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Effect of temperature, pH, and ions on sweet taste

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Abstract

The purpose of this experiment was to determine the effects of temperature (50°C and 6°C), pH (pH 3.0, 4.0, 5.0, 6.0, and 7.0) and the addition of monovalent and divalent cations (5 mM Na^+ , 5 mM K^+ , and 5 mM Ca^{2+}) on the sweetness intensity ratings of sweeteners ranging widely in chemical structure. A trained panel provided intensity evaluations for prototypical tastes (sweet, bitter, sour, and salty) as well as aromatic and mouth-feel attributes. The following sweeteners were included in this experiment: three sugars (fructose, glucose, sucrose), three terpenoid glycosides (monoammonium glycyrrhizinate, rebaudioside-A, stevioside), two polyhydric alcohols (mannitol, sorbitol), two dipeptide derivatives (alitame, aspartame), two *N*-sulfonylamides (acesulfame-K, sodium saccharin), one sulfamate (sodium cyclamate), one protein (thaumatin), one dihydrochalcone (neohesperidin dihydrochalcone), and one chlorodeoxysugar (sucralose). Two to five levels of each sweetener reflecting a range of sweetness intensities were tested, using formulae developed by DuBois et al. The main finding from this three-part study was that temperature, pH, and ions had little effect on perceived sweetness intensity. Even when significant differences were found in the temperature study, the effects were very small.

Keywords: Sweetness; Temperature; pH; Ions; Taste

1. Introduction

During the process of eating and drinking, taste receptors are exposed to foods and beverages that range widely in temperature, pH, and chemical composition. Comprehensive studies have not been performed, however, to determine the effect of these factors on the taste of sweeteners that vary widely in chemical structure.

Previous studies of the effect of temperature on sweetness have used only a limited number of sweeteners, and the results have been equivocal. Calvino [2] reported that the sweetness of sucrose solutions was greater at 37°C or 50°C than at 7°C for low concentrations (3% w/v), but this effect tended to disappear with increasing concentrations. Paulus and Reisch [3] found that thresholds for sucrose were lowest in the temperature range from 20°C to 40°C; there was a slight increase in threshold at 10°C with larger increases in thresholds (up to 35%) at 60°C. However, at higher concentrations (18% w/v), sweetness intensity was largely independent of concentration. McBurney et al. [3] found that taste thresholds for dulcin were lowest between 22°C and 32°C, and rose above and below this temperature range. Bartoshuk et al. [4] concluded that lower concentrations of sucrose were judged to gain in sweetness as temperature increased; this effect finally became negligible at about 0.5 M. Green and Frankmann [5] found that low concentrations of aspartame were perceived as less intense at 20°C than 36°C. In other studies, however, solution temperature was not found to affect the sweetness intensity of glucose [5,6] or fructose [5,6].

The effect of pH on sweetness has received little systematic treatment. Stone et al. [6] found that reducing pH from 5.8 to 4.0 had little effect on the sweetness of glucose and fructose, but reduction from 4.0 to 2.7 caused a 50% reduction in relative sweetness. McBride and Finlay [7] found that citric acid (0.006 M and 0.05 M) mixed with fructose (0.259 M and 1.2 M) suppressed the perceived sweetness; however, citric acid had no effect when mixed with sucrose (0.172 M and 0.8 M). The pH of the mixtures was not given. Stevens [8] found that weak concentrations of citric acid (e.g., below 10^{-3} M) had little effect on sucrose thresholds, while higher concentrations (2×10^{-3} M and above) slightly elevated sucrose thresholds; pH values were not reported.

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The effect of ions on sweet taste is also unclear. The sweetness of weak concentrations of sucrose have been reported to be enhanced when mixed with weak concentrations of NaCl; this may be explained, in part, by the fact that weak concentrations of NaCl are sometimes perceived as sweet [9]. Moskowitz [10], however, found in human psychophysical studies that mixtures of higher concentrations of sweet (glucose or fructose) or salty (NaCl) compounds developed an "unblended" or "clashing" taste in which the components alternated in attempting to dominate the perception. The biochemistry of taste suggests that salts should play some role in sweet taste because channels in taste cell membranes that transport sodium [11,12] or K^+ [13–15] play a role in sweet taste transduction. It was hypothesized in the present study that Na^+ and Ca^{2+} ions, which are found in high concentrations in extracellular fluid, would have different effects than K^+ , which is found intracellularly in high concentrations, on the flavor profiles of 11 sweeteners.

One reason that the experiments described above did not reach consistent conclusions may be that most subjects were untrained tasters, i.e., subjects who were not trained to make fine discriminations between taste qualities and intensities. The purpose of this three-part study was to investigate the effect of temperature (part 1), pH (part 2), and the addition of mono- and divalent cations, Na^+ , K^+ , and Ca^{2+} (part 3), on perceived sweetness using a trained taste panel. The effect of these factors on a variety of sweeteners that ranged widely in chemical structure was examined.

2. Materials and methods

2.1. Subjects

A trained panel of 18 subjects, 9 males and 9 females, participated in the study. The minimum number of subjects who participated in any given session was 8, and the maximum number of subjects was 17. The mean age of the subjects was 48 ± 4 years. All subjects were from the Duke University or Durham, NC, community. Subjects were paid for their participation.

2.2. Stimuli

In this three-part experiment, a total of 16 sweeteners were tested: three sugars (fructose, glucose, sucrose), three terpenoid glycosides (monoammonium glycyrrhizinate, rebaudioside-A, stevioside), two dipeptide derivatives (alitame, aspartame), two *N*-sulfonylamides (acesulfame-K, sodium saccharin), two polyhydric alcohols (mannitol, sorbitol), one dihydrochalcone (neohesperidin dihydrochal-

Table 1
Concentrations of 15 sweetners tested in the temperature and pH studies

Sweetener	2.5 Sweet level	5 Sweet level	7.5 Sweet level	10 Sweet level
Acesulfame-K	129.12 ppm (6.42×10^{-4} M)	356.06 ppm (1.77×10^{-3} M)	859.76 ppm (4.27×10^{-3} M)	2,937.50 ppm (0.0146 M)
Alitame	5.78 ppm (1.74×10^{-5} M)	14.58 ppm (4.40×10^{-5} M)	29.57 ppm (8.92×10^{-5} M)	60.87 ppm (1.84×10^{-4} M)
Aspartame	103.70 ppm (3.52×10^{-4} M)	254.55 ppm (8.65×10^{-4} M)	494.12 ppm (1.68×10^{-3} M)	933.33 ppm (3.17×10^{-3} M)
Fructose	19,400 ppm (0.108 M)	39,100 ppm (0.217 M)	58,700 ppm (0.326 M)	78,400 ppm (0.435 M)
Glucose	42,000 ppm (0.233 M)	83,700 ppm (0.465 M)	125,300 ppm (0.695 M)	167,000 ppm (0.927 M)
MAG	109.38 ppm (1.30×10^{-4} M)	456.52 ppm (5.44×10^{-4} M)	[a]	[a]
Mannitol	43,700 ppm (0.24 M)	78,300 ppm (0.43 M)	113,000 ppm (0.62 M)	149,000 ppm (0.82 M)
Na cyclamate	894.62 ppm (4.45×10^{-3} M)	1,583.10 ppm (7.87×10^{-3} M)	2,626.10 ppm (0.0131 M)	5,591.50 ppm (0.0278 M)
Na saccharin	48.51 ppm (2.36×10^{-4} M)	112.59 ppm (5.49×10^{-4} M)	303.06 ppm (1.48×10^{-3} M)	[a]
Neo-DHC	18.15 ppm (2.96×10^{-5} M)	55.21 ppm (9.01×10^{-5} M)	172.83 ppm (2.82×10^{-4} M)	[a]
Rebaudioside-A	66.67 ppm (6.91×10^{-5} M)	200.00 ppm (2.07×10^{-4} M)	600.00 ppm (6.22×10^{-4} M)	[a]
Sorbitol	56,500 ppm (0.31 M)	85,600 ppm (0.47 M)	118,000 ppm (0.65 M)	159,000 ppm (0.87 M)
Stevioside	138.51 ppm (1.72×10^{-4} M)	418.37 ppm (5.20×10^{-4} M)	1,281.30 ppm (1.59×10^{-3} M)	[a]
Sucrose	25,000 ppm (0.0730 M)	50,000 ppm (0.146 M)	75,000 ppm (0.219 M)	100,000 ppm (0.292 M)
Thaumatin	1.18 ppm (5.36×10^{-8} M)	3.53 ppm (1.60×10^{-7} M)	10.38 ppm (4.72×10^{-7} M)	360.00 ppm (1.64×10^{-5} M)

