SW.B6-*Soa*^{b} Nontaster Congenic Strains Completed and a Sucrose Octaacetate Congenic Quartet Tested with other Bitters

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Abstract

Ten SW.B6 SOA nontaster strains congenic with the SWR/J SOA taster inbred strain were bred via repeated backcross–intercross cycles, with selection for nontasting in each cycle. Preference ratio distributions and phenotypic proportions across cycles at 0.1 mM SOA were consistent with monogenic predictions. The SW.B6 mice completed a congenic quartet with the SWR/J, B6.SW SOA taster and C57BL/6J SOA nontaster strains. The *Soa* locus controlled avoidance differences within the quartet for SOA, raffinose undecaacetate, glucose pentaacetate and brucine. Background genes not linked to *Soa* controlled avoidance differences for L-phenylalanine and ethanol. Avoidance of bitter picric acid was influenced by the *Soa* locus, but avoidance of acetic acid was not. The quartet pattern for quinine HCl was unclear, with indications of both *Soa* and background effects. Two forms of ribose tetraacetate yielded different patterns. Avoidance differences controlled by the *Soa* locus were found for the pyranose form; however, all four strains avoided the furanose form. The pleiotropic effects of *Soa* allele substitution within the quartet were limited to a subset of bitter compounds. **Chem. Senses 21: 507–517, 1996.**

Introduction

A monogenic polymorphism in mice (*Mus domesticus*) for avoidance of the bitter compound sucrose octaacetate (SOA) is well documented (Warren and Lewis, 1970; Lush, 1981; Harder *et al.*, 1984; Whitney and Harder, 1986; Gannon and Whitney, 1989). Three alleles (*Soa*^{a}, *Soa*^{b} and *Soa*^{c}) leading to three levels of SOA sensitivity (taster, nontaster and demitaster) have been identified (Harder *et al.*, 1992). The *Soa* gene has been located on chromosome 6 ~62 cM from the centromere (Capeless *et al.*, 1992). The gene product is not known, but several peripheral allele-difference correlates have been reported (Shingai and Beidler, 1985; Miller and Whitney, 1989; Spielman *et al.*, 1992, 1994; Capeless *et al.*, 1994).

SWR/J (SW) and C57BL/6J (B6) inbred strains have different alleles at the *Soa* locus and show contrasting phenotypes in two-bottle preference tests (*Soa*^{a} taster and *Soa*^{b} nontaster respectively). Like any unrelated strains, they have a myriad of other genetically based differences as well. Isolating the SOA-sensitivity mechanism(s) would be much easier if these extraneous differences were eliminated. To

this end, B6.SW (Soa^a taster) strains congenic with the B6 strain were bred (Whitney and Harder, 1986; Whitney *et al.*, 1989). Phenotypic differences between the B6.SW and B6 mice reflect the sole genetic difference within this congenic pair—a segment of chromosome 6 containing the Soa^a allele transferred from the SW strain. Avoidance differences between B6.SW and B6 mice have been found for SOA and various other bitter compounds (Whitney *et al.*, 1990, 1991; Harder *et al.*, 1992; Whitney and Harder, 1994). When tested with sweet, sour or salty substances the congenic pair has not differed (with one apparent exception: sour acetic acid). The B6.SW mice not only differed from the B6 mice for several bitter substances but also closely resembled the SW mice tested at the same time. The B6.SW and SW strains both have the Soa^a allele, but have different background genomes. Their phenotypic similarity indicated no effect of background variation on taster allele expression.

The two types of comparison described above are only half the combinations possible starting from the B6 and SW strains. *Soa* allele difference effects could also be examined on a constant SW background, and effects of background variation on expression of the nontaster allele could be assessed. A second set of congenic strains, SW.B6 (Soa^b nontaster), was bred to enable these additional comparisons to be made. The first part of the present paper describes the development of the SW.B6 set and compares predictions of the monogenic model (and two alternative models) to the phenotypic ratios observed in segregating generations produced along the way. The SW and B6 inbred strains, together with the B6.SW and SW.B6 congenic strains, form a complete congenic quartet. The second part of the paper presents results from initial tests of this congenic quartet with other, mainly bitter, substances. The *Soa* allele-determined avoidance pattern seen in an extended SOA concentration series is compared with the patterns found with these other substances.

Part I: Congenic strain development

A set of SW.B6 nontaster strains congenic with the SW taster strain was bred via the cross–intercross protocol described by Flaherty (1981). The objective was to produce mice that had a genome essentially that of the SW strain, but with a chromosome segment carrying the nontaster allele from the B6 donor strain. To accomplish this, ten serial crosses to the SW strain were made in each congenic line. Each cross was expected to reduce the donor genetic material not linked to the target locus (*Soa*) by half. Linked material (the chromosome six segment) would be reduced more slowly. After the tenth cross < 0.2% of unlinked donor material was expected to remain, and the chromosome segment transferred from the B6 strain, carrying the Soa^b allele, was expected to average 18.1 cM in length (1.1% of the mouse genome) (Bartlett and Haldane, 1935; Flaherty, 1981).

