Effect of Ethanol on Interactions of Bitter and Sweet Tastes in Aqueous Solutions

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Abstract

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Experimental samples simulated the composition of vermouths. In all experiments, 0.01% quinine was used as a standard bitter substance. Sucrose increased the acceptability in the concentration range of up to 14%, remaining constant at higher concentrations, both in aqueous and 16% ethanolic solutions. A decrease of bitterness was observed in water but not in 16% ethanol. Ethanol did not affect the sweetness appreciably at the concentrations of up to 16%, but 32% ethanolic solutions appeared less sweet. Ethanol enhanced the bitterness only at high concentrations; interactions were similar in the samples containing 10% and 16% sucrose. Aspartame and Neotame sweetness increased the acceptability and decreased the bitterness similarly to sucrose, both in aqueous and in 16% ethanolic solutions.

Keywords: sweetness; bitterness; ethanol; sucrose; Aspartame; Neotame; sensory analysis

Taste interactions, both antagonistic and synergistic, were intensively studied as they are important both from scientific and technologic aspects (Baryłko-Pikielna 1975), especially in those papers, where the interactions of sweet, acidic, and salty tastes were examined in detail. Even the sensory acceptability of foods and beverages is affected by the taste interactions (Pokorný 2004). The interaction of bitter and sweet tastes is a typical example of antagonism. The bitterness of quinine was suppressed by the sweetness of sucrose, and on the contrary, the sweetness of sucrose was suppressed by quinine in aqueous solutions (Schiffman *et al.* 1995). These taste interactions are probably due to transduction (Walters 1996). Synthetic sweteners are amphiphilic in nature, therefore they can cross the cell membrane by electrophoretic transfer mechanism (Eggers *et al.* 2000). As they have an ionic character, contrary to sucrose, different sweeteners can act at different sites of the transduction system (Brand & Feigin 1996), and they may directly affect the behaviour of ionic channels. The topic was reviewed by Keast and Breslin (2002) in detail.

Another important example of the taste interactions is the reaction of tastes with ethanol. The interaction is particularly important in the formulation of alcoholic beverages. Dessert wines contain both sugar and 14–20% vol. ethanol, added in order to stop the fermentation and to modify the flavour. Sherry wines (Bakker 2003), port wines (Cristovam & Paterson 2003) or madeira wines (Goswell 2003) belong to this category. However, the prevailing sweet taste is not appreciated by

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some consumers. Vermouths have similar concentrations of ethanol and sugar as dessert wines, but the sweetness is reduced by the addition of bitter plant extracts, called botanicals (Liddie & Boero 2003). The sweetness perception proceeds via a mechanism similar to that of the bitterness perception, thus interactions between the two tastes are likely to occur (Cardello & Maller 1987). The sweetness and bitterness interactions have been well known as more than 300 years ago, bitter ethanolic extracts were consumed on a piece of sugar. However, the effects of ethanol on the interactions of sweetness and bitterness have not yet been studied, even though several studies have been published on the subject. Therefore, we investigated these interactions; some preliminary experiments were presented at the 6^{th} Wartburg Aroma Symposium in Eisenach (Panovská *et al.* 2004). In this paper, we present more detailed results of our experiments.

MATERIALS AND METHODS

Materials. Sucrose p. a. (Lachema, a. s., Brno, Czech Republic), Aspartame 98% (Urseta, s. r. o., Doksy u Kladna, Czech Republic), Neotame (The Nutrasweet Co., Augusta, USA), quinine hydrochloride anhydrous (Fluka Chemie, Buchs, Switzerland), ethanol extra fine 96% (Lihovary, Kolín, Czech Republic), tap water (Czech Standard, – ČSN 75 711, and Guidelines of the Ministry of Health of the Czech Republic, 2004), containing Ca > 40 mg/l, Mg > 20 mg/l, hardness degree 1.6 mmol/l, pH 7.71.

Sensory analysis. The test room was equipped according to the international standard (ISO 8589), and the sample serving was in agreement with the recommended standard procedure (ISO 6658). The panel of sensory assessors consisted of persons selected, trained, and monitored in agreement with the respective international standard (ISO 8586). Pairs of 15 ml samples were served in coded 25 ml beakers. Tap water was used for washing the mouth before and between the tastings. Short sippings of the sample solutions and minimum intervals of 15–60 s between tastings were allowed (Meilgaard *et al.* 1978). Unstructured graphical 100 mm scales (ISO 4121), orientated by the description at both ends, were used for the evaluation of the sensory acceptability (0% = unacceptable, 100% = excellent), sweetness (0% = imperceptible, 100% very strong), and bitterness (0% = imperceptible, 100% = very strong); the evaluation conformed to the international standard (ISO 6564). The results obtained with the use of graphical scales are more easily interpreted than those obtained using ordinal category scales (Cardello & Maller 1987).

