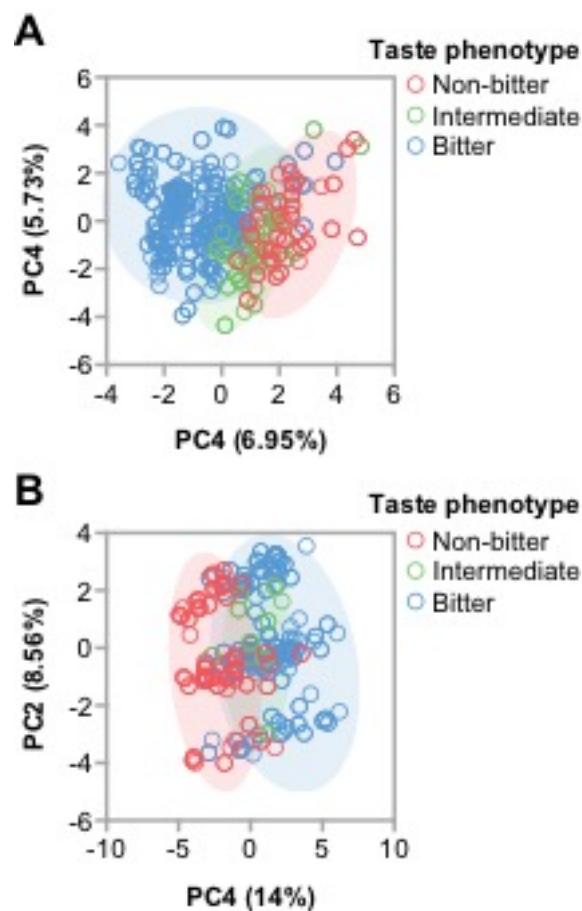
704 **Figure 3: Vegetable intake at baseline and after 6-months categorized by intervention**
705 **intensity.** Bar plots of the predicted vegetable intake adjusted for sex, ancestry, age, education,
706 income, and smoking status represented by the mean \pm 95% confidence intervals at either the
707 onset of the study (baseline) or at the 6-month follow up, grouped into the minimal or enhanced
708 intervention group. The *p* value of the interaction between intervention intensity and time is
709 indicated.

710



711 **Figure 4: Vegetable intake at baseline and after 6-months in either intervention group**
712 **categorized by *TAS2R38* bitter taste phenotype.** Bar plots of the predicted vegetable intake
713 adjusted for sex, ancestry, age, education, income, and smoking status represented by the mean \pm
714 95% confidence intervals at either the onset of the study (baseline) or at the 6-month follow up,
715 grouped by non-bitter (non), intermediate-bitter (inter), or bitter tasting phenotype within each
716 intervention (minimum or enhanced). The p value of the three-way interaction between taste
717 phenotype, time, and intervention intensity is indicated.

718



719 **Figure 5. Multivariate analysis of *TAS2R* polymorphisms in the HHL cohort.** Principal
720 component scatter plots of (A) AA and (B) CAU groups colored by the *TAS2R38* phenotype. The
721 percent variance explained by the indicated principal component (PC) is indicated.

722 **Tables**

723

724 **Table 1: Study participant demographics at baseline and after 6-months of dietary**

725 **intervention.** Data presented as the frequency in each category for the indicated time point and

726 intervention: *, **, and *** correspond to $p < 0.05$, < 0.01 , or < 0.001 via a chi-squared

727 comparing intervention intensity at baseline (†). The p value of a chi-squared test comparing

728 baseline to 6-month follow-up is also indicated (‡).