However, as the allele to be transferred was recessive, the ten serial crosses to the inbred partner strain could not be made consecutively (as in the B6.SW congenic development). The heterozygous progeny of each cross, if crossed again with the homozygous-dominant SW mice, would have produced 1/2 heterozygous and 1/2 homozygous-dominant offspring indistinguishable in behavioral tests (all SOA avoiders). There would have been no way to select individuals carrying the recessive allele for the next cross. Instead, the heterozygotes were intercrossed (siblings paired) to produce segregating generations expected to include 1/4 nonavoider individuals. These homozygous- recessive mice could be selected for the next cross to the SW strain. The intercrosses doubled the time needed to complete the set (relative to the B6.SW set), but provided generations whose phenotypic ratios could be compared with the ratios expected from the standard one-locus model. The adequacy of this genetic model of SOA-sensitivity determination was thus examined.

Method

The cross–intercross breeding protocol is depicted in Figure 1. Crosses to the SW strain (backcrosses) and intercrosses alternated after the F_1 generation. The backcross offspring (B_2–B_{10}) were verified to be consistent SOA avoiders before siblings were intercrossed (see testing procedure below). Intercross offspring (F_2–F_{10}) were similarly tested, and only consistent nonavoiders were backcrossed to SW mice. F_{11} nonavoider siblings were intercrossed to complete the congenic development. Their M_{12} offspring were all expected to be nonavoiders.

The SW and B6 parents of the F_1 generation were bred in our laboratory from mice purchased from The Jackson Laboratory (Bar Harbor, ME). Lab-bred SWs were used in five of the ten backcrosses. Purchased SWs were used in the other five to keep the SW.B6 background genome as close to the standard SWR/J genome as possible. Reciprocal

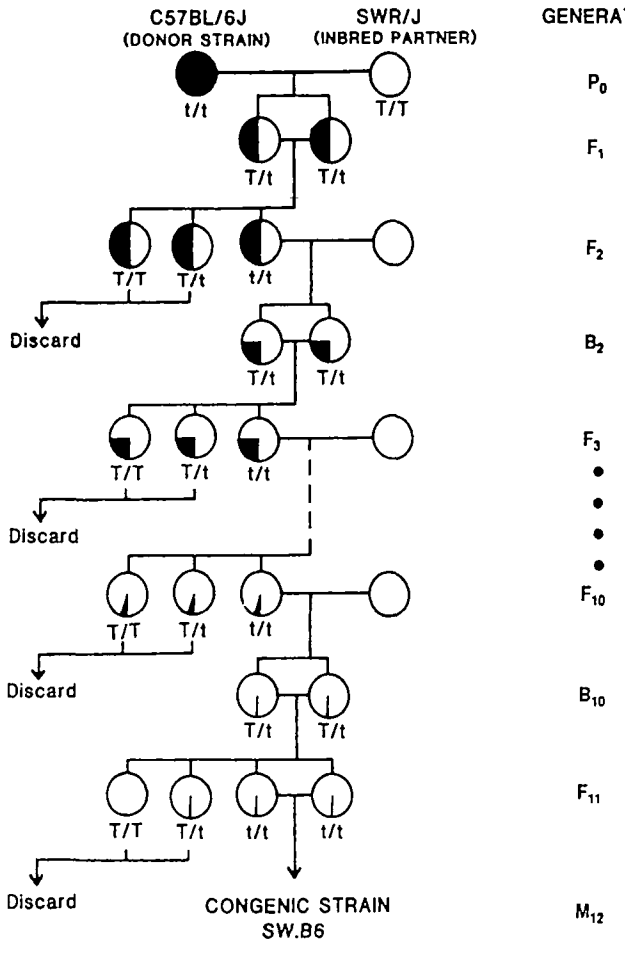

Figure 1 Cross-intercross breeding system used to develop SW.B6 SOA nontaster congenic strains. Intercross generations (F_2–F_{11}) alternated with backcross generations (B_2–B_{10}). Only nonavoiders (recessive homozygotes) were selected for breeding from each intercross generation, thereby retaining the *Soa*^{b} allele from the donor strain (modified from Flaherty, 1981).

pairings of the inbred strains produced two F_1 types (B6×SW and SW×B6) (by convention, the female parent is given first). Reciprocal intercrosses among the F_1 types produced four F_2 types [(B6×SW)(B6×SW), (B6×SW)(SW×B6), (SW×B6)(B6×SW) and (SW×B6)(SW×B6)]. Reciprocal backcrosses to the SW strain of each F_2 type produced eight B_2 types [e.g. SW×(B6×SW)(B6×SW) and (B6×SW)(B6×SW)×SW]. These eight B_2 types were then intercrossed within type (no between type pairings) to produce the F_3 generation. Four of the eight types had duplicate pairs established for a total of twelve B_2×B_2 pairs. Each of these twelve pairs founded an independent lineage leading to an SW.B6 congenic strain. Ten of the lineages survived to completion (generation M_{12}).