Statistical evaluation. The variance was calculated using the two-way F-test, and the statistical significance of variances was calculated using the two-way t-test. The distribution of values obtained by using the graphical scales followed the application of parametric method for the determination of mean values and standard deviations, conformed with the results presented by McBride (1985). Average differences between the results of the individual assessors were calculated after Gini (1912). The probability level was P = 0.95, unless otherwise stated. The software MS STATISTICA 7.0 was used. The rank test was evaluated after Kramer as indicated in the textbook by Baryłko-Pikielna (1975) at the probability level of P = 0.95.

RESULTS AND DISCUSSION

Design of experiments

In our model experiments simulating vermouths, sugars were represented by sucrose. Extracts of botanicals consist of several bitter substances of varying properties and efficiencies, so that the results of sensory experiments could be applied to the particular case only. Therefore, quinine hydrochloride was used as a well defined standard substance. The concentration of quinine hydrochloride was kept constant (0.01%) in all experiments in order to reduce the number of variables. All samples were evaluated by a group of assessors (total of 20 to 40 repeated determinations of the same sample on different days), chosen in random using the respective tables of random numbers (Baryłko-Pikielna 1975). It was acceptable that the same assessor could taste several times the same sample in random as differences found between the assessors in preliminary tests were not statistically significant (see the following paragraph). Neotame was included among synthetic sweeteners as a representative of modern and very efficient substances (Nofre & Tinti 2000). Two concentrations of synthetic sweeteners were used as the sweetness efficiency depended on the content of ethanol; therefore, the concentration corresponding to the sweetness of 10% sucrose in aqueous solution only could not be considered as satisfactory.

Table 1. Average differences of ratings by different assessors (mm of the graphical scale)

Assessor's code	Acceptability A (mm)	Sweetness S (mm)	Bitterness B (mm)
A	15	18	17
B	12	10	18
C	11	6	8
D	14	15	17
E	14	11	21
F	10	11	13
G	9	10	9
H	11	7	21
Mean and standard deviation	12 ± 2.1	11 ± 3.9	16 ± 5.0

Performances of individual assesssors

Average differences between the individual assessors in two ratings of the same samples on different days are shown in Table 1. The average differences and the respective standard deviations could be calculated as the position of ratings on unstructured graphical scales agreed with the suggestions outline by McBride (1985) in his critical paper. The differences were higher in the evaluation of bitterness than in the case of acceptabilities and evaluation of sweetness. This phenomenon is natural because receptors of the bitter taste are located at the end of the oral cavity (Baryłko-Pikielna 1975) so that they are available with difficulty; a longer time is necessary to reach the taste buds, and to wash the taste buds prior to receiving the next sample. The lack of correlation between the differences proves that there was no substantial difference in the performance in the evaluation of individual descriptors among the assessors. No statistically significant difference between the assessors was observed using the rank test (P = 0.95) on the basis of the results given in Tables 2 and 3, so that they could be regarded as a homogenous group.

Regressions between ratings of the individual sensory descriptors were essentially semilogarithmic, but very close to linear in the range studied (Table 2). The ratings between the descriptors were closely correlated (Table 3), being statistically significant (P = 0.02 or lower) in the case of all assessors.

Effect of sucrose on the sensory profile of aqueous and ethanolic solutions

All samples contained 0.01% quinine hydrochloride, differing only in the content of sucrose. Sucrose improved the acceptability in aqueous solutions in a certain range only (Table 4), connected with increasing sweetness, and decreasing

Table 2. Regression equations between sensory parameters in the case of individual assessors

Assessor's code	Regression between S and A	Regression between B and A	Regression between B and S
A	S = 61 log A – 37	B = 199 – 38 log A	B = 115 – 33 log S
B	S = 42 log A – 22	B = 99 – 23 log A	B = 105 – 26 log S
C	S = 75 log A – 71	B = 159 – 71 log A	B = 128 – 52 log S
D	S = 87 log A – 71	B = 112 – 44 log A	B = 92 – 30 log S
E	S = 54 log A – 26	B = 122 – 45 log A	B = 94 – 26 log S
F	S = 59 log A – 31	B = 143 – 65 log A	B = 154 – 66 log S
G	S = 80 log A – 64	B = 157 – 68 log A	B = 102 – 31 log S
H	S = 81 log A – 92	B = 157 – 59 log A	B = 117 – 37 log S