[a] Indicates sweetener does not achieve this sweetness intensity level.

Table 2
Concentrations (and expected sweetness intensity responses[a]) of the five levels of 11 sweeteners tested in the ion study

Sweetener	Level 1	Level 2	Level 3	Level 4	Level 5
Acesulfame-K	97.9 ppm (2) (4.87×10^{-4} M)	247.2 ppm (4) (1.23×10^{-3} M)	503.6 ppm (6) (2.50×10^{-3} M)	1,044.4 ppm (8) (5.19×10 M)	2,937.5 ppm (10) (0.0143 M)
Aspartame	180 ppm (4) (6.12×10^{-4} M)	340 ppm (6) (1.16×10^{-3} M)	560 ppm (8) (1.90×10^{-3} M)	900 ppm (10) (3.06×10^{-3} M)	1,600 ppm (12) (5.44×10^{-3} M)
Fructose	23,300 ppm (3) (0.129 M)	46,900 ppm (6) (0.260 M)	62,700 ppm (8) (0.348 M)	78,400 ppm (10) (0.435 M)	94,200 (12) (0.523 M)
Glucose	50,300 ppm (3) (0.279 M)	100,300 ppm (6) (0.557 M)	133,700 ppm (8) (0.742 M)	167,000 ppm (10) (0.927 M)	200,300 ppm (12) (1.11 M)
Mannitol	50,700 ppm (3) (0.278 M)	92,700 ppm (6) (0.509 M)	121,000 ppm (8) (0.662 M)	149,000 ppm (10) (0.816 M)	162,000 ppm (11) (0.890 M)
Na cyclamate	1,022.7 ppm (3) (5.08×10^{-3} M)	1,583.1 ppm (5) (7.87×10^{-3} M)	2,359.6 ppm (7) (0.0117 M)	3,841.0 ppm (9) (0.0191 M)	13,314.1 ppm (11) (0.0662 M)
Na saccharin	40 ppm (2) (1.95×10^{-4} M)	80 ppm (4) (3.90×10^{-4} M)	120 ppm (5) (5.85×10^{-4} M)	240 ppm (7) (1.17×10^{-3} M)	375 ppm (8) (1.83×10^{-3} M)
Rebaudioside-A	50 ppm (2) (5.18×10^{-5} M)	133 ppm (4) (1.38×10^{-4} M)	300 ppm (6) (3.11×10^{-4} M)	800 ppm (8) (8.29×10^{-4} M)	1,800 ppm (9) (1.87×10^{-3} M)
Sorbitol	62,500 ppm (3) (0.343 M)	98,800 ppm (6) (0.542 M)	126,000 ppm (8) (0.694 M)	158,000 ppm (10) (0.869 M)	197,000 ppm (12) (1.080 M)
Sucralose	46.55 ppm (3) (1.17×10^{-4} M)	98.53 ppm (6) (2.48×10^{-4} M)	153.88 ppm (8) (3.87×10^{-4} M)	259.94 ppm (10) (6.54×10^{-4} M)	648.99 ppm (12) (1.63×10^{-3} M)
Sucrose	30,000 ppm (3) (0.0876 M)	60,000 ppm (6) (0.175 M)	80,000 ppm (8) (0.234 M)	100,000 ppm (10) (0.292 M)	120,000 ppm (12) (0.351 M)

[a] DuBois et al. [16].

cone), one protein (thaumatin), one chlorodeoxysugar (sucralose), and one sulfamate (sodium cyclamate). Fifteen of the 16 sweeteners were evaluated in parts 1 and 2. Eleven of the 16 sweeteners were evaluated in part 3. All compounds tested in this experiment were dissolved in deionized water. Abbreviations are used for some of the sweeteners in this article: monoammonium glycyrrhizinate (MAG), sodium cyclamate (Na cyclamate), sodium saccharin (Na saccharin), and neohesperidin dihydrochalcone (neo-DHC).

For the evaluations of the effects of temperature and pH, each sweetener was tested at concentrations equivalent with 2.5%, 5%, 7.5%, and 10% sucrose using formulae developed by DuBois et al. [16]. Five sweeteners (MAG, neo-DHC, rebaudioside-A, Na saccharin, and stevioside), however, were not tested at concentrations isointense with 10% sucrose because they do not reach this sweetness intensity level. Also, MAG was not tested at the concentration equivalent with 7.5% sucrose for this same reason. Table 1 gives all the sweetener concentrations tested in both temperature and pH evaluations.

In the temperature evaluation (part 1), each sweetener was titrated to pH 5.0 using aqueous solutions of hydrochloric acid (HCl) or sodium hydroxide (NaOH). Warm-temperature solutions were presented at 50°C, and cold-temperature solutions were presented at 6°C. Warm and cold solutions were maintained at the appropriate temperatures by a warm-water bath and the laboratory foodstuffs refrigerator, respectively. In evaluations of the effect of pH (part 2), each sweetener was titrated to pH 3.0, 4.0, 5.0, 6.0, and 7.0, also using aqueous solutions of HCl or NaOH. Initial pH values for the 15 sweeteners tested ranged from 3.84 to 7.67. Solutions in the pH study were presented at room temperature (22°C).

For evaluations of the effect of ions (part 3), sweeteners were tested alone and in combination with three salts, each tested individually at 5 mM: sodium chloride (NaCl), potassium chloride (KCl), and calcium chloride ($CaCl_2$). Each sweetener was tested at five concentrations that covered the range of sweetness intensity achieved by each sweetener. Expected sweetness intensities reached for a given concentration of sweetener were determined according to the formulae developed by DuBois et al. [16]. Table 2 lists the concentrations of sweeteners tested in part 3 of this study; for each concentration, the expected sweetness intensity level expressed in sucrose equivalents is given in parenthesis.

2.3. Procedure

Prior to receiving the experimental samples, each subject tasted references at room temperature (22°C) acording to the methods used by DuBois et al. [16]. The sweet taste references were as follows: 2 sweet (2% sucrose), 5 sweet (5% sucrose), 7.5 sweet (7.5% sucrose), 10 sweet (10% sucrose), 12 sweet (12% sucrose), and 15 sweet (16% sucrose). Panelists also tasted bitter taste references labeled 2 bitter (0.02% caffeine) and 4 bitter (0.03% caffeine), a sour reference labeled 2 sour (0.01% citric acid) and a salty reference labeled 2 salty (0.2% NaCl). Bitter, sour, and salty references were based upon previous evaluations made by the trained panel. In addition, panel members have been trained to evaluate the quality and intensity of a variety of aromatic and mouth-feel attributes.

