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Preference for Sucralose Predicts Behavioral Responses to Sweet and Bittersweet Tastants

Gregory C. Loney, Ann-Marie Torregrossa, Chris Carballo and Lisa A. Eckel

Department of Psychology and Program in Neuroscience, Florida State University, 1107 West Call Street, Tallahassee, FL 32306-4301, USA

Correspondence to be sent to: Lisa A. Eckel, Department of Psychology, Florida State University, 1107 West Call Street, Tallahassee, FL 32306-4301, USA. e-mail: eckel@psy.fsu.edu

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Abstract

Rats can be classified as either sucralose avoiders (SA) or sucralose preferrers (SP) based on their behavioral responses in 2-bottle preference, 1-bottle intake, and brief-access licking tests. The present study demonstrates that this robust phenotypic variation in the preference for sucralose predicts acceptance of saccharin, an artificial sweetener with a purported concentration-dependent "bitter" side taste and a 0.25 M sucrose solution adulterated with increasing concentrations of quinine hydrochloride (QHCl). Specifically, SA displayed decreased preference for and intakes of saccharin (≥41.5 mM) and sucrose–QHCl (>0.5 mM QHCl) solutions, relative to SP. In a second experiment involving brief-access (30-s) tests, SP and SA did not differ in their unconditioned licking responses across a range of sodium chloride or QHCl solutions (0.03–1 mM). However, the acceptability threshold for sucrose was lower in SA, relative to SP (0.06 and 0.13 M, respectively). Our findings suggest that phenotypic differences in sucralose preference are indicative of a more general difference in the hedonic processing of stimuli containing "bittersweet" or "sweet" taste qualities.

Key words: artificial sweetener, individual differences, rats, taste

Introduction

Individual differences in taste sensitivity influence dietary choice, leading to phenotypic variation in the acceptance and avoidance of a number of foods (Drewnowski and Rock 1995; Duffy and Bartoshuk 2000). Humans, for example, display genetic variation in their ability to taste low concentrations of 6-*n*-propylthiouracil (PROP), reporting either a moderate to strong bitter taste or no taste at all (for review, see Bartoshuk et al. 1994). Some studies have shown that this ability to taste PROP is associated with greater sensitivity to certain bitter and sweet tasting compounds, oral irritants, and the perception of creaminess in fats (e.g., Bartoshuk et al. 1994; Lucchina et al. 1998; Prescott and Swain-Campbell 2000; Chang et al. 2006; Hayes and Duffy 2007). There is also some evidence that PROP tasters report decreased hedonic evaluation of certain sweet and bitter tastants, high concentrations of saccharin, cruciferous vegetables, coffee, and alcohol, relative to PROP nontasters (Bartoshuk 1979; Kaminski et al. 2000; Keller et al. 2002; Lanier et al. 2005; Yeomans et al. 2007). Although the degree to which this variation in taste sensitivity can be uniquely attributed to genetic variation in PROP sensitivity has been questioned (see Lim et al. 2008), the existing human literature supports the notion that variation in the perception of one tastant can affect the hedonic evaluation of other tastants, even those differing in taste quality. In particular, genetic variation in the genes encoding receptors responsible for bitter taste perception (*Tas2R* receptors) is associated with variation in responsiveness to multiple tastants (Pronin et al. 2007; Roudnitzky et al. 2011). Additionally, rats that have been selectively bred for either high or low preference for saccharin (HiS and LoS, respectively) differ not only in their acceptance of saccharin solutions but also in their consumption of sucrose solutions adulterated with quinine hydrochloride (QHCl) and ethanol (Dess et al. 1998; Dess 2000).

Artificial sweeteners, including saccharin and acesulfame-K, elicit a bitter side taste that is particularly evident at high concentrations (Bartoshuk 1979; Wiet and Beyts 1992; Schiffman et al. 1995; Pronin et al. 2007). Recent studies have shown that individual differences in the perception of this bitter side taste are associated with genetic variation in Tas2Rs. For example, allelic variation in *Tas2R31* accounts for phenotypic variation in the response to both

saccharin and acesulfame-K (Roudnitzky et al. 2011). This perception of an aversive side taste has likely contributed to the growing trend to replace these artificial sweeteners with newer products such as sucralose, the primary sweeting agent in Splenda. As a trichlorinated sucrose molecule, sucralose might be expected to have a more acceptable sucrose-like taste quality than saccharin and acesulfame-K. Despite sucralose's widespread use in the food industry, a systematic examination of its perceived taste quality in humans appears to be limited to a single study. A mixed-gender sample reported that the bitterness rating of sucralose is about one-third that of saccharin, acesulfame-K, and stevioside but about 4 times greater than that of sucrose (Schiffman et al. 1995). More is known about the taste quality and acceptance of sucralose in rats. Sclafani and Clare (2004) were the first to report that female rats display indifference to mild avoidance of sucralose solutions during 2-bottle preference tests. However, a closer examination of their data revealed that individual rats either avoided sucralose across the entire range of concentrations (0.6–10 mM) or preferred sucralose to water at all but the highest concentration. Individual variation in sucralose preference has also been reported in male and HiS/LoS rats (Bello and Hajnal 2005; Dess et al. 2009).

