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and body mass index in adults*

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Running head: Facial affective reaction and body weight

Title: Facial affective reactions to bitter-tasting foods and body mass index in adults

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23

24 **Abstract**

25 Differences in food consumption among body-weight statuses (e.g., higher fruit intake
26 linked with lower body mass index (BMI) and energy-dense products with higher BMI) has
27 raised the question of why people who are overweight or are at risk of becoming overweight eat
28 differently from thinner people. One explanation, in terms of sensitivity to affective properties of
29 food, suggests that palatability-driven consumption is likely to be an important contributor to
30 food intake, and therefore body weight. Extending this approach to unpalatable tastes, we
31 examined the relationship between aversive reactions to foods and BMI. We hypothesized that
32 people who have a high BMI will show more negative affective reactions to bitter-tasting
33 stimuli, even after controlling for sensory perception differences. Given that hedonic reactions
34 may influence consumption even without conscious feelings of pleasure/displeasure, the facial
35 expressions were included in order to provide more direct access to affective systems than
36 subjective reports. Forty adults (28 females, 12 males) participated voluntarily. Their ages
37 ranged from 18 to 46 years ($M=24.2$, $SD=5.8$). On the basis of BMI, participants were classified
38 as low BMI ($BMI < 20$; $n=20$) and high BMI ($BMI > 23$; $n=20$). The mean BMI was 19.1 for low
39 BMI ($SD=0.7$) and 25.2 for high BMI participants ($SD=1.8$). Each subject tasted 5 ml of a
40 grapefruit juice drink and a bitter chocolate drink. Subjects rated the drinks' hedonic and
41 incentive value, familiarity and bitter intensity immediately after each stimulus presentation. The
42 results indicated that high BMI participants reacted to bitter stimuli showing more profound
43 changes from baseline in neutral and disgust facial expressions compared with low BMI. No
44 differences between groups were detected for the subjective pleasantness and familiarity. The

45 research here is the first to examine how affective facial reactions to bitter food, apart from taste
46 responsiveness, can predict differences in BMI.

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48 Key words: Affective facial reaction; bitter food; body mass index; overweight; taste
49 responsiveness

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70 Introduction

71 Research on obesity has revealed that overweight/obese people display different eating
72 behaviours to lean people (for review, cf. French, Epstein, Jeffery, Blundell & Wardle, 2012;
73 Mesas, Muñoz-Pareja, López-García & Rodríguez-Artalejo, 2012). Several studies with both
74 children and adults agree that individuals with a higher body mass index (BMI, kg/m²) not only
75 consume larger amount of, e.g., energy-dense snacks (Berteus Forslund, Torgerson, Sjostrom &
76 Lindroos, 2005; Nicklas, Yang, Baranowski, Zakeri & Berenson, 2003), soda/sweetened
77 beverages (Blum, Jacobsen & Donnelly, 2005; Malik, Schulze & Hu, 2006; Nicklas et al., 2003)
78 and fast food (Bowman & Vinyard, 2004; Schröder, Fito & Covas, 2007) compared to those with
79 a lower BMI; but also less fruit and vegetables (Alinia, Hels & Tetens, 2009; Kahn et al, 1997;
80 Lin & Morrison, 2002; Mohindra, Nicklas, O'Neil, Yang & Berenson, 2009). Dietary patterns or
81 changes in patterns of food choice over time have also been linked to BMI status (e.g.,
82 Maskarinec, Novotny & Tasaki, 2000; Pachucki, 2012). Pachucki using cluster analysis with
83 dietary data showed that transitions to lower diet quality clusters (e.g., from fruits and legumes to
84 low/high-fat meat and soda) were associated with a higher BMI. Since excessive fat versus
85 inadequate vegetable and fruit intake have been identified as risk factors for developing obesity
86 and major diseases (e.g., Bray & Popkinand, 1998; Boeing et al., 2012), there is an urgent need
87 to understand why people at risk of obesity choose and eat differently from thinner people.

88 Among the determinants of food preferences related to weight status, some studies have
89 considered whether a predisposition to overeating might be related to hedonic processes (cf.
90 Blundell & Finlayson, 2004; Mela, 2001). In terms of taste preference (Drewnowski, 1997),
91 these studies are based on the assumption that differences in the perceived pleasantness of foods
92 (hedonic sensitivity), and not only in sensory perception (taste responsiveness), should explain

93 the individual variability in BMI. Given that pleasure “comprises the positive dimension of the
94 more general category of hedonic processing [...], which also includes other negative and
95 unpleasant dimensions” (Berridge & Kringelbach, 2008), an attractive possibility is to extend
96 this hedonic eating-based approach of overweight people to aversive tastes. Thus, subjects with a
97 higher sensitivity to the affective value of food might be likely to have a stronger drive to eat
98 pleasurable food as well as a higher avoidance of aversive tastes, promoting the
99 overconsumption of palatable energy-dense products and the rejection of unpalatable healthy
100 bitter substances. Although the results are mixed, the view that BMI is increased by a heightened
101 liking for highly palatable foods has received support from several sources of evidence,
102 including data from longitudinal (e.g., with the obesity-prone Pima Indian population; Salbe,
103 DelParigi, Pratley, Drewnowski & Tataranni, 2004) and cross-sectional studies (e.g., with the
104 distribution of BMIs among the high-fat phenotypes; Blundell et al., 2005). Particularly
105 interesting are the studies on the relationship between sensitivity to food reward and BMI (e.g.,
106 Davis & Fox, 2008; Franken & Muris, 2005). For instance, Davis, Strachan and Berkson (2004)
107 pointed out that overweight women were significantly more sensitive to the hedonic reward of
108 food, when comparing the self-reported scores on the *Physical Anhedonia Scale* with those of
109 their normal weight counterparts. Franken and Muris (2005) also found that reward sensitivity,
110 as indexed by *Sensitivity to Punishment and Sensitivity to Reward Questionnaire*, was positively
111 associated with BMI in young women.

112 Regarding the assumption that body mass is affected by variations in reactivity to
113 unpleasant tastes, to date no study has specifically investigated the relation between negative
114 affective (aversive) responses to bitter-tasting foods and body weight. The available studies
115 which have explored weight differences as a function of taste are based on sensory (e.g.,

116 threshold or intensity; for review, cf. Donaldson, Bennett, Baic & Melichar, 2009) but not
117 affective variations. Although this line of evidence does not directly address our question, results
118 seem to point out that the perception of taste intensity of bitter compounds (e.g., 6-n-
119 propylthiouracil [PROP]) could ultimately impact body weight (e.g., Lumeng, Cardinal, Sitto &
120 Kannan, 2008; Tepper & Ullrich, 2002). These studies suggest that greater responsiveness to the
121 bitter taste of PROP is positively associated with a higher BMI in children, but this relationship
122 is negative in adults. Unfortunately, it is not obvious from the gustatory perception data how the
123 individual differences in aversive reactions may influence the body weight status, especially
124 when sensory versus affective aspects of taste stimuli have been separated via physiological,
125 psychological and pharmacological manipulations in animals and humans (Berridge, 2000; e.g.,
126 keeping the sensory properties of a taste unchanged, while altering its pleasantness). Therefore,
127 the purpose of the present experiment was to compare the aversive responses to bitter-tasting
128 stimuli, measured by subjective ratings and behavioural observations in a taste reactivity
129 paradigm, between two healthy adult groups of varying BMI. Taking into account the evidence
130 for a non-linear relationship between sensitivity to reward and BMI, indicating a positive
131 relationship only in the normal and overweight range of BMI (Davis & Fox, 2008), the present
132 study was limited to the BMI range of 17.7-29.9. We anticipated that individuals at risk of
133 becoming overweight (BMI \approx 25) would be more responsive to the unpleasant properties of food
134 than those with a low body weight (BMI \approx 19). That is, high BMI should show lower scores on
135 hedonic ratings and higher intensity of disgust facial expressions compared to low BMI, even
136 after controlling the differences in taste responsiveness.

