

1 **Studies of human twins reveal genetic variation that affects dietary fat perception**

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22 **Author Contributions**

23 DRR designed the study. LC, FG, IM, and PJ collected data. PW, PASB, NER, LS, DB,

24 and JEH contributed to data interpretation. LF contributed reagents and also

25 contributed to data interpretation, and AS and AM assisted in stimuli preparation and

26 data collection. All authors were involved in drafting the article or revising it for

27 intellectual content, and have read and approved the final version of the manuscript.

28

## 29 Abstract

30 To learn more about the mechanisms of human dietary fat perception, 398 human twins  
31 rated fattiness and liking for six types of potato chips that differed in triglyceride  
32 content (2.5, 5, 10, and 15% corn oil); reliability estimates were obtained from a subset ( $n$   
33 = 50) who did the task twice. Some chips also had a saturated long-chain fatty acid  
34 (hexadecanoic acid, 16:0) added (0.2%) to evaluate its effect on fattiness and liking. We  
35 computed the heritability of these measures and conducted a genome-wide association  
36 study (GWAS) to identify regions of the genome that co-segregate with fattiness and  
37 liking. Perceived fattiness and liking for the potato chips were reliable ( $r = 0.31-0.62$ ,  $p <$   
38  $0.05$ ) and heritable (up to  $h^2 = 0.29$ ,  $p < 0.001$ , for liking). Adding hexadecanoic acid to  
39 the potato chips significantly increased ratings of fattiness but decreased liking. Twins  
40 with the G allele of *rs263429* near *GATA3-AS1* or the G allele of *rs8103990* within  
41 *ZNF729* reported more liking for potato chips than did twins with the other allele  
42 (multivariate GWAS,  $p < 1 \times 10^{-5}$ ), with results reaching genome-wide suggestive but not  
43 significance criteria. Person-to-person variation in the perception and liking of dietary  
44 fat was (a) negatively affected by the addition of a saturated fatty acid and (b) related to  
45 inborn genetic variants. These data suggest liking for dietary fat is not due solely to  
46 fatty acid content and highlight new candidate genes and proteins within this sensory  
47 pathway.

48 **Keywords:** taste, genetics, sensory, fat perception, oleogustus, taste receptors

## 49 Introduction

50 Sensory nutrition is a research area that investigates how the taste, smell, and flavor of  
51 food and drink affect food intake and diet quality, and how food choice in turn affects  
52 human health and disease (Forde 2018; Hayes 2015). While food is essential to our  
53 survival, and eating may be pleasant, it can also be dangerous, especially for those who  
54 “dig their grave with a spoon” (Card 2013) and die from heart disease or diabetes,  
55 health conditions that arise in whole or in part from dietary choices (Reed and Knaapila  
56 2010). Some of the pleasure of food arises from its dietary fat and sugar content. The  
57 sweetness of sugar is well understood from a sensory perspective (Nelson *et al.* 2001),  
58 with direct links between taste cells and brain areas of reward (e.g., (Veldhuizen *et al.*  
59 2017)). In contrast, the initial sensory steps responsible for the perception of dietary fat  
60 are less well understood, and what is known is contentious: whether there is a distinct  
61 taste quality for fat or fatty acids, and which of the chemical and texture components of  
62 fat are responsible for the sensations it evokes (Reed and Xia 2015; Running *et al.* 2015;  
63 Running and Mattes 2016).

64 One unresolved conundrum is mounting evidence that, while triglycerides and  
65 fatty acids both impart fatty sensations in foods, triglycerides tend to have a positive  
66 hedonic valance, e.g.,(Bakke *et al.* 2016) whereas fatty acids typically have a negative  
67 hedonic valence, e.g., scratchy (Voigt *et al.* 2014) or otherwise “bad” (Running and  
68 Mattes 2016). These data suggest multiple sensory pathways are involved in the

69 perception of fats in foods (Drewnowski 1992). One method to learn more about these  
70 multiple pathways is to evaluate origins of person-to-person or animal-to-animal  
71 differences—this type of genetics-driven approach helped identify the bitter and sweet  
72 receptors (Reed and Knaapila 2010; Reed *et al.* 2006). Here, we reasoned that people  
73 differ in their response to fat in food, that these differences are heritable, and that  
74 genome-wide methods are likely useful to identify the relevant genes.

75 To establish heritability, we selected a classic twin design, comparing  
76 monozygotic (MZ) and dizygotic (DZ) twins for their response to fat in foods. We also  
77 had to choose appropriate test stimuli that would generalize to real foods (vs. model  
78 systems) and appropriate behavioral methods. No one standard method has been  
79 adopted, with investigators in this area using many different stimuli to measure fat  
80 perception, including oil-and-water mixtures (Heinze *et al.* 2017); oil in salad dressing  
81 (Keller *et al.* 2012); fat in puddings (Mennella *et al.* 2012), in scrambled eggs or mashed  
82 potatoes (Mela and Sacchetti 1991), or in ice cream (Rolon *et al.* 2017) or added fatty  
83 acids in chocolate (Running *et al.* 2017). Here we used potato chips that varied in  
84 amounts of corn oil and an added fatty acid, capitalizing on our technical expertise in  
85 their production and practical constraints of our testing environment (an annual  
86 convention of twins; see below). We also tested the twins' ability to discriminate high-  
87 and low-fat milk samples.

## 88 **Materials and Methods**

89 **Participants.** We tested adult MZ and DZ twins who attended an annual convention of  
90 twins, the Twin Days Festival in Twinsburg, OH. This event is held each August, and  
91 all data reported here were collected during the 2018 convention. The exclusion criteria  
92 for participation were age less than 18 years, pregnancy, or an allergy or sensitivity to  
93 milk. All data were collected under protocols approved by the University of  
94 Pennsylvania Institutional Review Board (#701426).

95 **Stimuli.** Three types of stimuli were used: potato chips that differed in triglyceride and  
96 fatty acid content, multiple prototypical tastants, and milk that was either high (18.00%)  
97 or low (2.35%) in fat. Six types of potato chips were prepared, following standard  
98 methods at Pepsico research laboratories: chips that contained 2.5, 5.0, 10, or 15% corn  
99 oil and chips with 2.5% or 5.0% corn oil with added 0.2% (w/w) hexadecanoic acid, a  
100 saturated long-chain fatty acid (16:0). Time constraints prevented us from testing all  
101 combinations of triglycerides and fatty acids. Ascending amounts of corn oil were  
102 chosen to minimize carryover effects across samples; the fatty acid was added to gauge  
103 its impact on ratings of fattiness and liking.

104 The second type of stimuli comprised standard solutions (5 mL) used in taste  
105 psychophysics: plain deionized water, sucrose (12% w/v, 350 mM), sodium chloride  
106 (1.5% w/v, 256 mM), and the bitter compound phenylthiocarbamide (PTC;  $1.8 \times 10^{-4}$  M),  
107 all purchased from Sigma (St. Louis, MO). (We also tested menthol [1 mM] and  
108 capsaicin [3  $\mu$ M] for an unrelated project; those results are not reported here.) The third

109 stimuli comprised milk with 18.00% or 2.35% fat mixed at the Monell Chemical Senses  
110 Center using Shop Rite brand instant nonfat dry milk (SKU/UPC 041190010189)  
111 purchased at a local grocery store and anhydrous dairy fat (**Table 1**). All ingredients  
112 were combined in a homogenizer (GEA, Düsseldorf, Germany) and processed with five  
113 passes at 250 bars of pressure; resulting particle sizes were within the expected range.

