Published in final edited form as:
Mamm Genome. 2001 September ; 12(9): 695–699.

High-resolution genetic mapping of the sucrose octaacetate taste aversion (*Soa*) locus on mouse Chromosome 6

Alexander A. Bachmanov[1], **Xia Li**[1], **Shanru Li**[1], **Mauricio Neira**[2], **Gary K. Beauchamp**[1,3], and **Edwin A. Azen**[2]

[1]Monell Chemical Senses Center, Philadelphia, PA 19104, USA

[2]Departments of Medicine and Medical Genetics, University of Wisconsin, Madison, WI 53706, USA

[3]Department of Psychology and School of Veterinary Medicine, University of Pennsylvania, Philadelphia, PA 19104, USA

Abstract

An acetylated sugar, sucrose octaacetate (SOA), tastes bitter to humans and has an aversive taste to at least some mice and other animals. In mice, taste aversion to SOA depends on allelic variation of a single locus, *Soa*. Three *Soa* alleles determine 'taster' (Soa^a), 'nontaster' (Soa^b), and 'demitaster' (Soa^c) phenotypes of taste sensitivity to SOA. Although *Soa* has been mapped to distal Chromosome (Chr) 6, the limits of the *Soa* region have not been defined. In this study, mice from congenic strains SW.B6-Soa^b, B6.SW-Soa^a, and C3.SW-$Soa^{a/c}$ and from an outbred CFW strain were genotyped with polymorphic markers on Chr 6. In the congenic strains, the limits of introgressed donor fragments were determined. In the outbred mice, linkage disequilibrium and haplotype analyses were conducted. Positions of the markers were further resolved by using radiation hybrid mapping. The results show that the *Soa* locus is contained in a ~1-cM (3.3–4.9 Mb) region including the *Prp* locus.

Presumably, sensitivity to bitter compounds arose as a means of detecting toxic agents. Consequently, many structurally diverse compounds taste bitter to humans and other animals. An ability to detect bitter is now thought to be based on specific recognition by a family of putative bitter taste receptors (Adler et al. 2000; Matsunami et al. 2000) and perhaps other mechanisms (Lindemann 1996).

One important model system for investigating the bitter taste mechanism involves the acetylated sugar, sucrose octaacetate (SOA), which tastes bitter to humans and has an aversive taste to at least some mice and other animals. In mice, taste aversion to SOA depends on allelic variation of a single locus, *Soa*, with three known alleles. The Soa^a allele determines a 'taster' phenotype of strong SOA avoidance, the Soa^b allele determines a 'nontaster' phenotype of indifference to SOA, and the Soa^c allele determines a 'demitaster' phenotype of intermediate SOA sensitivity (demitasters are indifferent to 0.1 mM SOA, but avoid 1 mM SOA). An order of a phenotypic dominance of these alleles is taster > nontaster > demitaster (Harder et al. 1992). Three *Soa*-congenic strains originating from the SWR/J (SWR; Soa^a), C57BL/6J (B6; Soa^b), and C3HeB/FeJ (C3He; Soa^c) inbred strains have been selected: homozygous B6.SW-Soa^a (B6.SW) and SW.B6-Soa^b (SW.B6) strains, and a heterozygous segregating C3.SW-$Soa^{a/c}$ (C3.SW) strain (Whitney et al. 1989; Boughter and

Correspondence to: Alexander A. Bachmanov, Monell Chemical Senses Center, 3500 Market Street, Philadelphia, PA, 19104, USA. Phone: (215) 573-8094. Fax: (215) 898-2084. bachmanov@monell.org..

Whitney 1995; Harder et al. 1996). Within an outbred CFW strain, there is a phenotypical variation in SOA avoidance, which is due to segregation of taster and demitaster *Soa* alleles (Gannon and Whitney 1989; Harder et al. 1992).

The *Soa* locus has been mapped to distal Chr 6 (Azen 1991; Capeless et al. 1992; Lush et al. 1995). This region contains several candidate genes thought to be involved in bitter taste perception. A recent study has shown that one of these genes, *Prp*, is a less likely candidate for *Soa* (Harder et al. 2000). Several genes encoding G protein-coupled receptors expressed in taste tissue were also found in the *Soa* region (Adler et al. 2000; Matsunami et al. 2000). However, there is still no evidence that one or some of these receptors bind SOA as a ligand. A mechanism for transduction of SOA bitter taste could be identified by using positional cloning of the *Soa* locus. This requires high-resolution genetic and physical mapping of the *Soa* region. The chromosomal region proximal to *Prp* has been physically mapped (Brown et al. 1999; Depatie et al. 2000), but it does not include the whole *Soa* interval.