729

	Baseline characteristics (N=497)		p	Characteristics at 6-month follow-up (N=387)		p
Intervention intensity	Minimal	Enhanced		Minimal	Enhanced	
Total participants	238 (48%)	259 (52%)		176 (46%)	210 (54%)	
Phenotype						
Non-bitter taster	45 (19%)	48 (19%)	0.203	31 (18%)	42 (20%)	0.987
Intermediate taster	21 (9%)	36 (14%)		14 (8%)	29 (14%)	
Bitter taster	172 (72%)	175 (68%)		131 (74%)	139 (66%)	
*Sex						
M	78 (33%)	62 (24%)	0.029	62 (35%)	45 (21%)	0.883
F	160 (67%)	197 (76%)		114 (65%)	165 (79%)	
**Race (ancestry)						
White (CAU)	110 (46%)	90 (35%)	0.009	81 (46%)	71 (34%)	0.795
Black (AA)	128 (54%)	169 (65%)		95 (54%)	139 (66%)	
Age (y)						
18-29	3 (1%)	4 (2%)	0.078	1 (1%)	3 (1%)	0.258
30-44	31 (13%)	33 (13%)		12 (7%)	23 (11%)	
45-65	134 (56%)	171 (66%)		103 (59%)	137 (65%)	
> 65	70 (29%)	51 (20%)		60 (34%)	47 (22%)	
***Education						
Grade 12 or less	171 (72%)	148 (57%)	0.003	121 (69%)	113 (54%)	0.628
1- 2 y post high school	35 (15%)	46 (18%)		27 (15%)	36 (17%)	
3- 4 y post high school	20 (8%)	46 (18%)		19 (11%)	42 (20%)	
≥ 5 y post high school	12 (5%)	19 (7%)		9 (5%)	19 (9%)	
Total household income						
≤ \$14,999	70 (29%)	79 (31%)	0.409	49 (28%)	59 (28%)	0.900
\$15,000 – 29,000	53 (22%)	62 (24%)		41 (23%)	54 (26%)	
\$30,000 – 49,000	33 (14%)	30 (12%)		22 (13%)	24 (11%)	
≥ \$50,000	41 (17%)	63 (24%)		26 (15%)	56 (27%)	
Did not report	41 (17%)	25 (10%)		38 (22%)	17 (8%)	
**Smoking status						
Never	180 (76%)	220 (85%)	0.009	144 (82%)	179 (85%)	0.221
Some days or everyday	58 (24%)	39 (15%)		32 (18%)	31 (15%)	

730

731 **Table 2: TAS2R38 linkage disequilibrium and haplotype frequencies.** Statistical analyses of
 732 linkage disequilibrium (LD) are represented by R-squared (R^2), D, and Dprime values of the
 733 pairwise comparisons of the indicated SNPs from the AA and CAU participants. The plus strand
 734 haplotype sequence (HAPLO), the count of each haplotype, and the resulting amino acid
 735 sequence of the allele are indicated from the AA and CAU participants.

736

AA (N = 304)					
LD analysis	SNP1	SNP2	R²	D	Dprime
	rs10246939	rs1726866	0.49	-0.16	-1.00
	rs10246939	rs713598	0.95	0.24	0.99
	rs1726866	rs713598	0.46	-0.16	-0.98
HAPLO	C:G:G:307 PAV	T:A:C:190 AVI	T:G:C:104 AAI	C:G:C:5 AAV	T:A:G:2 PVI
CAU (N = 201)					
LD analysis	SNP1	SNP2	R²	D	Dprime
	rs10246939	rs1726866	0.98	-0.25	-0.99
	rs10246939	rs713598	0.84	0.23	1.00
	rs1726866	rs713598	0.84	-0.23	-1.00
HAPLO	C:G:G:170 PAV	T:A:C:214 AVI	T:G:C:1 AAI	C:G:C:16 AAV	C:A:C:1 AVV

737

738 **Table 3: TAS2R38 diplotype frequencies and associated phenotype.** The distribution of
739 diplotypes within the AA and CAU participants. The indicated bitter tasting phenotype for each
740 diplotype is indicated.