Recently, we provided the first systematic examination of the rat's variable response to sucralose (Loney et al. 2011). In this study, a large cohort of rats was tested in a series of 24-h 2-bottle preference tests involving water versus an ascending series of sucralose solutions (0.0025–5 mM). Approximately one-third of the rats preferred sucralose over water across the entire concentration range and were classified as sucralose preferrers (SP). The remaining two-thirds of rats avoided concentrations of sucralose ≥0.025 mM and were classified as sucralose avoiders (SA). This bimodal preference/avoidance profile was observed in both sexes and 2 strains of rats (Long–Evans and Sprague–Dawley). Rats were also given a series of brief-access (30-s) tests involving random presentations of varying concentrations of sucralose solutions. Licking was reduced in SA, relative to SP, at sucralose concentrations ranging from 0.625 to 5 mM. Because this paradigm minimizes postingestive feedback, the decline in sucralose-elicited licking in SA is likely the result of taste-guided behavior, with reduced licking reflecting a decrease in palatability (Davis 1973). Finally, we conducted a series of 1-h 1-bottle intake tests in 23-h water-deprived (i.e., highly motivated) rats. Intakes were significantly reduced in SA, relative to SP, at concentrations of sucralose >0.25 mM, suggesting that sucralose avoidance is a robust trait in SA. Taken together, these findings suggest that sucralose avoidance is driven by an aversive side taste that is detected by SA, but not by SP.

To investigate this hypothesis, the current study examined whether SP and SA differ in their preference for taste solutions characterized as sweet at low concentrations but having an aversive side taste at higher concentrations. First, we conducted 2-bottle preference tests involving water and an ascending series of saccharin solutions. Saccharin was chosen because it activates T2R receptors associated with the perception of bitter taste and elicits reports of a bitter or metallic off taste at the high end of the range of saccharin concentrations tested here (Schiffman et al. 1995; Kuhn et al. 2004). We also conducted 2-bottle preference tests involving water and a 0.25 M sucrose solution adulterated with increasing concentrations of QHCl. Sucrose–QHCl solutions were chosen in order to directly manipulate the bitter taste component of the test solution. Unlike saccharin solutions, which increase in perceived sweetness and bitterness as a function of increasing concentration (Schiffman et al. 1995), only the perceived bitterness of the sucrose–QHCl mixture should increase as a function of increasing QHCl concentration. In a second experiment, we examined whether SP and SA differ in their unconditioned licking responses to basic taste stimuli, presented during brief-access tests that provide estimations of the relative palatability of varying concentrations of a given taste stimulus (Davis 1973; Davis and Smith 1988). The affective licking responses of SP and SA were monitored during presentations of a prototypical sweet stimulus (sucrose), a prototypical bitter stimulus (QHCl), and a stimulus that is typically preferred at low concentrations and then avoided at higher concentrations in the absence of a change in taste quality (sodium chloride [NaCl]).

Materials and methods

Animals

Male Long–Evans rats (Charles River Breeding Laboratory, weighing 225–250 g) were individually housed in a temperature and humidity controlled room maintained on a 12:12 h lighting cycle. All rats had ad libitum access to chow (Purina, 5001) water (i.e., reverse-osmosis deionized water) throughout the study, unless otherwise noted. Animal usage and experimental protocols were approved by The Florida State University Institutional Animal Care and Use Committee.

Test cages

Two-bottle preference tests were conducted in custom polycarbonate home cages. The front panel of these cages allows simultaneous access to 2 fluid bottles equipped with ball-tip spouts. A food cup, positioned equidistant from the 2 drinking spouts, provides access to powdered chow. Brief-access licking tests were conducted in a Davis rig (Davis MS80 Rig; Dilog Instruments and Systems). This apparatus consists of a plastic cage with a wire-mesh floor. An opening at the front of the cage allows access to 1 of 8 spill-proof glass drinking tubes positioned on a sliding platform. A mechanical shutter opens and closes to allow access to each of the 8 tubes for a user-specified length of time. A computer controls both the movement of the platform, which determines the order of tube presentation, and the opening and closing of the

shutter, which determines the duration of tube access and the interval between tube presentations. Each individual lick is detected by a contact lickometer and recorded on a computer via DavisPro collection software (Dilog Instruments and Systems).