Table 3
Mean sweetness intensity ratings (with standard errors) for each concentration of 15 sweeteners at two temperatures in the temperature study

Sweetener	Concentration	Warm	Cold	Warm versus Cold
Acesulfame-K				
(2.5)[a]	129.12 ppm (6.42×10^{-4} M)	3.37 (0.79)	1.86 (0.42)	
(5)	356.06 ppm (1.77×10^{-3} M)	5.65 (0.84)	4.59 (0.60)	
(7.5)	859.76 ppm (4.27×10^{-3} M)	6.78 (1.07)	7.09 (0.78)	
(10)	2,937.50 ppm (0.0146 M)	7.80 (1.07)	7.89 (1.14)	
Alitame				
(2.5)	5.78 ppm (1.74×10^{-5} M)	2.68 (0.40)	1.06[b] (0.24)	[c]
(5)	14.58 ppm (4.40×10^{-5} M)	5.12 (0.43)	4.38 (0.37)	
(7.5)	29.57 ppm (8.92×10^{-5} M)	7.61 (0.54)	7.18 (0.47)	
(10)	60.87 ppm (1.84×10^{-4} M)	10.56 (0.67)	10.32 (0.63)	
Aspartame				
(2.5)	103.70 ppm (3.52×10^{-4} M)	3.68[b] (0.55)	1.63 (0.35)	[c]
(5)	254.55 ppm (8.65×10^{-4} M)	6.63[b] (0.79)	3.80 (0.65)	[c]
(7.5)	494.12 ppm (1.68×10^{-3} M)	9.55[b] (0.83)	7.6 (0.75)	
(10)	933.33 ppm (3.17×10^{-3} M)	10.13 (0.85)	10.20 (0.96)	
Fructose				
(2.5)	19,400 ppm (0.108 M)	2.91 (0.42)	2.21 (0.45)	
(5)	39,100 ppm (0.217 M)	4.48 (0.52)	4.50 (0.43)	
(7.5)	58,700 ppm (0.326 M)	6.24[b] (0.61)	7.61 (0.49)	
(10)	78,400 ppm (0.435 M)	7.61[b] (0.55)	8.79[*] (0.57)	
Glucose				
(2.5)	42,000 ppm (0.233 M)	2.95 (0.54)	1.45[b] (0.27)	[c]
(5)	83,700 ppm (0.465 M)	5.17 (0.52)	3.78[b] (0.44)	
(7.5)	125,300 ppm (0.695 M)	7.89 (0.59)	7.13 (0.84)	
(10)	167,000 ppm (0.927 M)	10.29 (0.71)	9.12 (0.79)	
MAG				
(2.5)	109.38 ppm (1.30×10^{-4} M)	3.99[b] (0.48)	2.45 (0.39)	[c]
(5)	456.52 ppm (5.44×10^{-4} M)	7.75[b] (0.56)	5.84 (1.04)	
Mannitol				
(2.5)	43,700 ppm (0.24 M)	2.38 (0.46)	0.95[b] (0.35)	[c]
(5)	78,300 ppm (0.43 M)	4.35 (0.43)	3.08[b] (0.39)	[c]
(7.5)	113,000 ppm (0.62 M)	7.01 (0.40)	6.25[b] (0.45)	
(10)	149,000 ppm (0.82 M)	8.61[b] (0.49)	8.65[b] (0.44)	
Na cyclamate				
(2.5)	894.62 ppm (4.45×10^{-3} M)	3.12 (0.47)	2.56 (0.45)	
(5)	1,583.10 ppm (7.87×10^{-3} M)	5.12 (0.54)	4.86 (0.48)	
(7.5)	2,626.10 ppm (0.0131 M)	6.64 (0.65)	8.02 (0.60)	
(10)	5,591.50 ppm (0.0278 M)	8.68 (0.64)	11.16 (0.67)	
Na saccharin				
(2.5)	48.51 ppm (2.36×10^{-4} M)	2.61 (0.40)	2.04 (0.38)	
(5)	112.59 ppm (5.49×10^{-4} M)	4.05 (0.43)	4.70 (0.62)	
(7.5)	303.06 ppm (1.48×10^{-3} M)	6.56 (0.53)	7.60 (0.71)	
Neo-DHC				
(2.5)	18.15 ppm (2.96×10^{-5} M)	4.55[b] (0.77)	4.28[b] (0.50)	
(5)	55.21 ppm (9.01×10^{-5} M)	7.61[b] (0.95)	7.50[b] (0.96)	
(7.5)	172.83 ppm (2.28×10^{-4} M)	9.08 (0.84)	10.20[b] (0.79)	
Rebaudioside-A				
(2.5)	66.67 ppm (6.91×10^{-5} M)	3.96[b] (0.53)	2.40 (0.27)	[c]
(5)	200.00 ppm (2.07×10^{-4} M)	8.18[b] (0.64)	5.43 (0.71)	[c]
(7.5)	600.0 ppm (6.22×10^{-4} M)	9.44[b] (0.53)	8.90[b] (0.71)	
Sorbitol				
(2.5)	5,6500 ppm (0.31 M)	3.79[b] (0.52)	2.40 (0.32)	[c]
(5)	85,600 ppm (0.47 M)	5.51 (0.54)	4.82 (0.62)	
(7.5)	118,000 ppm (0.65 M)	7.82 (0.71)	7.36 (0.74)	
(10)	159,000 ppm (0.65 M)	8.87 (0.57)	9.68 (0.60)	
Stevioside				
(2.5)	138.51 ppm (1.72×10^{-4} M)	5.38[b] (0.64)	3.94[b] (0.60)	
(5)	418.37 ppm (5.20×10^{-4} M)	6.90[b] (0.65)	7.20[b] (0.60)	
(7.5)	1,281.30 ppm (1.59×10^{-3} M)	8.27 (0.83)	8.72 (0.85)	
Sucrose				
(2.5)	25,000 ppm (0.0730 M)	3.54[b] (0.48)	2.26 (0.39)	
(5)	50,000 ppm (0.146 M)	5.80 (0.53)	5.05 (0.43)	
(7.5)	75,000 ppm (0.219 M)	6.98 (0.41)	7.38 (0.55)	
(10)	100,000 ppm (0.292 M)	8.90[b] (0.60)	8.33[b] (0.43)	
Thaumatin				
(2.5)	1.18 ppm (5.36×10^{-8} M)	3.43 (0.56)	4.31[b] (0.81)	
(5)	3.53 ppm (1.60×10^{-7} M)	6.79[b] (0.87)	6.06 (0.79)	
(7.5)	10.38 ppm (4.72×10^{-7} M)	8.83 (0.98)	7.20 (1.08)	
(10)	360.00 ppm (1.64×10^{-5} M)	13.39[b] (0.54)	12.56[b] (0.90)	

[a] Values in parentheses indicate sucrose equivalencies.
[b] Indicates that ratings are significantly different from the expected response.
[c] Significant difference between ratings of warm and cold temperature conditions.