114 **Sample presentation.** Single potato chips of roughly equivalent size and weight were  
115 placed in clear 3-5 oz plastic souffle cups with plastic lids (Universal Product Code  
116 [UPC] #742010492467). Participants were given potato chips in a predetermined order  
117 and asked to rate the potato chips for “fattiness” and “liking” on visual analog scales  
118 presented on an Apple iPad Air (9.7-inch display; Apple Inc., Cupertino, CA). Liking  
119 scales were anchored with “do not like at all” on the left and “like extremely” on the  
120 right. Similarly, the fattiness scale was anchored on the left with “not fatty at all” and on  
121 the right with “extremely fatty.” We also asked about “crispiness” and “saltiness,” to  
122 prevent a halo-dumping effect, a bias in sensory ratings which can occur when subjects  
123 are provided too few salient rating options (Clark and Lawless 1994). Participants were  
124 instructed to rinse their mouth with water (Nestle Pure Life, UPC 068274934711) after  
125 each sample. For logistical reasons and enhanced ecological validity, participants did  
126 not wear nose clips and chewed and swallowed all potato chip samples.

127 For the taste solutions, participants rated each for the qualities of “liking,”  
128 “saltiness,” “sweetness,” “sourness,” “bitterness,” and “burn” on visual analog scales,

129 with the left side anchored with “no [quality] at all” and the right side anchored with  
130 “extreme [quality],” as previously described (Knaapila *et al.* 2012). To focus on taste and  
131 reduce odor cues, participants wore nose clips (GENEXA LLC, UPC 708981350007).  
132 Participants were asked to hold each solution in their mouth for 5 s, rate it on the scale  
133 provided, spit out the solution, and rinse their mouth with water afterward.

134 For the milk fat discrimination test, a two-alternative forced choice task was  
135 used. Before testing began, each participant was given two references as warm-up  
136 samples; these were verbally identified to participants as “low-fat” and “high-fat”  
137 samples, respectively. Participants were then given 10 pairs of opaque bottles (EP-  
138 34434, Berry Global Group, Inc.). Each pair contained one low-fat and one high-fat  
139 sample (each 5 mL) presented in a fixed order. Participants wore nose clips; they were  
140 instructed to hold each sample in their mouth for 5 s, spit out the sample, and rinse  
141 their mouth with water afterward. For each pair, participants were asked, “Which  
142 solution tastes fattier?” If they were unsure, they were instructed to guess.

143 Discrimination ability was defined as the number correct across all 10 trials (i.e., perfect  
144 discrimination would be 10 out of 10 trials correct).

145 **Saliva collection and DNA extraction.** We obtained saliva samples from all participants  
146 by asking them to expectorate into collection tubes; DNA was extracted from the saliva  
147 using procedures recommended by Oragene (DNA Genotek, Kanata, Canada). We



148 measured and recorded DNA concentration and quality scores using a Nanodrop 1000  
149 Spectrophotometer (Thermo Fisher Scientific, Waltham, MA).

150 **Genotyping.** We conducted both single-marker and high-throughput based  
151 genotyping. Using the single-marker method, we typed three variant sites in the  
152 *TAS2R38* gene in all twins, as a quality-control step (a) to ensure that the DNA extracted  
153 from saliva could be genotyped, (b) to confirm that the genotype matched the  
154 psychophysical ratings of PTC bitterness, and (c) to get preliminary confirmation of  
155 twin zygosity (each pair of MZ twins is expected to have the same genotype). For these  
156 assays, DNA samples were diluted to a concentration of 10 ng/ $\mu$ L and used as  
157 templates in Taqman assays (*rs713598*, C\_\_8876467\_10; *rs1726866*, C\_\_9506827\_10;  
158 and *rs10246939*, C\_\_9506826\_10; Applied Biosystems, Foster City, CA) using  
159 previously established methods.

160 For the DNA high-throughput genotyping, we sent the DNA samples to the  
161 Center for Inherited Disease Research (CIDR; Baltimore, MD), which typed them for the  
162 Illumina OmniExpress panel (Infinium OmniExpressExome-8, v1.6; Illumina, San  
163 Diego, CA) following the manufacturer's procedures and the CIDR's standard quality-  
164 control methods. For 176 MZ twin pairs, we used high-throughput genotyping for only  
165 one twin of each pair and imputed the genotype of the other member of the pair  
166 because of their presumed identical genomes.

167 **Twin zygosity.** Twin zygosity was measured in three ways. Twins self-reported their  
168 zygosity status as (a) monozygotic (MZ; identical), (b) dizygotic (DZ; fraternal), or (c)  
169 uncertain; photographs were taken of each twin and rated for physical similarity by a  
170 research assistant blind to self-reported zygosity, and all twins were genotyped for the  
171 three markers described above. In rare cases where zygosity status was still uncertain,  
172 both members of the pair were genotyped using the high-throughput-based genotyping  
173 method (see above).

174 **Data analysis.** We conducted four types of statistical analysis: (a) descriptive statistics  
175 of the psychophysical data, (b) calculation of heritability, (c) tests of genome-wide  
176 association between genetic variants and the measures of fat perception, and (d) gene  
177 expression (RNASeq) and bioinformatics (enrichment) analyses. All descriptive  
178 statistics, such as means, standard deviations (SDs), and correlations among variables,  
179 were computed using R (v. 3.53) and R-Studio (v. 1.1.456).

180 *Sensory analyses.* For descriptive analyses, we plotted the probability density of  
181 the data (smoothed by a kernel density estimator) by a violin plot, calculated mean and  
182 SD, and checked for sex, race, and age effects on the sensory measures in a general  
183 linear model (GLM) using race and sex as fixed effects and age as a covariate. For all  
184 GLM analyses, individual group means were evaluated for difference using Tukey post  
185 hoc tests (honestly significant difference [HSD]). If race and sex had a significant effect

186 in the GLM analysis, to better understand their effects on psychophysical outcomes we  
187 grouped participants by these factors and compared the mean ratings. For age and its  
188 relationship to the psychophysical measures, we computed Pearson correlations.

189 To evaluate whether there were consistent person-to-person differences in the  
190 rating of the potato chips overall, Pearson correlations of intensity and liking measures  
191 among the six types of potato chip were calculated. In addition, we calculated  
192 Cronbach's alpha for psychophysical measures across all six types of potato chips. To  
193 understand the reliability of the measures, we assessed test-retest correlations among  
194 the same measures taken twice in a subset of participants ( $n = 50$ ).

195 To gauge the effect of corn oil concentrations and hexadecanoic acid on the  
196 sensory measures, we reconducted a linear mixed-model analysis with corn oil  
197 concentration (2.5% and 5.0%) and hexadecanoic acid (added or not) as two separate  
198 factors and treated the psychophysical data as repeated measurements, with race and  
199 age as covariates in the model. (We did not include sex in this model because results  
200 indicated that male and females were similar in their ratings.) In a complementary  
201 analysis, we reconducted the analysis using potato chip type as a single factor (with six  
202 levels, one for each type of potato chip). These complementary analyses were included  
203 because of the unbalanced design: not all concentrations of corn oil were presented with  
204 and without the added 0.2% hexadecanoic acid.

205           *Heritability.* For the heritability analysis, the Cholesky model was used to  
206 evaluate the magnitude of genetic and environmental influences on the traits, and the  
207 phenotypic variance was decomposed into additive genetic component ( $a^2$ ), shared  
208 environmental factors ( $c^2$ ), and nonshared environmental or individual-specific factors  
209 ( $e^2$ ), as described previously (Wise *et al.* 2007). Variance accounted for by each of these  
210 components was calculated by comparing MZ twin correlations to DZ twin correlations.  
211 The computation of the heritability was conducted using R package OpenMx (v. 2.13)  
212 (Boker *et al.* 2011).