137 In view of the importance of providing a relatively pure indication of affect (isolated
138 from the sensory and motivational properties of tastes), facial expressions were used here (cf.

139 Berridge, 2000). This way of assessing responses to food, beyond self-report measures alone,
140 was hoped to obtain a more exact evaluation of the relationship between the aversive reactions
141 and BMI, given that objective measures of liking reactions may sometimes provide more direct
142 access to hedonic systems than subjective reports (Berridge, Robinson & Aldridge, 2009). In
143 addition, it should be noted that many studies investigating taste preferences have found no
144 hedonic differences as a function of body weight (for review, cf. Bartoshuk, Duffy, Hayes,
145 Moskowitz & Snyder, 2006), the methods used to compare sensory and affective experiences
146 across groups being one possible explanation for these conflicting results. Concretely,
147 psychophysical errors derived from subjective measures (e.g., visual analogue or category scales)
148 have been suggested as a factor masking the relationship between orohedonic response and
149 obesity (Bartoshuk et al., 2006). Moreover, unlike facial patterns, self-ratings might not represent
150 accurate measures of pleasure/displeasure, because they may often conflate affective and
151 motivational (i.e., desire to eat) components of food and be too overlaid with cognitions to pick
152 up underlying core differences in food liking (Mela, 2001). In this sense, the present study
153 additionally sought to extend prior findings (e.g., Danner, Sidorkina, Joechl & Duerrschmid,
154 2013) on the contribution of facial expressions to sensory evaluation and affective testing of
155 bitter food; as well as explore the validity of hedonic self-report measures as assessment
156 instruments of the affective experience when they are employed with bitter tastes.

157

158 **Materials and Method**

159 **Participants**

160 **Forty healthy adults (28 females, 12 males) from the Faculty of Agrarian Sciences**
161 **(Pontificia Catholic University of Argentina, Argentina) were selected from a pool of people.**

162 Their ages ranged from 18 to 46 years ($M = 24.2$, $SD = 5.8$). Participants were asked to report
163 their height and weight. On the basis of their BMI, two groups were formed: low BMI,
164 consisting of lean subjects ($BMI < 20$; $n = 20$); and high BMI, encompassing participants that
165 were at risk of becoming overweight and overweight ($BMI > 23$; $n = 20$). The BMI values of 20
166 and 23 corresponded to percentile 40 and 60 respectively of the reference sample and were
167 deliberately selected in these ranges in order to establish a clear separation between BMI groups.
168 The mean BMI was 19.1 for low BMI ($SD = 0.7$) and 25.2 for high BMI participants ($SD = 1.8$),
169 being statistically different ($p < .05$). Exclusion criteria were aversions, smoking (more than 5
170 cigarettes per week; Sato, Endo & Tomita, 2002), illnesses, a history of eating disorders, diabetes
171 and allergy for the foods offered. Specially, participants who described themselves as being on
172 weight-loss diets or actively losing weight were excluded; this factor might be associated to bias
173 in reporting of sensory and affective perceptions of stimuli or influence the relationship between
174 bitter responsiveness and body weight (Tepper & Ullrich, 2002). Subjects were contacted by e-
175 mail and asked to participate in a research study investigating preferences for bitter foods. The
176 experiment was approved by the Ethics Committee of the Pontificia Catholic University of
177 Argentina. Participants were informed about the purpose of the study and that the experimental
178 procedure would be video recorded. All subjects gave their written consent, including agreement
179 to be recorded on video, and participated voluntarily.

180

181 Food solutions

182 Subjects received solutions of liquor chocolate (Natural Cocoa Liquor Refined NA760,
183 Cargill Agricola S.A., Brazil) and grapefruit juice, which were selected by their different bitter
184 compounds. The energy density was 28.9 and 0.4 kcal/g for the chocolate and the grapefruit,

185 respectively. The chocolate drink presented a high aromatic intensity at tasting temperature
186 (55°C; data not shown), a strong bitter taste and a high viscosity (viscosity > 1000). The
187 grapefruit juice exhibited a more neutral sensory profile (aroma and bitter taste; viscosity < 10)
188 at tasting temperature (20°C), which was prepared from natural pink grapefruit obtained from a
189 local store using an electric citrus juicer. Viscosity was measured by means of a rotational
190 viscometer (Brookfield DV-LVT; Brookfield Engineering Laboratories, Inc., Middleboro, MA,
191 USA) using the UL/Y adapter with S-00 spindle (chocolate) and S-38 spindle (grapefruit). The
192 sample chamber was placed in a water jacket connected to a bath (TC-502 Brookfield) to
193 perform the determinations at tasting temperature. PH values were 6.0 for the chocolate and 3.1
194 for the grapefruit. The pH was measured using a pH-meter (HANNA-pH 210, Germany), except
195 for the chocolate (determined by method IOCCC, 9/1972, in 10% solution; Gerken's Cacao,
196 Brazil). No sugar or sweeteners were added to the solutions.

197

198 Dependent variables

199 *Eating behaviour questionnaires and caloric intake assessment*

200 Preference and consumption of bitter substances were measured with a food preference
201 questionnaire (FPQ; with Cronbach's alpha (α) of .88), a food frequency questionnaire (FFQ; α =
202 .51) and a reduced version of the Spanish translation of Diet History Questionnaire (DHQ,
203 National Cancer Institute; α = .83). Although these instruments might not include all possible
204 dietary sources of bitter substances, they were meant to cover most bitter items in the
205 Argentinian diet. Factors that are thought to influence people's dietary choices were examined
206 with a version of the Food Choice Questionnaire in Spanish population (FCQ-SP; Jáuregui-
207 Lobera & Bolaños Ríos, 2011; α = .88). The size and the nature of the last meal before each

208 testing session were measured with a food record to obtain the amount of calories consumed.
209 Caloric intake was calculated by consultation with the USDA National Nutrient Database for
210 Standard Reference, Release 25 (December, 2011).