A = acceptability; S = sweetness; B = bitterness

Table 3. Correlation coefficients R^2 between ratings by individual assessors and the respective significant probability level (in parantheses)

Assessor's code	Number of cases	Regression between		
		A and S	A and B	S and B
A	21	0.589 (< 0.001)	0.533 (< 0.001)	0.548 (< 0.0001)
B	36	0.436 (< 0.001)	0.185 (< 0.02)	0.449 (< 0.001)
C	41	0.303 (< 0.01)	0.563 (< 0.001)	0.563 (< 0.0001)
D	42	0.518 (< 0.001)	0.706 (< 0.0001)	0.490 (< 0.0001)
E	34	0.260 (< 0.001)	0.410 (< 0.001)	0.325 (< 0.001)
F	36	0.504 (< 0.001)	0.672 (< 0.0001)	0.846 (< 0.00001)
G	35	0.504 (< 0.001)	0.640 (< 0.0001)	0.397 (< 0.001)
H	33	0.348 (< 0.001)	0.260 (< 0.01)	0.640 (< 0.0001)

A = acceptability; S = sweetness; B = bitterness

bitterness. The interactions are easily evident from the data given in Table 2 because the regression lines intersect in the case of interactions as explained by Meilgaard *et al.* (1978). The critical range was between 10–14% sucrose in the solution. In solutions containing 16% ethanol, the critical sucrose concentration was lower than 10% (Table 4). The acceptability of both aqueous and ethanolic solutions was affected by the ration of sweetness and bitterness, the regression being semilogarithmic (Figure 1).

Effect of ethanol on the sensory profile of sucrose and quinine hydrochloride solutions

In solutions containing 10% sucrose and 0.01% quinine hydrochloride, the rising concentration of ethanol did not much affect the ratings, except at higher concentrations not common in vermouths, but more approaching to those of liqueurs (Table 5). Therefore, we have not included the effect of the trigemical sense of ethanol in this study. Its effect was studied in another series of experiments (concentrations up to 45% ethanol), which is now in preparation for publication. Analogous behaviour was observed in solutions containing 18% sucrose (Table 5).

Comparison of different sweeteners in aqueous and ethanolic solutions

Sucrose, Aspartame, and Neotame were tested in aqueous solutions in the presence of 0.01% quinine hydrochloride (Table 6). In all cases, the increased

Table 4. Effect of sucrose on sensory parameters in solutions containing 0.01% quinine hydrochloride (N = 24; mean values and standard deviations of the mean values in parentheses)

Alcohol content (% vol.)	Concentration of sucrose (%)	Acceptability	Sweetness	Bitterness
		(mm of scale)		
0	0	10 (2)	4 (1)	84 (1)
	10	10 (4)	57 (3)	47 (4)
	14	65 (2)	68 (4)	32 (4)
	18	63 (2)	79 (2)	28 (3)
16	0	16 (3)	9 (1)	81 (3)
	10	52 (3)	67 (3)	41 (4)
	14	59 (3)	61 (3)	34 (2)
	18	55 (3)	70 (3)	40 (4)