213           *Genome-wide association studies.* For GWAS we expanded variants from ~720,000  
214 to 11,315,231 by imputation using the Michigan Imputation Server (Das *et al.* 2016) with  
215 the reference genome HRCr1.1 (McCarthy *et al.* 2016). We filtered out markers with a  
216 low minor allele frequency (<5%) and removed markers that had  $p$ -values associated  
217 with Hardy-Weinberg disequilibrium  $< 1e-6$ , genotype call rate  $< 0.9$ , and imputation  
218 score  $< 0.3$ . The remaining 4,234,798 variants on the 22 autosomes were used for GWAS  
219 for each trait (univariate GWAS [uvGWAS]), with genetic relatedness matrix (20  
220 eigenvalues) calculated by principal components analysis, and sex and age used as  
221 covariates (Hwang *et al.* 2019; Liu *et al.* 2018; Wu *et al.* 2018). The genome-wide  
222 significance threshold was  $p = 5.0e-8$ , and for suggestive associations it was  $p = 1e-5$   
223 (International HapMap 2005; Pe'er *et al.* 2008).

224           We reasoned that there would be more statistical power to detect associations if  
225 we considered the liking and fattiness ratings from all potato chips simultaneously,  
226 especially because, as the results indicated, these measures were correlated (e.g., people  
227 with high liking ratings for the 5% corn oil chip also liked the 10% chip more). Thus, we  
228 conducted multivariate GWAS (mvGWAS) using the correlated ratings for all the  
229 potato chips. The covariates are the same as uvGWAS procedure; the computation was  
230 done using GEMMA (Zhou and Stephens 2012), and regional associational plots were  
231 created using LocusZoom (Pruim *et al.* 2010). For the mvGWAS, GEMMA adjusted for  
232 testing multiple phenotypes and applied a correction for multiple phenotypes (Fatumo  
233 *et al.* 2019). For the milk discrimination task, the trait was not heritable (see Results), so  
234 we did not conduct GWAS.

235           *Candidate gene analyses.* We extracted variants from the candidate genes that were  
236 previously implicated in the sensory signaling of fat taste from either animal models  
237 (mouse and rat) or human studies: *CD36* (Abumrad 2005; Gaillard *et al.* 2007; Keller *et*  
238 *al.* 2012; Laugerette *et al.* 2005; Pepino *et al.* 2012; Sclafani *et al.* 2007a), *GNAT3* (Sclafani  
239 *et al.* 2007b), *GPR120* (Cartoni *et al.* 2010; Matsumura *et al.* 2007; Tsuzuki 2007), *GPR40*  
240 (Cartoni *et al.* 2007; Cartoni *et al.* 2010; Matsumura *et al.* 2007), *TRPM5* (Liu *et al.* 2011;  
241 Sclafani *et al.* 2007b), *GPR41* and *GPR43* (Brown *et al.* 2003), *GPR84* (Wang *et al.* 2006),  
242 and *KCNA2* (Gilbertson *et al.* 1998; Liu *et al.* 2005). In addition, we looked at genes for  
243 salivary enzymes (lipase, lysozyme, and amylase) and protein (lipocalin, mucin, and

244 protein rich in proline) because these proteins change in response to dietary fat  
245 consumption (Feron G 2013; Mounayar *et al.* 2014).

246 To extract the results of genotype-phenotype association for these candidate  
247 genes, we conducted analyses using two methods. In method 1 we identified the most  
248 significant variant within each candidate genes for each trait and extracted the relevant  
249 *p*-value and other test statistics. In method 2 we chose the most significant variant for  
250 traits of the potato chip with 5% corn oil (with no added fatty acid) and examined all  
251 the sensory measures for the same variant; that is, we chose the 5% corn oil chip as the  
252 baseline from which to compare the other associations. These methods are  
253 complementary because method 1 detects associations that are specific to a particular  
254 concentration of triglyceride and fatty acid combination, while method 2 detects  
255 common variants affecting the intensity and liking measures across the potato chip  
256 types. We also examined the effect of the variant *rs1761667* within *CD36* because it was  
257 previously associated with fat sensory perception in humans (Keller *et al.* 2012; Mrizak  
258 *et al.* 2015; Pepino *et al.* 2012; Sayed *et al.* 2015).

259 *Gene expression in human taste tissue using the RNASeq method.* To understand  
260 whether the genes identified by GWAS might be acting at the level of the receptors in  
261 taste tissue (as opposed to in the brain or in other tongue tissue, e.g., the filiform  
262 papillae), we compared the mRNA expression of these genes to those previously  
263 implicated in the peripheral aspects of fat taste perception (e.g., the candidate gene

264 *CD36*) in human taste tissue. To do so, we collected fungiform papillae from subjects  
265 recruited for our previous study (Douglas *et al.* 2019) using published procedures  
266 (Spielman *et al.* 2010) and isolated the RNA following the manufacturer's directions,  
267 processing the taste tissue with Quick-RNA MiniPrep R1054 (Zymo Research, Irvine,  
268 CA). We evaluated RNA quality expressed as an RNA integrity number (RIN) using the  
269 Agilent 2200 TapeStation system (Agilent Technologies, Santa Clara, CA). The six  
270 samples with sufficient RNA quality as determined by the Next-Generation Sequencing  
271 Core of the University of Pennsylvania (RIN > 7; 5 males and 1 female) were used to  
272 perform library preparation and sequencing (100 bp single-end) on the HiSeq 4000  
273 sequencer (Illumina, San Diego, CA) following the manufacturer's sequencing  
274 protocols. We mapped reads to the reference genome (GRCh38.p10) after the raw  
275 sequence data in fastq format passed standard quality filters equipped in Trimmomatic  
276 (Bolger *et al.* 2014), and then normalized the counts using the R package Ballgown  
277 (Frazee *et al.* 2014). The expression level in RPKM (reads per kilobase per million  
278 mapped reads) of each gene for each sample was used to compare their expression  
279 level.

280 *Pathway and gene set enrichment analysis.* We reasoned that genes identified  
281 through GWAS may be partners with other genes that code for proteins in related  
282 sensory pathways. Thus, we conducted pathway analyses of the genes identified by  
283 uvGWAS and mvGWAS. Using the background of the genes from the database of Gene

284 Ontology annotations (Thomas *et al.* 2003) and Reactome annotations (Fabregat *et al.*  
285 2018; Fabregat *et al.* 2017), we used Fisher's exact test to examine whether there was  
286 enrichment of these pathways versus all annotated human genes using  
287 GENEVESTIGATOR (Hruz *et al.* 2008).

## 288 **Results**

289 **Participant characteristics.** The twins ( $N = 398$ ) were predominantly female (72%,  $n$   
290 = 285; and 28% male,  $n = 113$ ), middle-aged ( $38.6 \pm 16.7$ , mean  $\pm$  SD), and members of MZ  
291 twin pairs ( $n = 360$  twins, 90.4%). Most were of European descent ( $n = 331$ , 83.2%), but  
292 some participants were of African descent ( $n = 50$ , 12.6%). The remaining racial groups  
293 (e.g., Asian) were grouped into an "other" category for the analyses described below ( $n$   
294 = 19, 4.8%). A total of 213 individual subjects were genotyped using the chip-based  
295 platform (MZ,  $n = 184$ ; DZ,  $n = 29$ ), and 176 MZ twins had their genotypes imputed.

## 296 **Liking and intensity measures**

297 *Liking and fattiness ratings differed across potato chips with variable fat content.*

298 Overall, participants liked the potato chips and were able to accurately rate them for  
299 fattiness. Adding 0.2% hexadecanoic acid to the potato chips increased fattiness at both  
300 corn oil concentrations tested (**Figure 1A**). The effect of added hexadecanoic acid on  
301 liking was less straightforward: for the 5% corn oil chips, adding a 16:0 fatty acid did  
302 not alter liking, while for the 2.5% corn oil chips, adding the fatty acid decreased liking



303 (Figure 1A). For chips with no added fatty acid, there was a mostly linear increase in  
304 ratings of fattiness as corn oil concentration increased, although a plateau was reached  
305 above 10% oil (Figure 1B). For liking, there was a J-shaped curve: participants liked the  
306 2.5% and 15% corn oil potato chips best (Figure 1B). See Supplemental Figures 1 and 2  
307 and Supplemental Table 1 for additional details.