211

212 *Bitter taste responsiveness and time-intensity measurements*

213 To determine how responsive the subjects were to the taste of PROP (Sigma Chemical
214 Company, St Louis, USA), three concentrations were used: 0.010, 0.032 and 0.600 mmol/L
215 (belonging to the regular PROP series for taste detection thresholds; e.g., Drewnowski,
216 Henderson & Shore, 1997a). All solutions were prepared in distilled water \geq 1 day before testing.
217 The perceived bitter sensations of the PROP solutions over time (recorded every 0.35 s) were
218 characterized using a computerized time-intensity (T-I) software program by moving a cursor
219 along a 500-pixel line that represented a 20 cm unstructured line scale anchored at both extremes
220 0 - 100 on the monitor (cf. Galmarini, Zamora & Chirife, 2009), after receiving verbal
221 instructions: 0 = not at all bitter and 100 = extremely bitter. The software provided the T-I curve
222 as well as the parameters that described it: maximum intensity reached (I_{max} ; 0-100), time
223 elapsed to maximum intensity (T_{max} ; in seconds), area under curve (AUC; representing the
224 overall bitterness perception of the whole stimuli perceived over the total time of recording) and
225 rate of increase of bitter (Rinc). The question asked was as follows: “How bitter do you find this
226 solution now in your mouth?” The subjects also rated the bitterness of the two food solution. The
227 rating method, question and software were the same as those for the PROP solutions.

228

229 *Self-report measures of food attributes*

230 Hedonic value (i.e., subjective pleasure) was rated on a 9-point hedonic scale with
231 opposing extremes of liking from 1 (dislike extremely) to 9 (like extremely), and with a neutral
232 point at 5 (neither like nor dislike), by answering the following question: “How pleasant is this
233 food now in your mouth?” In addition, given the importance of incentive value (i.e., desire to
234 eat) and familiarity (i.e., knowledge of and experience with the taste of stimuli) to the people’s
235 daily food and beverage choices, these attributes were examined as well. To account for this,
236 subjects rated the incentive value and familiarity of each food stimulus using 9-point category
237 scales, where 1 was “not at all” and 9 was “extremely”. The questions were as follows: “How
238 much do you want to eat this food?” and “How familiar are you with this food?” respectively.

239

240 *Facial expressions to foods*

241 A behavioural measure of taste-elicited affective reactions was provided by the analysis
242 of the facial patterns. Facial reactions were videotaped with a digital video camera (JVC GZ-
243 MS150SU), which was located in a hole of the booth wall, directly above the computer screen
244 and in front of the subject at a distance of 1.5 m. The illumination of the participant’s face was
245 optimized by using daylight lamps (6500 k), in addition to the ceiling lights. The participants sat
246 on a wooden school chair and were kept from turning their head by answering the questions and
247 rating the bitterness of the food solution on a computer screen. The cups used were transparent
248 so that they did not interfere with the recording. In addition, the camera had face detection
249 technology which identified people's faces following their movements and made adjustments to
250 achieve the optimum focus, exposure and white balance. The experimenter followed the facial
251 expressions in real time watching the camera screen without being seen by the subjects. The
252 video files were run through the FaceReader 4 software (Noldus Information Technology,

253 Wageningen, The Netherlands) and processed frame-by-frame at 50 Hz, scaling the facial
254 expressions from 0 (not present at all) to 1 (maximum intensity of the fitted model).
255 Approximately 85% of the video frames were analyzable by the software. This software
256 distinguished between seven facial reaction patterns or expressions (happy, sad, angry, surprised,
257 scared, disgusted and neutral) using the Active Appearance Modelling (cf. van Kuilenburg,
258 Wiering & den Uyl, 2005). In order to standardize the measurements and to compare the facial
259 expressions (of different duration and latency), the ten seconds before and after tasting the food
260 stimuli were taken for analysis. The facial analysis before tasting served as baseline. The
261 intensity of each facial expression was calculated by subtracting the average intensity of the
262 baseline period from the average intensity after tasting.

263

264 Procedure

265 Before starting the experimental session, participants completed the questionnaires and
266 were also presented with the PROP solutions in 10-ml plastic cups and asked to rate the bitter
267 intensity, rinsing between samples. PROP solutions were presented from lower to higher
268 intensity in order that the receptors were not saturated. The experimental session took place in an
269 individual booth kept at 22 ± 2 °C. The booth was equipped with a computer (Samsung
270 NP300E4AH) and software for the presentation of the instructions and recording subjects'
271 responses. The session lasted about 25 minutes and was subdivided into (1) a record of food
272 eaten for the evaluation of caloric intake; (2) presentation of neutral pictures (from the Geneva
273 Affective Picture Database; Dan-Glauser & Scherer, 2011) for a total time of 10 min in order to
274 minimize differences in motivational state; (3) delivery of food samples and tasting; and (4)
275 rating of hedonic, incentive, familiarity and bitter dimensions of each food sample. During the

276 tasting stage, subjects received 5 ml of a grapefruit juice drink and a bitter chocolate drink in 10-
277 ml plastic cups in counterbalanced order. There was a period of 120 seconds between the
278 presentation of one sample and the presentation of the next sample. They took each sample into
279 the mouth and tasted it using whole mouth tasting, but were instructed not to swallow the
280 solutions (sip-and-spit technique). Subjects were told to rinse with mineral water (presented in
281 120-ml thermal cups) before each food sample. PROP solutions and mineral water were offered
282 at room temperature. The experimenter was not visible to the subjects.

283

284 Data Analysis

285 Comparisons between BMI conditions for the eating behaviour questionnaires (FPQ,
286 FFQ, DHQ, FCQ-SP) and caloric intake were tested using independent samples t-tests. T-I
287 curves were first analyzed visually in order to remove the irrelevant points on the graph caused
288 by the use of the mouse. These points corresponded to small regions of the curves with abrupt
289 changes to very low or very high value, and were replaced by an average of the preceding and
290 following points (Lallemand, Giboreau, Rytz & Colas, 1999; Le Berre, Boucon, Knoop &
291 Dijksterhuis, 2013). The data for the T-I curve for each solution was separately averaged by low
292 and high BMI. Differences in T-I parameters for PROP and food solutions (Imax, Tmax, AUC,
293 Rinc), self-ratings of food attributes (hedonic, incentive, familiarity) and intensity of facial
294 expressions (angry, disgusted, happy, neutral, sad, scared, surprised) were analyzed using a two-
295 way repeated measures ANOVAs. Independent variables included BMI (Low vs. High) and
296 Food (Chocolate vs. Grapefruit) or PROP (0.010 vs. 0.032 vs. 0.600 mmol/L). Greenhouse-
297 Geisser correction was used in case of violation of the assumption of Sphericity. All pairwise
298 comparisons of individual means for effects found to be significant in the ANOVA were carried

299 out by using Tukey's multiple comparison tests to control for Type I error. Pearson's or
300 Spearman's correlations were used, when appropriated, to assess associations among taste
301 responsiveness, hedonic ratings or facial expressions and BMI status and between hedonic
302 ratings and facial expressions. Regression models were calculated to predict BMI using intensity
303 of the disgust facial expression and to predict the facial expression intensity using hedonic self-
304 report.

305

306 **Results**

307 Eating behaviour questionnaires and caloric intake

308 The mean values of the eating behaviour questionnaires and the caloric intake are shown
309 in Table 1. The total scores from the FPQ, FFQ, DHQ and total caloric consumption did not
310 differ between the BMI conditions (largest $t [38] = 1.24, p = .22$). Regarding FCQ-SP, analyses
311 indicated significant differences on one of the factors ($t [38] = 2.33, p < .05$). Specifically, the
312 low BMI rated sensory appeal (e.g., taste, smell or appearance) as more important in their food
313 choices than did the high BMI group.

