Subthreshold Olfactory Stimulation Can Enhance Sweetness

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Abstract

The impact of olfactory perception on sweetness was explored in a model solution using odorants at subthreshold concentrations. First, the impact of 6 odorants, previously described in the literature as congruent with sweetness, was investigated at suprathreshold level in a sucrose solution. Ethyl butyrate and maltol were selected as they had the highest and the lowest sweetness-enhancing properties, respectively. Second, the impact on sweetness of the 2 odorants was investigated at subthreshold concentrations. A system delivering a continuous liquid flow at the same sucrose level, but with varying odorant concentrations, was used. At a subthreshold level, ethyl butyrate but not maltol significantly enhanced the sweetness of the sucrose solution. This study highlights that olfactory perception induced by odorants at a subthreshold level can significantly modulate taste perception. Finally, contrary to results observed with ethyl butyrate at suprathreshold levels, at subthreshold levels, the intensity of sweetness enhancement was not proportional to ethyl butyrate concentration.

Key words: cognitive integration, familiarity, olfaction, psychophysics, taste

Introduction

Pioneer studies on interaction at a suprathreshold level between odor and taste perception using static sensory measurement (Murphy et al. 1977; Murphy and Cain 1980) or time–intensity evaluation (Cliff and Noble 1990; Bonnans and Noble 1993) showed that subjects attribute a taste to aqueous solutions flavored with an odorant. Retronasal olfactory perception can also be modulated by taste perception (Hort and Hollowood 2004).

Perceptual similarity between odorant and tastant in a mixture seemed to be a good predictor of taste intensity change (Frank et al. 1991). Indeed, an odor can acquire a taste quality when the odor–taste pair is perceived in food commonly experienced by consumers. Congruency is defined by Schifferstein and Verlegh (1996) as ''the extent to which 2 stimuli are appropriate for combination in a food product.'' For instance, pineapple flavoring enhances perceived sweetness of a model solution (Prescott 1999). In a real food context, strawberry odor enhances whipped cream sweetness, whereas peanut butter does not affect sweetness rating (Frank and Byram 1988). Another study showed that vanilla flavoring enhances sweetness perceived by children and adults when added to milk (Lavin and Lawless 1998). Furthermore, Stevenson et al. (1999) reported that an odor can decrease taste intensity when the odor–taste pair is not congruent in the food. In their experiment, caramel odor, related to sweet taste, decreased sour taste intensity. Functional magnetic resonance imaging also provides evidence for the convergence of taste and olfactory stimuli in the lateral anterior part of the orbitofrontal cortex to produce flavor in humans (de Araujo et al. 2003).

Stimuli at a subthreshold concentration also have an impact on perception. Integration at a subthreshold level of congruent taste and olfactory stimuli presented orthonasally was demonstrated using a variant of the 2-alternative forced-choice method (Dalton et al. 2000). Indeed, the orthonasal olfactory threshold of benzaldehyde significantly decreased with the presence of a saccharin solution in mouth at a subthreshold concentration. The same experiment repeated with monosodium glutamate did not lead to any change in benzaldehyde sensitivity. As for olfactory/taste interaction at a suprathreshold level, interaction at a subthreshold level seems to occur only with familiar odorant/tastant pairs. These results about the impact of familiarity were confirmed by repeating the same experiment with another panel (Pfeiffer et al. 2005). In addition, the authors also highlighted that olfactory/taste interaction occurred when the olfactory stimulus was delivered retronasally. But in both cases (orthonasal and retronasal), integration with taste only occurred when stimuli were presented at the same time. The same authors also showed a lack of integration for the benzaldehyde/saccharin pair for 4 subjects. This result may be explained by a lack of familiarity with this taste/aroma pair. Delwiche and Heffelfinger (2005) also demonstrated an integration of odor and taste at a subthreshold level. Contrary

to the results of Dalton and Pfeiffer, the authors showed that odor/taste integration is not dependent on familiarity. This study concludes that the impact of tastant and odorant is additive, regardless of the harmony of the taste/odor pair.

The goal of the present study was to investigate whether an odorant at a subthreshold level could enhance sweetness of a sucrose solution. The odorant was presented orally in a sucrose mixture clearly perceived as sweet. The aim of this protocol was to mimic everyday consumption of sweet food where synchrony of the odorant/tastant delivery is more likely to induce olfactory/taste integration (Pfeiffer et al. 2005). Indeed, there is a need for the food industry to extend understanding about the impact of olfactory/taste interaction on consumer perception.

Benzaldehyde, ethyl butyrate, furaneol, vanillin, maltol, and isoamyl acetate odorants were selected because of their reported enhancing properties on sweetness at a suprathreshold level (Lavin and Lawless 1998; Hollowood et al. 2002; Kato 2003; Baldwin et al. 2004; Cerf-Ducastel and Murphy 2004; Hort and Hollowood 2004).

Two experiments were designed to fulfill our objectives. The first experiment aimed at selecting the odorants having the highest and the lowest enhancing properties on sweetness at a suprathreshold concentration. The second experiment was carried out to quantify and compare the impact of the 2 selected odorants at a subthreshold level on the sweetness of a sucrose solution.

A liquid delivery system was used during the second experiment. Compared with standard in-cup tasting, this system continuously delivered a liquid flow with a constant sucrose concentration but with different odorant concentrations and without tasting interruption and sample change. Subjects were therefore less disrupted and influenced than with "cup tasting." This system prevented aroma evaporation during tasting and saved preparation time, even though the evaluation was conducted individually. The dilution error risk was also minimized. Hort and Hollowood (2004) explained that using the Dynataste system allowed to mimic beverage consumption over a realistic time period.

To avoid confusion we will use the term "odor" to refer to the orthonasal olfactory perception evaluated above the cup and "aroma" to refer to the retronasal olfactory perception (i.e., the organoleptic attribute perceptible by the olfactory organ via the back of the nose (NF ISO 5492 1995). The term "olfactory" regroups both odor and aroma perception.