308 *Relationship between liking and fattiness relative to benchmarks.* Within each type of  
309 potato chip, the ratings of liking and fattiness were only slightly or not at all related  
310 (Figure 2A). This relationship between liking and sensory quality differed from those  
311 for the benchmark taste solutions; for example, participants liked sucrose better if they  
312 rated it as sweeter (Figure 2B).

313 *Reliability of liking and fattiness relative to benchmarks.* The ratings of both fattiness  
314 and liking for the potato chips were reliable ( $r = 0.31-0.62, p < 0.05$ ; Supplemental  
315 Figure 3), slightly lower than (but mostly similar to) those for the benchmark taste  
316 solutions (sucrose, NaCl, and PTC;  $r = 0.54-0.74, p < 0.0001$ , except for NaCl saltiness;  
317 Supplemental Figure 3).

318 *Age, race, and sex effects on fattiness and liking.* Men and women were similar in  
319 their ratings of all sensory stimuli (Supplemental Table 2). Race and age had significant  
320 effects on some sensory ratings ( $p < 0.01$ ; Supplemental Table 2). Younger participants  
321 liked some of the potato chip types more than older participants ( $r = -0.17$  to  $-0.14, p <$   
322  $0.001$ ; Supplemental Figure 4). People of European ancestry rated some potato chips as

323 less fatty than did people of African ancestry (5.0% corn oil without added fatty acid;  $p$   
324  $< 0.05$ , GLM analysis followed by post hoc Tukey HSD tests; **Supplemental Figure 5**).  
325 There were also race effects for the other sensory stimuli, for example, for the liking of  
326 sucrose and PTC. **Supplemental Figure 5** summarizes all sensory results that differed  
327 by race.

328 *Relationships of ratings across potato chip type.* Each participant tasted and rated six  
329 potato chips, and there were correlations among each participant's ratings of fattiness  
330 (Cronbach's alpha = 0.75, 95% confidence boundaries = 0.72-0.79) and liking (Cronbach's  
331 alpha = 0.77, 95% confidence boundaries = 0.74-0.81). Fattiness correlations tended to be  
332 higher among the chips without added FA than with the chips with added FA. A scatter  
333 matrix of pairwise correlations between potato chips types is shown in **Figure 3**.

334 *Discrimination of milk fattiness.* On average, participants could discriminate the  
335 high- and low-fat milk samples (exact binomial test, one-tailed,  $p < 0.0001$ ), but only  
336 slightly above chance (probability of success = 0.53; **Supplemental Figure 6A**). This  
337 ability to discriminate was only somewhat reliable when testing the same participant  
338 twice (retest correlation,  $r = 0.36$ ;  $p > 0.05$ ; **Supplemental Figure 3**). We had expected  
339 based on our pilot data collected in our sensory laboratory that about 30% of  
340 participants would perform this discrimination perfectly every time, with 10 out of 10  
341 samples correctly identified, but our results showed that only 3% of subjects could do  
342 so.

343 **Heritability.** Between about 10% and 30% of the variation in potato chip liking arose  
344 from genetics ( $h^2$ ), but only about 5-15% for ratings of fattiness (**Table 2**). For  
345 comparison, for the bitter compound PTC, the most heritable taste trait currently  
346 known, liking heritability was 53%, and for sucrose, which has a midrange heritability,  
347 it was 46%. The pattern of heritability for NaCl was similar to that for potato chips, as  
348 rating of NaCl liking has more genetic variation than does rating of NaCl saltiness. We  
349 did not calculate heritability for the milk fat discrimination because there was no  
350 similarity in milk discrimination scores between the twins (**Supplemental Figure 6B**).

351 **Genome-wide association.** No associations met the commonly accepted genome-wide  
352 significance threshold, but we did identify suggestive variants using the univariate and  
353 multivariate methods. uvGWAS identified nine associations for fattiness and eight for  
354 liking (**Table 3**). All these associations were specific for potato chip type. The mvGWAS  
355 detected two variants for chip fattiness and five variants for chip liking (**Table 4**). We  
356 reasoned that associations detected with both uvGWAS and mvGWAS would be most  
357 valid. Of the seven genotype associations detected by mvGWAS, two (*GATA3-AS1* and  
358 *ZNF729*) were also detected by uvGWAS (**Figure 4**): twins with the G allele of *rs263429*  
359 (*10:8085050*, near *GATA3-AS1*) reported more liking for the potato chips than did twins  
360 with the other allele and the same was true for the G allele of *rs8103990* (*19:22476027*,  
361 within *ZNF729*) (mvGWAS,  $p < 1 \times 10^{-5}$ ; **Table 4**). We show the allelic effects for these two

362 variants in **Figure 5**. The effects of the novel variants were larger than those for *CD36*,  
363 the candidate gene previously associated with fat perception (**Supplemental Figure 7**).

364 **Candidate genes.** None of the candidate genes consistently met a genome-wide  
365 statistical threshold, but some candidate genes were more often associated with potato  
366 chip fattiness or liking than others at a nominal significance threshold ( $p < 0.05$ ; **Figure**  
367 **6A**). The most notable results were significant variants within *CD36* and *TRPM5*  
368 associated with potato chop liking and fattiness (**Figure 6B, C**; **Supplemental Figure 8**,  
369 **Supplemental Tables 3 and 4**). For *CD36*, the variant *rs1761667* (which was associated  
370 with fat perception in previous studies) did not pass the quality-control filters, but we  
371 examined a nearby variant, *rs1722501*, that was in nearly perfect linkage disequilibrium  
372 ( $R^2 > 0.99$ ) with *rs1761667*. However, participants did not differ in ratings of potato chip  
373 fattiness or liking for this proxy marker (**Supplemental Table 5**), although there were  
374 many associations for other variants within *CD36*, as noted above (see **Figure 6**).

375 **Gene expression, pathway, and gene enrichment analysis.** We reasoned that  
376 expression of fat candidate genes (those that have a proposed role in peripheral fat or  
377 fatty acid signaling) would be a benchmark to compare the taste-tissue expression of the  
378 novel genes identified from the GWAS results. Compared with receptor and other  
379 signaling candidate genes (*GPR40*, *GPR41*, *GRP43*, *GPR84*, *GPR120*, *TRPM5*, *CD36*,  
380 *KCNA2*, and *GNAT3*), the novel genes have relatively higher expression levels in

381 fungiform papillae, especially for *RAPGEF2*, *GLI3*, *MCTP1*, and *MLLT3* (**Figure 7**).  
382 *ZNF729* and *GATA3-AS1* had a similar expression abundance as the candidate genes  
383 *GPR40*, *GPR41*, *GPR84*, *GPR120*, *KCNA2*, and *TRPM5* but much lower than the  
384 candidate genes *GRP43*, *CD36*, and *GNAT3*. The presence of many of the novel genes in  
385 taste tissue is consistent with a role in peripheral perception, but some candidate genes  
386 had a very low abundance. This subset of low-abundance novel genes may be nearly  
387 undetectable in the taste tissue sampled because only a few of the relevant cells may  
388 have been present in the tissue sample or because the genes may act at different times  
389 (e.g., early development) or in different tissues (e.g., the filiform papillae or the brain).

390 We conducted pathway analysis to understand the function of as many of the  
391 novel genes identified as possible. In the GENEVESTIGATOR analysis, 21 of the 22  
392 associated genes identified by GWAS (*RP11-575F12.1* is not found in the database) were  
393 tested against the 74,727 background genes. Three gene sets were enriched using the  
394 associated genes as bait ( $p < 0.001$ , Fisher's exact test; **Supplemental Figure 9**,  
395 **Supplemental Table 6**), from the Gene Ontology categories synapse GO:0045202, cell-  
396 cell signaling GO:0007267, and positive regulation of neurogenesis GO:0050769. Overall,  
397 these results point to a role of these genes and their protein products in sensory  
398 signaling and perhaps regulation of sensory cell types.