314 (Table 1 about here)

315 Bitter taste responsiveness and time-intensity measurements

316 The sum of bitter ratings for the three PROP solutions was used to assess the PROP taster
317 status (Kaminski, Henderson & Drewnowski, 2000; Ly & Drewnowski, 2001). The participants
318 whose summed responses were 59 or less (i.e., 10th percentile or less) were classified as non-
319 tasters, while those with summed ratings in excess of 59 were classified as tasters. Only four
320 participants were PROP non-taster, two with high BMI and two with low BMI. As can be seen,
321 the average bitterness T-I curves of the three PROP solutions for the two BMI conditions over

322 the time course of 20 sec are shown in Figure 1. The PROP concentrations were differently
323 perceived by the subjects according to I_{max} ($F [2, 68] = 36.83, p < .001, \eta^2 = .52$), T_{max} ($F [1.5,$
324 $52.7] = 6.67, p < .01, \eta^2 = .16$) and R_{inc} ($F [1.7, 57.7] = 19.25, p < .001, \eta^2 = .36$). There were
325 no significant main effects of BMI or interactions between BMI and PROP concentration (largest
326 $F [1.5, 52.7] = 3.33, p = .085$). Post hoc comparisons showed that the 0.600 mmol/L presented
327 lower T_{max} and higher I_{max} and R_{inc} values compared to 0.010 mmol/L ($ps < .05$); and higher
328 I_{max} and R_{inc} than 0.032 mmol/L ($ps < .05$). On the other hand, the 0.032 mmol/L showed
329 lower T_{max} and higher R_{inc} values than 0.010 mmol/L ($ps < .05$). In contrast, a significant
330 interaction between BMI and PROP concentration on AUC was found ($F [1.8, 59.9] = 6.91, p <$
331 $.001, \eta^2 = .17$). This interaction revealed that only the lowest PROP concentration was perceived
332 differently by the BMI conditions, for which the subjects with high BMI perceived 0.010
333 mmol/L to be bitterer than those with low BMI ($t [28.66] = -2.78, p < .01$). On the other hand,
334 both BMI groups showed differences in the AUC among PROP stimuli (smallest $F [2, 34] =$
335 $5.20, p < .05, \eta^2 = .23$), with higher values in the highest PROP compared with the intermediate
336 PROP concentration ($p < .05$).

337 Regarding food solutions, the average bitterness T-I curves of the chocolate and
338 grapefruit for the two BMI conditions over the time course of 20 sec are shown in Figure 2. A
339 visual inspection of curves showed that the solutions were perceived differently according to
340 BMI. Concretely, all subjects with low BMI started the curves with zero or very close to zero
341 values for the grapefruit and chocolate, whereas approximately 32% of high BMI subjects
342 presented values higher than 30 for the chocolate. However, the statistical analyses revealed no
343 effect of BMI or their interaction with Food (largest $F [1, 37] = 3.23, p = .085$) on I_{max} , T_{max} ,
344 AUC and R_{inc} . There was a Food effect on I_{max} ($F [1, 37] = 114.06, p < .001, \eta^2 = .75$), T_{max}

345 (F [1, 37] = 156.47, $p < .001$, $\eta^2 = .81$), AUC (F [1, 37] = 77.23, $p < .001$, $\eta^2 = .68$) and Rinc (F
346 [1, 37] = 12.25, $p < .01$, $\eta^2 = .25$), showing higher values on I_{max}, AUC and Rinc for chocolate
347 compared with grapefruit. T_{max} showed a lower value for chocolate compared with grapefruit.

348 Additionally, the question of whether BMI could be related to bitter taste responsiveness
349 was examined. Results of the analysis showed that BMI was not correlated with the bitter taste
350 perception of PROP concentrations or food solutions determined by the I_{max}, T_{max}, AUC and
351 Rinc parameters ($p_s \geq .13$).

352

353 (Figure 1 about here)

354 (Figure 2 about here)

355

356 Self-report measures of food attributes

357 Ratings on the hedonic value varied between the food solutions (F [1, 38] = 69.7, $p <$
358 $.001$, $\eta^2 = .65$), reflecting higher pleasure ratings for the grapefruit (rating = 6.3) than for the
359 chocolate (rating = 2.8; which perceived as strongly unpleasant). Although foods' hedonic scores
360 of high BMI were smaller than those of low BMI (4.1 vs. 4.9) and inspection of data revealed
361 that 70% of the low BMI compared with scarcely 40% of the high BMI subjects evaluated
362 grapefruit with values 7-9 on the hedonic scale or 65% of the low BMI compared with 80% of
363 the high BMI participants evaluated chocolate with values 1-3, there was no significant effect of
364 BMI or their interaction with Food (largest F [1, 38] = 3.00, $p = .91$). Additionally, the question
365 of whether BMI could be related to hedonic ratings was examined. Results of the analysis
366 showed that BMI was not associated with hedonic scores for chocolate ($r = -.077$, $p = .64$) or
367 grapefruit ($r = -.157$, $p = .33$).

368 Incentive ratings varied significantly between foods ($F [1, 38] = 72.59, p < .001, \eta^2 = .66$)
369 and BMI groups ($F [1, 38] = 7.83, p < 0.01, \eta^2 = .17$), but there was no a significant BMI x Food
370 interaction ($F [1, 38] = 1.96, p = .17$). These effects revealed that low BMI wanted to drink more
371 bitter foods (rating = 4.9) than high BMI (rating = 3.5), and that the desire to eat was higher for
372 the grapefruit (rating = 5.9) than chocolate (rating = 2.5). Familiarity varied between the food
373 solutions ($F [1, 38] = 11.78, p = .001, \eta^2 = .24$). There were no significant main effect of BMI or
374 their interaction with Food (largest $F [1, 38] = 2.56, p = .12$), indicating that the grapefruit
375 solution was rated as more familiar that the chocolate. Familiarity ratings for the both food
376 solutions were in the moderate-to-high range (ratings > 7.5).

377

378 Facial expressions to foods

379 On average, the times to reach the maximum intensity of negative emotions after tasting
380 the food stimuli were 1.67 seconds for ‘disgusted’, 2.43 seconds for ‘sad’ and 3.28 seconds for
381 ‘angry’. The mean changes from baseline in intensity of facial expressions for BMI conditions
382 and food solutions are shown in Figure 3 and Figure 4, respectively. Analyses on taste-elicited
383 facial patterns revealed a main effect of Food concerning the facial expression “disgusted” ($F [1,$
384 $38] = 14.47, p < .01, \eta^2 = .28$), “angry” ($F [1, 38] = 5.30, p < .05, \eta^2 = .12$) and “neutral” ($F [1,$
385 $38] = 6.24, p < .05, \eta^2 = .14$); and a main effect of BMI concerning the expressions “disgusted”
386 ($F [1, 38] = 4.90, p = .053, \eta^2 = .11$) and “neutral” ($F [1, 38] = 4.03, p = .052, \eta^2 = .10$). No
387 significant BMI x Food interactions were observed (largest $F [1, 38] = 3.15, p = .09$). Regarding
388 the main effect of Food, the chocolate produced an increase in expressions “disgusted” and
389 “angry”, and a decrease in “neutral” compared to the grapefruit.