Experiment 1: selection of 2 odorants having the highest and the lowest enhancing properties on sweetness among the 6 odorants tested at suprathreshold concentration

Subjects

Nine untrained students, 5 females and 4 males between 18 and 25 years old, were recruited for experiment 1. They had never participated in any sensory tasting, and no training session was conducted for this study.

Sample preparation

Mineral water (Vittel, France) with sucrose was used for odorant evaluation at a suprathreshold level for all solutions. As the aim was to obtain a solution perceived as sweet, a sucrose concentration of 5 g/l, and therefore, above the detection threshold was chosen according to Hong et al. (2005). We validated that this concentration was above threshold through a triangle test with 24 subjects (10 females and 14 males between 30 and 40 years old). The results showed that a 5 g/l sucrose solution prepared with Vittel water was significantly perceived as different from a pure Vittel water solution (P value < 0.0001). The triangle test was conducted using a nose clip to ensure that subjects discriminated samples based on the sweet taste of sucrose and not on a possible olfactory stimulation by the tastant as already reported by Mojet et al. (2004, 2005).

Each odorant (Sigma-Aldrich Chemie GmbH, Munich, Germany) was evaluated at low, medium, and high concentrations in the sweetened water solution. For each odorant, the 3 suprathreshold concentrations were defined on the basis of a preliminary tasting with 8 project members to obtain a perceptible 3-step enhancement of the olfactory note compared with the unflavored sweetened water (reference). This preliminary tasting conducted in pure Vittel water consisted for each compound in a ranking–scaling task of 8 odorant solutions on 2 attributes (overall odor intensity, overall aroma intensity). A linear scale whose extremities were defined as "not intense" and "extremely intense" was used. For each odorant, 3 concentrations (low, medium, and high) were selected according to the following sensory criteria: 1) the 3 concentrations for each odorant had clearly discriminable odor and aroma intensities and 2) for each level the selected concentrations of the 8 odorants were isointense. In addition, each solution was compared with pure water using a nose clip to ensure that the odorant did not induce any other perception than olfaction (taste, trigeminal, or texture). Odorant concentrations were as follows (ppm): benzaldehyde 10, 50, 100; ethyl butyrate 5, 10, 20; maltol 100, 500, 1300; furaneol 10, 75, 150; vanillin 100, 200, 400; isoamyl acetate 10, 50, 70. For all odorants, the highest concentration was still soluble in water (Chemfinder database, Cambridge Soft Corporation, Cambridge, UK, 2004). Sample preparation was carried out 1 h prior to evaluation using volumetric flasks with a magnetic stirring bar and covered with Parafilm M Barrier Film (Structure Probe, Inc., West Chester, PA).

Sensory procedure

For each odorant, four 30-ml samples (the unflavored sample and 3 flavored samples) were presented simultaneously. Samples were coded with 3-digit random numbers and served at room temperature in a 50-ml cup. The subjects

assessed the samples in a predefined order, according to a replicated balanced experimental design over subject, and could retaste if needed. Each of the 6 sets of 4 samples was assessed during a 30-min session over 6 days. Due to technical constraints, the odorant order was not balanced over the 6 sessions and was the same for all subjects. The subjects ranked samples from the least to the most intense according to their odor, aroma, and sweetness intensity. Equality between samples was permitted. Vittel mineral water and unsalted crackers were used for rinsing between samples. Tests were conducted in an air-conditioned room (18 °C), under white light in individual booths. Data acquisition was carried out on a computer screen with FIZZ software (Biosystèmes 1990).

Data analyses

All data were analyzed using FIZZ software. For each attribute, the global difference between the 4 sums of ranks was tested using a nonparametric 2-way analysis on ranks (Friedman test, significance level $\alpha = 0.05$). For attributes showing significant differences, a multiple comparison test was applied (paired comparison with a Bonferonni adjustment with $\alpha = 0.00833(0.05/C_4^2)$, (Wolfe 1998).

Results

For each set of 4 samples, all odorants significantly enhanced odor (P value < 0.0001) and aroma (P value < 0.0001) compared with the reference. However, the olfactory impact on sweetness differed according to the odorant. Ethyl butyrate had the highest enhancing impact on sweetness, and maltol had the lowest enhancing impact on sweetness (See Figure 1a,b). They were therefore selected for the next step of the study. The magnitude of the sweetness-enhancing effect in-

duced by the 4 other odorants was intermediate (benzaldehyde P value: 0.001, furaneol P value: 0.002, isoamyl acetate P value: 0.012, and vanillin P value: 0.520).

Experiment 2: odorant impact at subthreshold concentration on sweetness

Subjects

A new panel of 9 untrained subjects (6 females and 3 males between 18 and 25 years old) participated in parts A and B of the second experiment.

A. Determination of the lowest threshold value within the panel ([LTP]) for the 2 odorants selected from the first experiment

Sample preparation

Fifteen 1-l solutions of each odorant were prepared at room temperature with pure mineral water (Vittel, France) with odorant concentrations ranging from 0.1 to 5.94E-06 ppm for ethyl butyrate and from 2.5 to 1.5E-04 ppm for maltol. Each solution was prepared 1 h prior to the tasting.