Panelists were asked to give sweetness intensity ratings as well as ratings of the other prototypical tastes (bitter, sour, and salty), and other aromatic and mouth-feel notes of experimental samples at each taste session. The panelists were served 15 mL of solution of each sample in 30-mL plastic medicine cups. Samples were coded with random three-digit numbers for identification. Subjects rinsed their mouths thoroughly with deionized water and, if necessary, ate unsalted top crackers to rid their mouths as much as possible of lingering attributes of previous samples. In addition, panelists refrained from smoking, eating, or drinking anything other than water for 30 min prior to the tasting session.

2.4. Temperature study (part 1)

At a given taste panel, 1 sweetener of the 15 sweeteners (see Table 1) at all possible intensity levels (2.5%, 5%, 7.5%, and 10% sucrose equivalencies) was tested at either the warm (50°C) or the cold (6°C) temperature. The order of the samples presented in a given taste session was randomized across panelists. Panelists were instructed to evaluate samples immediately upon receiving them to ensure the accuracy of the temperature at the time of evaluation.

During a given taste session, panelists were instructed to swirl the samples around in their mouths for 10 s and then to expectorate. Immediately following expectoration of a sample, panelists completed a full flavor profile of the sample on laptop computer units using CSA software (Computerized Sensory Analysis, Version 4.3, Compusense Inc., 1994). In doing a flavor profile of an experimental sample, subjects rated the intensity of sweet, bitter, sour, salty, licorice, papery, metallic, and astringent attributes. All other notes, including aromatic and mouth-feel attributes, as well as comments, were entered on a comments screen. Subjects also entered the time of onset of maximum sweetness intensity by choosing either none (no sweetness perceived), early, early-middle, middle, middle-late, or late. Intensity of attributes was noted by making a mark with a light-pen on a line scale that was anchored at 0, 1.5, 3, 4.5, 6, 7.5, 9, 10.5, 12, 13.5, and 15. Between experimental samples, there was a 2-min time delay before the computer program continued to the next sample.

2.5. pH Study (part 2)

At a given taste session, 1 of the 15 sweeteners (see Table 1) at one of the four intensity levels (2.5%, 5%, 7.5%, and 10% sucrose equivalencies) was tested at all five pH levels—pH 3.0, 4.0, 5.0, 6.0, and 7.0. The order of the five samples in a given taste session was randomized across panelists. Panelists were asked to give sweetness intensity ratings in addition to ratings of other taste, flavor, and mouth-feel notes of every sample at each session. Samples were presented at room temperature (22°C).

During a given taste session, panelists were instructed to swirl the samples around in their mouths for 10 s, and then to expectorate. Immediately following expectoration of a sample panelists completed a full flavor profile of the sample on a paper answer sheet (this study was completed prior to obtaining Computerized Sensory Analysis, Version 4.3, Compusense Inc., 1994). In doing a flavor profile of an experimental sample, subjects rated all tastes, aromatics, and mouth-feel factors. Subjects indicated all factors (as well as intensity) that they perceived. Intensity was noted by making a mark on a 15-cm line scale that was anchored at 0, 5, 10, and 15 cm. Panelists would then measure the mark using a 15-cm ruler. Subjects also indicated the time of onset of maximum sweetness intensity by circling either early, middle, or late. Between experimental samples, panelists were instructed to wait at least 2 min before continuing to the next sample.

2.6. Ions study (part 3)

Five concentrations of eleven sweeteners (see Table 2) were tested in solutions alone and in combination with chloride salts of Na^+ , K^+ and Ca^{2+}. At a given taste panel, one sweetener (at five concentrations) by itself or mixed with one of the salts was presented. The order of the five samples in a given taste session was randomized across panelists. Subjects also evaluated each of the three salts alone (not mixed with a sweetener).

During a given taste session, panelists were instructed to swirl the samples around in their mouths for 10 s and then to expectorate. Immediately following expectoration of a sample, panelists completed a full flavor profile of the sample on Toshiba laptop computer units using CSA software (Computerized Sensory Analysis, Version 4.3, Compusense Inc., 1994). In doing a flavor profile of an experimental sample, subjects rated the intensity of sweet, bitter, sour, salty, papery, fruity, licorice, metallic, viscous, and astringent attributes. All other notes, including aromatic and mouth-feel factors, as well as comments, were entered on a comments screen. Subjects also entered the time of onset of maximum sweetness intensity by choosing either none (no sweetness perceived), early, early-middle, middle, middle-late, or late. Intensity of attributes was noted by making a mark with a light-pen on a 15-point line scale that was anchored at 0, 1.5, 3, 4.5, 6, 7.5, 9, 10.5, 12, 13.5, and 15. Between experimental samples, there was a 2-min time delay before the CSA program continued to the next sample.

3. Results

3.1. Temperature study (part 1)

An analysis of variance (ANOVA) was performed to determine the effect of temperature (warm, cold) on the sweetness intensity ratings of a trained panel. The observed sweet responses at each concentration of sweetener in both temperature conditions were compared with the expected responses (i.e., 2.5, 5, 7.5, or 10) for each concentration of sweetener based upon the aforementioned formulae [16].