To begin positional cloning of *Soa*, we conducted this study aimed to determine a *Soa* nonrecombinant interval. Replicate *Soa*-congenic strains were used to define an overlapping part of the donor fragments as a *Soa* critical region. Outbred CFW mice were used to conduct linkage-disequilibrium analyses similar to those in human population studies. Finally, marker order and physical distances were estimated by using radiation hybrid (RH) mapping.

Materials and methods

Mouse genotyping

The tail tissues were collected from the following inbred, congenic, and outbred mice:

- **i.** Mice from SWR/J (SWR), C57BL/6J (B6), and C3HeB/FeJ (C3He) inbred strains (n = 3 for each strain).
- **ii.** Mice from SW.B6-Soa^b (SW.B6) congenic strain in generation $NE_{10}F_{23}$ (n = 3). In all tested SWR, B6, C3He, and SW.B6 mice, genotypes for all tested markers were identical within the strains.
- **iii.** Mice from 11 substrains (# 1–6, 8–12; n = 1 for each strain) of an extinct B6.SW-Soa^a (B6.SW) congenic strain in generation $N_{11}F_4$. These B6.SW mice were previously phenotyped (they all were SOA tasters), and their *Prp* RFLP genotypes were examined (substrains # 1, 2, 6, and 8 were homozygous for an SWR allele, substrains # 3–5 and 9–12 were heterozygotes with one allele from the SWR strain; G. Whitney and E.A. Azen, unpublished).
- **iv.** Mice from three substrains (# 3, 5, and 6) of a segregating C3.SW-$Soa^{a/c}$ (C3.SW) congenic strain in generations N_{29}–N_{30}. All C3.SW mice were phenotyped in 96-h, two-bottle tests with 0.1 m_M SOA (methods are described in Bachmanov et al. 1996). The mice could be divided into two nonoverlapping groups: approximately one half strongly avoided 0.1 m_M SOA (phenotype of SOA tasters; $Soa^{a/c}$ heterozygotes), and the other half was indifferent to 0.1 m_M SOA (phenotype of demitasters; homozygotes for C3He Soa^c allele; data not shown). Genotyping of these mice confirmed that the tasters had one copy of a donor chromosome fragment from the SWR strain, whereas the demitasters were homozygous for alleles of the C3He inbred partner strain for all tested markers. Within substrains # 3 and 5, there were two groups of mice with different sizes of donor fragments; these groups are referred to as substrains # 3a, 3b, 5a, and 5b. Numbers of genotyped SOA taster mice from each substrain were 1 (# 3a), 4 (# 3b), 5 (# 5a), 12 (# 5b), and 6 (# 6).

v. Mice from CFW outbred strain (n = 41). In these 41 mice, SOA avoidance and Prp RFLP haplotypes had been characterized previously (Capeless et al. 1992; Harder et al. 1992). Among them, 21 were SOA tasters and had one or two copies of an SWR-type *Prp* allele (allele C, Table 1), and the other 20 were demitasters and had no SWR-type *Prp* alleles (a summary of these data, but not individual genotypes, was published in Harder et al. 1992). Linkage disequilibrium was analyzed by using an approach similar to one described in Peissel et al. (2000). Frequencies of alleles for each marker were compared between taster and demitaster groups by using Pearson Chi-square tests. The frequency of each allele was calculated as a ratio of a total number of alleles (one for heterozygotes and two for homozygotes) to total number of Chr 6 chromosomes (number of mice times two). The p values from the Chi-square tests were transformed into their negative base-10 logarithms. To correct for multiple (n = 28) comparisons, we used a Bonferroni correction, setting the level of statistical significance at $p < 0.05/28 \approx 0.0018$, corresponding to $-\log_{10}(p) > 2.75$.

Genomic DNA was purified from mouse tails with the NaOH/Tris method (Truett et al. 2000). Genotyping was performed with methods described elsewhere (Bachmanov et al. 1997). Briefly, microsatellite (simple sequence length polymorphisms, or SSLP) markers were amplified by using PCR with primers purchased from Research Genetics, Inc. (Huntsville, Ala.). The denatured PCR products were electrophoresed on a polyacrylamide sequencing gel and visualized by autoradiography.