399 **Discussion**

400 Dietary fat is added to food to increase its flavor and palatability, but whether fat is  
401 sensed by chemical cues (e.g., from fatty acids), textural cues, or both is contentious.  
402 The data from this study support previous observations that fatty acids provide a  
403 chemical cue for fattiness but that this component of fattiness is not desirable (Running  
404 *et al.* 2017). When hexadecanoic acid (a saturated 16-carbon fatty acid) was added to the  
405 potato chip lowest in fat, it was rated as fattier but was less liked than a potato chip  
406 with a comparable amount of fat but without the added fatty acid. Thus, presumably,  
407 taking a broader view and generalizing, this result suggests that increasing “fattiness”  
408 by adding fatty acids to foods would not make them better liked, and raises the  
409 possibility that recently discovered antagonists to the fatty acid receptors (Milligan *et al.*  
410 2017) might improve fat flavor. These data support the hypothesis that there are at least  
411 two sensory inputs for fat perception, a chemical cue and presumably a textural cue,  
412 with the texture conveying perhaps the pleasant aspects of fattiness.

413 In addition to studying the relationship between fattiness and liking, we also  
414 attempted to study fat discrimination, asking participants to choose the fattier milk  
415 solution from a pair of high- and low-fat samples. This task was difficult for the  
416 participants, and almost no one correctly identified the high-fat sample 10 times out of  
417 the 10 trials. This result came as a surprise because our preliminary testing suggested  
418 this task was easy; however, most preliminary testing was conducted with  
419 commercially available low- and high-fat milk samples and in a quiet sensory

420 laboratory, making discrimination easier. The prepared milk samples used for testing  
421 here were the same in all aspects except the amount dietary fat added, and for many  
422 people the oral cues alone (as opposed to visual or olfactory cues) are insufficient to  
423 discriminate low-fat from high-fat samples. One additional concern about data was the  
424 effect of transportation on the stimuli: the milk was prepared and then driven by truck  
425 several hundred miles to the test location – conceivably, vibration may have caused  
426 coalescence of the fat globules that altered the ability to discriminate between samples.

427         The main focus of this study was to examine whether person-to-person  
428 differences in the liking or perception of fattiness are due in part to individual genetic  
429 variation. To establish the heritability of a trait, it is essential to have a reliable  
430 measurement, that is, a trait that can be measured reproducibly; accordingly,  
431 demonstrating that the measures used were reliable was an essential precondition for  
432 the heritability calculations. We learned from the reliability and heritability analyses  
433 that liking for this solid food matrix, potato chips, with differing fat concentrations was  
434 more similar among genetically identical (MZ) twins than among nonidentical (DZ)  
435 twins. Ratings of fattiness were also heritable, but less so, aligning with results from our  
436 studies of other taste modalities, which, for example, demonstrated that liking for a  
437 concentrated salt solution is more heritable than are salty intensity ratings (Knaapila *et*  
438 *al.* 2012). Our results are in contrast to a recent study of the effect of diet on fatty acid  
439 perception in twins, which reported few or no genetic effects (Costanzo *et al.* 2018);

440 however, these two studies differed in methods, as did the number of twins  
441 investigated, 88 in (Costanzo *et al.* 2018) vs. 398 here.

442 Thus, despite the logistical challenges posed by measuring percepts from dietary  
443 fat, there is evidence for a genetic determinant on par with other traits that have been  
444 studied using GWAS methods (Clarke *et al.* 2017). Building on the heritability analysis,  
445 we also performed two types of GWAS, which are agnostic to prior information about  
446 which genes and variants might be previously known or suspected to contribute to the  
447 perception of dietary fat. This part of the study was underpowered and returned no  
448 results that met the classic statistical threshold for GWAS results but did provide, in  
449 tandem with the bioinformatic analysis, clues about which genes and pathways might  
450 be worth pursuing in future work, specifically in the realm of cell-to-cell  
451 communication and perhaps cell type.

452 Of particular interest is the association between fat liking and variants in the  
453 transcription factors that contribute to the development of taste cells (Ermilov *et al.*  
454 2016; Qin *et al.* 2018). The transcriptome was not helpful in interpreting the novel genes  
455 in part because taste tissue from fungiform papillae is unlikely to be involved in the  
456 textural aspects of fat perception, and in part because the abundance of even the known  
457 genes is very low to undetectable in fungiform taste tissue. Single-cell studies from all  
458 regions of the oral cavity would be a step forward, which is increasingly more feasible  
459 as methods improve, although the most complete experimental paradigm would also



460 include the sensory pathways, including brain regions that process the sensory  
461 properties of dietary fat information (Grabenhorst and Rolls 2014).

462         The results of the candidate gene analyses were more compelling in the sense  
463 that, although none of the results were individually very striking, multiple methods of  
464 analysis have repeatedly indicated a role for *CD36* and *TRPM5* in the perception of  
465 dietary fat, in both human and animal studies (Chamoun *et al.* 2018), especially gene  
466 knockout studies. Parenthetically, we did not see associations with the proxy marker we  
467 used to try to replicate the previous studies exactly (Keller *et al.* 2012; Mrizak *et al.* 2015;  
468 Pepino *et al.* 2012; Sayed *et al.* 2015), but *CD36* is a large gene with many potentially  
469 functional variants, and therefore a fine-mapping study in multiple populations is  
470 warranted. There may be multiple variants that cause a spectrum of effects that differ  
471 by ancestral population, e.g., (Gurdasani *et al.* 2019).

472         We speculate that sensory nutrition and taste perception offer a way to reduce  
473 nutrition-related human diseases, by studying the nuanced and often misunderstood  
474 relationship between liking and intake (Hayes in press). GWAS allows us to screen and  
475 identify common genetic variants associated with fat consumption (Tanaka *et al.* 2013),  
476 and our findings, combined with future functional genomic analyses, especial single-  
477 cell profiling, will delineate the causal genetic variants and biological mechanisms  
478 underlying the observed statistical associations with fat perception (Gallagher and  
479 Chen-Plotkin 2018).

480 **Conflict of Interest**

481 The authors declare no conflicts of interest.

482

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723 **Figure 1.** Corn oil and corn oil spiked with 0.2% hexadecanoic acid (FA) modify ratings  
724 of fattiness and liking of potato chips. (A) Potato chips with more corn oil plus added  
725 FA increased fattiness and decreased liking. (B) As corn oil concentration (2.5%, 5.0%,  
726 10%, and 15%, without added FA) increased, fattiness ratings increased linearly but  
727 liking changed in a J-curve: participants liked potato chips more with corn oil at the  
728 lowest and highest concentrations (2.5% and 15%). The points and bars show least  
729 square mean (LSM) and standard error of rating scores, and different letters (a, b, and c)  
730 indicate a significant LSM difference between groups.

731 **Figure 2.** Pearson correlations between sensory measures indicate multiple mechanisms  
732 underlying dietary fat perception ( $N = 398$ ). (A) No or weak correlations between  
733 ratings of liking and fattiness depending on the type of potato chip. FA=fatty acid  
734 (hexadecanoic acid). (B) Strong correlations between liking and other taste ratings  
735 (sweetness, saltiness, and bitterness) for the standard taste solutions sucrose, NaCl, and  
736 phenylthiocarbamide (PTC).

737 **Figure 3.** Strong and positive interrelated correlations of ratings of fattiness (A) and  
738 liking (B) across the six types of potato chips: scatter plots (lower left), density  
739 distributions (diagonal line), and correlations (upper right). FA=fatty acid  
740 (hexadecanoic acid).



741 **Figure 4.** Venn diagram comparing loci identified by uvGWAS and mvGWAS (see  
742 Methods for details). Two variants were detected by both methods: *10:8085050* near the  
743 gene *GATA3-AS1* and *19:22476027* within *ZNF729*.