390 Regarding the BMI effect, the results showed that the bitter foods elicited significantly
391 more intense reactions of “disgusted” and strongest reduction of “neutral” in high BMI than in
392 low BMI. As an additional check for the possibility that sensory but not affective responsiveness
393 to bitter taste might have contributed to the observed between-group differences on facial
394 expressions, two-way analyses of covariance (BMI x Food) were performed on both “disgusted”
395 and “neutral” expressions using as covariate the overall bitterness perception (AUC) for the
396 0.010 mmol/L PROP concentration (that was significantly different across BMI groups in
397 previous analyses). The results showed that the ANCOVA and ANOVA produced similar
398 conclusions –with a significant effect of BMI on the expressions “disgusted” ($F [1, 35] = 3.41, p$
399 $= .07, \eta^2 = .09$) and “neutral” ($F [1, 35] = 4.54, p < .05, \eta^2 = .11$). Additionally, the relationship
400 between BMI and disgust facial expression was examined, showing a significant positive
401 correlation for chocolate ($r = .304, p < .05$), though the percent of variance explained was very
402 low, with a coefficient of determination (R^2) of .092 ($F [1, 39] = 3.87, p = .056$). There was no
403 significant linear correlation between BMI and “disgusted” for grapefruit or between BMI and
404 the neutral facial expression (for chocolate or grapefruit) and BMI ($ps > .2$).

405 (Figure 3 about here)

406 (Figure 4 about here)

407

408 Association between self-reported hedonic value and facial expressions

409 Correlations of hedonic ratings with the different facial expressions indicated that the
410 hedonic scores were negatively associated with the intensity of disgust facial expression ($r = -$
411 $.463, p < .01$) for chocolate. Results for grapefruit showed a negative relationship between the
412 hedonic scores and the disgust facial expression ($r = -.323, p < .05$) and a positively correlation

413 related to the neutral facial reactions ($r = .405$, $p < .01$). To further examine the significant
414 associations, linear regressions analyses were performed (on primary data, but means were
415 plotted on the graphs for each point of the hedonic scale for clarity; Figure 5). The results
416 showed low R^2 values; $R^2 = .133$ for “disgusted” to chocolate ($F [1, 39] = 5.83$, $p < .05$); $R^2 =$
417 $.055$ for “disgusted” to grapefruit ($F [1, 39] = 2.22$, $p = .14$); and $R^2 = .164$ for “neutral” to
418 grapefruit ($F [1, 39] = 7.43$, $p < .05$). It should be noted that the point 5 of the hedonic scale,
419 which is the neutral value to pass from liking to disliking, matched with the inflection point
420 changing positive to negative facial expression values.

421 (Figure 5 about here)

422

423 **Discussion**

424 According to recent hedonic eating theories of obesity, we hypothesized that overweight
425 individuals would be more reactive to unpleasant tasting food than lean people. Facial expression
426 results were consistent with our hypothesis, showing that bitter-tasting stimuli (grapefruit and
427 chocolate) elicited significantly more intense disgust reactions and less neutral state reactions in
428 the high BMI than in the low BMI condition. Furthermore, the disgust intensity response to
429 strong bitter (chocolate) was positively related to BMI, though the percent of variance explained
430 was very low ($\approx 10\%$). To our knowledge, this is the first study which has revealed a link
431 between aversive patterns of taste reactivity and weight status. Partial support was also obtained
432 by hedonic ratings, which showed a trend toward lower preference scores for bitter foods in high
433 BMI; but failed to provide significant differences between BMI groups. Although the reasons for
434 this difference are unclear, some possibilities may be suggested (see below).

435 One interpretation is that these different aversive reactions were related to an enhanced
436 perception of bitter intensity in the high BMI compared with the low BMI participants. If we

437 consider that overweight individuals had a heightened acuity for bitterness, it should not be
438 surprising that they reflected increased dislikes for bitter-tasting foods (Drewnowski, Henderson
439 & Shore, 1997b) and therefore a higher facial reactivity compared with their normal weight
440 counterparts. It is well established that functional or structural differences (e.g., number of taste
441 buds and density of taste buds per papilla) in the gustatory system may affect taste preferences
442 and, ultimately, body weight (cf. Donaldson, Bennett, Baic & Melichar, 2009). For instance,
443 higher BMI and higher propensity to be overweight was found in individuals with a genetically
444 mediated ability to taste PROP (tasters) compared with nontasters (Fischer, Griffin, England &
445 Garn, 1961; Lumeng, Cardinal, Sitto & Kannan, 2008; but see Keller, Steinmann & Nurse,
446 2002). This interpretation cannot be completely ruled out given the complexity of taste
447 perception; however, it seems unlikely in view of our sensory evaluation data using time-
448 intensity methodology. In fact, no effect of BMI status on sensory response to bitter food
449 samples was detected when the comparison was done in terms of I_{max} , T_{max} , AUC and R_{inc} .
450 This lack of sensory difference for bitter compounds between overweight and normal weight
451 subjects is not new, and it has been reported in adults and children (e.g., Drewnowski, Henderson
452 & Cockroft, 2007; Goldstein, Daun & Tepper, 2007; Nasser, 2001). Responsiveness to PROP
453 concentrations also did not differ between the BMI groups, except for a slight variation in the
454 AUC of the low PROP concentration (0.010 mmol/L) which was higher in high BMI. Even so,
455 after treating this parameter as a confounding factor and covariant, the observed differences in
456 disgust and neutral facial reactions between the groups remained at least marginally significant.
457 Therefore, the greater reactivity to affective component of taste in high BMI could not be
458 attributed to differences in bitterness intensity alone.

459 That the high BMI participants expressed an enhanced sensitivity to the affective
460 properties of taste compared with the low BMI participants is an alternative interpretation of the
461 current data. Thus, pleasantness of taste could be considered a mediator variable of the
462 relationship among the bitter taste perception and food selection, dietary patterns, and ultimately
463 body weight. As pointed by Tepper, White, Koelliker, Lanzara, d'Adamo & Gasparini (2009),
464 variations in bitterness perception may not be sufficient to alter food acceptability, since
465 bitterness represents only one facet of the complex sensory profile of a food. It is also important
466 to consider the role of other factors, such as hedonic processes. Our findings supported this
467 explanation: overweight participants experienced a similar bitterness perception to those of lean
468 participants; further, the overweight people disliked bitterness more. A number of observations
469 seem to indicate a heightened affective response to bitter compounds in overweight individuals.
470 For instance, Bartoshuk et al. (2006) found that the maximum disliking for the food/beverages
471 (including dark chocolate and grapefruit juice) rose with BMI. Interestingly, a stronger hedonic
472 response to palatable food has recently been implicated as a factor for weight gain (e.g., Salbe et
473 al., 2004). Since reward and aversion might be mediated by overlapping neural systems and
474 constitute an affective continuum (Umberg & Pothos, 2011), it is possible that liking of
475 sweetness and disliking of bitterness express the same psychobiological trait in the risk for
476 weight gain and overeating. Confirmation of this possibility might have considerable
477 implications for nutritional, health and weight status.