In each generation, all offspring in the first two surviving litters from each pair were tested individually for SOA avoidance or nonavoidance. Consecutive 48 h, two-bottle preference tests with 0.01 mM and 0.1 mM SOA solutions (versus dH_2O) were given at 40–60 days of age. A few B6 and SW mice (usually three per strain) were also tested with each group of offspring as phenotypic standards. Testing was done in a separate temperature-controlled room on a 12 h light/12 h dark cycle. Purina Mouse Diet #5015 was available *ad libitum* at the back of the cages throughout the tests. The SOA solutions were prepared by dissolving SOA crystals (Sigma Chemical Co., St Louis, MO) in heated deionized water, then cooling to room temperature in a cold-water bath. Two inverted 25 ml graduated cylinders, with neoprene stoppers and curved stainless-steel spouts, were placed on the front of each testing cage. Clips on these suspended stainless-steel cages held the cylinders 6 cm apart, with the spouts protruding through the wire-mesh fronts 1–2 cm above the cage floor. SOA cylinders were placed on the right side and dH_2O cylinders on the left for the first 24 h of each test. Cylinder positions were reversed for the second 24 h to control for any side biases. Amounts consumed from the cylinders were recorded for each 24 h period and a preference ratio was calculated [preference ratio = amount solution/(amount solution + amount dH_2O)]. The animal's preference ratio for a given SOA concentration was the mean of its two 24 h ratios. The second 48 h test immediately followed the first.

In previous testing with 0.01 and 0.1 mM SOA, a preference ratio criterion of 0.15 best distinguished taster from nontaster mice. Nearly all SW (taster) mice had ratios <0.15 and nearly all B6 (nontaster) mice had ratios >0.15, at both concentrations (Whitney *et al.*, 1989). Therefore, this criterion was used to classify mice tested during the SW.B6 development. Mice were considered avoiders only if both 48h preference ratios were <0.15. Mice were eligible for selection as nonavoiders only if both 48 h preference ratios were ≥0.15. A few mice had one ratio above the criterion and one ratio below. These inconsistent mice were never selected for pairing. The mice actually chosen for pairing, from among those eligible, were the avoiders with ratios nearest 0.0 (complete avoidance) and the nonavoiders with ratios nearest 0.5 (complete indifference).

The ratio of individuals avoiding to not-avoiding 0.1 mM SOA in each intercross generation (F_2–F_{11}) was compared (χ^2 goodness-of-fit tests) to the ratios expected from three genetic models compatible with the inbred and F_1 results. The standard monogenic model (one autosomal gene with

two alleles, the taster allele completely dominant) predicted 75% tasters:25% nontasters in each intercross generation. The additive two-locus model (two unlinked autosomal genes with equal effects, combining linearly, each with two alleles, and the SW strain having both dominant alleles) predicted 58% tasters:42% nontasters. The epistatic two-locus model (the same as the previous model except that the effects combine non-linearly, a dominant allele at one locus overshadowing two recessives at the other locus) predicted 94% tasters:6% nontasters. All three models predicted 100% tasters in the backcross generations and 100% nontasters in the M_{12} final generation.