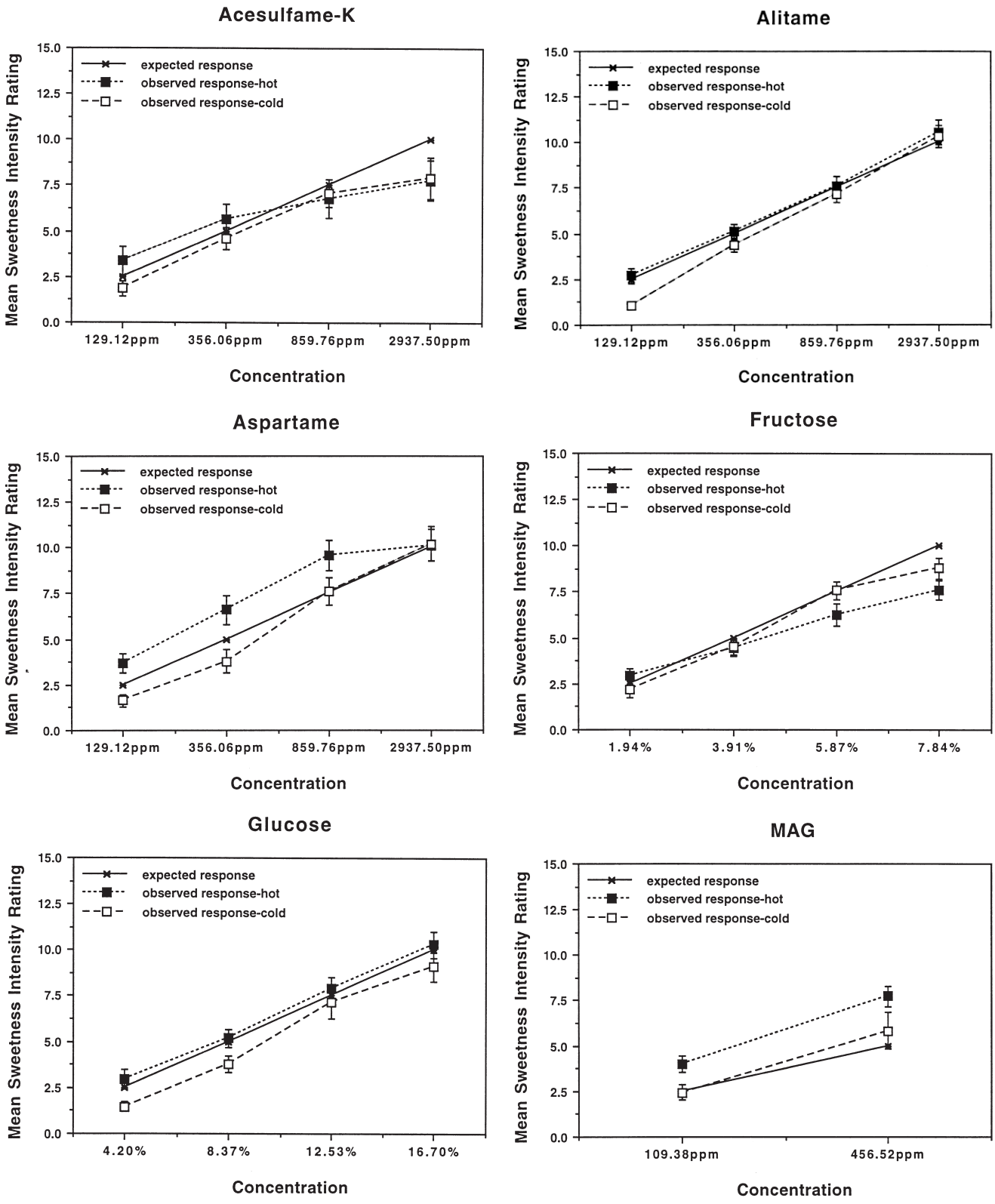

Fig. 1. Sections a–o depict the mean sweet responses for both warm and cold conditions, as well as the expected response, at each concentration of all 15 sweeteners, respectively.

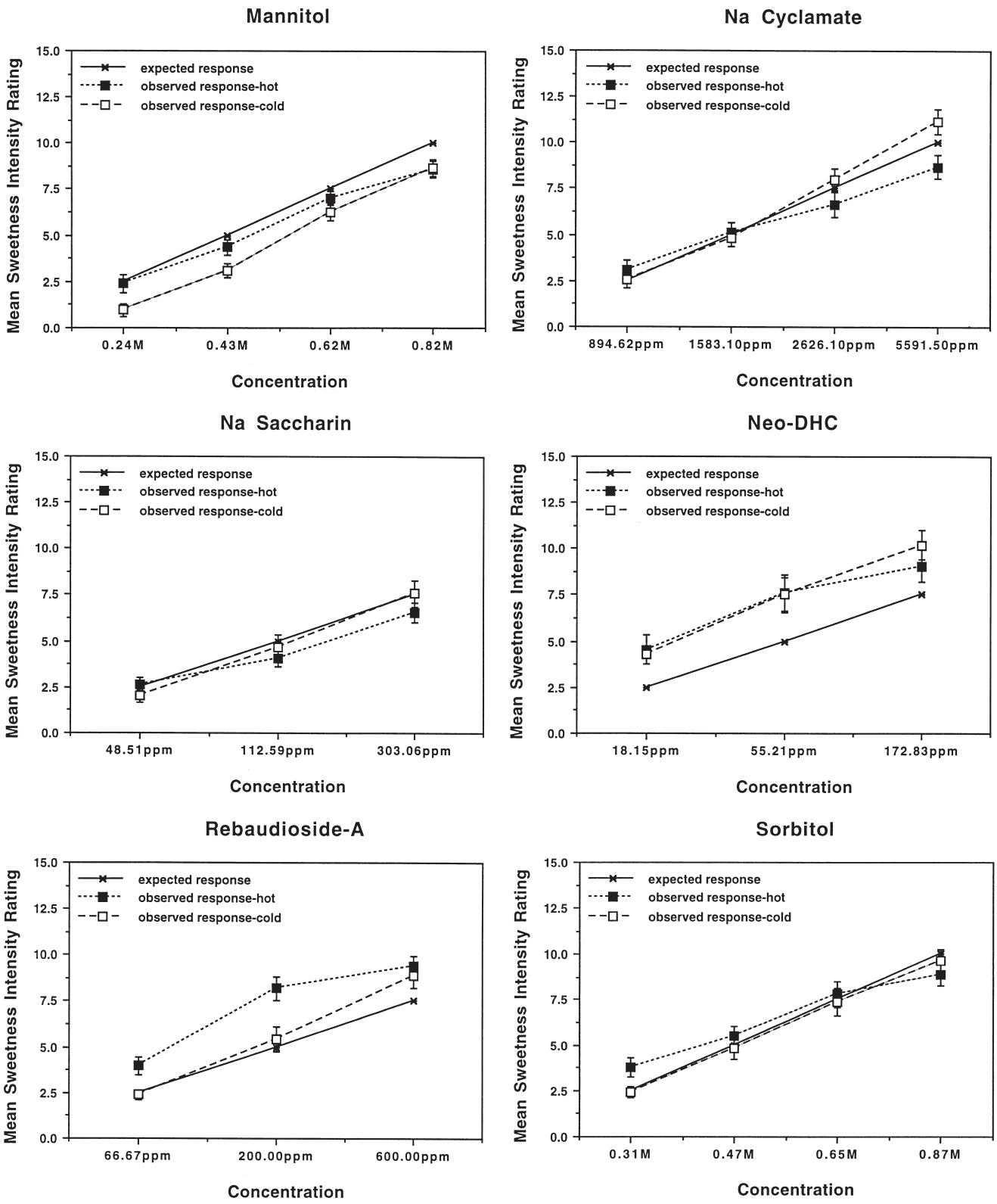

Fig. 1. (*continued*).

