Learning and Retention Time Effect on Memory for Sweet Taste in Children Laureati M^{*}, Pagliarini E

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5 Abstract

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This study investigated the effect of learning and retention time on memory for fruit purée varying 6 7 in sweetness among 214 children aged 8-10 yrs. During a first session, all children received a snack 8 including a target fruit purée. Half children tasted the snack without any mention to memory 9 (incidental group), whereas the other children (intentional group) were explicitly asked to taste and 10 remember it. During a second session, children of each learning group were divided in two groups, 11 which were tested for memory after respectively one day and one week. Children were confronted with a series of samples consisting of the same target previously tasted and variants of it slightly 12 13 modified in sweetness. Children performed also a hedonic and a perceptive test. Memory was better 14 under incidental rather than intentional conditions. Recognition was based more on the correct 15 rejection of the distractors rather than on the identification of the target. No clear evidence for a retention time effect on memory was found. The relationship between sweetness perception and 16 17 memory is discussed.

18 Keywords: Incidental learning; Intentional learning; Retention time; Food memory; Children

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19 **1. Introduction**

20 Over the last decade, a remarkable amount of studies have been performed in an attempt to 21 delineate the mechanisms involved in incidental learning and memory for food. If we consider the 22 way we learn, store and retrieve sensory food input, it is fairly evident that we rarely pay attention 23 to what we eat or drink, unless something differs from our expectations. Nevertheless, sensory 24 information is unconsciously retained by the brain and remains "hidden" until the time when a new 25 food is experienced (Köster, Prescott & Köster, 2004). At this moment, sensory memory becomes a 26 determinant factor in food choice, since it enables the comparison of sensory information with 27 stored information obtained in previous experiences with the same or a similar product, thus 28 influencing food sensory and hedonic perception through expectations generation and cognitive 29 associations expression. The resulting sensory image is added to memory and may in turn play a 30 role in subsequent food experiences (Sulmont-Rossé, Issanchou, & Köster, 2003).

Food learning is almost never intentional or explicit and memory for food is also to a very large extent implicit (Morin-Audebrand et al., 2012). There are very few examples of explicit learning related to food, one of these is when subjects make an effort to remember the food sensory properties in sensory tests but this situation is pretty rare in everyday life. Indeed, when eating or drinking, it is extremely unusual to consciously decide "I have to remember this food" (Issanchou, Valentin, Sulmont, Degel, & Köster, 2002).

37 Since the nature of food memory is basically implicit, an implicit recognition paradigm was 38 proposed and validated in order to investigate learning and memory for food in an ecologically 39 valid way (Mojet & Köster, 2002). This paradigm includes two phases: (1) an acquisition phase and 40 (2) a retrieval phase, which is carried out after a given retention interval. During the acquisition 41 phase, participants are incidentally exposed to a target food (*i.e.* the only food to be remembered 42 later) which is administrated in a natural eating situation. During the retrieval phase following the 43 retention interval, participants are unexpectedly asked to recognize the target food among a series of 44 samples slightly different in one or more sensory aspects (*i.e.* the distractors). This paradigm - 45 which differs from that used in almost all other (implicit and explicit) memory experiments in the 46 literature – focuses on the recognition of minor changes of a target food provided in a real eating 47 context. All other previous experiments have been directed to the identification of clearly different 48 stimuli presented out of their natural context which must be later identified among other clearly 49 distinct new stimuli.

50 Through the application of this paradigm, some food memory features have been delineated. First of 51 all, a number of studies (Mojet & Köster, 2002, 2005; Köster et al., 2004; Morin-Audebrand, 52 Laureati, Sulmont-Rossé, Issanchou, Köster & Mojet, 2009) have shown that memory for food 53 occurs, but it is extremely product-dependent. For example, sweetness might be the crucial feature 54 for the memorization of a custard dessert (Morin-Audebrand et al., 2009) but not for the recognition of an orange juice or a yoghurt, which are actually better remembered for their bitterness and 55 56 sourness respectively (Köster et al., 2004). Furthermore, there is general agreement in the literature 57 suggesting that memory for food is modulated by novelty (Morin-Audebrand et al., 2012). This 58 means that, when a memory effect occurs, it is mainly based on consumers' ability to reject 59 something not previously tasted (i.e. the distractors) rather than to identify a food already 60 experienced (*i.e.* the target). Another common finding is that food memory seems to be independent 61 from age; despite the recognised assumption that memory declines over the lifespan, it should be 62 considered that this loss of memory ability is remarkable in explicit memory but not in implicit 63 memory (that being the case of food memory), which is almost unaffected by age (Balota, Dolan, & 64 Duchek, 2000). Accordingly, food studies carried out on differently aged consumers groups showed 65 that adults (age 18-45) and elderly people (age > 60) have comparable memory indices (Møller, 66 Wulff & Köster, 2004; Møller, Mojet & Köster, 2007; Laureati, Morin-Audebrand, Pagliarini, 67 Sulmont-Rossé, Köster & Mojet, 2008; Sulmont-Rossé, Møller, Issanchou, & Köster, 2008). 68 Very few studies have attempted to compare incidental and intentional learning for sensory and 69 food stimuli. Møller et al. (2004, 2007) compared incidental and intentional learning and memory in