The tested markers spanned a region of Chr 6 between 36.5 and 74.1 cM from the centromere (chromosomal positions were obtained from the Mouse Genome Database, MGD, http://www.informatics.jax.org). In the inbred and *Soa*-congenic mice, 38 SSLP markers were tested (Fig. 1). In the CFW outbred mice, 31 SSLP markers were tested (Fig. 2).

Radiation hybrid (RH) mapping

The T31 mouse-hamster radiation hybrid panel (Research Genetics) was genotyped according to a standard protocol (http://www-shgc.stanford.edu/Mapping/rh/procedure/rhassaynew.html) with markers *D6Mit13.1, 110, 111, 219, 290, 370*, and *374*. All markers were tested at least twice and scored in ethidium bromide-stained agarose gels. The data were submitted for analysis to The Jackson Laboratory RH Database.

Results and Discussion

Haplotypes of the Soa-congenic strains

Of the 38 genotyped SSLP markers, 27 (71%) were polymorphic between the SWR and C3He strains, and 34 (89%) were polymorphic between the B6 and SWR strains. The results of genotyping are presented in Fig. 1. The *Soa*-containing region is limited to an overlapping part of all donor fragments, which is flanked proximally by *D6Mit370* and distally by a group of markers, *D6Mit111, D6Mit110*, and *D6Mit290* (their positions could not be ordered based on aligning donor fragments, but the RH mapping described below and the MGD map placed *D6Mit111* as the closest distal flanking marker for the *Soa* region). The distance between the *D6Mit370* and *D6Mit111* markers flanking the *Soa*-containing region is ~1 cM (MGD map). The overlapping part of the donor fragments includes three markers, *D6Mit219* and two markers within the *Prp* locus, *D6Mit13* and *47.MMPRPMPB.*

Allele frequencies in SOA-taster and -demitaster outbred CFW mice

Of the 31 genotyped SSLP markers, 27 (87%) were polymorphic, with two to four alleles each. Four markers were in linkage disequilibrium with the SOA taste aversion phenotype: *D6Mit370, D6Mit219, D6Mit13*, and the *Prp* RFLP (Fig. 2). Genotypes for three markers, *Prp* RFLP, *D6Mit13* and *D6Mit219*, were found in combinations, suggesting that they are parts of nonrecombinant allelic haplotypes (Table 1). These haplotypes were consistent with the dominance of the taster Soa^a-allele: all tasters (expected to have one or two Soa^a-alleles) had one or two copies of alleles C (*Prp* RFLP) and B (*D6Mit13* and *D6Mit219*), whereas nontasters had none of these alleles (exemplified in Fig. 3). This demonstrates that *Soa, Prp*, and *D6Mit219* have not been separated by recombinations during many generations of existence of the CFW strain, and therefore they are tightly linked.

D6Mit370 was also in linkage disequilibrium (Fig. 2), but 7 of 41 mice did not retain *D6Mit370* as a part of a haplotype formed by the other three markers (Table 1). Although allele B of *D6Mit370* was predominantly represented among SOA tasters, one taster mouse did not have this allele, and one demitaster mouse was a B/C heterozygote. Thus, *D6Mit370* can be excluded from the *Soa*-containing nonrecombinant interval.

The results of genotyping the CFW mice were consistent with the *Soa*-congenic data. In both experiments, *D6Mit219* and *Prp* were within the *Soa* region, and *D6Mit370* appeared to be the closest proximal flanking marker. However, analyses of the CFW mice did not help with ordering markers within the distal flanking group: *D6Mit111* was not polymorphic in the CFW strain, and linkage disequilibrium for *D6Mit290* and *D6Mit110* was not significant.

RH mapping

The genetic analyses described above have shown that the proximal boundary of the *Soa* region is *D6Mit370*, and the distal boundary is defined by a group of markers, *D6Mit111, D6Mit110*, and *D6Mit290*. However, the order of markers within this distal group could not be established. To resolve the order of the markers and to estimate the physical size of the *Soa* region, we genotyped these markers using the T31 RH panel. The following order and distances were obtained: *D6Mit370*–45.2 cR– *D6Mit13.1*–9.0 cR–*D6Mit219*–20.0 cR–*D6Mit111*–3.1 cR– *D6Mit290*/*D6Mit110*/*D6Mit374*. Therefore, the two closest markers flanking the *Soa* nonrecombinant interval are *D6Mit370* and *D6Mit111* (separated by 74.2 cR).