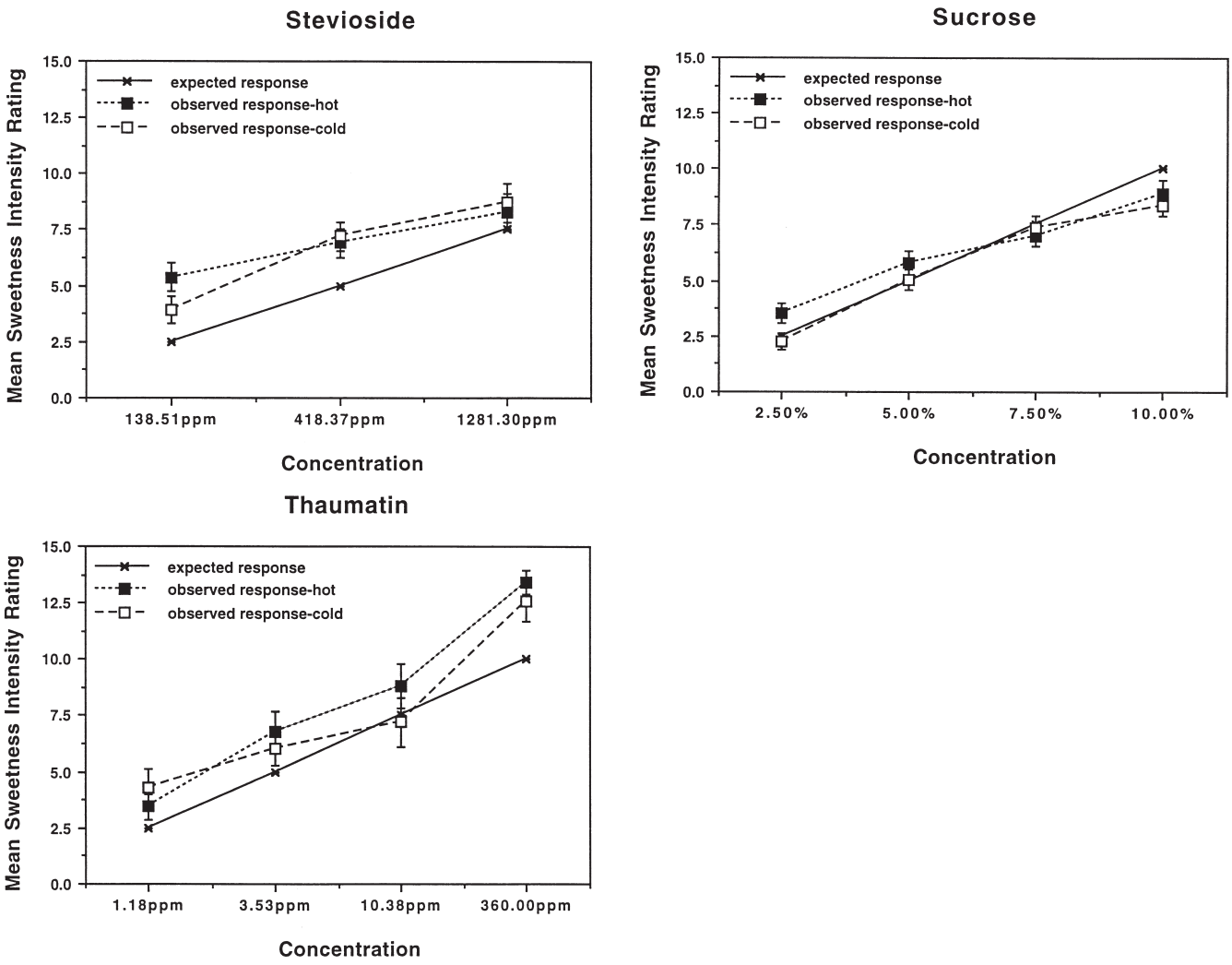

Fig. 1. (*continued*).

The analysis also determined whether there were significant differences in sweetness intensity between the warm versus cold conditions. Table 3 gives the observed mean sweetness intensity ratings and standard errors of each concentration tested of all 15 sweeteners in warm and cold conditions. Means with an "b" indicate that a statistically significant difference was found between those sweet responses and the expected response given in column 1 in parentheses. In addition, a "c" is given for those cases where the sweet responses for warm and cold conditions of a given concentration of sweetener are statistically different from each other. Figure 1a–o depicts the mean sweet responses for both warm and cold conditions, as well as the expected response, at each concentration of all 15 sweeteners, respectively.

Sweet ratings for samples presented at the warm temperature were significantly greater (at $p < 0.05$) than those served at the cold temperature at the lowest level (equivalent to 2.5% sucrose) for alitame, aspartame, glucose, monoammonium glycyrrhizinate (MAG), mannitol, rebaudioside-A, and sorbitol. This was also true at the next lowest sweet level (equivalent to 5% sucrose) for aspartame, mannitol, and rebaudioside-A. In only one instance was a sweet rating higher at the cold temperature, and that was for Na cyclamate at the highest level (equivalent to 10% sucrose). Sweetness intensity ratings tended to be enhanced (i.e., greater than expected) by warm temperatures at the lower levels of sweetness for aspartame, MAG, neo-DHC, rebaudioside-A, and stevioside. Enhancement of sweet responses was also seen at the cold temperatures for neo-DHC, stevioside, and thaumatin. Significant suppression of sweet responses was found at cold temperatures for some levels of alitame, fructose, glucose, and mannitol.

3.2. pH study (part 2)

An analysis of variance (ANOVA) was performed to determine the effect of pH (3.0, 4.0, 5.0, 6.0, or 7.0) on the sweetness intensity ratings and other descriptors by the trained panel. The analysis looked for significant differences in sweetness intensity between the pH levels. No sta-

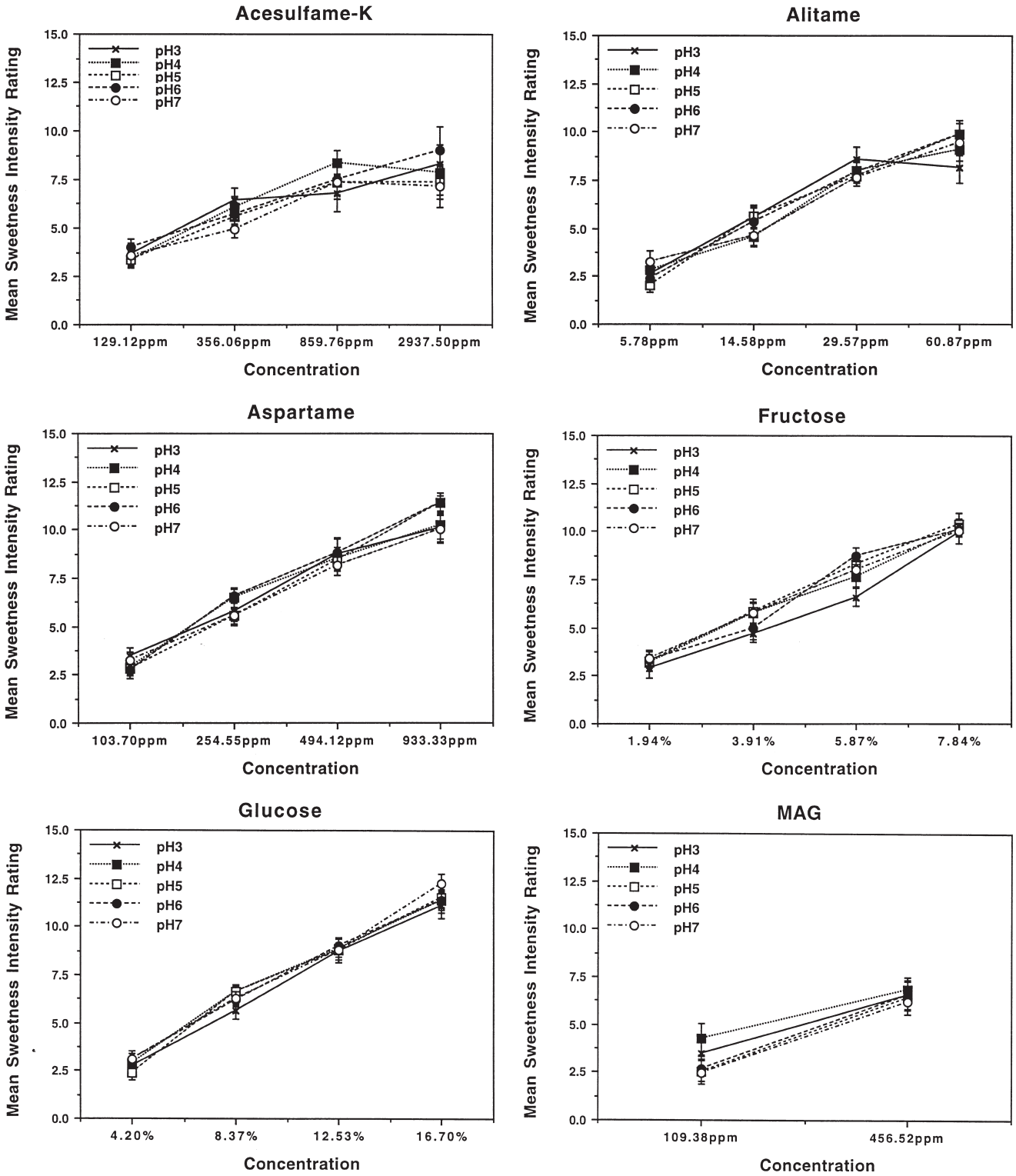

Fig. 2. Sections a–o depict the mean sweet responses for each pH level at each concentration of all 15 sweeteners, respectively.