478 The tendency to avoid bitter vegetables and fruits, which contain water, dietary fibre
479 (Howarth, Saltzman & Roberts, 2001), human health-bioactive compounds (Drewnowski &
480 Gomez-Carneros, 2000) and have a low fat content, could reduce satiety and increase energy
481 (palatable) intake, body weight (cf. Rolls, Ello-Martin & Tohill, 2004) and the risk of some of

482 the diet-related chronic diseases (Slavin & Lloyd, 2012). This tendency to avoid bitter foods was
483 confirmed by our results; ratings of desire to eat revealed that high BMI participants wanted less
484 to drink bitter stimuli compared with the low BMI group. Comparing the responses given with
485 regards to the motivational factors that underlie the food choices (FCQ-SP scales), the sensory
486 appeal was rated as more important by the low BMI than high BMI group. No differences related
487 to mood, health and natural content, weight control, convenience, familiarity and price were
488 observed. In contrast, reported total dietary intake of vegetables and fruits from food frequency
489 and diet history questionnaires did not support a reduced consumption of these substances among
490 overweight subjects. Bitter foods included grapefruit juice, spinach, kale, coleslaw, broccoli,
491 cauliflower, brussels sprouts, as well as beer, wine, tea and coffee. A more accurate assessment
492 of the relationship between body weight and bitter food consumption would require a study of
493 the effects of particular fruit/vegetable intake and explore how these foods are eaten by
494 separating them by preparation: fresh, baked, or fried; in mixtures; or with other
495 accompaniments (Lin & Morrison, 2002).

496 The use of taste reactivity also provided insights of interest for the sensory and consumer
497 evaluation. Although facial reactivity has been used in infants and adults to study the hedonic
498 function of taste (e.g., de Wijk, Kooijman, Verhoeven, Holthuyzen & de Graaf, 2012; Steiner,
499 Glaser, Hawilo & Berridge, 2001), it has not previously been applied to overweight adults.
500 Similar to other studies (e.g., Danner et al., 2013), FaceReader technology was a sufficiently
501 suitable and accurate method, in our case for differentiating between the two bitter foods:
502 stimulus perceived as more bitter (i.e., chocolate, $I_{max} = 69.4$ vs. grapefruit, $I_{max} = 34.3$) was
503 more strongly disliked (on the basis of the intensity of the elicited disgust and angry expressions)
504 in both BMI groups. Moreover, although the study was not addressed to the question of whether

505 the relationship between aversive taste sensitivity and BMI could be mediated by the energy
506 density of foods, it should be noted that the patterns of aversive reactions were not affected by
507 the energy content (chocolate = 28.89 vs. grapefruit = 0.39 kcal/g). As noted in Epstein,
508 Truesdale, Wojcik, Paluch and Raynor (2003), hedonics and the reinforcing value of high-calorie
509 foods measured by subjective ratings and behavioural observations in a taste reactivity paradigm
510 seem to be separate processes in humans. Still, the influence of energy density on aversive taste
511 processing currently remains unexplored to the best of our knowledge.

512 Regarding the validity of hedonic self-reports, we explored whether these reports
513 reflected a genuine affective response to aversive value of foods, rather than a cognitive or
514 motivationally determined response. As an affective measure, hedonic self-reports should be
515 highly related to facial patterns (a well-established measure of the hedonic evaluation of taste
516 stimuli; Berridge, 2000). Investigations have demonstrated the reliability and the validity of the
517 nine-point hedonic scale in assessing product likes and dislikes (cf. Stone & Sidel, 2004).
518 However, moderate associations ($\approx -.40$) of subjects' facial expressions of disgust to bitter foods
519 with their hedonic ratings of these same solutions were found. This is a level generally
520 considered acceptable, though we cannot rule out the fact that the nine-point hedonic scale
521 measured other aspects, not only pleasure/displeasure, but also intensity of sensation, social
522 desirability or cognitions regarding bitter foods. It can be seen that this potential bias might have
523 blunted the differences on hedonic ratings between high and low BMI. Some other explanations
524 of the failure of hedonic self-reports to provide significant BMI group differences can be
525 suggested. For example that the relatively small sample size limited the ability to detect an
526 effect; that the 9-point hedonic scales provided invalid group comparisons for bitterness because
527 of psychophysical errors (assuming erroneously that intensity perception is the same for subjects

528 in different BMI groups; as pointed out by Bartoshuk et al., 2006, for sweet taste in the obese vs.
529 non-obese). Further studies are needed to confirm these possibilities. In addition, it is worth
530 considering that these potential sources of error were not sufficient to make BMI differences in
531 incentive motivation disappear, reporting that low BMI participants showed a stronger desire to
532 eat the bitter stimuli than high BMI participants. It would seem that the question “How much do
533 you want to eat this food?” is more sensitive than “How pleasant is this food now in your
534 mouth?” for measuring differences between conditions.

535 Several limitations of this study should also be discussed. First, our study tested bitter
536 perception with time-intensity methodology, a tool for fundamental research on bitterness (cf.
537 Cliff & Heyman, 1993). Because of the complexity of the measurements, participants should be
538 trained (Dijksterhuis & Piggott, 2000). However, the participants only had a relatively short
539 training in order to learn how to move the mouse on the scale on the screen. It might be asked,
540 would we have found more differences (and in which direction?) if both BMI groups had been
541 trained? Another detail to note was the beginning of the time-intensity curves, especially for the
542 chocolate, in which all low BMI subjects started the curves with zero values and, approximately,
543 32% of high BMI subjects started with values higher than 30. Would it be possible to consider
544 that high BMI subjects can perceive bitter tastes faster? This question requires further
545 investigation and another approach such as reaction time methodology could be used (Bonnet,
546 Zamora, Buratti & Guirao, 1999; Guirao & Zamora, 2000). Finally, as pointed by Danner et al.,
547 (2013), it is also important to recognize that motor artefacts caused by drinking could be
548 misinterpreted by the FaceReader as expression. In order to minimize artefacts, liquid samples
549 which need less processing in the mouth were used.

550 In summary, although BMI is a complex variable for which aversive reactions explain
551 only a small portion, hedonic (appetitive or aversive) over-responding may be one factor
552 contributing to the susceptibility to weight gain also through avoidance of health-promoting
553 food. Additional research is therefore needed to examine affective mechanisms that control
554 dietary selection and food consumption, given the increasing incidence of obesity.

555

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564

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728

729 **Table 1.** Scores of the eating behaviour questionnaires and caloric intake for the BMI groups.