Taste solutions

All solutions were prepared fresh daily by dissolving sucralose (Tate & Lyle), reagent-grade chemicals (Sigma Aldrich), and sucrose (Publix) in deionized water. Preference test stimuli consisted of sucralose solutions (0.0025, 0.025, 0.25, and 2.5 mM), saccharin sodium solutions (4, 14.5, 21, 31, 41.5, and 52 mM), and a 0.25 M sucrose base solution adulterated with QHCl (0.015, 0.03, 0.06, 0.13, 0.25, 0.5, and 1 mM). Brief-access test stimuli consisted of sucrose and NaCl solutions (0.015, 0.03, 0.06, 0.13, 0.25, 0.5, and 1 M for both) and QHCl solutions (0.015, 0.03, 0.06, 0.13, 0.25, 0.5, and 1 mM). Solution concentrations were chosen to overlap those used in previous research (e.g., Smith and Sclafani 2002; Sclafani and Clare 2004; Loney et al. 2011).

Classification of rats as either SA or SP

Rats were housed in preference-test cages for 1 week prior to collecting behavioral data. During this adaptation period, both drinking bottles contained water. Following adaptation to the test cages, rats were assigned to 1 of 3 groups. Groups 1 and 2 ($n = 16$ and 20, respectively) were used in Experiment 1 and group 3 ($n = 16$) was used in Experiment 2. At the beginning of each experiment, preference for sucralose was assessed via 24-h 2-bottle preference tests involving water and an ascending series of sucralose solutions (0.0025–2.5 mM). Each concentration of sucralose was presented for 2 days. Bottle position was alternated each day. Intakes of water and each concentration of sucralose were monitored daily and averaged across the 2 test days. Preference for each concentration of sucralose was calculated by dividing average sucralose intake by average total fluid (sucralose and water) intake and expressing the scores as a percentage. As validated in our earlier work (Loney et al. 2011), individual rats were classified as SP if they displayed a preference for sucralose (consumed >50% of daily fluid as sucralose) at the 2 highest concentrations; the remaining rats were classified as SA. Using these criteria, ~40% of rats were classified as SP and ~60% were classified as SA. Representative preference scores for SP and SA are shown in Table 1. Additional behavioral testing (2-bottle preference tests involving other tastants in Experiment 1 or brief-access licking tests in the Davis rig in Experiment 2) began 1 week following assessment of sucralose preference.

Table 1 Classification of rats as either SP or SA

	Concentration of sucralose (mM)			
	0.0025	0.025	0.25	2.5
SP	78 ± 6%	83 ± 3%	97 ± 1%	91 ± 7%
SA	60 ± 6%	61 ± 8%	31 ± 11%	5 ± 1%

Data are presented as means ± standard error of the mean. Rats were given a series of 2-bottle preference tests involving water versus an ascending series of sucralose solutions. Rats were classified as SP if they displayed a preference (consumed >50% of daily fluid as sucralose) at the 2 highest concentrations of sucralose; the remaining rats were classified as SA.

Procedure

Experiment 1

Preference for saccharin and sucrose–QHCl solutions was assessed via a series of 24-h 2-bottle preference tests. One group (7 SP and 9 SA) was given access to water and an ascending series of saccharin solutions (4–52 mM). A second group (7 SP and 13 SA) was given access to water and a 0.25 M sucrose base solution adulterated with an ascending series of QHCl concentrations (0.015–1 mM). Water and taste solutions (saccharin or sucrose–QHCl) were presented for 2 days before testing the next concentration in the series. Water and taste solution intakes were monitored daily and bottle position was alternated each day.