741

AA (N = 304)		
Diplotype	Freq	Phenotype
PAV / PAV	0.286	bitter
PAV / AVI	0.270	bitter
AAI / PAV	0.155	bitter
AVI / AVI	0.118	non
AAI / AVI	0.115	intermediate
AAI / AAI	0.033	intermediate
AAV / PAV	0.013	bitter
AVI / PVI	0.007	intermediate
AAV / AAI	0.003	intermediate

CAU (N = 201)		
Diplotype	Freq	Phenotype
AVI / PAV	0.438	bitter
AVI / AVI	0.289	non
PAV / PAV	0.184	bitter
AAV / AVI	0.040	intermediate
AAV / PAV	0.040	bitter
AVI / AAI	0.005	intermediate
AVV / AVI	0.005	intermediate

742

743 **Table 4: Regression coefficients for vegetable intake frequency at baseline (Model 1) and**
 744 **mixed effects coefficients at 6 months (Model 2).** The coefficient of variation, standard error
 745 (SE), *t* statistic (Model 1), *z* score value (Model 2), 2-tailed *p* values ($P > |t|$ or $P > |z|$), and
 746 95% confidence intervals (CI) are provided: *, **, and *** correspond to $p < 0.05$, < 0.01 , or $<$
 747 0.001

748

MODEL 1

Variables	Coefficient	SE	<i>t</i>	$P > t $	95% CI
Intermediate taster	-0.10	0.337	-0.28	0.777	-0.76 – 0.57
Bitter taster	0.01	0.231	0.03	0.979	-0.45 – 0.46
Non-smoker	0.14	0.231	0.58	0.562	-0.32 – 0.59
**Female	0.63	0.199	3.15	0.002	0.24 – 1.02
Age	0.01	0.008	1.88	0.061	-0.001 – 0.03
*Education	0.08	0.037	2.08	0.038	0.004 – 0.15
***Income	0.14	0.034	4.11	<0.001	0.07 – 0.21
Race	0.14	0.195	0.74	0.459	-0.24 – 0.53
Constant	0.80	0.766	0.97	0.335	-0.77 – 2.25

MODEL 2

Variables	Coefficient	SE	<i>z</i>	$P > z $	95% CI
Intermediate taster	0.06	0.501	0.13	0.899	-0.92 – 1.05
Bitter taster	0.37	0.313	1.17	0.242	-0.25 – 0.98
Enhanced intervention group	0.19	0.387	0.49	0.621	-0.57 – 0.95
Inter.: Enhanced	-0.17	0.644	-0.27	0.791	-1.43 – 1.09
Taster: Enhanced	-0.70	0.434	-1.54	0.123	-1.52 – 0.18
6-month follow-up	0.46	0.338	1.36	0.174	-0.20 – 1.12
Inter.: 6-months follow-up	-0.26	0.601	-0.43	0.671	-1.43 – 0.92
Taster: 6-months follow-up	-0.89	0.376	-2.38	0.018	-1.63 – -0.16
Enhanced: 6-months	0.25	0.450	0.56	0.573	-0.63 – 1.13
Inter.: Enhanced: 6-month follow-up	0.40	0.758	0.53	0.598	-1.09 – 1.89
Taster: Enhanced: 6-month follow-up	0.68	0.505	1.35	0.177	-0.31 – 1.67
Non-smoker	0.30	0.198	1.54	0.123	-0.08 – 0.69
***Female	0.70	0.166	4.22	<0.001	0.38 – 1.02
Age	0.01	0.007	1.55	0.122	-0.003 – 0.02
**Education	0.09	0.031	2.89	0.004	0.03 – 0.15
***Income	0.14	0.028	4.93	<0.001	0.08 – 0.19
Race	-0.01	0.164	-0.01	0.994	-0.32 – 0.32
Constant	0.75	0.164	1.12	0.264	-0.56 – 2.05

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750

751 **Table 5: SNP and SNP-time associations with vegetable intake.** The *p* values of the
752 association of either the indicated SNP or the SNP : time interaction with reported vegetable
753 intake. The location of the gene is indicated by chromosome (Chr) and position (Pos).

754

SNP	Gene	Chr	Pos	p (SNP)	p (SNP:time)
rs713598	<i>TAS2R38</i>	7	141673345	0.0659	0.0147
rs10246939	<i>TAS2R38</i>	7	141672604	0.0659	0.0147
rs1726866	<i>TAS2R38</i>	7	141672705	0.1208	0.1452
rs10772408	<i>TAS2R49</i>	12	11151599	0.4936	0.7443
rs1376251	<i>TAS2R49</i>	12	11138852	0.3534	0.9068
rs7301234	<i>TAS2R49</i>	12	11150884	0.2838	0.7276

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