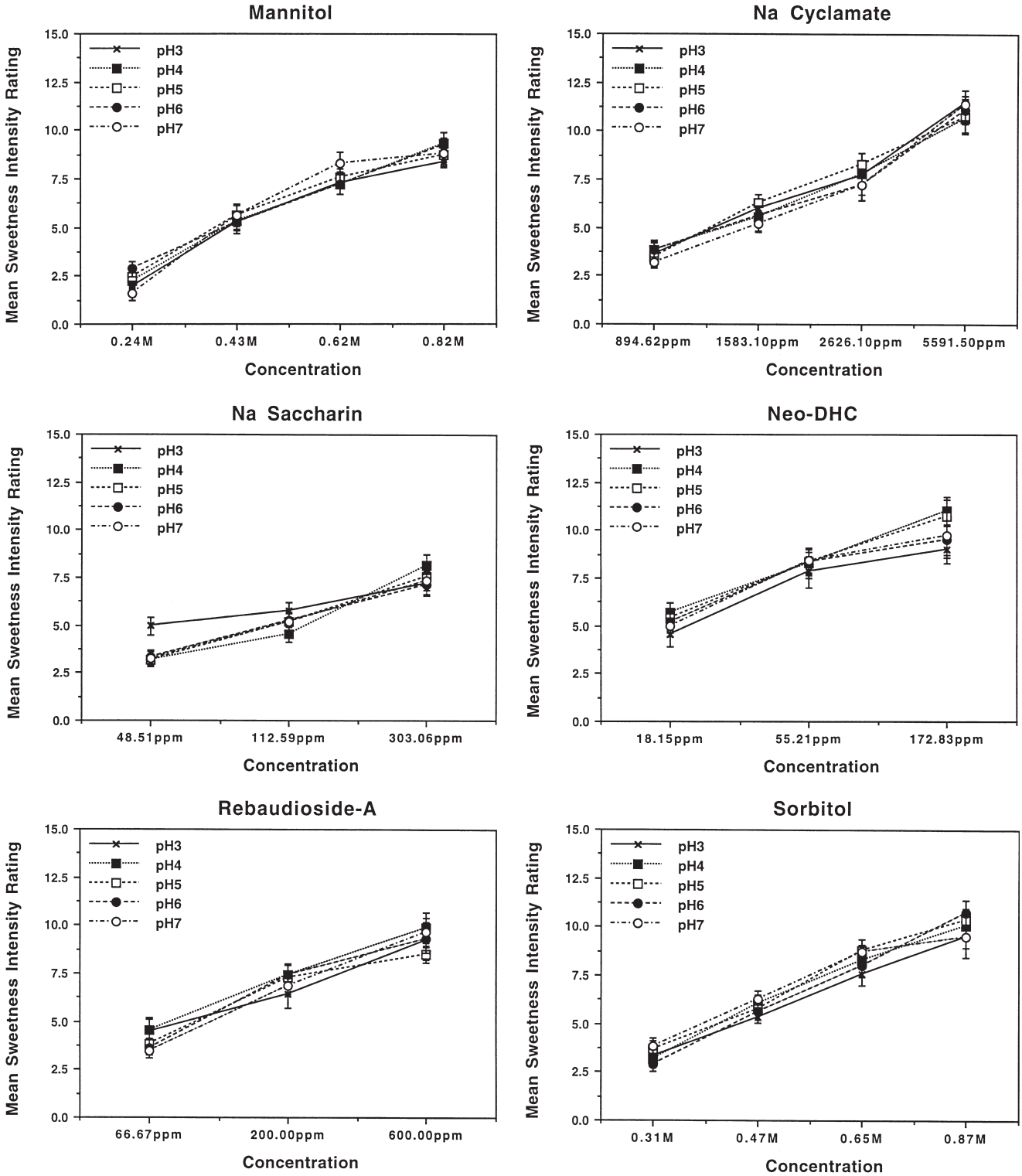

Fig. 2. (*continued*).

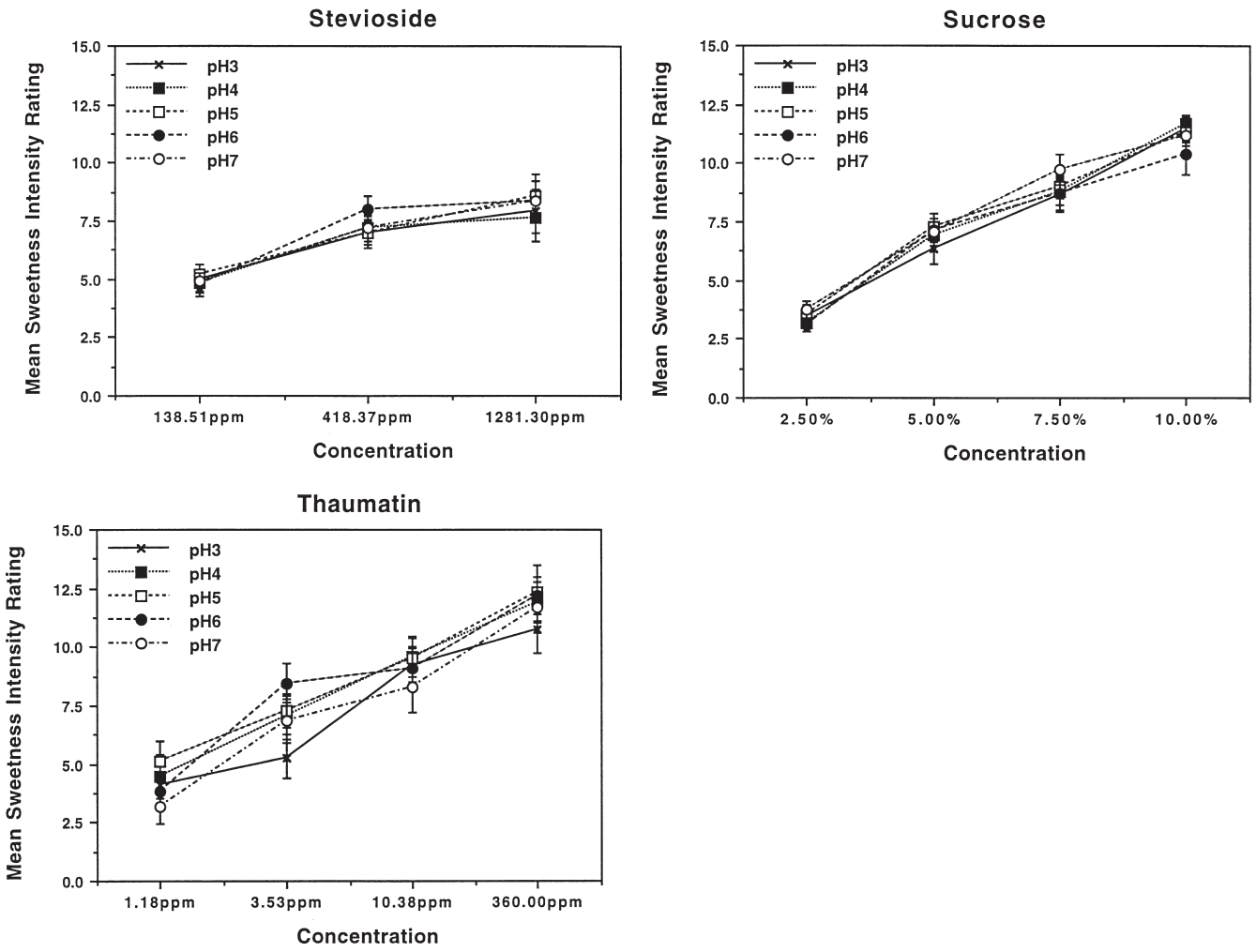

Fig. 2. (*continued*).

tistical differences were found between the sweetness intensity ratings of the five pH levels. Figure 2a–o depict the mean sweet responses for each pH level at each concentration of all 15 sweeteners. For most sweeteners significant differences were found in intensity ratings for sour and astringency between the pH levels. Both sourness and astringency increased with decreasing concentration.