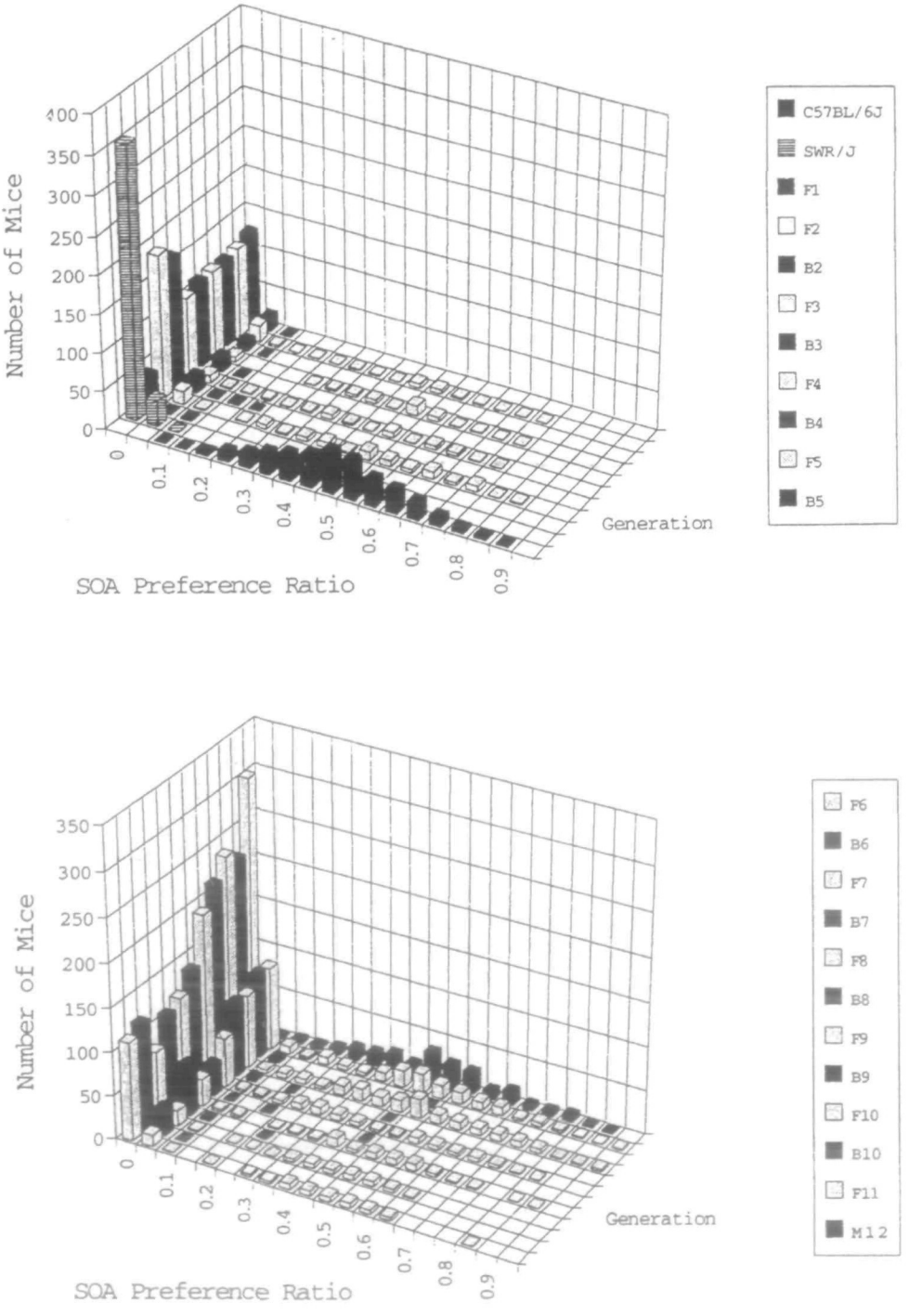

Figure 2 Distributions of individual 0.1 mM SOA preference ratios in 48 h, two-bottle tests across generations. Ratios near 0.5 indicate approximately equal dH_2O and SOA solution consumptions. Ratios near zero indicate strong SOA solution avoidance.

Results

The distributions of individual 0.1 mM SOA preference ratios across generations are shown in Figure 2. [The 0.01 mM distributions (not shown) were similar in all generations, though most were shifted slightly higher.] The virtually non-overlapping distributions of the inbred strains illustrate the taster–nontaster phenotypic contrast. The C57BL/6J preference ratios were approximately normally distributed around 0.5, the ratio produced by equal water and solution consumptions. The SWR/J ratios were grouped near zero, indicating strong avoidance of the SOA solution. The heterozygous F_1 mice closely resembled the homozygous SWR/J mice, indicating taster dominance. The backcross mice (B_2–B_{10}) also resembled the SWR/Js, except for 1–3 outliers in some generations. The intercross distributions (F_2–F_{11}) were bimodal, consistent with allelic segregation. In most intercross generations, ~3/4 of the scores fell within the SWR/J range, while the remaining scores were spread over the C57BL/6J range. The final M_{12} distribution resembled the C57BL/6J distribution, with a mean not significantly different from 0.5 (mean = 0.482, SD = 0.177, $t = 1.50$, df = 221, $P > 0.10$). Seven of the M_{12}s had scores <0.15; however, they appeared to be just the lower tail of a symmetrical nontaster distribution. Five mice had scores as extreme at the upper end of the distribution (i.e. >0.85). None of the seven mice avoided 0.01 mM. These inconsistent mice were excluded from contributing to the next generation (M_{13}).

Table 1 χ^2 goodness-of-fit comparisons of observed 0.1 mM SOA phenotypic ratios in SW.B6 intercross generations to ratios expected from one-locus and two-locus genetic models

Generation	*n*	Observed %		One-locus	Two-locus	
		Tasters	Nontasters	Expected % 75:25	Additive % 58:42	Epistatic % 94:6
C57BL/6J	360	0.3	99.7			
SWR/J	387	100	0			
F_1	46	100	0			
F_2	283	73.2	26.8	0.52	25.99***	208.45***
B_2	198	98.5	1.5			
F_3	157	74.5	25.5	0.02	17.16***	100.88***
B_3	141	100	0			
F_4	159	75.5	24.5	0.02	19.55***	91.22***
B_4	142	100	0			
F_5	169	83.3	16.6	6.41*	54.06***	31.10***
B_5	150	100	0			
F_6	169	75.2	24.8	0.00	26.40***	100.76***
B_6	156	100	0			
F_7	133	74.4	25.6	0.02	14.42***	86.51***
B_7	182	99.5	0.5			
F_8	210	72.9	27.1	0.51	18.57***	158.75***
B_8	203	99.0	1.0			
F_9	320	78.4	21.6	2.01	53.94***	130.32***
B_9	324	99.4	0.6			
F_{10}	467	69.6	30.4	7.57**	24.91***	469.46***
B_{10}	349	99.7	0.3			
F_{11}	543	75.1	24.9	0.01	64.07***	324.65***
M_{12}	222	3.2	96.8			
F_2–F_{11}	2610	74.6	25.4	0.18	289.85***	1648.55***

*$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

The total number tested in each generation, and the percentages of mice with scores <0.15 (tasters) and ≥0.15 (nontasters) are shown in Table 1. The χ^2 values for the comparisons of observed to expected phenotypic ratios are also shown, and deviations from the ratios expected with each genetic model are indicated. The observed ratios were different from the expected ratios for both two-locus models in every generation ($P < 0.001$). Two of the ten observed ratios (F_5 and F_{10}) differed from the one-locus expectation ($P < 0.05$ and $P < 0.01$ respectively). The other eight observed ratios were not different from the one-locus expectation ($P > 0.05$). The observed ratio for all intercross mice combined (74.6:25.4%) was in close agreement with the 75:25% monogenic expectation. These results yielded no support for either two-locus model. The monogenic model was, in the main, supported. The two apparent deviations from the monogenic expectation are less troubling when it is noted that the *P*-values given above were for single comparisons. As ten such comparisons were made for each genetic model, overall $\alpha s < 0.05$ would require individual *P*-values of <0.005 for statistical significance. With this correction, none of the observed ratios differed from the one-locus expectation, though all still deviated from the two-locus expectations.