Although 1 cR corresponds to approximately 100 kb on average throughout the genome (Van Etten et al. 1999), this ratio varies from region to region (Rowe et al. 2000) and could be best estimated empirically. The physical distance between *D6Mit370* and *D6Mit13.1* estimated from two independent studies (Brown et al. 1999; Depatie et al. 2000) is 2–3 Mb. (Although these two publications do not show the exact position of *D6Mit370*, they describe YACs 52A6 and 95E6, which contain *D6Mit370* according to the MIT database, http://www-genome.wi.mit.edu). The distance between these two markers in our RH mapping experiment was 45.2 cR, which gives us an estimation of 1 cR = 44–66 kb. Thus, a 74.2-cR distance between *D6Mit370* and *D6Mit111* corresponds to 3.3–4.9 Mb.

In conclusion, we have defined the nonrecombinant *Soa* interval, which has a size of ~1 cM (74.2 cR, or 3.3–4.9 Mb). Frequency of recombinations within this interval appears to be lower than the average for the genome (1 cM/2 Mb; Silver 1995). This region includes the *Prp* locus and probably some of the putative bitter-taste receptor genes (Adler et al. 2000; Matsunami et al. 2000). This study sets up a stage for physical mapping of the *Soa* region and positional cloning of the *Soa* gene.

Acknowledgments

We thank Glayde Whitney and David B. Harder for providing the SW.B6-*Soa*^{b} and C3.SW-*Soa*$^{a/c}$ congenic mice, John D. Boughter for providing tissues of mice from the C3.SW-*Soa*$^{a/c}$ congenic substrain # 3, Lucy Rowe (The Jackson Laboratory) for assistance with RH data analysis, Ed Amberger for technical help, Tatiana M. Foroud for advice on linkage disequilibrium analysis, and Danielle R. Reed for valuable comments on an earlier version of the manuscript. Supported by NIH grants R03 DC03854 (A.A.Bachmanov), R01 DC00882 (G.K.Beauchamp) and 5 R37 DEO3658 (E.A.Azen).

References

Adler E, Hoon MA, Mueller KL, Chandrashekar J, Ryba NJP, Zuker CS. A novel family of mammalian taste receptors. Cell. 2000; 100:693–702. [PubMed: 10761934]

Azen, EA. Linkage studies of genes for salivary proline-rich proteins and bitter taste in mouse and human. In: Wysocki, CJ.; Kare, MR., editors. Genetics of Perception and Communication. Marcel Dekker; New York: 1991. p. 279-290.

Azen EA, Davisson MT, Cherry M, Taylor BA. *Prp* (proline-rich protein) genes linked to markers *Es-12* (esterase-12), *Ea-10* (erythrocyte alloantigen), and loci on distal mouse chromosome 6. Genomics. 1989; 5:415–422. [PubMed: 2613230]

Bachmanov AA, Tordoff MG, Beauchamp GK. Ethanol consumption and taste preferences in C57BL/6ByJ and 129/J mice. Alcohol Clin Exp Res. 1996; 20:201–206. [PubMed: 8730208]

Bachmanov AA, Reed DR, Ninomiya Y, Inoue M, Tordoff MG, et al. Sucrose consumption in mice: major influence of two genetic loci affecting peripheral sensory responses. Mamm Genome. 1997; 8:545–548. [PubMed: 9250857]

Boughter JD, Whitney G. C3.SW-*Soa*^{a} heterozygous congenic taster mice. Behav Genet. 1995; 25:233–237. [PubMed: 7598666]

Brown MG, Zhang J, Du Y, Stoll J, Yokoyama WM, Scalzo AA. Localization on a physical map of the NKC-linked *Cmv1* locus between *Ly49b* and the *Prp* gene cluster on mouse chromosome 6. J Immunol. 1999; 163:1991–1999. [PubMed: 10438936]

Capeless CG, Whitney G, Azen EA. Chromosome mapping of *Soa*, a gene influencing gustatory sensitivity to sucrose octaacetate in mice. Behav Genet. 1992; 22:655–663. [PubMed: 1290451]

Depatie C, Lee SH, Stafford A, Avner P, Belouchi A, et al. Sequence-ready BAC contig, physical, and transcriptional map of a 2-Mb region overlapping the mouse chromosome 6 host-resistance locus *Cmv1*. Genomics. 2000; 66:161–174. [PubMed: 10860661]

Gannon KS, Whitney G. Sucrose octaacetate tasting in a heterogeneous population of CFW mice. Behav Genet. 1989; 19:417–431. [PubMed: 2757593]

Harder DB, Capeless CG, Maggio JC, Boughter JD, Gannon KS, et al. Intermediate sucrose octa-acetate sensitivity suggests a third allele at mouse bitter taste locus *Soa* and *Soa-Rua* identity. Chem Senses. 1992; 17:391–401.