70 young and elderly subjects. They found that young adults remembered odors and flavors better

under intentional than incidental learning conditions, whereas the elderly remembered these stimuli
equally well under both conditions and as well as the young under the incidental condition.

73 There have been very few studies investigating the retention time effect on food memory, and the 74 results of such studies are contradictory. Frijters (1977) explored the ability to discriminate odors 75 within very short delay intervals (0, 5, 8 and 12 s) and did not find a retention time effect on 76 subjects' performance. Barker and Weaver (1983) showed that through lengthening the time 77 interval between the presentations of two explicitly learned stimuli, a decrease in the ability to 78 remember odors occurred, whereas taste stimuli memory was less influenced by retention time. 79 Cubero, Avancini de Almeida & O'Mahony (1995) and Avancini de Almeida, Cubero & 80 O'Mahony (1999) showed that citrus flavored beverage discrimination was better when stimuli 81 were experienced subsequently and that performance deteriorated as interstimulus interval 82 increased. Similarly, Degel, Piper & Köster (2001) found that memory for unconsciously learned 83 odors decreased with increased delay interval (from 60 min to 120 min). Contrasting results were 84 obtained by Harker, Gunson, Brookfield & White (2002) who investigated the ability to detect 85 differences in apple firmness when presented with fruit at 1 day and 1 min interstimulus delays. 86 They reported that subjects encountered more difficulties in detecting texture differences after a 1 87 min interval as compared to a 1 day interval, but their results could be criticised on the basis of their 88 testing procedure that demanded people to test apples at a very high rate in the one minute interval 89 condition allowing no time for recovering from adaptation or even muscular fatigue.

Quite surprisingly, little research has been conducted on these topics despite their importance. Actually, when performing sensory testing, products are usually assessed subsequently with time intervals between tasting sessions as short as possible. These circumstances do not necessarily reflect real life conditions. In most cases, foods may only be tasted and compared days, weeks or months apart. The time interval depends on the specific foods and consumers involved in the study. Therefore, a more ecological approach to the consumers' food learning and memory investigation is important to be considered.

97 The aim of the present study was to investigate how the learning type (incidental *versus* intentional) 98 and the retention interval length (one day versus one week) might influence food memory in 99 children. Based on our knowledge and information, this topic has never been investigated so far. 100 Given that sweetness has a powerful hedonic appeal, especially among children and young people, 101 sweet foods and beverages have been indicated as potential contributors to the obesity epidemic 102 worldwide (Drewnoski, Mennella, Johnson & Bellisle, 2012). There is currently considerable 103 research on the biological mechanisms that influence sweet taste preferences and drive the 104 consumption of sweet tasting foods but very few studies were addressed to memory for sweet taste. 105 Studying food memory may provide with an indication about the way in which incidental memory 106 for food works and about sensory impressions' role in this memory. In addition, the study of sweet 107 taste perception and memorization could provide food companies with strategic information on new 108 low calories formulations development. This is especially important considering the growing and 109 widespread children's obesity phenomenon.

110 **2. Materials and Method**

111 2.1. Subjects

Two-hundred-fourteen children (106 girls and 108 boys), aged between 8-10 years were recruited in two Milan schools. One school was tested for incidental learning and the other one for intentional learning of a food stimulus. The children were divided into two groups, which were tested for memory respectively after one day and one week after the learning phase. The two schools shared the same refectory and had the same lessons schedule. Children from the two schools were matched for gender (χ^2 =0.02; p=0.89) and age (χ^2 =0.74; p=0.69).