Experiment 2

Unconditioned licking responses to 3 basic tastants (NaCl, QHCl, and sucrose, in order of testing) were examined in the Davis rig. Rats (7 SP and 9 SA) were maintained on a 23-h fluid-deprivation schedule throughout the training period and during tests involving NaCl and QHCl (1-h water access was provided 2 h after testing in the Davis rig). During sucrose testing, rats were given ad libitum access to water. On the first day of training, individual rats were placed in the Davis rig for 15 min and given free access to one tube containing water. On the second day of training, each of the 8 tubes contained water and were presented one at a time across 3 randomized blocks. This was done to accustom the rats to the sound of the shutter and the movement of the platform. Each rat was given 60 s to initiate a lick to each tube presentation. Following the initial lick, each tube was available for 30 s before the next tube was presented. If a rat did not make a lick during the initial 60 s that presentation was counted as a 0 in the data analysis. During testing, rats were exposed to the day 2 training protocol, but each of the 8 tubes contained either water or 1 of 7 concentrations of the basic tastant presented one at a time across 3 randomized blocks. This procedure was repeated daily for a total of 5 days per tastant, and the number of licks to water and each tastant concentration were recorded and averaged. Rats were given a 5-day rest period before testing the next tastant in the series.

Data analysis

Intakes during 2-bottle preference tests (Experiment 1) were monitored daily and then averaged across the 2 test days of each solution concentration. Average preference for each

solution concentration was calculated by dividing average solution intake by average total fluid (solution and water) intake and expressing the scores as a percentage. Preference scores were analyzed by 2-factor mixed-design (SP/SA group × concentration) analyses of variance (ANOVAs). Fluid intake was analyzed by 3-factor (SP/SA group × fluid × concentration) mixed-design ANOVAs. Data collected in the Davis rig (Experiment 2) are presented as the average number of licks during all 3 presentations across all 5 testing days (i.e., data represent an average of 15 presentations per concentration). The mean number of licks was analyzed by 2-factor mixed-design (SP/SA group × concentration) ANOVAs. For both experiments, significant main or interactive effects ($P < 0.05$) were examined using Tukey's honestly significant difference tests.

Results

Experiment 1

Two-bottle preference tests: saccharin

Preference for saccharin decreased as a function of increasing concentration in all rats ($F_{5,70} = 40.78$, $P < 0.0001$). However, this decline in saccharin preference differed in SP and SA ($F_{1,14} = 5.44$, $P < 0.05$, Figure 1). Post hoc tests revealed that the first reliable decrease in preference occurred at a lower saccharin concentration in SA, relative to SP (31 vs. 41.5 mM, respectively, Ps < 0.05). In addition, preference for the 2 highest concentrations of saccharin was decreased in SA, relative to SP (Ps < 0.05). Additional analyses at the 2 highest saccharin concentrations revealed that fluid intake was differentially affected by sucralose preference ($F_{1,14} = 5.12$, $P < 0.05$, Figure 2). At both concentrations, SA consumed more water and less saccharin than SP (Ps < 0.05).

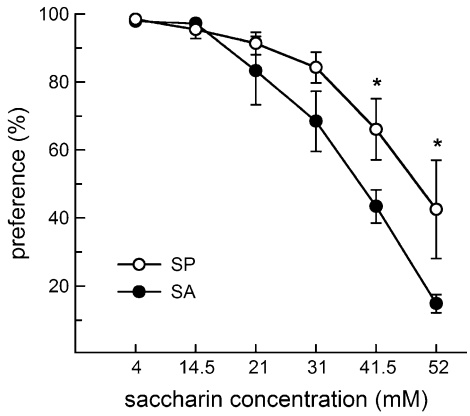

Figure 1 Preference for saccharin over water in SP and SA. Data are mean (±standard error of the mean) preference scores (saccharin intake divided by total fluid intake, expressed as percentages). All rats displayed a decrease in preference as a function of increasing saccharin concentration. This decrease in saccharin preference was first detected at a lower concentration of saccharin in SA, relative to SP (31 vs. 41.5 mM, respectively). In addition, preference for saccharin at the 2 highest concentrations was reduced in SA, relative to SP. *SA less than SP, Ps < 0.05.

Two-bottle preference tests: sucrose–QHCl

Preference for sucrose–QHCl solutions decreased as a function of increasing QHCl adulteration, however, the magnitude of this effect was differentially influenced by sucralose preference ($F_{6,108} = 2.30$, $P < 0.05$, Figure 3). Post hoc tests revealed that the first reliable decrease in the preference for sucrose–QHCl solutions occurred at a lower concentration of QHCl adulteration in SA, relative to SP (0.13 vs. 0.5 mM, respectively, Ps < 0.05). In addition, preference for sucrose–QHCl solutions adulterated with the 2 highest concentrations of QHCl was decreased in SA, relative to SP (Ps < 0.05). An additional analysis involving sucrose–QHCl solutions containing the 2 highest concentrations of QHCl adulteration revealed that fluid intake was differentially affected by sucralose preference ($F_{1,18} = 6.67$, $P < 0.05$, Figure 4). At both concentrations, SA consumed more water and less of the sucrose–QHCl solutions than SP (Ps < 0.05).