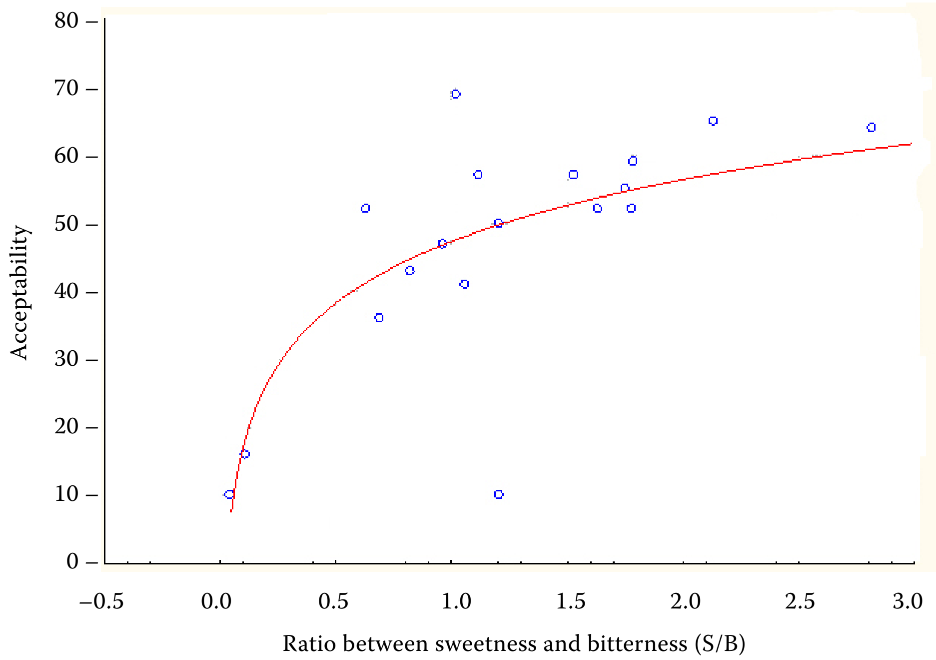

Figure 1. Relations between the ration of sweetness and bitterness and the acceptability – A = 47 + 30 log(S/B)

concentrations of the synthetic sweeteners, studied in these experiments, depressed the bitterness and increased the acceptability, similarly but not equally to sucrose. In the solutions containing 16% ethanol, the increase of acceptabilities with increasing sweetness (Table 6) was the same as in aqueous solutions, but the decrease of bitterness was about the same as in sucrose only in the case of Aspartame. On the contrary, in the solutions containing Neotame, the decrease of bitterness and the increase of sweetness were negligible, if the two concentrations of the sweeteners are compared. The difference could be attributed to the bitter taint of the sweetener Neotame, compared with sucrose of Aspartame (Šedivá *et al.* 2006).

CONCLUSIONS

The acceptability of solutions containing quinine hydrochloride as the bitter substance and a sweetener is better than in the case of a flavouring substance only, while the concentrations of ethanol common in vermouths have no substantial effect, contrary to the effect of high concentrations of ethanol (32%), common in bitter liqueurs. Sugar decreased the bitterness of quinine hydrochloride,

Table 5. Effect of ethanol on sensory parameters of aqueous solutions containing sucrose and 0.01% quinine hydrochloride (N = 24; mean values and standard deviations of the mean values in parantheses)

Concentration of sucrose (%)	Concentration of ethanol (% vol.)	Acceptability	Sweetness	Bitterness
		(mm of scale)		
10	0	50 (4)	54 (3)	47 (4)
	8	52 (1)	56 (3)	43 (3)
	16	52 (3)	67 (3)	41 (4)
	32	64 (5)	58 (5)	71 (6)
18	0	63 (2)	79 (2)	28 (3)
	8	63 (3)	71 (2)	32 (3)
	16	55 (2)	70 (3)	40 (4)
	32	68 (5)	66 (5)	67 (7)

Table 6. Effect of different sweeteners on sensory parameters of aqueous solutions containing 0.01% quinine hydrochloride (N = 24; mean values and standard deviations of the mean values in parantheses)

Content of ethanol (% vol.)	Sweeteners added	Acceptability	Sweetness	Bitterness
		(mm of scale)		
0	10% sucrose	50 (4)	57 (3)	47 (4)
	0.05% Aspartame	43 (4)	47 (3)	57 (3)
	0.09% Aspartame	57 (3)	66 (4)	43 (4)
	0.0012% Neotame	36 (4)	44 (3)	64 (4)
	0.0020% Neotame	57 (3)	46 (3)	49 (3)
16	10% sucrose	52 (3)	67 (3)	41 (4)
	0.05% Aspartame	47 (3)	52 (4)	54 (4)
	0.09% Aspartame	52 (4)	71 (2)	40 (4)
	0.0012% Neotame	41 (3)	57 (3)	54 (4)
	0.0020 % Neotame	69 (6)	57 (5)	56 (6)

and quinine hydrochloride decreased the sweetness of sucrose. Synthetic sweeteners improved the acceptability of samples, but the effect of Neotame was more moderated than the effect of Aspartame. The difference can be explained by different sensory profiles of Neotame as compared with Aspartame or sucrose (Šedivá *et al.* 2006).

References

Bakker J. (2003): Sherry. In: Caballero B., Trugo L.C., Finglas P.M. (eds): Encyclopedia of Food Sciences and Nutrition. 2nd Ed. Academic Press, Amsterdam: 5252–5257.