Characteristic	Low BMI	High BMI
FPQ	108.3 ± 6.7	101.1 ± 6.4
FFQ	2.9 ± 0.5	3.8 ± 0.6
FCQ-SP: Health	16.4 ± 0.9	16.1 ± 1.7
FCQ-SP: Mood	11.7 ± 0.8	12.8 ± 0.9
FCQ-SP: Convenience	12.4 ± 0.8	10.8 ± 1.0
FCQ-SP: Sensory appeal	14.3 ± 0.3	12.8 ± 0.6*
FCQ-SP: Natural content	5.6 ± 0.4	6.3 ± 0.5
FCQ-SP: Price	7.0 ± 0.4	7.2 ± 0.5
FCQ-SP: Weight control	6.5 ± 0.5	6.3 ± 0.5
FCQ-SP: Familiarity	6.3 ± 0.4	5.7 ± 0.5
DHQ	48.0 ± 4.1	46.3 ± 4.8
Intake before testing session (Kcal)	470.4 ± 42.1	445.1 ± 57.9

730 Note. Values are means (± SEM). FPQ: food preference questionnaire; FFQ: food frequency
731 questionnaire; FCQ-SP: Food Choice Questionnaire in Spanish population; DHQ of Diet History
732 Questionnaire (Spanish translation). Kcal: kilocalories. *p < .05, significant differences for comparisons
733 between low and high BMI conditions.

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736 **Figure Captions**

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738 **Figure 1.** Time-intensity curves for average bitterness obtained of the three PROP solutions
739 (0.010, 0.032 and 0.60 mmol/L) for the two BMI conditions (low and high).

740 **Figure 2.** Time-intensity curves for average bitterness obtained of the chocolate and grapefruit
741 solutions for the two BMI conditions (low and high).

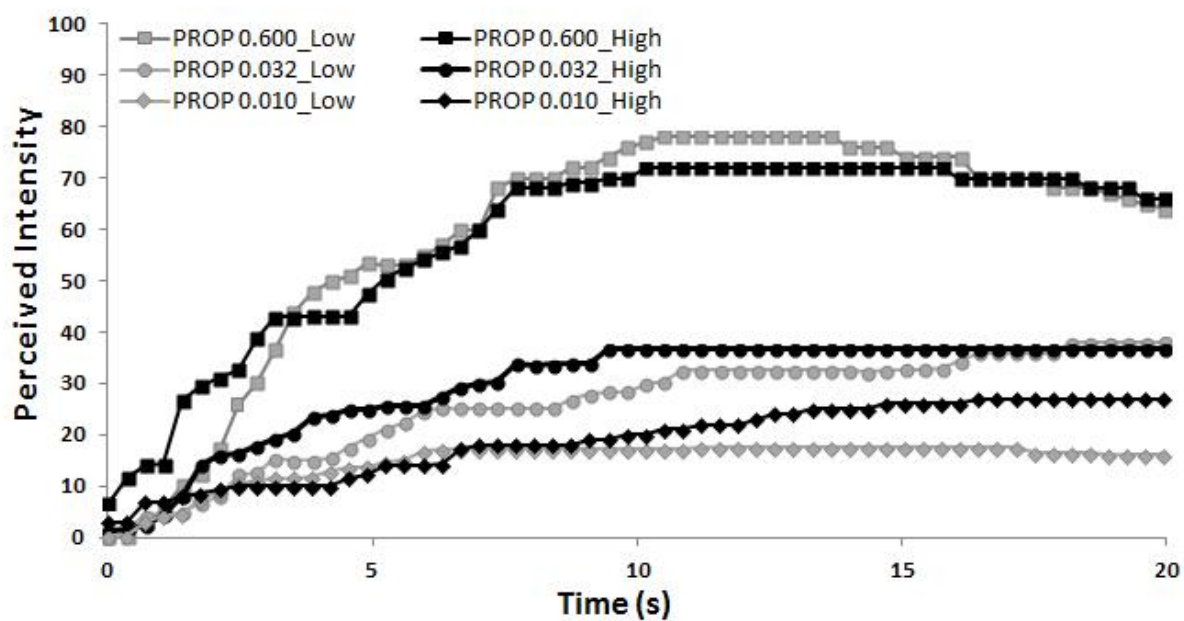
742 **Figure 3.** Changes from baseline in intensity of facial expressions for BMI conditions (low and
743 high). Bars express the mean changes from baseline (\pm SEM).

744 **Figure 4.** Changes from baseline in intensity of facial expressions for food solutions (chocolate
745 and grapefruit). Bars express the mean changes from baseline (\pm SEM).

746 **Figure 5.** Regression of facial expression intensity of disgusted (A: chocolate; B: grapefruit) or
747 neutral (C: grapefruit) against self-reported hedonic rating (1-dislike extremely to 9- like
748 extremely).

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751 **Figure 1**

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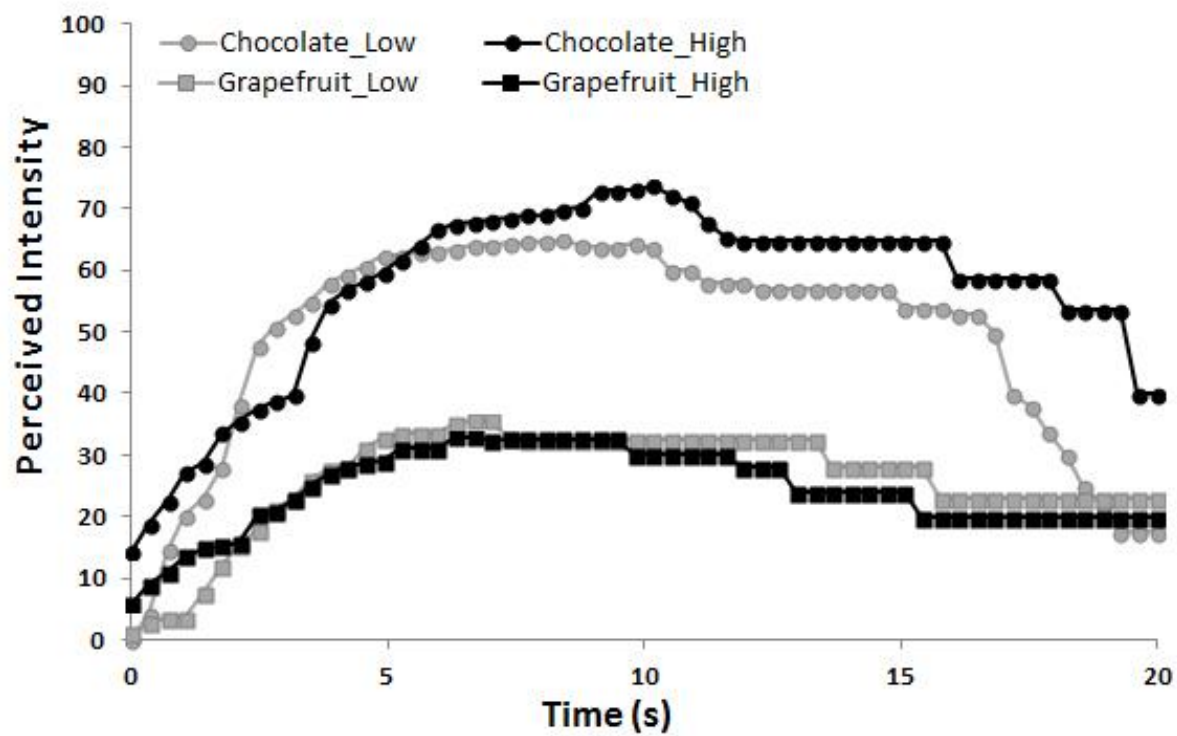
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758 **Figure 2**