Discussion

Ten SW.B6-*Soa*^{b} nontaster strains congenic with the SWR/J taster inbred strain were bred. The M_{12} final generation was verified to be similar in SOA phenotype to the original C57BL/6J nontaster inbred strain. All SW.B6 mice in subsequent generations are expected to be homozygous-recessive nontasters.

The *Soa*^{b} allele was retained across generations by phenotypic testing and selection. The B6 chromosome segment linked to the *Soa* locus, however, was expected to vary somewhat in length and position from one SW.B6 line to another. Because of this variation, only loci very close to *Soa* would also be likely to have a consistent B6 genotype across lines.

The outliers seen in some backcross generations suggested slightly (<2%) incomplete penetrance of the taster phenotype in heterozygotes (no outliers were seen among the 387 homozygous-dominant SWR/Js). The phenotypic ratios observed in segregating generations generally conformed to those predicted by the standard one-locus model. They were decidedly inconsistent with both the less-extreme and more-extreme ratios predicted by the two-locus models. Overall, the segregation data were consistent with previous reports.

Part II: Sensitivity to other compounds

The SW.B6 mice completed a congenic quartet for SOA sensitivity. The quartet consists of two congenic pairs, SW (taster)–SW.B6 (nontaster) and B6 (nontaster)–B6.SW (taster). The members of each congenic pair differ only in a portion of chromosome 6 containing the *Soa* locus. Within-pair comparisons allow the behavioral, physiological and/or anatomical effects of *Soa* allele variation to be assessed without 99% of the extraneous variation existing between the two inbred strains. Each congenic strain can also be compared with the inbred strain of the other pair (e.g. SW.B6 versus B6) to determine the effects of background genome variation on a given *Soa* allele. With the complete quartet, allele-difference effects can be assessed against both background genomes, and background-difference effects can be assessed for both alleles.

In the present study all four members of the quartet were compared to determine effects of *Soa* and background variation on avoidance of other tastants. Previous studies have reported effects of *Soa* allele differences on avoidance of several bitter compounds and one sour compound. *Soa* pleiotropic effects have been reported for ten other acetylated sugars (Lush, 1986; Whitney *et al.*, 1990, 1991; Harder *et al.*, 1992). Such effects have also been reported for bitter compounds with quite different structures and widely varying toxicities. These include denatonium benzoate, strychnine HCl, brucine, isohumulone and quinine sulfate (Lush, 1982; Whitney *et al.*, 1990, 1991; Boughter *et al.*, 1992; Harder *et al.*, 1992; Whitney and Harder, 1994). An effect of *Soa* variation has been reported for only one concentration of one non-bitter substance, 10 mM acetic acid (Whitney *et al.*, 1990).

Soa pleiotropic effects would be strongly indicated in the congenic quartet comparisons when: (i) the inbred strains differ; (ii) the congenic strains differ; and (iii) each congenic strain closely resembles the inbred strain with the same *Soa* allele. If the congenic strains differ but each resembles the inbred strain with the same background genome, effects of loci not on the transferred chromosome segment (i.e. background genes) would be indicated. Patterns combining both effects, or simply unclear, are possible as well. The pattern expected when the *Soa* locus alone controls

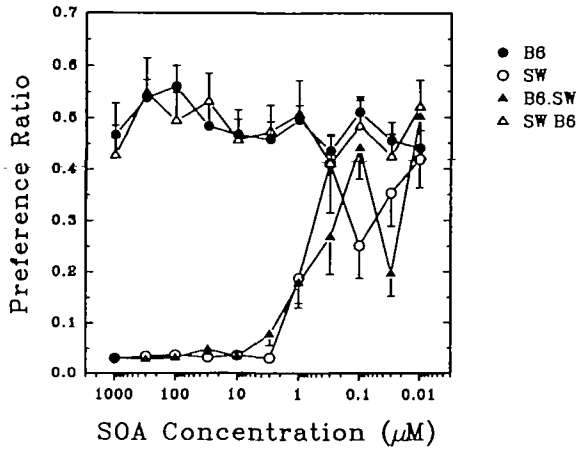

Figure 3 SOA concentration response functions (mean ± SE) in consecutive 48 h, two-bottle tests for the congenic quartet members (from Whitney and Harder, 1994). From 1000 to 1 μM, the inbred strains differed and each congenic strain closely followed the inbred with the same *Soa* allele. This indicated phenotypic control by the *Soa* locus.

avoidance differences can be seen in Figure 3 (from Whitney and Harder, 1994). In this descending SOA series, the SW inbred and B6.SW congenic strains (with the Soa^a taster allele) avoided concentrations down to 1 μM. The B6 inbred and SW.B6 congenic strains (with the Soa^b nontaster allele) were indifferent at all concentrations. The quartet patterns for eight other bitter compounds, acetic acid and ethanol are compared with this pattern.