Experiment 2

Brief-access tests: NaCl and QHCl

The number of licks elicited by NaCl and QHCl solutions decreased as a function of increasing concentration ($F_{7,98} = 28.18$ and 124.40, respectively, Ps < 0.0001; Figure 5). During tests

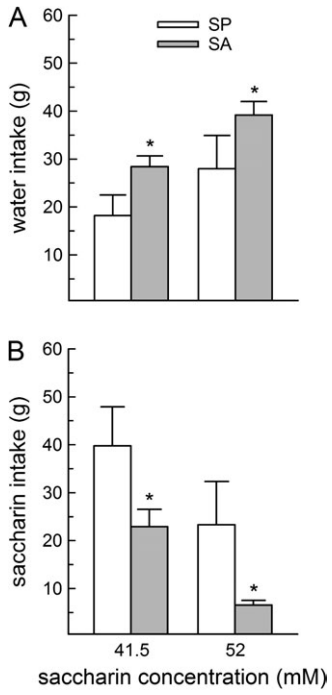

Figure 2 Fluid intake during 2-bottle preference tests at the 2 highest concentration of saccharin in SP and SA. At both concentrations, water intake was increased (**A**) and saccharin intake was decreased (**B**) in SA, relative to SP (A) *SA different from SP, Ps < 0.05.

involving NaCl, all rats displayed decreased licking at the 2 highest concentrations of NaCl, relative to water and the lower NaCl concentrations (Ps < 0.05). During tests involving QHCl, all rats displayed a progressive decrease in the number of licks to QHCl, relative to water (Ps < 0.05). The number of licks elicited by NaCl and QHCl solutions was not influenced by main or interactive effects of sucralose preference (F values = 0.81–2.63, not significant).

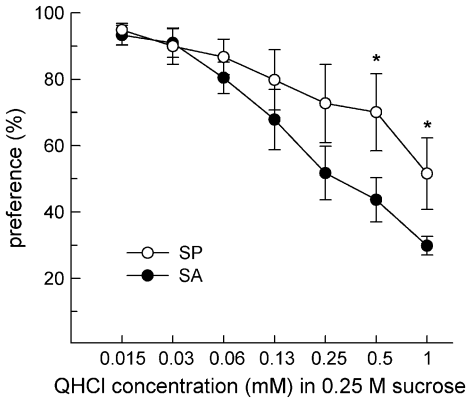

Figure 3 Preference for 0.25 M sucrose solutions adulterated with increasing concentrations of quinine hydrochloride (QHCl) over water in SP and SA. Data are mean (±standard error of the mean) preference scores (sucrose–QHCl intake divided by total fluid intake, expressed as percentages). All rats displayed a decrease in preference as a function of increasing QHCl adulteration. The decline in solution preference was first detected at a lower concentration of QHCl adulteration in SA, relative to SP (0.13 vs. 0.5 mM, respectively). In addition, preference for the sucrose–QHCl solution at the 2 highest QHCl concentrations was reduced in SA, relative to SP. *SA less than SP, Ps < 0.05.

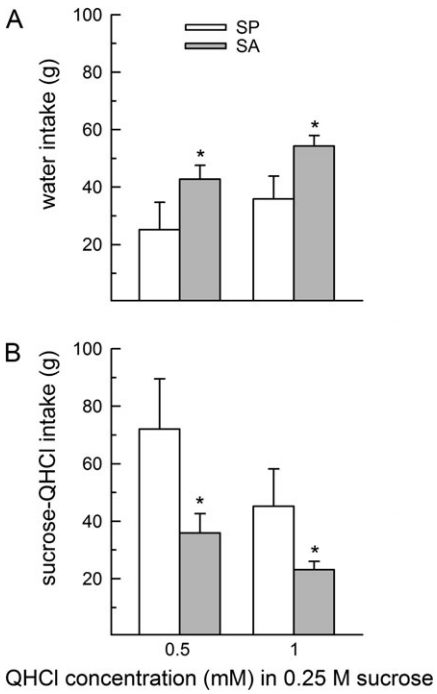

Figure 4 Fluid intake during 2-bottle preference tests involving water and 0.25 M sucrose solutions adulterated with 0.5 and 1 mM QHCl in SP and SA. At both concentrations, water intake was increased (**A**) and sucrose–QHCl intake was decreased (**B**) in SA, relative to SP (A). *SA different from SP, Ps < 0.05.