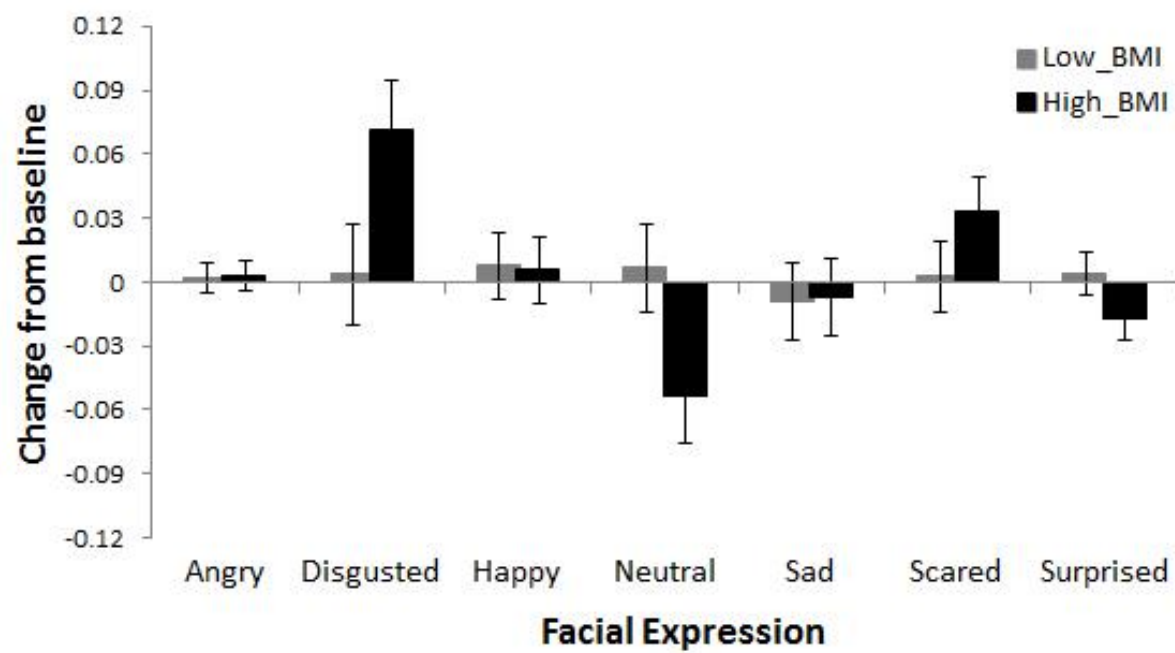
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764 **Figure 3**

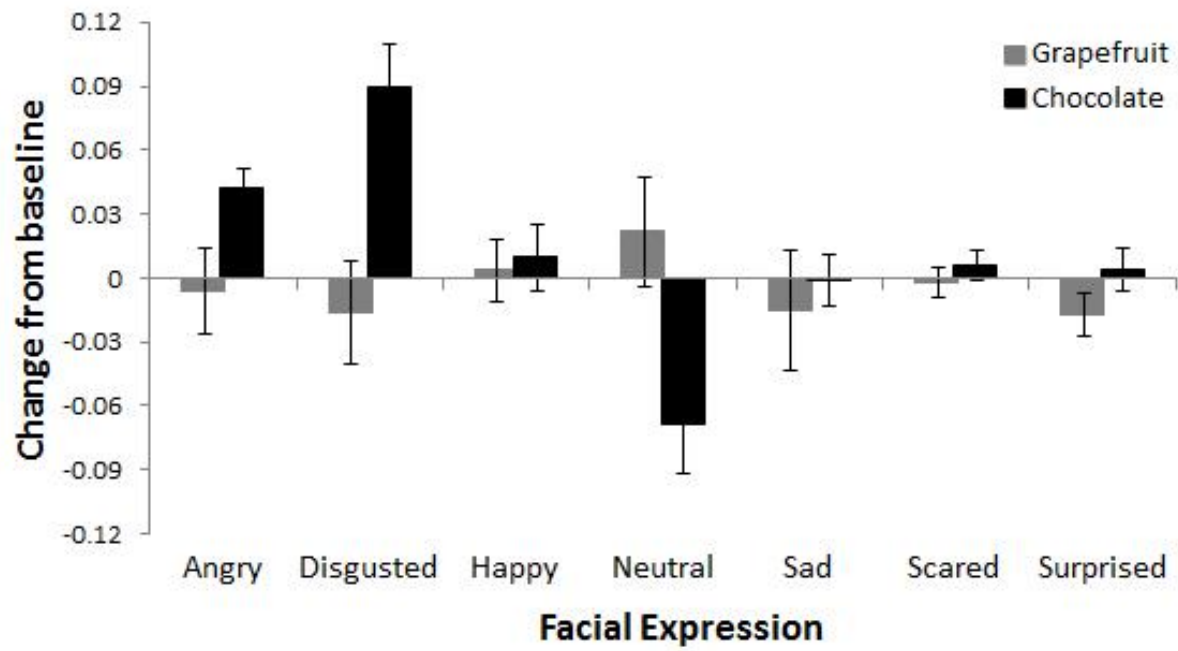
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770 **Figure 4**

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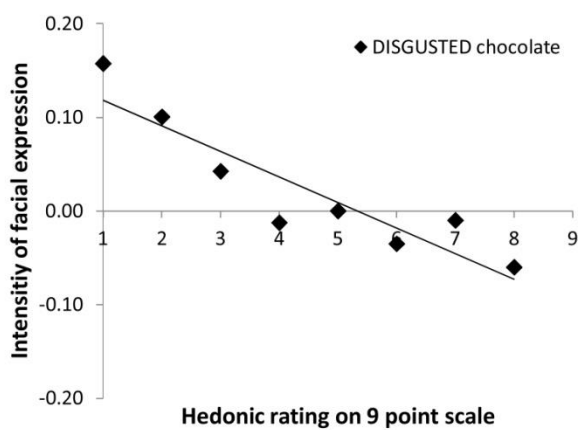
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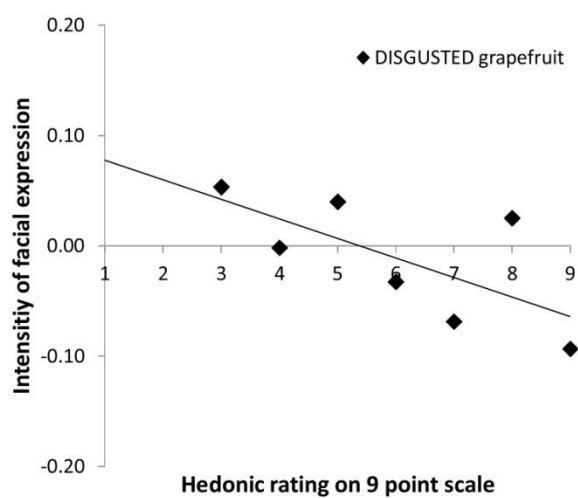
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775 **Figure 5**

A)



B)



C)