Method

The SOA avoidance phenotypes of all mice used were confirmed in preliminary tests similar to those conducted during the congenic development. Panels of 10–20 mice per strain were then given series of 48 h, two-bottle tests with one or two other compounds. Two or three concentrations of each compound (except ethanol) were presented in descending order. Descending series were used because mice sometimes avoid concentrations presented in descending order to which they appear indifferent when presented in ascending order (Harder *et al.*, 1989). After completing a compound, the mice were given a 48 h period with one dH_2O cylinder in the center position before starting the next compound. Concentrations within a descending series were tested consecutively. As before, the inbred mice were either purchased from the Jackson Laboratory or bred from such mice. All mice were at least 40 days old at the start of SOA testing. Roughly equal numbers of males and females of each strain were tested.

All four members of the quartet were tested together with acetic acid, picric acid, ribopyranose tetraacetate (RPTA), ribofuranose tetraacetate (RFTA) and ethanol. Raffinose undecaacetate (RUA), glucose pentaacetate (GPA), brucine, L-phenylalanine and quinine HCl had already been tested on panels consisting of the two inbred strains and the B6.SW congenics. Additional panels consisting of the inbred strains and the SW.B6 congenics were tested with these substances when the congenic set was completed. Potential sequence effects were minimized to the extent possible given that mice were exposed to more than one substance. Each panel of mice was tested with only two substances after the SOA pre-test (for the ethanol panel, just one substance), and toxic compounds (brucine and picric acid) were the final substances in their respective sequences.

Avoidance differences and similarities among the strains at individual concentrations were assessed via single-factor analyses of variance, followed by Tukey-HSD multiple comparisons. For compounds where Cochran's tests indicated significant heterogeneity of variance, the logarithms of the raw preference ratios were analyzed. As many analyses of variance were expected to be performed, a stringent *a priori* criterion for significant strain differences ($P < 0.01$) was adopted.

Results

Soa allele variation clearly controlled RUA and GPA avoidance (Figure 4a and b). The B6 mice were completely indifferent to both substances, the SW mice profoundly avoided both substances and for both substances the congenic strains closely followed the inbred strains with the same *Soa* alleles. The patterns were so clear for these acetylated sugars that statistical analysis seemed redundant. The pattern for brucine was equally clear (Figure 5a). Despite brucine's toxicity and structural dissimilarity to SOA, the *Soa* locus again controlled avoidance. As above, statistical analysis was omitted.

In contrast, no *Soa*-variation effect was found for sour 10 mM acetic acid (Figure 5b). The inbred strain means were not significantly different and both congenic means were intermediate [$F(3, 56) = 1.357$, $P > 0.25$]. More substantial strain differences were found for bitter 1 mM picric acid (Figure 5b) [$F(3, 46) = 13.207$, $P < 0.001$]. The strains with the taster allele avoided to a greater degree than the strains with the nontaster allele, but the SW mean was not significantly different from the SW.B6 mean (Tukey, $P > 0.01$). The pattern suggested *Soa* allele influence, but did not

rule out the effects of other genes. There were no significant strain differences at 0.1 mM [$F(3, 46) = 1.337$, $P > 0.25$].

The pattern expected when background genetic variation controls avoidance differences can be seen in Figure 6. In this extended test with 10% ethanol, the SW mice avoided while the B6 mice were indifferent, but the congenic strains closely followed the inbred strains with the same background genomes despite having different *Soa* alleles. Again, the pattern was so clear that statistical analysis seemed redundant. Background variation also affected avoidance of L-phenylalanine (Figure 7a), though the avoidance differences were not as striking as for ethanol. Significant strain differences were found in the SW.B6 panel at 10 mM [$F(2, 37) = 6.928$, $P < 0.003$]. The SW and SW.B6 means at this concentration differed from the B6 mean (Tukey, $P < 0.01$) but did not differ from each other (Tukey, $P > 0.01$). The congenic strain thus followed its background genome, not its *Soa* allele. At 100 mM in both panels, to the extent that the inbred strains diverged, the congenic mice tended to follow their background genomes as well.

The quinine HCl pattern was only minimally informative (Figure 7b). Although the SW mice avoided to a slightly greater extent than the B6 mice in both panels, in neither was the difference significant (all $P > 0.01$). In addition, while the B6.SW mice closely followed the B6 strain with the same background genome, the SW.B6 mice diverged from the SW strain, suggesting an *Soa* allele influence.

The SW, B6 and B6.SW mice were tested on two forms of

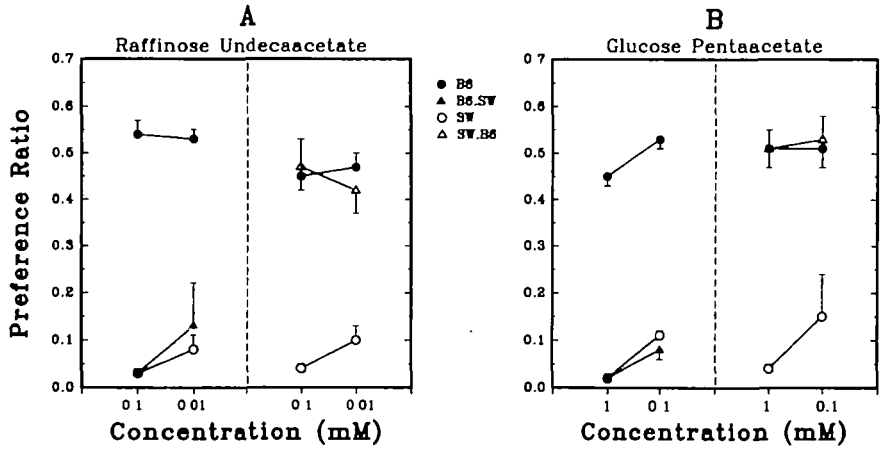

Figure 4 Quartet response functions to RUA **(a)** and GPA **(b)**. For each compound, the B6.SW and SW.B6 strains were tested in separate panels with the inbred strains (data in B6.SW panel for RUA from Whitney *et al.*, 1990). The *Soa* locus controlled avoidance of both acetylated sugars.