Sensory procedure

The threshold of each subject for each odorant was determined in Vittel water using forced-choice ascending concentration series method of limit (ASTM Sensory Testing Methods 1991). For each odorant, the subjects performed a series of 15 three-alternative forced-choice discrimination tasks (3-AFCs). Each 3-AFC comprised 2 unflavored water samples and 1 flavored water sample. An ascending odorant concentration range was defined with a dilution factor of 2. The appropriate concentration range was determined for
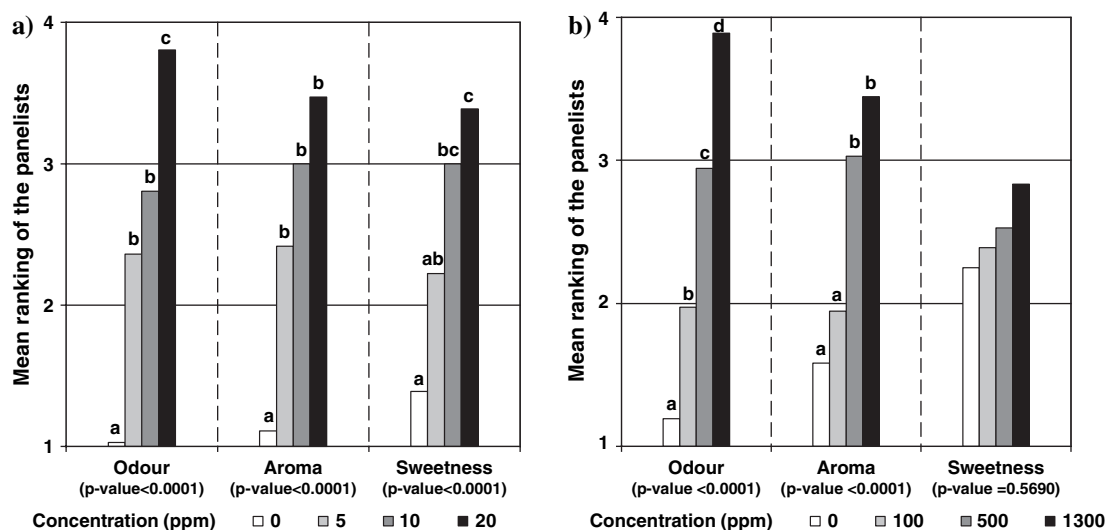

Figure 1 (a, b) Olfactory and sweetness modulation of aqueous sucrose solutions by a) ethyl butyrate and b) maltol. Samples with the same letter are not significantly different.

each odorant following benchscale preliminary trials including the range of threshold values reported in the literature. The 15 three-AFCs were evaluated in an ascending concentration order. The 30-ml solutions were tasted in plastic cups coded with 3-digit random numbers at room temperature in 50-ml cups. Samples were evaluated under the same conditions as previously described for experiment 1 with a rinsing between each of the 15 three-AFCs.

Data analyses

For each subject, the concentration above which all 3-AFC tests were correct was considered as the individual detection threshold concentration. However, when an incorrect response was given following at least 2 correct responses, the 15 three-AFC was repeated to obtain an unbiased threshold detection value. For each odorant, the lowest and the highest threshold concentration within the panel was defined and the geometric mean of individual threshold was calculated. The lowest individual threshold concentration within the panel [LTP] was chosen as a basis to determine the subthreshold concentrations evaluated in the next step of experiment 2. To prepare dilutions largely below any individual threshold, the 5 following concentrations were chosen: [LTP]/16, [LTP]/32, [LTP]/64, [LTP]/128, and [LTP]/256.

Results

The lowest and highest threshold concentrations within the 9 subjects were 1) 3.90E-04 ([LTP]) and 2E-01 ppm with a panel geometric mean of 1E-02 ppm for ethyl butyrate and 2) 1.21E-03 ([LTP]) and 1.25 ppm with a panel geometric mean of 1.24 ppm for maltol.

B. Investigation of the impact of odorant at a subthreshold level on sweetness intensity

Sample preparation (liquid delivery system)

To explore the impact of odorants at a subthreshold level on sweet perception, a liquid delivery system was developed, inspired by the Dynataste system of Hort and Hollowood (2004). The device was based on a programmable 4-channel preparative high-performance liquid chromatography pump (Merck-Hitachi, L 7150) and four 1-l reservoirs. The 4 reservoirs were linked to the high performance liquid chromatography (HPLC) mixing chamber, and the mixing chamber was linked to the subject's mouth with Teflon tubing. One reservoir (A) contained an aqueous sucrose solution, and the 2 other reservoirs (B and C) contained the same aqueous sucrose solution as reservoir A but with odorant at 2 concentrations. As explained by Hort and Hollowood (2004), by programming the flow rate of each pump, the composition of the delivered liquid can vary over time but the overall flow rate remains constant. In the present study, the device delivered online a solution with a constant in-mouth flow rate, thereby avoiding any variation of in-mouth tactile stimulation and a constant sucrose level but odorant concentration varied. The programming over time of the contribution of each channel to the final liquid flow delivered a solution with a 25-ml/s flow rate alternatively flavored and nonflavored. The 5 odorant concentrations were distributed into 2 delivery sequences as described in Table 1. The first sequence was divided into 6 steps (3 flavored and 3 unflavored), and the second sequence was divided into 4 steps (2 flavored and 2 unflavored stimuli) with a total of 10 steps for the 2 sequences. This flavored and unflavored liquid alternation ensured that tubing was rinsed between each odorant concentration delivery and therefore limited sensory adaptation. Each of the 10 steps was delivered for 18 s. Total duration of the 2 sequences was, therefore, 180 s.

Compared with the cup tasting in experiment 1 carried out for the odorant selection, the sucrose concentration of the liquid was increased from 5 to 15 g/l. This was because the subjects did not consider the stimulus as sweet enough during the familiarization sessions as, with this dynamic system, they had to swallow regularly and could not keep the liquid in mouth or retaste the samples.