Parents were asked to read a short study explanation, to sign a consent form and to complete a questionnaire where they were to indicate whether their child had any food allergy or followed a specific diet. Parents also answered questions concerning their child's preference and consumption frequency for some foods, including those under study. All children involved in the study met the following criteria: healthy; not on a specific diet; not suffering from food allergies or from smell and taste disorders. Children did not receive any particular reward for participation, but an
educational "taste lesson" at the end of the experiment.

125 2.2. Stimuli

A commercial apple fruit purée (Frutta Pura Mellin, SpA, Italy) was chosen for the study. The ingredients listed on the label were: apple and vitamin C. The experimental products, consisting of one target and two distractors (one less sweet and one sweeter than the target), were produced at the University of Milan sensory laboratory by adding different amounts of sucrose to 1000 g of fruit purée.

In order to obtain perceivable but subtle differences in sweetness intensity among the target and the 131 132 distractors, the Just Noticeable Difference (JND) (i.e. the smallest difference perceived by 50% of 133 the population) was calculated involving a separate group of 38 children. According to Köster et al. 134 (2004), five fruit purée samples which differed for equal sugar concentration steps (C1, C2, C3, C4, 135 C5) were produced. The middle concentration served as a reference (Ref=C3) and the other 136 concentrations were used as comparison stimuli (C1, C2, C4, C5). The reference was compared to 137 each of the other concentrations through a paired comparison test. Each pair contained at least one 138 reference (Ref), the other sample was a comparison sample (C1, C2, C4, C5). The reference was 139 also tested against itself. Each pair was presented twice: once with the reference presented in the 140 first tasting position and once with the reference presented in the second tasting position. Thus, 141 children received 10 fruit purées pairs. The pairs' presentation order was systematically varied over 142 children. For each pair, children were asked to state which of the two samples was the sweetest. 143 Children were instructed to rinse their mouth with water before the test and after each pair.

In order to calculate the JND of the whole group, we determined the percentage of the times when each comparison stimulus was judged to be more intense in sweet taste than the reference. These percentages were turned into z-scores under the normal probability curve and plotted against the concentration of sugar added. The function of the best fitting straight line through these points was determined and the concentration values corresponding to z-values of -0.675 and +0.675 (z-values

of 25% stronger and 75% stronger than the reference) were calculated from this function. The JND was found by taking half of the difference between these two concentration values. Although this method is slightly incorrect since the arithmetic mean was used instead of the geometrical mean between these two concentrations to determine the size of the JND's, it was considered that the difference was small enough to use it.

The distractors were respectively 1.5 JND lower (D-) and 1.5 JND higher (D+) in concentration than the target (T). The -1.5 JND distractor, the target and the +1.5 JND distractor were obtained by adding respectively 44 g, 87 g, and 130 g of sugar to 1000 g (1.5 JND=43 g/kg) of the base fruit purée, which reported on label a 96 g/kg concentration of carbohydrates naturally present in apples.

158 2.3. Procedure

159 2.3.1. Day 1: Learning Session

160 During the learning session (first day), children of both schools were offered a mid-morning snack 161 consisting of a biscuit, a fruit juice portion and a target fruit purée portion. In order to guarantee an 162 involuntary learning of food aspects, the incidental group of children was asked under a false pretense to eat the snack and to rate the liking degree of each food item. The false pretense was 163 164 conceived just with the purpose of distracting children's attention from the real aim of the study, 165 thus memory was never mentioned. Also the intentional learning group of children ate the snack 166 and rated the liking degree of each food item but – accordingly with the explicit learning methods – 167 they were asked to focus their attention on the features of food they would have consumed, since 168 they would have been asked to perform a memory test later.

169 2.3.2. Day 2: Test Session

170 Children belonging to each learning group were divided in two groups, which were tested for 171 memory after respectively one day and one week since the day of the learning phase. As previously 172 mentioned, children belonging to the incidental group were unaware of the study aim and were 173 unexpectedly asked to perform a memory test. All children were confronted with a series of fruit 174 purée samples consisting of the target tasted during the learning session and each of the two