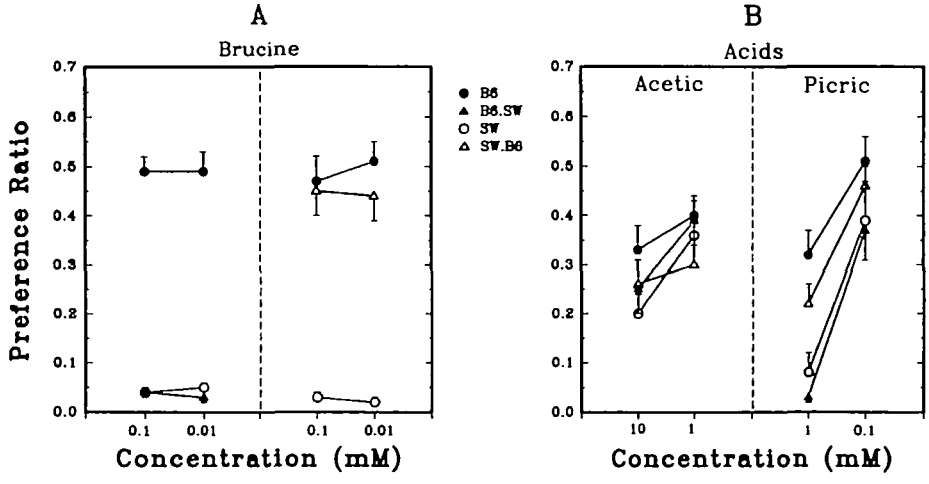

Figure 5 Quartet response functions to brucine **(a)** and two acids **(b)**. The congenic strains were tested with brucine in separate panels (data in B6.SW panel from Whitney and Harder, 1994). The *Soa* locus controlled brucine avoidance. All four quartet members were tested together with acetic acid and picric acid. No strain differences were found with acetic acid. A possible *Soa* effect was found at 1 mM picric acid.

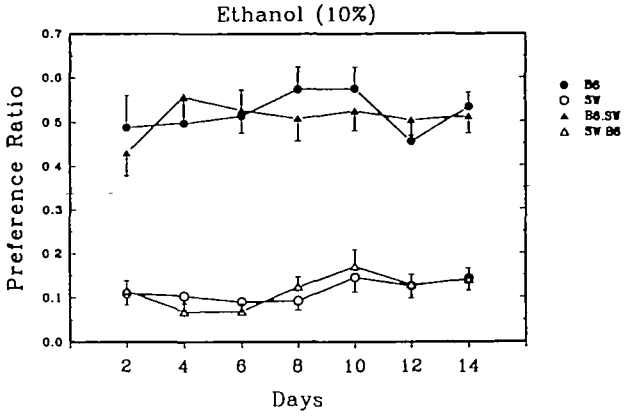

Figure 6 Quartet response functions in extended testing with 10% ethanol. The inbred strains differed across all tests, and each congenic strain closely followed the inbred with the same background genome. This indicated that *Soa* allele differences had no effect.

ribose tetraacetate (pyranose and furanose) with quite different outcomes (Figure 8b). An SOA-like pattern typical of acetylated sugars was found for 10 mM RPTA [$F(2, 41) = 56.982$, $P < 0.001$; Tukey, $P < 0.01$], but no avoidance differences were found for RFTA. The B6 nontaster mice avoided 10 mM RFTA as well as did the SW and B6.SW taster mice. A second panel including all four members of the quartet was also tested on both compounds (Figure 8a). An *Soa* effect (SW and B6.SW means < SW.B6 and B6 means) was again found for 10 mM RPTA [$F(3, 52) = 66.160$, $P < 0.001$; Tukey, $P < 0.01$], and also for 3 mM RPTA [$F(3, 52) = 35.835$, $P < 0.001$; Tukey, $P < 0.01$]. As in the previous RFTA panel, the B6 mice avoided 10 mM RFTA and were joined by the SW.B6 mice. The taster and nontaster means were slightly more separated than before,