Brief-access tests: sucrose

The number of licks elicited by sucrose solutions was influenced by an interactive effect of sucralose preference and concentration ($F_{7,98}$ = 2.88, P < 0.01; Figure 6). Although both groups displayed a progressive increase in the number of licks as the concentration of sucrose increased (Ps < 0.05), the acceptability threshold, defined as the lowest sucrose concentration that elicited significantly more licks than water, was lower in SA, relative to SP (0.06 vs. 0.13 M, respectively, Ps < 0.05). In addition, there was a tendency for SA to consume more of the 0.06 M and less of the 0.5 and 1 M sucrose solutions, relative to SP, although these group differences failed to reach statistical significance.

Discussion

Rats can be unambiguously classified as either SP or SA based on their relative intakes of water and sucralose solutions presented during 2-bottle preference tests. Specifically,

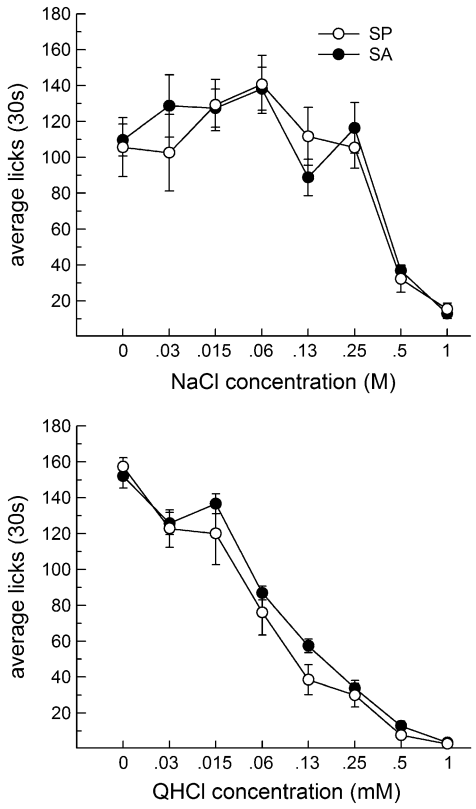

Figure 5 Unconditioned licking responses to NaCl (upper panel) and QHCl (lower panel) solutions in water-restricted SP and SA. For both stimuli, all rats displayed a decrease in licking behavior as a function of concentration (Ps < 0.05) with no differences between SP and SA.

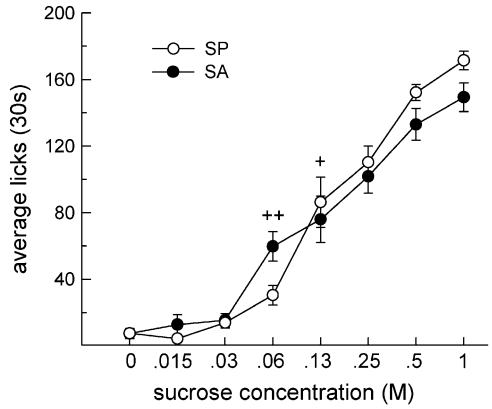

Figure 6 Unconditioned licking responses to sucrose solutions in water-replete SP and SA. All rats displayed a monotonic increase in licking as a function of concentration ($P < 0.05$). SA displayed a decreased acceptability threshold, defined as the first concentration in which licking was significantly greater than water, relative to SP. $^{++}$Acceptability threshold in SA, $P < 0.05$. $^{+}$Acceptability threshold in SP, $P < 0.05$.

SP consume more sucralose than water across a broad range of sucralose concentrations (0.0025–5 mM), whereas SA consume more water than sucralose at concentrations ≥0.025 mM (Loney et al. 2011). Here, we extend these findings by demonstrating that acceptance and intakes of saccharin and QHCl-adulterated sucrose solutions are decreased in SA, relative to SP. We also provide evidence that the acceptability threshold for sucrose solutions during brief-access licking tests is lower in SA, relative to SP.