Sensory procedure

For each of the odorants (ethyl butyrate and maltol), the 9 subjects evaluated during 180 s sequences 1 and 2 with a 180 s break between each sequence. The total duration of one session (sequence 1 and sequence 2) including the break was 360 s. The amount of liquid swallowed during a session

Table 1 Contribution over time of each channel of the delivery system to the in-mouth liquid flow

Stimuli n°	Delivery timing (s)	Odorant concentration	Contribution of each sucrose solution reservoir to the in-mouth liquid flow (%)		
			Reservoir A	Reservoir B	Reservoir C
Sequence 1					
1	0–18	0	100		
2	18–36	[LTP]/256	50		50
3	36–54	0	100		
4	54–72	[LTP]/64	75	25	
5	72–90	0	100		
6	90–108	[LTP]/16		100	
Break (180 s)					
Sequence 2					
7	72–90	0	100		
8	90–108	[LTP]/32	50	50	
9	108–126	0	100		
10	126–144	[LTP]/128			100

Reservoir A: pure 1.5% sucrose solution; Reservoir B: 1.5% sucrose solution + odorant [LTP]/16; Reservoir C: 1.5% sucrose solution + odorant [LTP]/128.

based on a 25-ml/min flow rate was 75 ml. Each session was duplicated. A total of 4 sessions per judge was therefore conducted for the evaluation of the 2 odorants. To avoid contamination between odorants inside the pump tubing, the session order was not randomized between subjects. All subjects started with the evaluation of ethyl butyrate. To standardize among the 9 subjects the liquid delivery in mouth and swallowing, subjects were trained to pinch the Teflon tube extremity between the top and bottom incisors with 1-cm tube into the mouth. The subjects were invited to swallow regularly and normally. They scored over time, on a 11-box scale anchored at the extremities "not sweet at all" to "very sweet," the sweet taste intensity at 10 time points corresponding to ten 18-s steps. A computerized FIZZ session was coupled to the HPLC pump and synchronized with the appearance on PC screen of the sweetness scale 10 s after the beginning of each step. This time period took into account the 3 s needed for the pump to make the mixing and deliver the required concentration and the 7 s allowing subjects to experience and evaluate the stimulus before scoring it. The subjects could then score the sweetness intensity during the 8 remaining seconds. During evaluation, subjects were also asked to report on a sheet of paper any perception other than sweetness (olfactory or gustatory). Moreover, a debriefing session was carried out at the end of the second experiment to collect general comments of subjects. After exploring the impact of the 2 odorants, an additional session was conducted to validate that the sweet enhancement was not due to the device or the procedure. The sweetness intensity of an unflavored sucrose solution (15 g/l) was scored over time at 10 different points.

Before evaluation sessions, subjects were familiarized with the device and protocol during 2 sessions and especially with the constant flow of liquid into the mouth and the scoring procedure. This familiarization was carried out with unflavored sucrose solution only.

Data analyses

For each of the 5 odorant concentrations ([LTP]/16 to [LTP]/256), sensory data were transformed according to the formula $SC_n = SO_n - S_n$, where SC is the sweetness change, SO the sweetness of sucrose solution with odorant, and S the sweetness of sucrose solution without odorant evaluated before SO (see Figure 2). A confidence interval at 95% was calculated for the 5 SC panel mean scores. For the familiarization test with sucrose only, perceived changes in sweetness were also calculated and plotted as described above.

Results

Figure 3 shows that all subthreshold concentrations of ethyl butyrate significantly increased the perceived sweetness of the sucrose solution. In addition, sweetness enhancement by ethyl butyrate was constant, whatever the odorant concentration. Maltol did not consistently modify perceived sweetness (See Figure 4). The lowest concentration significantly enhanced sweetness, whereas the highest concentration significantly reduced sweetness. Intermediate concentrations 2, 3, and 4 did not significantly modify sweetness.

Changes in sweetness induced by subthreshold levels of ethyl butyrate and maltol are detailed for the different subjects in Figure 5. Seven subjects out of 9 (See bottom right part of the Figure 5) perceived the sucrose solution more frequently sweeter with ethyl butyrate than with maltol. Subject 6 found an equivalent high enhancement of sweetness with both ethyl butyrate and maltol. Sweetness perception of subject 1 was weakly enhanced by both odorants.

The evaluation of the sucrose solution without odorant addition conducted at the end of the study validated that sweetness enhancement was not due to the device and procedure used. As expected, results showed that perceived sweetness did not significantly differ over 10 consecutive evaluations (See Figure 6).
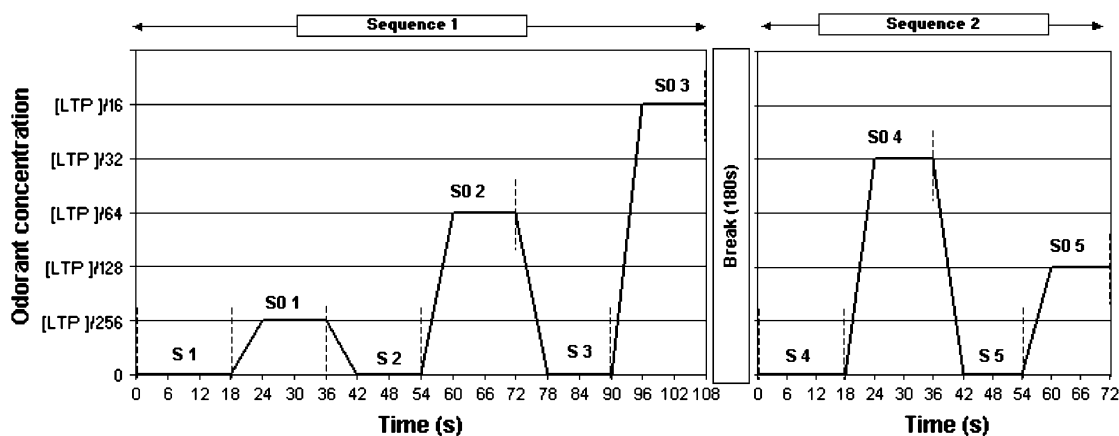