744 **Figure 5.** Allele effect of variants *10:8085050* near gene *GATA3-AS1* (A) and *19:22476027*  
745 within the gene *ZNF729* (B) on ratings of liking across types of potato chips. For both  
746 variants, participants with G allele rated higher liking for all potato chip types than did  
747 those with other allele. The standard residual scores for liking were calculated in the  
748 general linear model with covariates of sex, age, and 20 eigenvalues.

749 **Figure 6.** Candidate gene effect on fat perception for potato chips. (A) Total counts of  
750 nominal  $p < 0.05$  out of 28 tests for each candidate gene for the two methods of  
751 candidate gene analysis (method 1 and method 2; see Materials and Methods) in the  
752 outputs from uvGWAS and mvGWAS. (B, C) Associations of top variants within  
753 candidate genes *CD36* and *TRPM5* with ratings of liking (B) and fattiness (C) for each  
754 type of potato chip. x-Axes show effect size ( $\beta \pm SE$ ), obtained from uvGWAS, and y-axes  
755 show  $-\log(p\text{-value})$ , obtained from uvGWAS and mvGWAS, for the top variants within  
756 *CD36* and *TRPM5* (no  $\beta \pm SE$  data were available from mvGWAS; i.e.,  $\beta \pm SE = 0$  is not  
757 true). Red dashed lines indicate  $p = 0.05$ ; the points above this line indicate a nominal  
758 significant effect on the trait. FA=fatty acid (hexadecanoic acid). For other details of the  
759 data, see **Supplemental Tables 3 and 4.**

760 **Figure 7.** Box plots of taste tissue expression abundance of genes near the peak  
761 statistical associations from the GWAS (novel hits) and for candidate genes (shown in  
762 blue) known from prior studies to contribute to fat perception. Two genes, *ZNF729* and  
763 *GATA3-AS1* (shown in red), were commonly detected by both uvGWAS and mvGWAS  
764 in the present study. RPKM=reads per kilo base per million mapped reads. *RNU6-356P*  
765 had no expression in any sample. Outliers are not shown. Red asterisks indicate genes  
766 with statistically higher expression level compared with other genes in taste tissue ( $p <$   
767  $0.05/351 = 0.000142$ , Bonferroni corrections for multiple tests).  
768

769 **Supplemental Figure 1.** Changes in ratings of fattiness and liking by corn oil  
770 concentration across the six types of potato chips. FA=fatty acid (hexadecanoic acid).  
771 For other details, see **Figure 1**.

772 **Supplemental Figure 2.** Violin plots for ratings of the sensory traits. The violin area  
773 shows the estimated density of each rating score point. The dots and bars show means  
774 and SDs.

775 **Supplemental Figure 3.** Pearson correlations between test and retest of each rating ( $n =$   
776 50).

777 **Supplemental Figure 4.** Pearson correlations between age and sensory measures for  
778 potato chips and other taste stimuli. Young participants were more sensitive to taste  
779 stimuli than were older participants.

780 **Supplemental Figure 5.** Least square mean (LSM) and standard error of sensory  
781 measures by race. EA=European Americans, AA=African Americans, Oth=others  
782 (Asian, Hispanic, Native American, mixed). Different letters (a, b) show a significant  
783 LSM difference.

784 **Supplemental Figure 6.** Most participants had difficulty discriminating milk fat  
785 content, with near chance levels overall. **(A)** Histogram of milk fat discrimination  
786 scores. The dashed white line shows probability of success, which is near the chance

787 level of 5, but it is significantly different from the chance level,  $p < 0.001$ . (B) No  
788 significant correlations were observed between twin 1 and twin 2 for milk  
789 discrimination for either DZ or MZ twins; thus, no heritability for milk fat  
790 discrimination scores was calculated.

791 **Supplemental Figure 7.** Regional associational plots, based on mvGWAS results, for  
792 single-nucleotide polymorphisms in linkage disequilibrium ( $r^2$ ) with the peak variants  
793 10:8085050 near the gene *GATA3-AS1* (A) and 19:22476027 within the gene *ZNF729* (B)  
794 for ratings of liking, and for the fat perception candidate gene *CD36* for ratings of liking  
795 (C) and fattiness (D) for potato chips. The highlighted chromosome regions show the  
796 target genes.

797 **Supplemental Figure 8.** Associations of top variants within each candidate gene with  
798 ratings of liking (A) and fattiness (B) for each type of potato chip. For details see **Figure**  
799 **6.**

800 **Supplemental Figure 9.** Gene set enrichment analyses. Venn diagram visualizes  
801 overlapping genes among the top three gene sets and the target genes (21 out of 22  
802 GWAS hits; *RP11-575F12.1* is not found the database). All genes ( $n = 74,727$  total genes)  
803 were selected from Reactome annotations and Gene Ontology annotations as  
804 background collection. The top three gene sets identified are synapse GO:0045202, cell-

805 cell signaling GO:0007267, and positive regulation of neurogenesis GO:0050769 (see

806 **Supplemental Table 6**).

807

808 **Table 1.** High- and low-fat milk ingredients

<b>Milk Type</b>	<b>Fat Content (%)</b>	<b>Water (mL)</b>	<b>Dry Milk (g)</b>	<b>Dairy Fat (g)</b>	<b>Casein (g)</b>
Low fat	2.35	890	90.7	23.7	10.09
High fat	18.00	890	90.7	216.8	12.04

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812 **Table 2.** Heritability ( $h^2$ ) of fat sensory traits, with NaCl, sucrose, and PTC as a  
 813 benchmarks ( $n = 199$  twin pairs)

<b>Stimulus</b>	<b><math>h^2</math></b>	<b>CI</b>
<b>Liking (chips)</b>		
2.5% corn oil	0.21*	0.07 – 0.34
5.0% corn oil	0.10	0.00 – 0.24
10% corn oil	0.10	0.00 – 0.24
15% corn oil	0.29*	0.15 – 0.41
2.5% corn oil with 0.2% hexadecanoic acid	0.10	0.00 – 0.24
5.0% corn oil with 0.2% hexadecanoic acid	0.10	0.00 – 0.24
<b>Fattiness</b>		
2.5% corn oil	0.05	0.00 – 0.20
5.0% corn oil	0.11	0.00 – 0.25
10% corn oil	0.12	0.00 – 0.27
15% corn oil	0.07	0.00 – 0.22
2.5% corn oil with 0.2% hexadecanoic acid	0.03	0.00 – 0.17
5.0% corn oil with 0.2% hexadecanoic acid	0.15	0.00 – 0.29
<b>Other solutions</b>		
Sucrose sweetness	0.11	0.00 – 0.25
Sucrose liking	0.46*	0.33 – 0.56
NaCl saltiness	0.19*	0.05 – 0.32
NaCl liking	0.38*	0.25 – 0.49
PTC bitterness	0.49*	0.38 – 0.59
PTC liking	0.53*	0.42 – 0.62

814 CI=confidence interval.

815 \*Different from zero.

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**Table 3.** Suggestive associations for ratings of potato chip fattiness and liking identified by uvGWAS.