A growing literature suggests that sucralose may have an aversive side taste that some rats (i.e., SA) are more sensitive to than others (i.e., SP). First, SA display avoidance, rather than indifference, toward sucralose solutions that are highly preferred by SP in 2-bottle preference tests (Sclafani and Clare 2004; Bello and Hajnal 2005; Loney et al. 2011). Second, sucralose-elicited licking responses during brief-access (30-s) tests, which offer an indication of the palatability of the test solution (Davis 1973), are decreased in SA, relative to SP (Loney et al. 2011). Third, consumption of sucralose solutions is decreased in SA, relative to SP, even when sucralose is the only fluid available during 1-bottle intake tests conducted in highly motivated rats maintained on a 23-h water-deprivation schedule (Loney et al. 2011). In the current study, we investigated whether SP and SA also differ in their preference for saccharin solutions, which elicit reports of a bitter side taste at the upper end of the concentration range used here (Schiffman et al. 1995). Our findings replicate previous reports of an inverse relationship between saccharin concentration and preference in rats (Nachman 1959; Morrison and Jessup 1977; Smith and Sclafani 2002) and provide the first demonstration that the avoidance and decreased intake of saccharin solutions ≥41.5 mM are increased in SA, relative to SP. These findings suggest that SA are more sensitive than SP to an aversive (presumably bitter) taste component that decreases the acceptance of high concentrations of saccharin in rats.

The perceived sweet and bitter taste components of saccharin solutions increase as a function of increasing concentration (Schiffman et al. 1995). To provide a more direct measure of variation in the perception of the bitter taste component of a binary (bittersweet) solution, SP and SA were given a series of 2-bottle preference tests involving a 0.25 M sucrose solution adulterated with increasing concentrations of QHCl. Similar to our findings involving saccharin solutions, SA displayed decreased acceptance and intake of sucrose–QHCl solutions within the upper end of the concentration range, relative to SP. This provides additional support for the hypothesis that SA are more sensitive than SP to the bitter taste component of bittersweet solutions. Because both saccharin and sucrose–QHCl solutions elicit a dual-opponent taste in rats (Morrison and Jessup 1977; Hsiao and Fan 1993), our findings suggest that sucralose may also elicit a dual taste that is detected by SA, but not by SP. We acknowledge, however, that the 2-bottle preference testing paradigm used to categorize rats as SP or SA in the current study raises the possibility that any postingestive effects of sucralose consumption could have generalized to subsequently tested solutions. However, the likelihood that postingestive generalization mediated the decreased acceptance of saccharin and QHCl-adulterated sucrose solutions in SA is diminished by our earlier demonstration that sucralose avoidance can be detected in sucralose-naive rats within the first 30 s of a brief-access licking test, which minimizes any postingestive effects of the test solution (Loney et al. 2011). Thus, orosensory feedback alone appears sufficient to drive the differing behavioral phenotypes of SP and SA. Nevertheless, in order to eliminate the possibility of postingestive generalization, the acceptance of bittersweet solutions should be assessed in SP and SA following categorization via brief-access licking tests. Additionally, our findings do not rule out the possibility that SA may be less motivated than SP by the sweet component of the solutions tested here. However, we believe that a decreased hedonic response to sweet would result in an indifference to sucralose and other bittersweet solutions rather than the marked avoidance observed in the current and previous studies (Sclafani and Clare 2004; Bello and Hajnal 2005; Loney et al. 2011). Thus, the most parsimonious explanation is that SA perceive an aversive taste in sucralose solutions that SP are considerably less sensitive to and that this individual variation predicts the acceptance of solutions containing a bitter taste component.

Previous studies have shown that binary mixtures of bitter and sweet tastants exert a suppressive effect on the reported bitterness and sweetness of the resulting bittersweet solutions. In humans, the perceived sweetness and bitterness of sucrose–QHCl solutions applied to the lingual surface is less than the perceived intensities of either stimulus presented alone (Bartoshuk 1975). Interestingly, humans that

are sensitive to the bitter taste of PROP are not only less accepting of high concentrations of saccharin (Bartoshuk 1979), but they are also more sensitive to the sweetness suppression elicited by sucrose–QHCl mixtures, relative to PROP nontasters (Prescott et al. 2001). It has also been shown that activity in sucrose-responsive (S) fibers in the chorda tympani (CT) nerve is significantly suppressed by the addition of QHCl to a taste mixture perfused across the tongue of golden hamsters (Formaker et al. 1997). Thus, it is possible that individual variation in the processing of neural or sensory information may contribute to variation in the rat's sensitivity to aversive taste components of taste mixtures. In support of this notion, genetic variation in saccharin preference results in decreased avidity for sucrose–QHCl mixtures, sucralose, and ethanol solutions in LoS rats, relative to HiS rats (Dess et al. 1998; Dess 2000; Dess et al. 2009). Interestingly, ethanol solutions have been shown to generalize to sucrose–QHCl mixtures in conditioned taste aversion experiments (Kiefer and Mahadevan 1993), suggesting a similar bittersweet taste quality and perhaps a similar neural mechanism contributing to the decreased acceptance of these solutions. Taken together, these individual differences in behavioral and electrophysiological findings may provide a common physiological mechanism contributing to the increased sensitivity to the aversive properties of bittersweet solutions shared by human tasters of PROP, LoS rats, and SA.