Figure 2 Schematic representation of the liquid flow evaluated over time. [LTP]: lowest threshold of the panel (3.90E-04 ppm for ethyl butyrate and 1.21E-03 for maltol); S: sweetness of sucrose solution without odorant; SO: sweetness of sucrose solution with odorant.
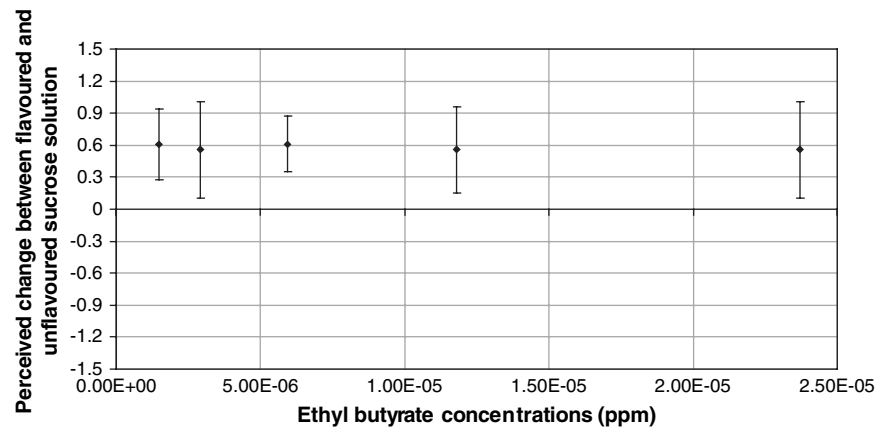
Figure 3 Effect of subthreshold ethyl butyrate concentrations on perceived sweetness of a 15 g/l sucrose solution. ⎪ 95% confidence interval of means.
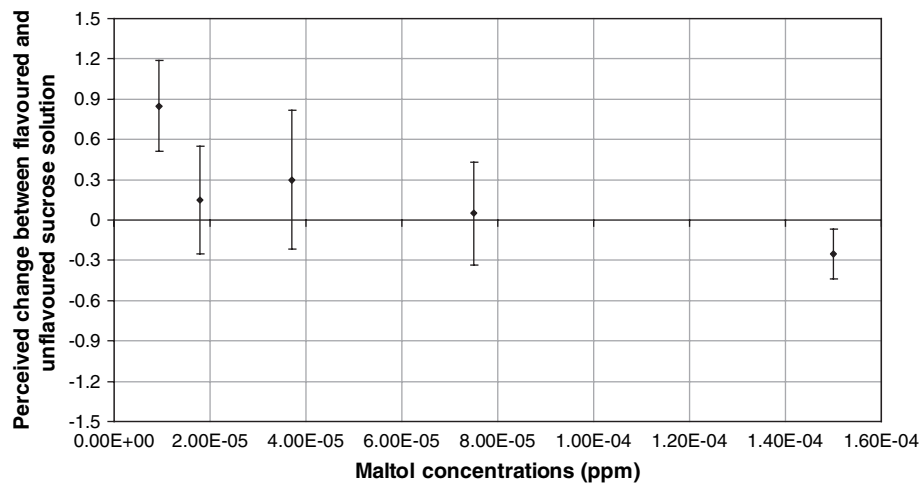

Figure 4 Effect of maltol concentrations on perceived sweetness of a 15 g/l sucrose solution. ⎪ 95% confidence interval of means.

Discussion

Impact of odorant at a suprathreshold level on sweetness

Results obtained for all odorants at suprathreshold levels highlighted a clear consensus and discrimination of the 4 odorant concentrations regarding their odor and aroma intensity. Our results were partially in agreement with previous studies that reported the sweetness-enhancing properties of the 6 odorants tested. Ethyl butyrate, often described in the literature as having a strawberry note (Miettinen et al. 2004), seemed, in the present study, to be the odorant the most consistently associated with sweetness by the panel. Furaneol, isoamyl acetate, and benzaldehyde also significantly enhanced sweetness of the sucrose solution. Vanillin and maltol did not significantly boost sweetness even though these 2 compounds have already been reported to enhance sweetness. Indeed, Lavin and Lawless (1998) showed that vanilla flavoring, which has olfactory characteristics close to those of vanillin, enhances milk sweetness compared with plain

milk. Kato (2003) showed that the maltol generated in roux (wheat flour and butter mixture) during heating enhances sweetness. These 2 studies were conducted in food, whereas in the present study, the odorant/tastant pairs were evaluated in water. The association between sweet taste and olfactory notes induced by vanillin and maltol may not be strong enough in a liquid system compared with more texturized food where these tastant/odorant pairs are usually experienced. Our hypothesis is in agreement with recent studies showing that odor/taste interaction results from associations experienced and memorized implicitly through food exposure (Köster et al. 2004; Köster 2005) and that food familiarity strongly modulates olfactory/taste interactions (Labbe et al. 2006). The role of food experience on sensory interaction at suprathreshold level has also been demonstrated at a neural level using neuroimaging (Small et al. 2004). Brain activation representing olfactory/taste interaction depends on the subject's previous experience with smell/taste combinations.

Impact of odorant at a subthreshold level on sweetness

The second part of the study was carried out with another panel of untrained subjects to avoid any bias induced by the first experiment. Indeed, previous odorant/sweet taste coexposure with odorant at a suprathreshold level may reinforce odor/taste cognitive association and therefore enhance the odorant's impact on taste perception (Labbe et al. 2006).