Baryłko-Pikielna N. (1975): Zarys analizi sensorycznej żywności. Wydawnictwa Naukowo-Techniczne, Warszawa: 171–172.

Brand J.G., Feigin A.M. (1996): Biochemistry of sweet taste transduction. Food Chemistry, **56**: 199–207.

Cardello A.V., Maller O. (1987): Psychophysical bases for the assessment of food quality. In: Kapsalis J.G. (ed.): Objective Methods in Food Quality Assessment. CRC Press, Boca Raton.

Cristovam E., Paterson A. (2003): Port. In: Caballero E., Trugo L.C., Finglas P.M. (eds): Encyclopedia of Food Sciences and Nutrition. 2nd Ed. Academic Press, Amsterdam: 4830–4844.

ČSN 75 711 (2004): Ukazatele jakosti pitné vody. Český normalizační institur, Praha.

Eggers S.C., Acree T.E., Shallenberger R.S. (2000): Sweetness chemoreception theory and sweetness transduction. Food Chemistry, **68**: 45–49.

Gini G. (1912): Variabilita e mutabilita contributo allo studio delle distributioni e delle relationi statistiche. Universita degli Studi, Bologna: 156.

Goswell R.W. (2003): Madeira and related products. In: Caballero E., Trugo L.C., Finglas P.M. (eds.): Encyclopedia of Food Sciences and Nutrition. 2nd Ed. Academic Press, Amsterdam: 3638–3640.

ISO 6658 (1985a): Sensory analysis – Methodology – General guidance. International Organization for Standardization, Geneva.

ISO 6564 (1985b): Sensory analysis –Flavour profile methods. International Organization for Standardization, Geneva.

ISO 4121 (1987): Sensory analysis – Methodology – Evaluation of food products by methods using scales. International Organization for Standardization, Geneva.

ISO 8589 (1988): Sensory analysis – General guidance for the design of test rooms. International Organization for Standardization, Geneva.

ISO 8586 (1989): Sensory analysis – General guidance for the selection, training and monitoring of assessors – selected assessors. International Organization for Standardization, Geneva.

Keast R.S.J., Breslin P.A.S. (2002): An overview of binary taste-taste interactions. Food Quality and Preference, **14**: 111–124.

Liddie P., Boero L. (2003): Vermouth. In: Caballero E., Trugo L.C., Finglas P.M. (eds): Encyclopedia of Food Sciences and Nutrition. 2nd Ed. Academic Press, Amsterdam: 5980–5184.

McBride R.L. (1985): Sensory measurement: an introductory review. CSIRO Food Research Quarterly, **45**: 59–63.

Meilgaard M., Civille G.V., Carr B.T. (1978): Sensory Evaluation Techniques. CRC Press, Boca Raton.

Norfe C., Tinti J.M. (2000): Neotame: discovery, properties, utility. Food Chemistry, **69**: 245–257.

Panovská Z., Šedivá A., Jedelská M., Pokorný J.: (2004): Interaction of bitter and sweet flavour in beverages. In: Hofmann T., Rothe M., Schieberle P. (eds): State-of-the-Art in Flavor Chemistry and Biology. Proceedings 7th Wartburg Symposium on Flavour Chemistry and Biology, 21–23 April 2004. Deutsche Forschungsanstalt für Lebensmittelchemie, Eisenach: 404–407.

Pokorný J. (2004): Effect of flavour interaction on the flavour acceptability of foods and beverages. In: Hofmann T., Rothe M., Schieberle P. (eds): State-of-the-Art in Flavor Chemistry and Biology. Proceedings 7th Wartburg Symposium on Flavour Chemistry and Biology, 21–23 April 2004. Deutsche Forschungsanstalt für Lebensmittelchemie, Eisenach: 146–151.

Schiffman S.S., Booth B.J., Losoe M.L., Pecore S.D., Warwick Z.S. (1995): Interactions of bitter and sweet tastes. Brain Research Bulletin, **36**: 505–513.

Šedivá A., Panovská Z., Pokorný J. (2006): Sensory profiles of sweetness in aqueous solutions. Czech Journal of Food Sciences, **24**: 283–287.

Vyhláška č. 252/20044 Ministerstva zdravotnictví ČR (Guideleines of the Ministry of Health of the Czech Republic) z 22. 4. 2004.

Walters D.E. (1996): How are bitter and sweet taste related? Trends in Food Science and Technology, **7**: 399–403.

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