3.3. Ions study (part 3)

It was hypothesized in the present study that Na^+ and Ca^{2+} ions, which are found in high concentrations in extracellular fluid, would have different effects than K^+, which is found intracellularly in high concentrations, on the flavor profiles of 11 sweeteners. For this reason, an analysis of variance (ANOVA) was conducted by collapsing the data for $CaCl_2$ and NaCl together (group 1—extracellular) and comparing it with data for KCl (group 2—intracellular), and data for the sweeteners with no salts added (group 3—sweetener alone). Contrasts between groups were determined as well. Table 4 indicates the attributes for which significant differences were found between the groups at $p < 0.05$. The addition of ions had no significant effect on sweetness intensity for any sweetener. However, sodium saccharin and sucralose had significantly longer times of onset of maximum sweetness intensity for the extracellular group than the sweetener alone group. For mannitol, both the sweetener alone group and the extracellular group had a longer time of onset of maximum sweetness intensity than the KCl (intracellular) group.

No significant differences were found for any attributes for glucose and sodium cyclamate. Bitter ratings were significantly higher for the KCl group than the extracellular salts group or the sweetener alone group (i.e., without ions) for acesulfame-K, aspartame, fructose, and sucralose. These slight but significant increases in bitter ratings for acesulfame-K, aspartame, fructose, and sucralose were not due to taste profile differences from the salts themselves; there were no significant differences in the taste profiles of the salts at 5 mM. Figure 3 shows the mean bitterness intensity ratings for these four sweeteners for all three groups. For sucrose, bitter ratings were significantly higher for the KCl

group and the sweetener alone group than with the extracellular salts. Additional significant effects were found for other attributes, such as sour, salty, and viscous; however, these effects involved intensity ratings considered to be below a meaningful level.

An analysis of variance was also performed without collapsing the data from $CaCl_2$ with that of NaCl to ascertain whether the combining of $CaCl_2$ and NaCl into one group diminished the power of the results of one or both salt mixtures. The results of this analysis were similar to those described above.

4. Discussion

Small but statistically significant effects of temperature and pH were found for some but not all sweeteners. Overall the large molecular weight (and highly potent) sweeteners were most frequently enhanced by hot or cold temperatures. Elevating the temperature of sweetener solutions from 22°C (room temperature) to 50°C produced increases in sweetness intensity ratings for some high potency sweeteners including aspartame, MAG, neo-DHC, rebaudioside-A, and stevioside, especially at low concentrations. The bulk sweeteners sucrose and sorbitol were also slightly but significantly enhanced by elevated temperature at the lowest concentration. These data are consistent with previous studies that have reported enhancement of sweetness by elevated temperatures at low concentrations for sucrose [1,4] and aspartame [5]. In our study, the only significant decreases in sweetness intensity ratings due to elevated temperature were for higher levels of bulk sweeteners (sugars and polyhydric alcohols) including fructose, mannitol, and sucrose. This finding is consistent with Paulus and Reisch [2], who reported elevated thresholds (reduced sensitivity) for sucrose at higher temperatures.

The colder temperature (6°C) had less effect on the perceived intensity of the sweeteners tested. Some concentrations of high potency sweeteners with the largest molecular weights (neo-DHC, rebaudioside-A, stevioside, and thaumatin) were significantly enhanced at the cold temperature. Decreases in perceived sweetness intensity at the cold temperature were found for some levels of the dipeptide alitame and four bulk sweeteners (fructose, glucose, mannitol, and sucrose). The two *N*-sulfonylamides (acesulfame-K, sodium saccharin) and the sulfamate (sodium cyclamate) showed no significant change in our study in sweetness intensity at any and all concentrations for either temperature variation (6° or 50°C).

No significant changes in perceived sweetness were found over a pH range from 3 to 7. However, sourness and astringency increased with decreasing pH values. These data indicate that trained panelists can separate sweetness intensity from the intensity of other taste attributes. Furthermore, Na^+, Ca^{2+}, and K^+ at 5 mM had no effect on sweetness intensity for any sweetener. However, the addition of KCl slightly increased bitter ratings for four sweeteners: acesulfame-K, aspartame, fructose, and sucralose.

Table 4
Attributes found to have significant differences between the three groups for each sweetener

Sweetener	Significant[a] attribute	Specific comparison
Acesulfame-K	bitter	group 2 > group 1, 3
	salty	group 1 > group 3
Aspartame	bitter	group 2 > group 1,3
	licorice	group 3 > group 1, 2
	metallic	group 2 > group 1
	viscous	group 1 > group 3
Fructose	bitter	group 2 > group 1, 3
	salty	group 1 > group 3
Glucose	—	—
Mannitol	salty	group 1 > group 2, 3
	time	group 1, 3 > group 2
Na cyclamate	—	—
Na saccharin	salty	group 1, 2 > group 3
	astringent	group 1, 3 > group 2
	time	group 1 > group 3
Rebaudioside-A	fruity	group 3 > group 1, 2
Sorbitol	salty	group 1, 2 > group 3
	metallic	group 2, 3 > group 1
	astringent	group 2, 3 > group 1
Sucralose	bitter	group 2 > group 1, 3
	time	group 1 > group 3
Sucrose	bitter	group 2, 3 > group 1
	sour	group 1 > group 3
	salty	group 1 > group 2, 3

[a] $\propto = 0.0500$.

The main finding from this three-part study was that temperature, pH, and ions had little effect on the perceived sweetness intensity of the sweeteners studied. Even when significant differences were found in the temperature study, the effects were very small.

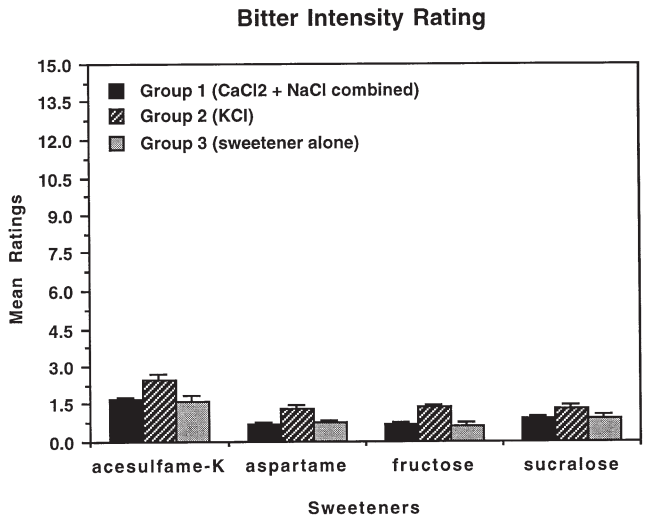

Fig. 3. Mean ratings of group 1 (sweetener with extacellular salts-$CaCl_2$ + NaCl- combined), group 2 (sweetener with intracellular salt-KCl), and group 3 (sweetener alone) for the sweeteners that were found to have significant differences in bitterness ratings between the three groups.

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