Stimuli	CHR	SNP (CHR:BP)	ALT	REF	MAF	Beta	SE	P	Gene	HIT_TYPE	SNP_TYPE
<b>Fattiness</b>											
5.0% NoFA	4	4:45361270	G	C	0.12	0.92	0.20	6.14E-06	<i>PRKRIRP9</i>	nearest	Imputed
5.0% NoFA	7	7:42032565	T	C	0.26	0.62	0.14	8.25E-06	<i>GLI3</i>	within	Genotyped
5.0% NoFA	8	8:105135473	A	T	0.15	0.86	0.18	1.92E-06	<i>RIMS2</i>	within	Imputed
5.0% NoFA	11	11:7476601	A	G	0.21	0.74	0.15	9.51E-07	<i>SYT9</i>	within	Imputed
10% NoFA	7	7:25514020	C	G	0.18	0.74	0.16	4.91E-06	<i>AC091705.1</i>	nearest	Imputed
15% NoFA	5	5:94138997	C	T	0.17	0.76	0.17	4.37E-06	<i>MCTP1</i>	within	Genotyped
15% NoFA	6	6:77345798	C	T	0.10	0.87	0.20	9.64E-06	<i>RP11-354K4.1</i>	nearest	Imputed
15% NoFA	11	11:86707909	G	C	0.09	0.88	0.20	9.45E-06	<i>RP11-736K20.6</i>	within	Imputed
15% NoFA	19	19:51226244	T	C	0.38	0.59	0.13	9.19E-06	<i>CLEC11A</i>	nearest	Imputed
<b>Liking</b>											
5.0% NoFA	2	2:215358759	C	T	0.06	-1.12	0.25	8.47E-06	<i>VWC2L</i>	within	Imputed
5.0% NoFA	20	20:50443885	T	C	0.25	0.63	0.13	1.84E-06	<i>RP5-1112F19.2</i>	nearest	Imputed
<b>10% NoFA</b>	<b>10</b>	<b>10:8085050</b>	<b>A</b>	<b>G</b>	<b>0.08</b>	<b>-0.92</b>	<b>0.20</b>	<b>5.27E-06</b>	<b>GATA3-AS1</b>	<b>nearest</b>	<b>Imputed</b>
10% NoFA	12	12:127482374	C	G	0.07	-0.95	0.21	9.08E-06	<i>RP11-575F12.1</i>	within	Imputed
2.5% with 0.2% FA	4	4:160267304	A	G	0.06	-1.32	0.30	8.02E-06	<i>RAPGEF2</i>	within	Imputed
2.5% with 0.2% FA	12	12:13948270	A	G	0.10	-1.13	0.24	3.58E-06	<i>GRIN2B</i>	within	Imputed
<b>2.5% with 0.2% FA</b>	<b>19</b>	<b>19:22476027</b>	<b>A</b>	<b>G</b>	<b>0.11</b>	<b>-1.07</b>	<b>0.23</b>	<b>4.92E-06</b>	<b>ZNF729</b>	<b>within</b>	<b>Imputed</b>
5.0% with 0.2% FA	13	13:95498608	T	C	0.09	-0.98	0.22	5.94E-06	<i>RPL21P112</i>	nearest	Imputed

NoFA=no added hexadecanoic acid; CHR=chromosome; SNP=single-nucleotide polymorphism; BP=base pair; ALT=alternative allele; REF=reference allele; MAF=minor allele frequency; SNP TYPE=individual's genotype was genotyped or imputed. Two genes (*PRKRIRP9* and *RP11-270L13.1*) on chr 4 for ratings of potato chip fattiness reached genome-wide suggestive threshold ( $1e-5$ ), but only one with a relatively lower  $p$ -value is reported in this table. Variants for potato chip liking shown in boldface were also detected by the mvGWAS across all corn oil concentrations; see **Table 4**.



**Table 4.** Suggestive genes for ratings of potato chip fattiness and liking identified by mvGWAS

Trait	CHR	SNP (CHR:BP)	ALT	REF	MAF	P	Gene	HIT_TYPE	SNP_TYPE
Fattiness	2	2:141755773	A	T	0.32	2.96E-06	<i>LRP1B</i>	within	Imputed
	10	10:116662107	C	T	0.05	8.97E-06	<i>RP11-106M7.4</i>	nearest	Imputed
Liking	7	7:34676953	T	A	0.06	3.05E-07	<i>NPSR1-AS1</i>	within	Imputed
	8	8:40998854	A	T	0.04	5.14E-07	<i>RNU6-356P</i>	nearest	Imputed
	9	9:20636973	T	C	0.48	1.15E-06	<i>MLLT3</i>	nearest	Genotyped
	<b>10</b>	<b>10:8085050</b>	<b>G</b>	<b>A</b>	<b>0.08</b>	<b>8.19E-07</b>	<b><i>GATA3-AS1</i></b>	<b>nearest</b>	<b>Imputed</b>
	<b>19</b>	<b>19:22476027</b>	<b>A</b>	<b>G</b>	<b>0.11</b>	<b>4.71E-06</b>	<b><i>ZNF729</i></b>	<b>within</b>	<b>Imputed</b>

See **Table 3** for abbreviations and other details. Two genes on chr 2 (*LINC00486* and *LRP1B*) and two genes on chr 10 (*FAM160B1* and *BP11-106M7.4*) for potato chip fattiness, and two genes on chr 7 (*EEPD1* and *NPSR1-AS1*) and two genes on chr 8 (*RNU6-356P* and *SULF1*) for potato chip liking reached genome-wide suggestive threshold ( $1e-5$ ), but only one gene with a relatively lower  $p$ -value on each chromosome is reported in this table. Variants for potato chip liking shown in boldface were also detected by the uvGWAS across all corn oil concentrations. For abbreviations, see the caption of Table 3.

823 **Supplemental Table 1. Summary statistics for linear mixed model analyses**

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825 FA, fatty acid; ICC, intraclass correlation. Boldface indicates the test statistic meets a  
826 significance threshold of  $p < 0.01$ .

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830 **Supplemental Table 2.** The effect of sex, race, and age on sensory measures for potato  
831 chips, taste stimuli, and milk discrimination

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833 PTC, phenylthiocarbamide. Highlighting indicates suggestive effects with a *p-value* <  
834 0.05.

835

836 **Supplemental Table 3.** The effect of the top variant within each candidate gene on  
837 ratings of potato chip fattiness and liking.

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843 FA, fatty acid; mvGWAS, multivariate genome-wide association study. Highlighting  
844 indicates suggestive effects with a  $p$ -value  $< 0.05$ .

845 \*For *GPR41* and *GPR84*, no variant within the genes was available from the association

846 data, so we expanded the region to 500 bp up- and downstream for each site when

847 extracting the variant to examine for association. For other details see **Supplementary**

848 **Tables 5 and 6.**

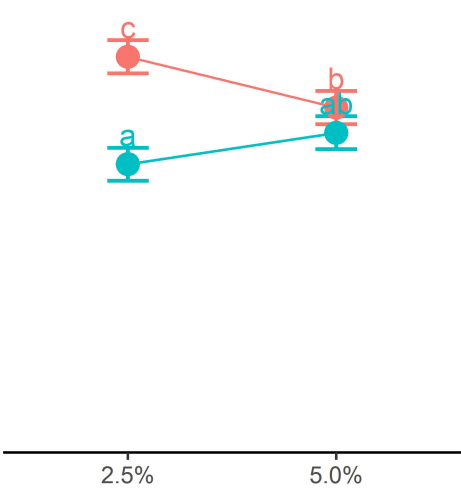
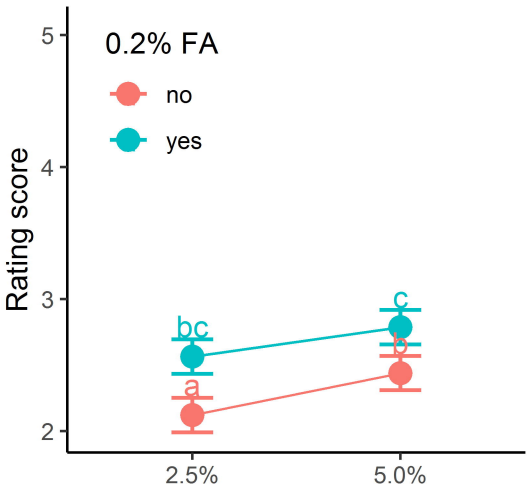
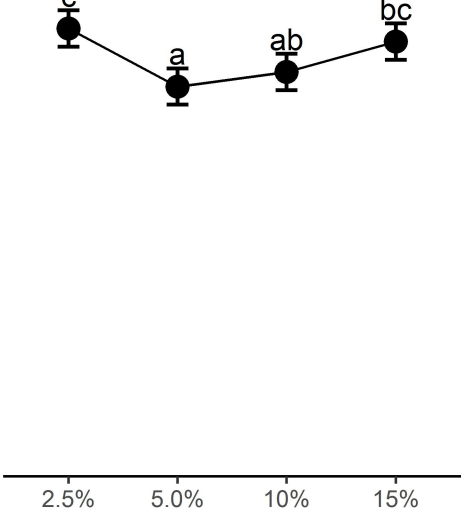
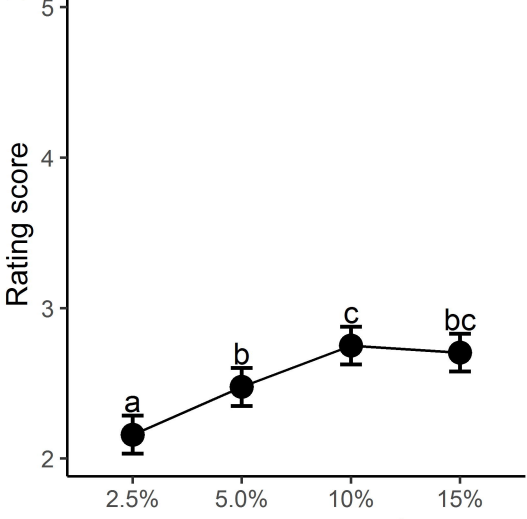
849 **Supplemental Table 4.** The top variant within each candidate gene with effects on  
850 ratings of potato chips with 5% corn oil (without added fatty acid) had effects on  
851 fattiness and liking for other types of potato chips. For details, see **Supplemental Table**  
852 **3.**  
853  
854