Harder DB, Gannon KS, Whitney G. SW.B6-*Soa*^{b} nontaster congenic strains completed and a sucrose octaacetate congenic quartet tested with other bitters. Chem Senses. 1996; 21:507–517. [PubMed: 8902280]

Harder DB, Azen EA, Whitney G. Sucrose octaacetate avoidance in nontaster mice is not enhanced by two type-A *Prp* transgenes from taster mice. Chem Senses. 2000; 25:39–45. [PubMed: 10667992]

Lindemann B. Taste reception. Physiol Rev. 1996; 76:719–766.

Lush IE, Hornigold N, King P, Stoye JP. The genetics of tasting in mice. VII. Glycine revisited, and the chromosomal location of *Sac* and *Soa*. Genet Res. 1995; 66:167–174. [PubMed: 8522158]

Matsunami H, Montmayeur JP, Buck LB. A family of candidate taste receptors in human and mouse. Nature. 2000; 404:601–604. [PubMed: 10766242]

Peissel B, Zaffaroni D, Zanesi N, Zedda I, Manenti G, et al. Linkage disequilibrium and haplotype mapping of a skin cancer susceptibility locus in outbred mice. Mamm Genome. 2000; 11:979–981. [PubMed: 11063253]

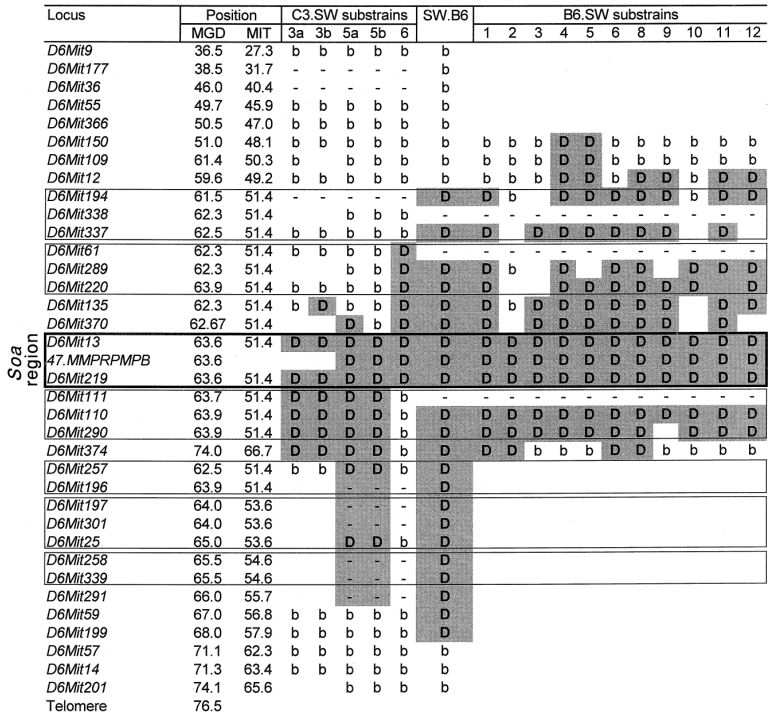
Rowe LB, Barter ME, Eppig JT. Cross-referencing radiation hybrid data to the recombination map: lessons from mouse chromosome 18. Genomics. 2000; 69:27–36. [PubMed: 11013072]

Silver, LM. Mouse Genetics: Concepts and Applications. Oxford University Press; New York: 1995.