The second goal of the current study was to determine whether SP and SA differ in their affective responses to basic tastants (NaCl, QHCl, and sucrose) presented during brief-access (30-s) licking tests. This particular paradigm was chosen because it minimizes postingestive effects and thus provides a direct measure of the palatability of a taste solution (Davis 1973). Moreover, our previous work (Loney et al. 2011) has shown that rats can be categorized as SP or SA based solely upon their hedonic licking responses during brief (30 s) presentations of concentrations of sucralose that are avoided by SA in 2-bottle preference tests (e.g., 2.5 mM). Thus, brief-access tests are sufficiently robust to elucidate differences in behavioral orosensory phenotypes. Both the NaCl and QHCl solutions elicited concentration-dependent licking responses that decreased as a function of increasing stimulus concentration, consistent with previous studies (Breslin et al. 1993; St John et al. 1994; Brasser et al. 2005). However, we did not detect any group differences in the number of licks displayed by SP and SA. Thus, under the current test conditions, individual variability in sucralose preference is not predictive of differential hedonic responding to NaCl or QHCl solutions. Although SA are more sensitive than SP to the bitter taste component of bittersweet solutions assessed during 24-h 2-bottle preference tests, this increased sensitivity did not translate to a difference in acceptability of solutions containing varying concentrations of QHCl alone. This discrepancy may be related to the fact that all rats were fluid deprived during the brief-access tests and thus were highly motivated to lick to any solution presented. This increased motivation may have masked any latent group differences in the hedonic responding to QHCl in SP and SA. Future studies involving intraoral delivery of tastants to fluid-replete SP and SA utilizing the taste-reactivity paradigm should prove useful in determining whether individual differences in sucralose preference affect the hedonic responses to purely bitter tastants like QHCl.

In the current study, brief-access tests involving sucrose solutions elicited concentration-dependent licking responses that increased as a function of increasing sucrose concentration, consistent with previous studies (Davis 1973, Spector and Smith 1984). However, the acceptability threshold, defined as the lowest sucrose concentration that elicited significantly more licks than water, was lower in SA, relative to SP. Thus, SA appear more sensitive than SP to dilute concentrations of sucrose. Our findings are similar to previous reports that C57BL/6J "taster" mice, which express a polymorphism in the *Tas1r3* gene, display increased sensitivity to the hedonic properties of dilute sucrose solutions, relative to 129P3/J "nontaster" mice (Dotson and Spector 2004; Glendinning et al. 2005). We also noted a tendency for a reduction in sucrose-elicited licking in SA, relative to SP, at the 2 highest concentrations tested here. This is similar to previous reports that LoS and C57BL/6J taster mice display decreased acceptance of concentrated sucrose solutions relative to HiS and 129P3/J non-taster mice, respectively (Dess 2000; Sclafani 2006). The emerging similarities in the behavioral response to sucrose among SA, LoS rats, and C57BL/6J taster mice highlights a need for additional investigations of the relative sensitivity to sweet taste in SP and SA rats.

The individual variation in sucralose preference identified here and in previous studies (Sclafani and Clare 2004; Bello and Hajnal 2005; Loney et al. 2011) appears to represent a biologically meaningful division. In the current study, we demonstrated that SA are not only more sensitive to the aversive component of bittersweet solutions that drives down their acceptance in 2-bottle preference tests but that they are also more sensitive to the reinforcing orosensory properties of dilute sucrose solutions. Interestingly, this increased sensitivity may be accompanied by reduced hedonic evaluation of more concentrated "sweet" solutions in a manner similar to human tasters of PROP, LoS rats, and taster mice. Our finding that SP and SA do not differ in their unconditioned licking responses to QHCl solutions suggests that sucralose avoidance is not mediated simply by heightened sensitivity to bitter taste (as assayed via QHCl). Rather, it appears likely that the differential acceptance of bittersweet mixtures is driven by a more complex interplay between bitter and sweet taste components. Additional studies are necessary to determine whether there is a shared physiological mechanism that results in the similar hedonic responses observed across species and rodent strains as a function of their acceptance and intakes of various tastants. There is a clear need to understand

the mechanism underlying individual differences in taste perception, which have been shown to predict a number of less obviously correlated behavioral phenomena such as impulsivity, drug seeking behavior, and risk taking behaviors (Dess et al. 1998; Perry et al. 2007; Anker et al. 2008; Carroll et al. 2008).

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