corresponding distractors modified in sweetness. Children performed a memory test, a liking test, 175 176 and a discrimination test. The memory test consisted in presenting a monadic series of 4 samples: 2 target samples and one sample of each of the 2 distractors. The ratio targets/distractors (1:1) was 177 178 chosen to avoid unwanted learning effects due to overrepresentation of the target in the memory 179 test. Children were asked to taste each sample and to answer the question: "Did you eat this sample 180 vesterday/one week ago? Yes/No". They were not informed about the exact number of targets and 181 distractors in the series, but they were told that some of the samples might be the same as the one 182 previously tasted. Then, the children completed a liking test. They received three new samples in a monadic series (one target and the two distractors) and for each of them they were asked to rate how 183 much they liked it on a seven-point hedonic-facial-vertical scale with the anchors "super bad" 184 185 (bottom of the scale) and "super good" (upper part of the scale) (Pagliarini, Ratti, Balzaretti & 186 Dragoni, 2003). Finally, a paired comparison test was conducted in order to check whether the 187 children perceived the expected sweetness differences between the target and the distractors. Each 188 child was given a tray consisting of three fruit purée pairs: one pair consisting of the less sweet 189 distractor and the target (D- vs T), one consisting of the sweeter distractor and the target (D+ vs T) 190 and one consisting of two identical samples of the target (T vs T). The pairs presentation was 191 randomized over children and the test was performed so that, at the time of comparing the target vs 192 a distractor, half of the children assessed first the distractor and then the target (D- vs T; D+ vs T), 193 whereas the other half was to assess first the target and then the distractor (T vs D-; T vs D+). For 194 each pair of fruit purée, children were asked to point out the sweeter sample.

195 2.4. Experimental Conditions

Sessions were performed in the classrooms, at 10 am mid-morning break in the presence of a teacher and an experimenter. The number of children in the classes ranged from 15 to 25. During the first session (learning session, day 1) children were invited to sit at their own table, thus ensuring real meal conditions as much as possible, and they were offered 100 mL of fruit juice, 80 g of fruit purée and 1 biscuit. During the test session (day 2), children received 20 g samples of fruit 201 purée for each sample for the memory test and 15 g samples of fruit purée for each sample for the202 liking and discrimination tests.

203 Children were provided with a booklet for each test, and they were given a short explanation about 204 the use of the scale and the instructions to complete the booklet before each test session. Children 205 were instructed to rinse their mouth with water before the beginning of each session and after the 206 tasting of each sample. Each experimenter had the instructions to read to the children for all the 207 tests. In order to ensure consistency of the instructions provided, the interviewers were instructed to 208 follow strictly the script.

The experimental samples were prepared the day before each session and stored at 4 °C. Samples were taken out from the cooling room 2 h before the session and served at room temperature in plastic cups covered with a plastic lid and coded with different three digit numbers in each test. Within each session, the design was balanced for order and carry-over effect (Macfie, Bratchell, Greenhoff & Vallis, 1989).

214 2.5. Data Analysis

215 Memory was tested by means of the Signal Detection Theory (SDT). According to the SDT, two 216 factors underlie the participants' responses in a memory test: (1) the participants' ability to identify 217 the target amongst distractors (memory strength) and (2) the participants' tilt toward one response 218 or the other (response bias). Two parametric indices, namely the d' and c indices, are usually used 219 to measure these two dimensions (Macmillan & Creelman, 2005). The index d' is commonly assessed by the proportion of "yes" responses to the targets (hits) corrected by the proportion of 220 "yes" responses to the distractors (false alarms), whereas c is assessed by the average of "yes" 221 responses relative to the average of "no" responses over all samples. To be computed, these 222 223 parametric indices require a response frequencies normal transformation. However, the computation 224 of these indices is questionable when the number of targets and distractors by participant is small 225 (Snodgrass & Corwin, 1988) as in this case, since it was impossible to ask children to eat a too large 226 number of samples. Therefore, according to Laureati et al. (2008), in the present study the

proportion of "yes" answers to the target (YesT) and to the distractors (YesD) and the proportion of "no" answers to the target (NoT) and to the distractors (NoD) were determined and used to calculate memory indices based on the same principle as the d' and the c, but non-parametric (*i.e.* the normal transformation is not required). For the targets, the "yes" responses correspond to the correct recognition, whereas for the distractors, the "no" responses correspond to the correct rejections.