Truett GE, Heeger P, Mynatt RL, Truett AA, Walker JA, Warman ML. Preparation of PCR-quality mouse genomic DNA with hot sodium hydroxide and tris (HotSHOT). Biotechniques. 2000; 29:52–54. [PubMed: 10907076]

Van Etten WJ, Steen RG, Nguyen H, Castle AB, Slonim DK, et al. Radiation hybrid map of the mouse genome. Nat Genet. 1999; 22:384–387. [PubMed: 10431245]

Whitney G, Harder DB, Gannon KS. The B6.SW bilineal congenic sucrose octaacetate (SOA)-taster mice. Behav Genet. 1989; 19:409–431. [PubMed: 2757592]



Locus	Position		C3.SW substrains					SW.B6	B6.SW substrains										
	MGD	MIT	3a	3b	5a	5b	6		1	2	3	4	5	6	8	9	10	11	12
D6Mit9	36.5	27.3	b	b	b	b	b	b											
D6Mit177	38.5	31.7	-	-	-	-	-	b											
D6Mit36	46.0	40.4	-	-	-	-	-	b											
D6Mit55	49.7	45.9	b	b	b	b	b	b											
D6Mit366	50.5	47.0	b	b	b	b	b	b											
D6Mit150	51.0	48.1	b	b	b	b	b	b	b	b	b	D	D	b	b	b	b	b	b
D6Mit109	61.4	50.3	b		b	b	b	b	b	b	b	D	D	b	b	b	b	b	b
D6Mit12	59.6	49.2	b	b	b	b	b	b	b	b	b	D	D	b	D	D	b	D	D
D6Mit194	61.5	51.4	-	-	-	-	-	D	D	b		D	D	D	D	D	b	D	D
D6Mit338	62.3	51.4			b	b	b	-	-	-	-	-	-	-	-	-	-	-	-
D6Mit337	62.5	51.4	b	b	b	b	b	D	D		D	D	D	D	D	D		D	
D6Mit61	62.3	51.4	b	b	b	b	D	-	-	-	-	-	-	-	-	-	-	-	-
D6Mit289	62.3	51.4			b	b	D	D	D	b		D		D	D		D	D	D
D6Mit220	63.9	51.4	b	b	b	b	D	D	D			D	D	D	D	D	D		D
D6Mit135	62.3	51.4	b	D	b	b	D	D	D	b	D	D	D	D	D	D		D	D
D6Mit370	62.67	51.4			D	b	D	D	D		D	D	D	D	D	D		D	
D6Mit13	63.6	51.4	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D
47.MMPRPMPB	63.6				D	D	D	D	D	D	D	D	D	D	D	D	D	D	D
D6Mit219	63.6	51.4	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D	D
D6Mit111	63.7	51.4	D	D	D	D	b	-	-	-	-	-	-	-	-	-	-	-	-
D6Mit110	63.9	51.4	D	D	D	D	b	D	D	D	D	D	D	D	D	D	D	D	D
D6Mit290	63.9	51.4	D	D	D	D	b	D	D	D	D	D	D	D	D		D	D	D
D6Mit374	74.0	66.7	D	D	D	D	b	D	D	D	b	b	b	D	D	b	b	b	b
D6Mit257	62.5	51.4	b	b	D	D	b	D											
D6Mit196	63.9	51.4			-	-	-	D											
D6Mit197	64.0	53.6			-	-	-	D											
D6Mit301	64.0	53.6			-	-	-	D											
D6Mit25	65.0	53.6			D	D	b	D											
D6Mit258	65.5	54.6			-	-	-	D											
D6Mit339	65.5	54.6			-	-	-	D											
D6Mit291	66.0	55.7			-	-	-	D											
D6Mit59	67.0	56.8	b	b	b	b	b	D											
D6Mit199	68.0	57.9	b	b	b	b	b	D											
D6Mit57	71.1	62.3	b	b	b	b	b	b											
D6Mit14	71.3	63.4	b	b	b	b	b	b											
D6Mit201	74.1	65.6			b	b	b	b											
Telomere	76.5																		

Fig. 1.

Haplotypes defining donor fragments in three *Soa*-congenic strains. Substrains are indicated by numbers. “D” and shading indicate donor strain genotype; “b”, background strain genotype; “-”, no polymorphisms; cells are blank if data are missing. All C3.SW mice had one copy of a donor fragment; all SW.B6 mice had two copies of donor fragments; among the B6.SW mice, substrains # 1, 2, 6, and 8 had two copies, and substrains # 3–5 and 9–12 had one copy of the donor fragment. Markers are arranged based on their positions on the MGD and MIT maps, our genetic and RH mapping data, and physical mapping data (Brown et al. 1999; Depatie et al. 2000). *D6Mit374* is mapped on the MGD/MIT maps more distally (at 74.0 and 66.7 cM respectively), but our genetic and RH mapping position this marker to a more proximal location, which corresponds to genetic and physical mapping by Depatie et al. (2000). Boxes indicate markers mapped to the same bins because their positions within a bin could not be ordered based on haplotypes of donor fragments (some of them could be ordered within the bins based on the MGD map). Of the 38 markers tested, only two (*D6Mit52* and *D6Mit113*) were not polymorphic among the three inbred strains, and they are not shown in the figure. Two C3.SW substrains (#5b and 6) had the smallest overlap of the donor fragments, and therefore they were the most informative for defining the limits of the *Soa* region.