All subthreshold concentrations of ethyl butyrate enhanced sweetness, whereas maltol induced a significant enhancement for 1 concentration out of 5 only. Our results with maltol partially agree with Bingham et al. (1990),
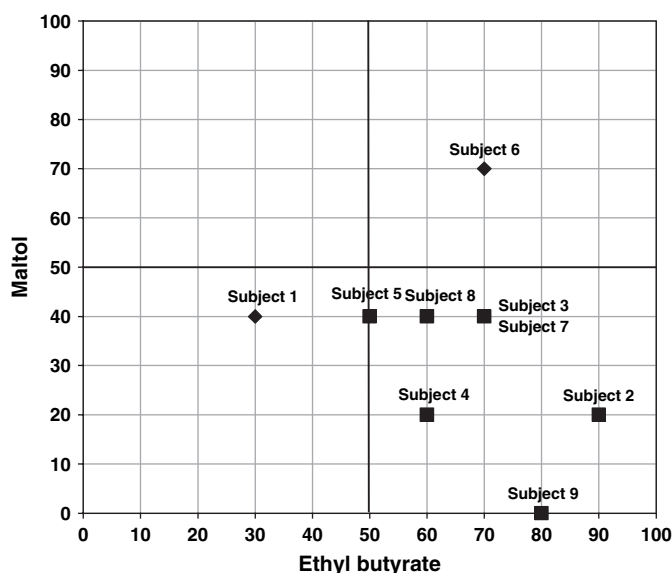

Figure 5 Percentage of times each subject judged the solution with odorant sweeter than the pure sucrose solution for maltol (vertical axis) and for ethyl butyrate (horizontal axis). $N = 10$ (5 concentrations × 2 replications).

who, using a triangle test, showed that a subthreshold concentration of maltol (15 ppm) did not significantly change lemonade flavor perception. Sweetness enhancement by ethyl butyrate was neither due to the device nor due to the protocol. Consequently, 2 hypotheses can be proposed to explain this observation. First, sucrose enhances odorant release into the headspace due to physical–chemical interaction and led to a suprathreshold level of odorant. This may result in a sweetness scoring increase because of a perceptual olfactory–taste interaction or a dumping of olfactory perception on the sweetness scale (Frank et al. 1993; Clark and Lawless 1994). This hypothesis is unlikely for 3 main reasons: 1) Nahon et al. (1998) demonstrated that release of ethyl butyrate present in orange aroma was not enhanced by sucrose (at a concentration similar to that used in the present study) compared with the aqueous control, 2) in the present study, subjects did not report any perceived olfactory notes during evaluation with the liquid delivery system, and 3) 2 replicated triangle tests were performed by a 24-subject panel (10 females and 14 males between 30 and 40 years old) to check by sniffing the impact of the highest subthreshold ethyl butyrate concentration (3.2 ppm) on olfactory perception. Tests were conducted in water (comparing an unsweetened Vittel water solution with an unsweetened flavored Vittel water solution) and also in sweet water (comparing an unflavored 15 g/l sucrose solution with a flavored 15 g/l sucrose solution) to highlight any odorant/taste interactions. Each triangle test result showed that samples were similar at a 10% significance level (β), where similarity was defined as a maximum of 20% of assessors recognizing the difference. Therefore, at the concentrations used, ethyl butyrate did not induce any orthonasal olfactory stimulation.

Our second hypothesis is that ethyl butyrate enhances sweetness at subthreshold level through perceptual integration. This hypothesis probably explains our phenomenon
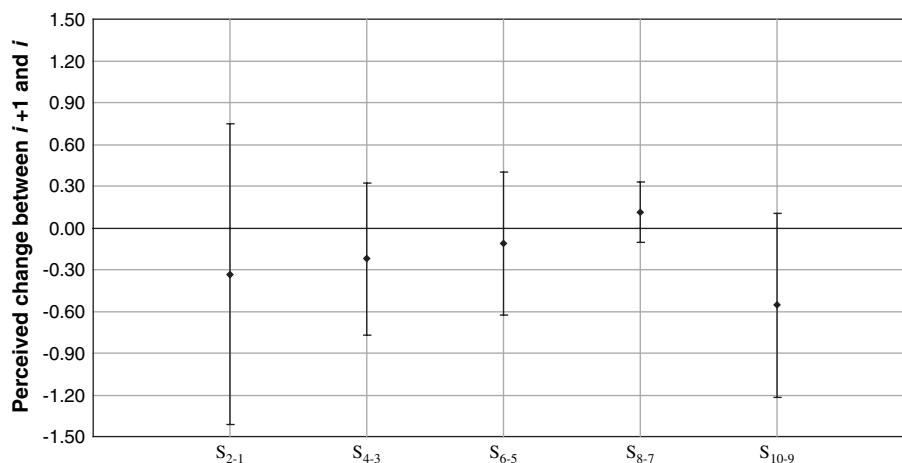

Figure 6 Sweetness evaluation of the unflavored sucrose solution over time (same calculations and representations as for Figures 4 and 6). ⊥ 95% confidence interval of means, S_{i+1-i} account for sweetness score difference between evaluation $i+1$ minus evaluation i.

and supports previous findings by Dalton et al. (2000) and Pfeiffer et al. (2005).

Results obtained with ethyl butyrate and maltol at sub-threshold levels were consistent with those at suprathreshold levels: significant effect of ethyl butyrate and nonsignificant effect of maltol on sweetness in both cases. Even if ethyl butyrate and maltol were both congruent with sweetness, the level of familiarity for ethyl butyrate/sucrose compared with maltol/sucrose pairs might explain the higher impact of ethyl butyrate on sweetness. Indeed, congruency between taste and olfaction has been reported to influence olfactory/taste central integration even with subthreshold concentrations of odorants (Dalton et al. 2000; Pfeiffer et al. 2005).