232 As regarding the memory strength, a recognition index was computed: recognition index (P0) =233 YesT- YesD. This index is equivalent to the index P0 proposed by Snodgrass & Corwin (1988). 234 The recognition index varies from -1 to +1. The more the recognition index is close to +1, the more 235 the participant managed to recognize the target amongst the distractors. On the contrary, a 236 recognition index equal to or lower than 0 reveals that the target incidental learning did not occur. 237 As regarding the response bias, a bias index was computed: bias index = 0.5*[(NoT + NoD) - (YesT)]+ YesD)]. This bias index varies from -1 to +1. A positive bias index indicates a bias to respond 238 239 "no", a zero bias index indicates no bias and a negative bias index indicates a bias to respond 240 "ves". Student *t*-tests were used to assess whether memory indices were different from zero or not. According to SDT, d' and C reflect two independent dimensions underlying participant's responses, 241 242 thus we can state that a memory effect occurs if the proportion of "yes" responses for the target is higher than the proportion of "yes" responses for the distractors, even if the participants had a bias 243 to answer "no" during the absolute memory test. 244

Analysis of variance (ANOVA) was performed considering type of *Learning* (2), *Retention time* (2), *Gender* (2), *Age* (3) and their two-way interactions as factors, and memory indices as dependent variables in the model. The paired comparison test results were analyzed through unilateral statistical test (p=1/2) according to the binomial distribution (ISO, 2005). For each pair, the correct answers number was calculated and compared with the minimum number of correct answers to affirm that there is a significant (p<0.05) difference between samples.

The hedonic test results were analyzed through ANOVA considering *Learning, Retention time,*JNDs, Gender, Age and their relevant two-way interactions as factors and hedonic values as

dependent variable. Statistical analyses were performed using SAS/STAT statistical software
package version 9.3.1. (SAS Institute Inc., Cary, USA).

255 **3. Results**

256 3.1. Discrimination Test

257 To check whether the children were able to perceive the differences between the target and the 258 distractors, a paired comparison test was performed. For this test, a reduced number (n=65) of 259 children was involved since not all children were available due to practical constraints. The number 260 of correct answers for each pair and the minimum number at which a response becomes significantly (p<0.05) higher than expected on the basis of chance guessing was computed from the 261 262 binomial distribution (p=1/2) and shown in Figure 1. Results showed that 49 out of 65 (75.4 %) of the children correctly perceived the D- distractor as less sweet than the target, whereas only 39 out 263 264 of 65 (60.0%) of the children perceived the D+ distractor as sweeter than the target. This seems to 265 suggest that the actual distance between the target and the D- distractor was 1.0 JND rather than 1.5 JND and that the actual distance between the target and the D+ distractor was even somewhat lower 266 267 than 1.0 JND. This might explain the difference in correct response in the memory test to be 268 reported below. Anyhow, considering that the minimum number to have a significant response is 269 equal to 41 for p<0.05 (represented by a line in Figure 1), it can be stated that children identified the 270 less sweet distractor more easily, whereas the answers for the sweeter distractor only tended to 271 reach significance. The control pair target-target was not significant, suggesting that children 272 correctly perceived the two target samples as equally sweet.

Results were also analyzed by learning and retention time in order to see whether children belonging to the incidental or intentional group or to the one day or one week group differed in their discrimination ability. It was found that the proportion of children who correctly judged the D- as less sweet and the D+ as sweeter than the target was comparable among groups (p-values always >0.05 based on a Chi-square analysis).

278 3.2. Memory Test: Learning and Time Retention Effect

279 Memory index P0 calculated by learning and by retention time is reported in Figure 2. As can be 280 seen memory improved with increased retention time in the incidental condition, whereas under 281 intentional learning conditions, memory declined with increased retention time. More specifically, 282 under incidental learning conditions, children showed positive memory indices both after one day 283 and after one week from the learning session, even if only after one week the index is significantly 284 higher than zero ($M_{(P0)}=0.21$; $t_{(43)}=2.86$; p<0.01). In the case of the intentional group, no memory 285 effects were found, whatever the retention time was.

ANOVA results highlighted a significant effect only for the main factor type of *Learning* ($F_{(1,199)}=3.61$; p<0.05) on children's ability (P0) to remember the stimulus previously experienced. According to the multiple range test, memory was better in incidental ($M_{(P0)}=0.16$) rather than intentional learning conditions ($M_{(P0)}=0.03$). No effect of *Age*, *Gender*, *Retention time* and of their two-way interactions on P0 has been found.