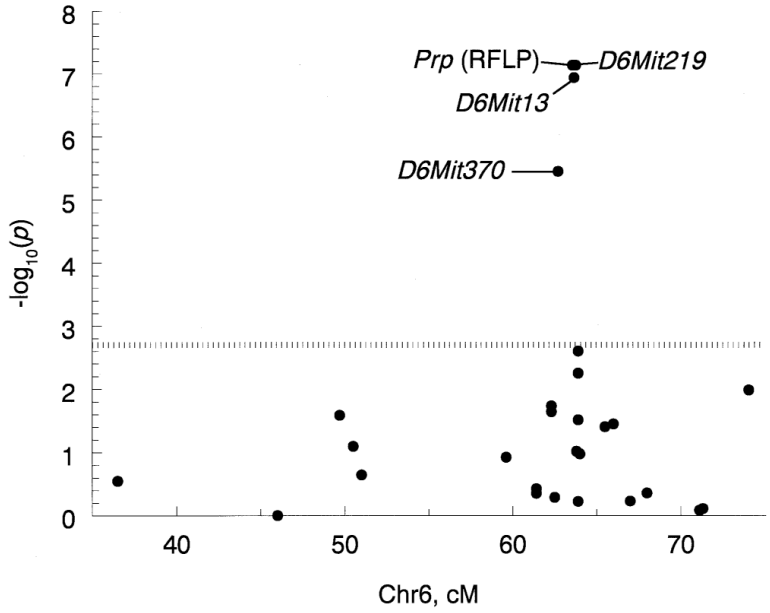


Fig. 2.

Linkage disequilibrium mapping of the *Soa* locus. Dots represent negative $\log_{10}$ of *p*-values in χ^2 tests comparing frequencies of alleles of polymorphic markers genotyped in SOA-taster and SOA-demitaster CFW mice (D6Mit9, 12, 13, 14, 36, 52, 55, 57, 59, 61, 109, 110, 135, 150, 195, 196, 199, 219, 220, 258, 290, 291, 301, 337, 366, 370, 374, and an RFLP of the *Prp* locus; *D6Mit111, 177, 257*, and *339* markers have been tested but were not polymorphic). Marker positions relative to the centromere are based on the MGD map. Dotted horizontal line shows threshold of statistical significance [$-\log_{10}(p) = 2.75$, corresponding to $p = 0.0018$].

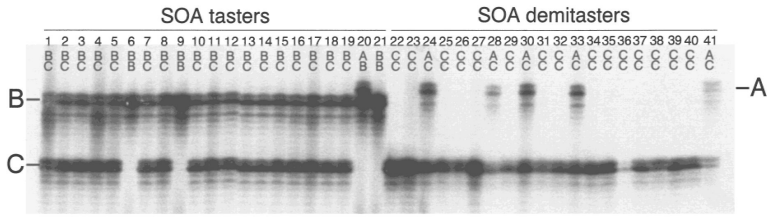


Fig. 3.

Genotypes of the CFW mice for *D6Mit219*. A, B, and C indicate alleles. Mouse numbers (corresponding to those in Table 1) and genotypes are shown at the top. Mice # 1–21 are SOA tasters; mice # 22–41 are SOA demitasters. All tasters have one (# 1–5, 7, 8, and 10–20) or two (# 6, 9, and 21) copies of allele B, whereas none of the SOA demitasters have this allele.

Table 1

Genotypes of 41 CFW mice at loci showing significant linkage disequilibrium with the SOA taste aversion phenotype.