Our results showed that 2 of the 9 subjects were not consistent with the panel. Subject 6 showed considerable taste enhancement both with maltol and with ethyl butyrate. This subject might be as familiar with ethyl butyrate/sucrose association as with maltol/sucrose association. In contrast, subject 1 showed a weakly enhanced sweetness perception for both odorants. He/she might be less familiar with these odorant/tastant associations. Another explanation for differences between these 2 subjects may be sensitivity. Because thresholds varied widely among subjects, the extent to which the stimulus was below subthreshold also varied very much. To better assess the impact of subject odorant sensitivity on sweetness enhancement, the ratio between subthreshold stimulus concentration and individual threshold was calculated and plotted against individual sweetness enhancement. Figure 7 shows the results for the above-mentioned subjects (1, 6). The level of the subject's odorant threshold cannot explain differences in the sweetness enhancement between subjects. Indeed, for subject 1, the concentrations of ethyl butyrate used were closer to his/her threshold than the concentrations of maltol (10 000 times below threshold), but sweetness enhancement was weak for both odorants. For subject 6, both odorants strongly enhanced sweetness even though the concentrations of maltol and ethyl butyrate were largely below the subject's threshold values (1000 and 10 000 times lower for maltol and ethyl butyrate, respectively). Based on these observations, the ratio subthreshold
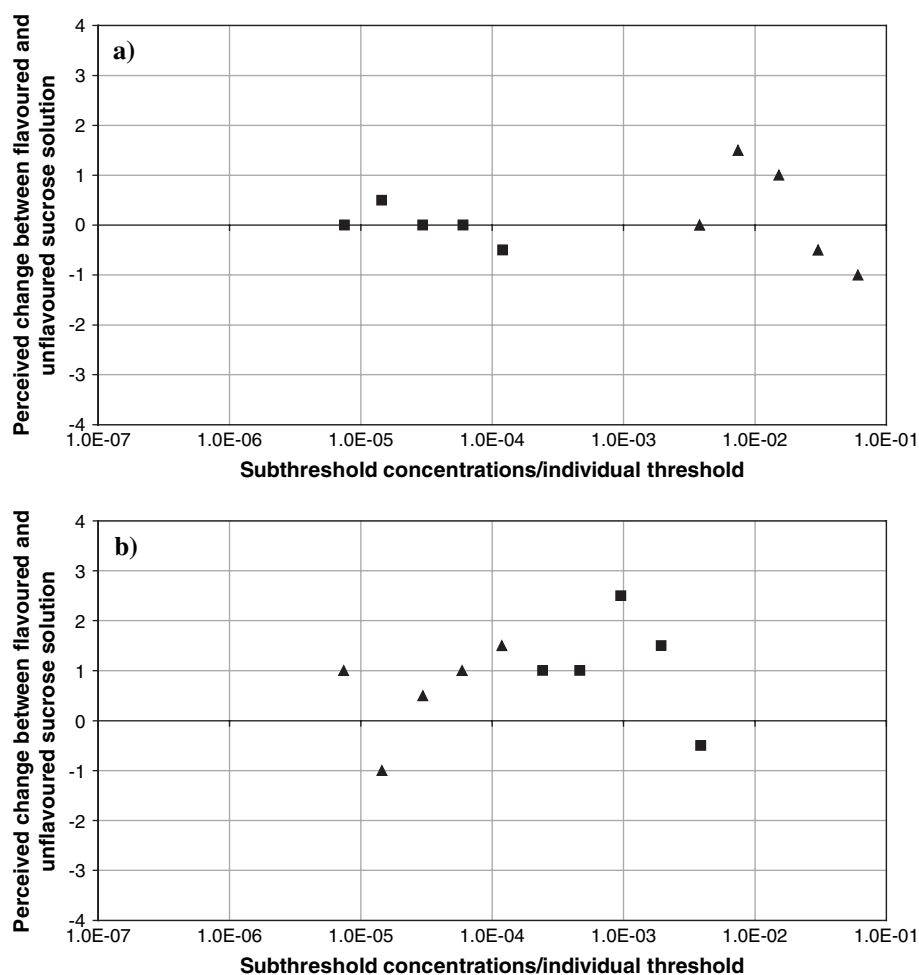

Figure 7   (a, b) Effect of the ratio subthreshold odorant concentration (5 concentrations × 2 replications)/individual threshold on the individual sweetness enhancement for ethyl butyrate (▲) and maltol (■). (a) subject 1, (b) subject 6.

concentration/individual threshold seems to have little impact on sweetness modulation. On the other hand, familiarity might explain the intersubject differences. Pfeiffer et al. (2005) also highlighted an absence of integration between saccharin taste and benzaldehyde odor for 4 out of 16 subjects. The authors assumed that these subjects were not familiar with the benzaldehyde/saccharin pair.

At subthreshold levels, the boosting impact of ethyl butyrate on sweetness does not seem to depend on its concentration: the sweetness enhancement was not proportional to ethyl butyrate concentration. This outcome suggests that odorant stimulation at subthreshold level led to an ''on–off'' taste modulation, which is different from taste modulation by an odorant at suprathreshold level. Conversely, results obtained in experiment 1 and reported in the literature on taste modulation by an odorant at suprathreshold level (Labbe et al. 2006) showed that sweetness enhancement was proportional to odorant concentration: the sweetness increased with increase in odorant concentration.

The main finding of the present study was that sweetness of a sucrose solution can be enhanced by subthreshold levels of an odorant. Given these results, it would be interesting to investigate the level of sucrose reduction that can be compensated by odorant addition while maintaining sweetness. In addition, the liquid delivery system should be improved in order to deliver 1) greater randomization within odorants, subjects, and sessions and 2) the same ratio between subthreshold concentration and individual threshold for each subject.

Acknowledgements

References

ASTM Sensory Testing Methods. 1991. Standard practice for determination of odor and taste thresholds by a forced-choice ascending concentration series method of limits. West Conshohocken (PA): ASTM International. E.679.

Baldwin EA, Goodner K, Plotto A, Pritchett K, Einstein M. 2004. Effect of volatiles and their concentration on perception of tomato descriptors. J Food Sci. 69:S310–S318.