P0 memory index was also calculated by JNDs in order to establish whether children remembered better an increase or a decrease in sweetness. Thus, a memory index for each distractor was calculated for each child in both learning conditions and both retention times. Results showed no memory effect for both distractors under intentional learning conditions neither after a one day nor one week interval, whereas under incidental learning conditions the less sweet distractor was recognized but only after a one week interval ($M_{(P0)}=0.32$; $t_{(43)}=3.91$; p<0.001).

297 Bias index calculated by type of learning, retention time, gender or age was always positive and 298 significantly (p<0.05) different from zero, suggesting the children's tendency to answer 'no' to the 299 recognition question. No effect of learning condition, retention time, age, gender or their interaction 300 was found on bias index, suggesting that children had the same tilt to answer to the recognition 301 question whatever the type of learning and retention time were, and regardless of gender and age. 302 This answer pattern is consistent with the data shown in Figure 3 where the proportion of correct 303 answers for both the targets (hits) and the distractors (correct rejections) are reported. Data were 304 averaged across retention time since ANOVA results highlighted no effect on memory for this variable. Chi-square results pointed out that, under both learning conditions, the correct rejections proportion was significantly higher than the hits proportion (χ^2 =3.28, p<0.01 for incidental learning; χ^2 =3.56, p<0.001 for intentional learning), suggesting that children identified something not previously experienced more easily than something already learned. Furthermore, results highlighted that the hits proportion was comparable between learning conditions, whereas the correct rejection proportion was significantly higher (χ^2 =4.73, p<0.05) with incidental rather than intentional learning conditions.

312 3.3. Hedonic Test

Lsmeans hedonic scores by stimulus are reported in Figure 4. ANOVA results showed a significant 313 effect for the main factor JNDs ($F_{(2, 615)}=6.49$; p<0.01). A tendency to prefer fruit purée with higher 314 315 sucrose concentration is observed in Figure 4. More specifically, children gave significantly lower 316 hedonic ratings to the less sweet distractor, as compared to the target and the sweeter distractor that 317 were comparable in terms of liking. The Age ($F_{(2, 615)}=6.22$; p<0.01) and Retention time ($F_{(1, 615)}=6.22$; p<0.01) 318 ₆₁₅₎=16.08; p<0.01) main factors were also significant. According to multiple comparison tests, 10 319 y.o. children gave significantly higher hedonic ratings than younger children. In addition, children 320 gave lower liking ratings after one day as compared to one week interval. The other main factors 321 and all their interactions were not significant.

322 **4. Discussion**

323 The present paper investigated a topic never considered so far: comparing children's incidental and324 intentional learning and memory for sweetness in a real food product.

The main research output are: 1) children's memory coming from a food stimulus involuntary learning is better than that originated by a voluntary learning effort of it; 2) the time elapsing in the interval between the food stimulus learning and the retrieval phase does not influence children's memory.

Results obtained confirm previous studies which showed that children are able to incidentally learn
and then memorize food stimuli (Laureati, Pagliarini, Mojet & Köster, 2011; Laureati et al. 2008).

331 Since no literature about the comparison between intentional and incidental children's food learning332 is available, our discussion is limited to research conducted on adults.

333 Møller et al. (2004) studied the voluntary (intentional) and involuntary (incidental) odor learning 334 memory and found that odor memory was higher when stimuli were learned intentionally for the 335 young, whereas the contrary was seen in the elderly, thus suggesting that intentional odor memory 336 performance declines with age. Comparable results were obtained in a following study (Møller et 337 al., 2007) aimed at comparing incidental and intentional learning in adults and elderly subjects 338 using real food. It was found that the adults remembered novel flavors added in soups better under 339 intentional than incidental learning condition, whereas the elderly remembered these stimuli equally 340 well under both conditions. The results of the present study contrast with those obtained by Møller 341 and colleagues, since the present authors found that children's memory was better under incidental 342 than intentional learning conditions. This divergence might be explained at least in two ways. First, 343 Møller and colleagues performed their studies in a laboratory context, which is somewhat different 344 from the present study's conditions (i.e. taste stimulus added to a real food and evaluated in a natural eating context). Thus, it might well be that a formal condition, such as a laboratory test, 345 346 might increase subjects' attention on the stimuli provided. Second, the age groups considered were 347 different. As concerning this point, the present authors suspect that the discrepancy's cause is 348 probably not the age-related differences but rather the other contextual factors. Indeed, there is 349 evidence that food learning and memory under ecological conditions is comparable among children, 350 adults and elderly people (Laureati et al., 2008).