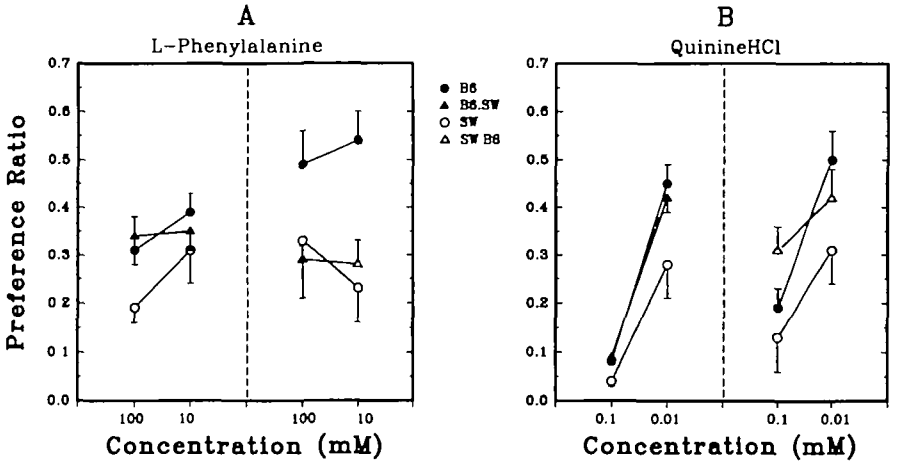

Figure 7 Quartet response functions to L-phenylalanine **(a)** and quinine HCl **(b)** [data in B6.SW panel for quinine HCl from Whitney *et al.* (1990)]. Background genome effects were seen in the SW.B6 panel for L-phenylalanine, with a similar trend in the B6.SW panel. No strain differences were found for quinine HCl.

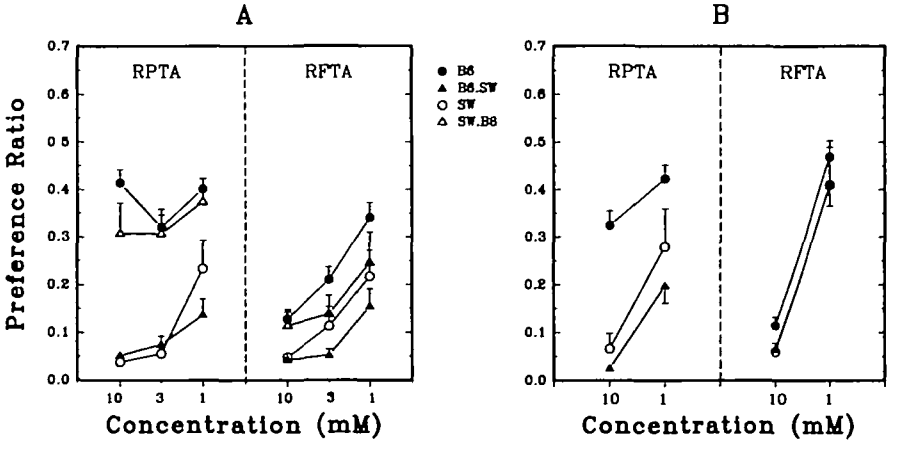

Figure 8 Quartet response functions to ribopyranose and ribofuranose tetraacetates. An *Soa* locus effect was found in both RPTA panels. Strain differences were absent **(b)** or greatly reduced **(a)** in the RFTA panels.

resulting in significant strain differences [$F(3, 52) = 10.250$, $P < 0.001$], but the complete SOA pattern was not found. The SW.B6 mean did not differ significantly from any of the other means (Tukey, $P > 0.01$).

Discussion

The *Soa* locus completely controls SOA avoidance in the congenic quartet. The *Soa* locus also controlled avoidance of the bitter substances RUA, GPA, RPTA, brucine and perhaps picric acid. *Soa* allele differences had no effect on L-phenylalanine or ethanol avoidance. The one reported Soa locus effect on avoidance of a non-bitter substance, acetic acid (Whitney *et al.*, 1990), was not confirmed. Together, these findings support the conclusion that the influence of the Soa locus is restricted to a subset of bitter compounds. This subset includes compounds other than acetylated sugars, but does not extend to all bitters. Soa allele differences have not been shown to reliably affect preference/avoidance differences for any non-bitter compounds.

Small inbred strain differences (i.e. similar phenotypic expression of background genomes) and inconsistent congenic strain relative sensitivities have characterized the quartet responses to not only acetic acid but also citric and hydrochloric acids (Whitney *et al.*, 1990; Harder *et al.*, 1994). The B6 and SW strains appear not to provide a useful behavioral contrast for sour acids in general. The reliability of the somewhat larger contrast found for picric acid needs to be confirmed before the relative influences of the Soa locus and background loci can be determined. No avoidance differences have been found for tannic acid, which is also bitter (Whitney *et al.*, 1991).

The B6 and SW strains provided a poor contrast for quinine HCl as well. Similar results have been seen with quinine sulfate (Whitney *et al.*, 1990) and free-base quinine (D.B. Harder, unpublished data). The C3HeB/FeJ inbred strain background provides a better contrast to the SW strain for quinine (Whitney and Harder, 1994). Both Soa locus and background locus effects on quinine avoidance have been demonstrated via crosses involving C3HeB/FeJ mice (Boughter *et al.*, 1992). The recently completed C3.SW-*Soa*^{a} congenic set should provide for a clearer assessment of quinine avoidance determination (Boughter and Whitney, 1995).

The RFTA findings constitute the first reported lack of Soa locus effect for an acetylated sugar. The observation that the taster–nontaster difference for ribose tetraacetate is at least greatly reduced, if not completely eliminated, by a specific change in its molecular configuration may aid in identifying the basis for Soa-influenced avoidance differences.

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