Bingham AF, Birch GG, Graaf Cd, Behan JM, Perring KD. 1990. Sensory studies with sucrose-maltol mixtures. Chem Senses. 15:447–456.

Biosystèmes. 1990. FIZZ: Logiciel de gestion de l'analyse sensorielle et des tests consommateurs. Couternon (France).

Bonnans S, Noble AC. 1993. Effect of sweetener type and of sweetener and acid levels on temporal perception of sweetness, sourness and fruitiness. Chem Senses. 18:273–283.

Cerf-Ducastel B, Murphy C. 2004. Validation of a stimulation protocol suited to the investigation of odor-taste interactions with fMRI. Physiol Behav. 81:389–396.

Clark CC, Lawless HT. 1994. Limiting response alternatives in time-intensity scaling: an examination of the halo-dumping effect. Chem Senses. 19:583–594.

Cliff M, Noble AC. 1990. Time-intensity evaluation of sweetness and fruitiness and their interaction in a model solution. J Food Sci. 55:450–454.

Dalton P, Doolittle N, Nagata H, Breslin PAS. 2000. The merging of the senses: integration of subthreshold taste and smell. Nat Neurosci. 3:431–432.

de Araujo IET, Rolls ET, Kringelbach ML, McGlone F, Phillips N. 2003. Taste-olfactory convergence, and the representation of the pleasantness of flavour, in the human brain. Eur J Neurosci. 18:2059–2068.

Delwiche JF, Heffelfinger A. 2005. Cross-model additivity of taste and smell. J Sens Stud. 20:512–525.

Frank RA, Byram J. 1988. Taste-smell interactions are tastant and odorant dependent. Chem Senses. 13:445–455.

Frank RA, Schaffer G, Smith DV. 1991. Taste-odor similarities predict taste enhancement and suppression in taste-odor mixtures. Chem Senses. 16:523.

Frank RA, van der Klaauw NJ, Schifferstein HN. 1993. Both perceptual and conceptual factors influence taste-odor and taste-taste interactions. Percept Psychophys. 54:343–354.

Hollowood TA, Linforth RS, Taylor AJ. 2002. The effect of viscosity on the perception of flavour. Chem Senses. 27:583–591.

Hong JH, Chung JW, Kim YK, Chung SC, Lee SW, Kho HS. 2005. The relationship between PTC taster status and taste thresholds in young adults. Oral Surg Oral Med Oral Pathol Oral Radiol Endod. 99:711–715.

Hort J, Hollowood TA. 2004. Controlled continuous flow delivery system for investigating taste-aroma interactions. J Agric Food Chem. 52:4834–4843.

Kato Y. 2003. Chemical and sensory changes in flavor of roux prepared from wheat flour and butter by heating to various temperatures. Food Sci Tech Res. 9:264–270.

Köster EP. 2005. Does olfactory memory depend on remembering odors? Chem Senses. 30(Suppl 1):i236–i237.

Köster MA, Prescott J, Köster EP. 2004. Incidental learning and memory for three basic tastes in food. Chem Senses. 29:441–453.

Labbe D, Damevin L, Vaccher C, Morgenegg C, Martin N. 2006. Modulation of perceived taste by olfaction in familiar and unfamiliar beverages. Food Qual Pref. 17:582–589.

Lavin JG, Lawless HT. 1998. Effects of color and odor on judgments of sweetness among children and adults. Food Qual Pref. 9:283–289.

Miettinen SM, Hyvonen L, Linforth RST, Taylor AJ, Tuorila H. 2004. Temporal aroma delivery from milk systems containing 0-5% added fat, observed by free choice profiling, time intensity, and atmospheric pressure chemical ionization-mass spectrometry techniques. J Agric Food Chem. 52:8111–8118.

Mojet J, Heidema J, Christ-Hazelhof E. 2004. Effect of concentration on taste-taste interactions in foods for elderly and young subjects. Chem Senses. 29:671–681.

Mojet J, Köster EP, Prinz JF. 2005. Do tastands have a smell? Chem Senses. 30:9–21.

Murphy C, Cain WS. 1980. Taste and olfaction: independence vs interaction. Physiol Behav. 24:601–605.

Murphy C, Cain WS, Bartoshuk LM. 1977. Mutual action of taste and olfaction. Sens Processes. 1:204–211.

Nahon DF, Koren PA, Roozen JP, Posthumus MA. 1998. Flavor release from mixtures of sodium cyclamate, sucrose, and an orange aroma. J Agric Food Chem. 46:4963–4968.

NF ISO 5492. 1995. Analyse sensorielle-Vocabulaire. In: AFNOR, editor. Contrôle de la qualité des produits alimentaires-Analyse sensorielle. Paris (France): Recueil de normes françaises. p. 27–51.

Pfeiffer JC, Hollowood TA, Hort J, Taylor AJ. 2005. Temporal synchrony and integration of sub-threshold taste and smell signals. Chem Senses. 30:539–545.

Prescott J. 1999. Flavour as a psychological construct: implications for perceiving and measuring the sensory qualities of foods. Food Qual Pref. 10:349–356.

Schifferstein HN, Verlegh PW. 1996. The role of congruency and pleasantness in odor-induced taste enhancement. Acta Psychol. 94:87–105.

Small DM, Voss J, Mak YE, Simmons KB, Parrish T, Gitelman D. 2004. Experience dependent neural integration of taste and smell in the human brain. J Neurophysiol. 92:1892–1903.

Stevenson RJ, Prescott J, Boakes RA. 1999. Confusing tastes and smells: how odours can influence the perception of sweet and sour tastes. Chem Senses. 24:627–635.

Wolfe DA. 1998. Non-parametric methods. In: Armitage P, Colton T, editors. Encyclopedia of biostatistics. Vol. 4. Chichester (UK): Wiley. p. 3030–3037.