The fact that involuntary food learning is more effective in generating memory should not be a surprising result if we consider that in everyday life we learn about food without any explicit effort. On the contrary, it is extremely rare that we pay attention to the food we eat or drink unless there is something unexpected. Another important consideration coming from this result is that explicit paradigms should be cautiously considered when applied for studying food learning and memory

since they are probably not appropriate, being less ecologically valid than implicit experimentalprocedures.

A more detailed account of memory responses in terms of hits, misses, correct rejections and false 358 359 alarms showed that children were not able to recognize the target previously tasted better than 360 chance, since the percentage of hits under both incidental and intentional conditions was 361 approximately 50%. The percentage of correct rejections was always higher than the percentage of 362 hits, showing that under both conditions rejection of the distractors contributed more to memory 363 performance than the target identification. However, the percentage of correct rejections was higher 364 under incidental than intentional conditions and this might explain the better memory performance 365 when children involuntarily learn food stimuli. This result is in agreement with Morin Audenbrand 366 et al. (2012), who analyzed the results obtained in several experiments differing for experimental 367 conditions, type of food and participants but sharing the same implicit paradigm used in the present 368 experiment and found that – at least for incidentally learned sensory stimuli – memory is based on 369 novelty or change detection (i.e. distractors) rather than on previously encountered stimuli 370 recollection and recognition.

371 Considering the time retention effect on sensory memory, mixed results are present in the literature 372 and none of them have been obtained involving children. In general, it is assumed that lengthening 373 the time interval elapsing from the learning and the retrieval phase might have a negative effect on 374 memory performance because of an increased possibility of fading and confusion of the stored 375 bases resulting from the stimulations. In accordance with this assumption, odor recognition tests 376 performed considering intervals between the stimuli initial and second presentation varying in terms 377 of seconds (Engen, Kuisma & Eimas, 1973), minutes (Barker & Weaver, 1983), days (Rabin & Cain, 1984) or weeks (Engen & Ross, 1973), found that recognition performance generally 378 379 deteriorated as the interval was longer. Taste stimuli are less influenced by time of retention (Barker 380 & Weaver, 1983): this seems in agreement with our results. However, these studies are based on

explicit or implicit paradigms which anyway considered simple sensory stimuli such as odors, andnone of them were conducted on children, thus they are hardly comparable to the present one.

383 The memory storage systems for food sensory properties are not well understood. From vision and 384 audition research (Baddeley, 1997), it would seem that the first memory stage is somewhat a wake 385 of the sensations elicited by the food. This immediate memory would explain superior recognition 386 skills at short retention time. Despite the contrasting opinions in literature, especially for olfactory 387 memory, it has been suggested that, in the same way as for vision and audition, the sensations 388 elicited by food would be expected to be held in a short term memory. Sometimes these would be 389 transferred into a long term, more permanent memory (Baddeley, 1997). How long exactly the 390 sensations elicited by a food are held in short term memory before being transferred to long term 391 memory is not known. For auditory and visual stimuli, it seems to be a matter of seconds or 392 minutes. For food sensations, it has never been investigated. In this context, we found no clear 393 evidence of retention time effect on children's memory for sweetened fruit purée even though 394 memory was better after one week than one day interval under incidental learning conditions. One 395 hypothesis that might be forwarded to explain this result is that better memory for incidental 396 learning after one week retention interval is due to a better food stimuli perception from children 397 belonging to the incidental-one week group. However, this is not the case since results of the paired 398 comparison test analyzed by learning and retention time showed no difference among children 399 groups in their ability to discriminate the target from the distractors. Another point that should be 400 considered is that different children were involved for incidental and intentional tests as well as for 401 tests after one day and one week retention interval. However, this choice has been forced by the 402 nature of the paradigm used. In this respect, care has been taken to balance each learning and 403 retention time group for age and gender.

404 As concerning hedonic data, children liked more the fruit purées with a higher sugar concentration 405 and clearly pointed out the less sweetened samples as the least pleasant, although pretty high liking 406 ratings were observed for all the products evaluated (D-, T, and D+). This result is particularly 407 significant in relation to the memory and discrimination tests findings. Indeed, when memory data 408 were analyzed by distractor type, it was found that the less sweet distractor was better recognized. 409 The less sweet distractor was also better discriminated from the target in terms of sweetness 410 perception. This would suggest that children seem more aware of sugar subtraction than addition 411 from a hedonic, perceptive, and a memory point of view.