855 **Supplemental Table 5.** The variant *rs1722501* (chr7:80244694) as proxy for *rs1761667*  
856 within *CD36* has no significant effect on ratings of potato chip fattiness and liking.

FA, fatty acid; mvGWAS, multivariate genome-wide association study. *rs1761667* (7:80244939) was not in Hardy-Weinberg disequilibrium ( $p=8.5e-15$ ) and thus did not pass the filter test statistics ( $p>1e-6$ ); therefore, we extracted the variant *rs1722501*, which had an  $R^2>0.99$  and linkage disequilibrium  $>0.99$  with *rs1761667*. For this marker, there was no significant effect on fatty and liking for any of the six potato chip types tested. For abbreviations, e.g., MAF, see **Tables 3**.

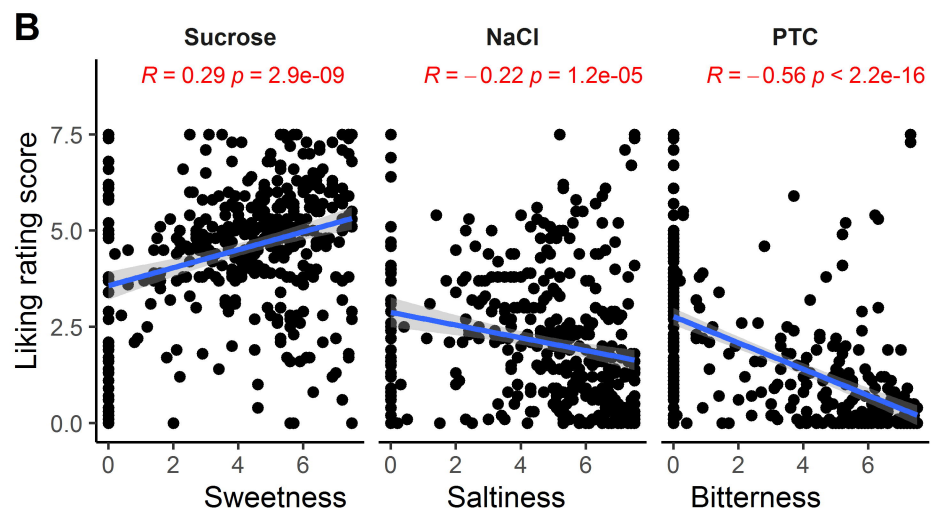
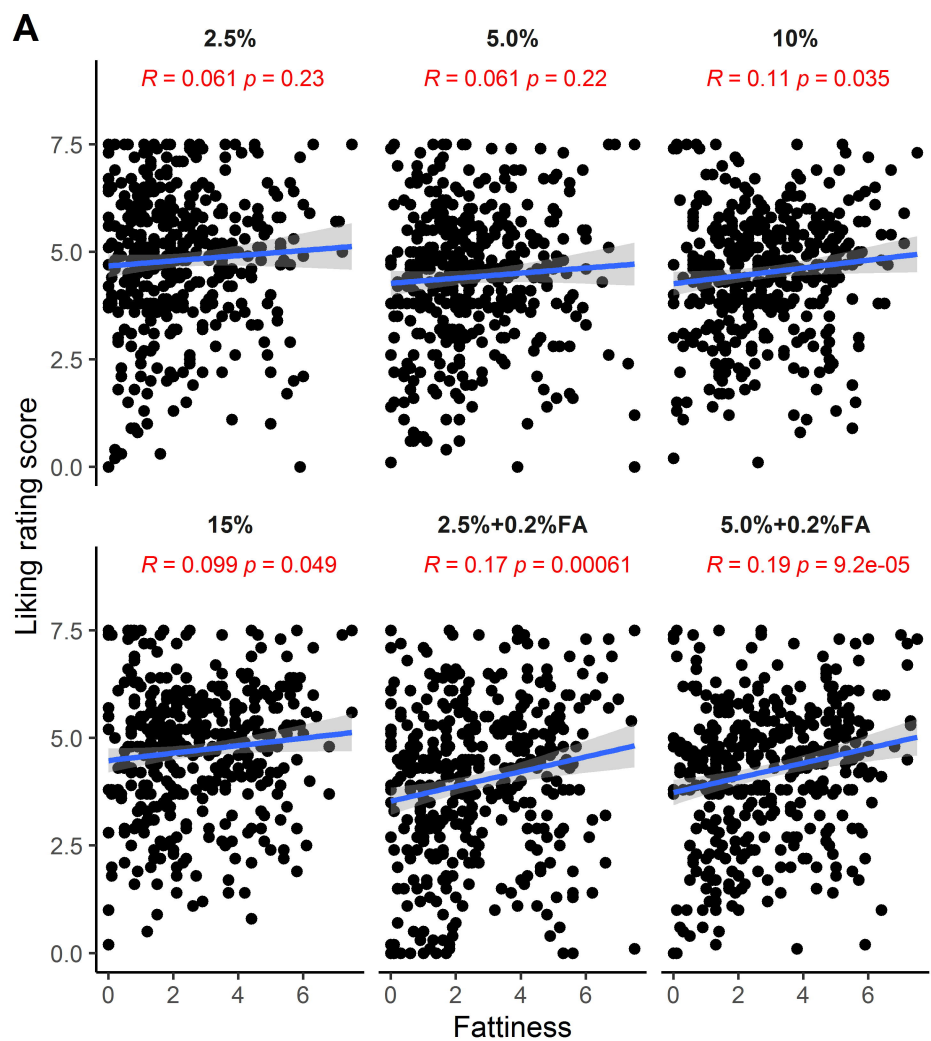
857  
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861 **Supplemental Table 6. Gene set enrichment analysis**  
862

**A****Fattiness****Liking****B**

Corn oil concentration







uvGWAS

mvGWAS

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*AC091705.1*  
*CLEC11A*  
*GLI3*  
*GRIN2B*  
*MCTP1*  
*PRKRIRP9*  
*RAPGEF2*  
*RIMS2*  
*RP11-354K4.1*  
*RP11-575F12.1*  
*RP11-736K20.6*  
*RP5-1112F19.2*  
*RPL21P112*  
*SYT9*  
*VWC2L*

*GATA3-AS1*  
*ZNF729*

*LRP1B*  
*MLLT3*  
*NPSR1-AS1*  
*RNU6-356P*  
*RP11-106M7.4*