412 The higher liking degree expressed by children for more sugary products is in accordance with 413 literature data (Liem & de Graaf, 2004) and could be explained by the sour taste of the product 414 chosen in the present experiment. This result is particularly interesting for food companies which 415 are required more and more to optimize children's products by reducing the sugar and fats contents 416 due to the growing and widespread phenomenon of children obesity. In this context, food 417 developers should keep in mind that young consumers can perceive even the smallest differences in 418 the sweetness of a given food product – especially in the case of a reduced amount of sugar. 419 Children would also be able to learn and memorize involuntarily such variations in sweetness.

420 A possible explanation for the better discrimination of the D- distractor from a perceptive, hedonic, and memory point of view might be that in order to get the same discrimination, the sweetness 421 422 difference between the D+ distractor and the target sample should have been larger than the 423 difference between the D- distractor and the target sample. In the present experiment the sensory 424 distance among the distractors and the target sample was equal. In other words, this would mean 425 that the sugar amount added to the target (43 g) to obtain the D+ distractor would have exerted less 426 influence on the perceived intensity than the sugar amount added to the D- distractor (43 g) to 427 obtain the target sample. Nevertheless, although the effects found in the present experiment both for 428 the hedonic and the perceptive tests seem to point into the same direction, it is unlikely that they 429 explain all the difference observed for the couple D- vs Target and the couple D+ vs Target.

430 It should also be considered that the perceptive and hedonic tests results could in turn explain the 431 outcome of a lower memory for the D+ distractor. A similar effect was found in Morin-Audebrand 432 et al.'s (2009) paper where the authors highlighted that young adults were able to discriminate a 1.5 433 JND less sweet custard dessert from the target custard but not custards which were 1.5 JND and 434 even 2.5 JND sweeter than the target. In Morin-Audebrand and colleagues' paper, a memory effect 435 for taste occurred and depended more on the correct rejection of the distractors rather than on the 436 identification of the target, as in the present case. Köster et al. (2004) also found the same type of 437 insensitivity to added sugar and showed a distorted memory for sweet taste. In their experiment, an 438 orange juice sample was varied in sweetness and bitterness and a voghurt sample in sweetness and 439 sourness. They used just noticeable differences to prepare distractors that varied from the target 440 sample by -1.0, +1.0, +1.5 and +2 JNDs for each varied taste. They found that for both orange juice 441 and yoghurt, varying sweetness had no effect on memory performance. Even the distractor that was 442 2 JNDs sweeter than the target sample was not recognized as different from the memory of the 443 target. On the contrary, the distractors that differed in bitterness and sourness were clearly 444 recognized as different from the memory of the target. They also assessed relative memory (asking 445 subjects whether the experimental target and distractors were more, less or equally sweet/sour/bitter 446 than the target eaten before) for the same foods and found that the memory for sweetness was 447 distorted and that only addition of 2 JND sugar came to be marginally different from the target, 448 whereas a deduction of -1 JND caused a very clear and significant difference from the target. The 449 same memory distortion for sweetness had earlier been found by Barker and Weaver (1983). 450 Further research is needed to better understand the relationship between human taste perception and 451 memory ability.

Finally, it should be borne in mind that the experiment results cannot be generalized due to the small number of children involved in the study. In this regard, in order to confirm our results, it is recommended to increase the number subjects and to extend the evaluation considering other food stimuli.

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530 Figure caption

- 531 **Fig.1** Results of the discrimination test: number of correct answers for each couple of stimuli: D- vs
- 532 T, T vs T, D+ vs T (D- = distractor 1.5 JND less sweet than the target; T = Target; D+ = distractor
- 533 1.5 JND sweeter than the target). The line indicates the minimum number at which an answer
- 534 becomes significant for p<0.05.
- 535 Fig.2 Results of the memory test: memory index by learning (incidental, intentional) and time of
- retention (day, week) and its significant difference from zero (** p<0.01; n.s. not significant).
- 537 Fig. 3 Proportion of correct answers for the target (hits) and the distractors (correct rejections) for
- 538 incidental and intentional learning conditions.
- 539 Fig. 4 Results of the hedonic test: Ismeans hedonic ratings by JND. D-=distractor 1.5 JND less
- 540 sweet than the target; T= Target; D+=distractor 1.5 JND sweeter than the target.
- 541