Locus: **cM Pos-n (MGD):** **−log10(*p*):** **Alleles[a]:**	***D6Mit370*** **62.67** **5.4** **A,B,C**	***Prp (RFLP)*** **63.6** **7.1** **A,C,E**	***D6Mit13*** **63.6** **6.9** **A,B,C,D**	***D6Mit219*** **63.6** **7.1** **A,B,C**	**Genotype[b]**	**SOA Sensitivity**	**Mouse[d]**
	B/C[c]	C/C	B/B	B/B	1	taster	6
	B/C[c]	C/C	B/B	B/B	1	taster	9
	B/C[c]	C/C	B/B	B/B	1	taster	21
	B/C	C/A	B/C	B/C	2	taster	1
	B/C	C/A	B/C	B/C	2	taster	2
	B/C	C/A	B/C	B/C	2	taster	3
	B/C	C/A	B/C	B/C	2	taster	4
	B/C	C/A	B/C	B/C	2	taster	5
	B/C	C/A	B/C	B/C	2	taster	7
	B/C	C/A	B/C	B/C	2	taster	8
	B/C	C/A	B/C	B/C	2	taster	10
	B/C	C/A	B/C	B/C	2	taster	11
	B/C	C/A	B/C	B/C	2	taster	12
	B/C	C/A	B/C	B/C	2	taster	13
	B/C	C/A	B/C	B/C	2	taster	15
	B/C	C/A	B/C	B/C	2	taster	16
	B/C	C/A	B/C	B/C	2	taster	17
	B/C	C/A	B/C	B/C	2	taster	18
	B/C	C/A	B/C	B/C	2	taster	19
	C/C[c]	C/A	B/C	B/C	2	taster	14
	B/B[c]	C/E	B/A	B/A	3	taster	20
	C/C	A/A	C/C	C/C	4	demitaster	22
	C/C	A/A	C/C	C/C	4	demitaster	23
	C/C	A/A	C/C	C/C	4	demitaster	25
	C/C	A/A	C/C	C/C	4	demitaster	26
	C/C	A/A	C/C	C/C	4	demitaster	27
	C/C	A/A	C/C	C/C	4	demitaster	29
	C/C	A/A	C/C	C/C	4	demitaster	31
	C/C	A/A	C/C	C/C	4	demitaster	32
	C/C	A/A	C/C	C/C	4	demitaster	34
	C/C	A/A	C/C	C/C	4	demitaster	35
	C/C	A/A	C/C	C/C	4	demitaster	36
	C/C	A/A	C/C	C/C	4	demitaster	37
	C/C	A/A	C/C	C/C	4	demitaster	38

Locus: **cM Pos-n (MGD):** **−log10(*p*):** **Alleles[a]:**	***D6Mit370*** **62.67** **5.4** **A,B,C**	***Prp (RFLP)*** **63.6** **7.1** **A,C,E**	***D6Mit13*** **63.6** **6.9** **A,B,C,D**	***D6Mit219*** **63.6** **7.1** **A,B,C**	**Genotype[b]**	**SOA Sensitivity**	**Mouse[d]**
	C/C	A/A	C/C	C/C	4	demitaster	39
	C/C	A/A	C/C	C/C	4	demitaster	40
	C/A	A/E	C/A	C/A	5	demitaster	30
	C/A	A/E	C/A	C/A	5	demitaster	33
	C/A	A/E	C/A	C/A	5	demitaster	41
	B/C[c]	A/E	C/A	C/A	5	demitaster	28
	C/C[c]	A/E	C/D	C/A	6	demitaster	24

[a]Details on RFLP alleles of *Prp* are given in Azen et al. (1989; 1991) and Harder et al. (1992). Relative allele sizes of the SSLP markers are: C > A > B (*D6Mit370*), D > A > B > C (*D6Mit13*), and A > B > C (*D6Mit219*).

[b]Genotypes consistent across the three markers, *Prp* RFLP, *D6Mit13*, and *D6Mit219*, respectively: (1) C/C - B/B - B/B; (2) C/A - B/C - B/C; (3) C/E - B/A - B/A; (4) A/A - C/C - C/C; (5) A/E - C/A - C/A; (6) A/E - C/D - C/A. Tasters have genotypes 1–3; demitasters have genotypes 4–6. Retention of these genotypes can be explained by four nonrecombinant allelic haplotypes: C-B-B, A-C-C, E-A-A, and E-D-A for *Prp* RFLP, *D6Mit13*, and *D6Mit219* respectively. Probably in a mouse # 24 with genotype 6, allele A of *D6Mit13* mutated into allele D, while being in a tight linkage with the rest of the *Prp* locus and with *D6Mit219*.

[c]*D6Mit370* genotypes suggesting recombinations with haplotypes of the distal group of markers (found in 7 of 41 mice).

[d]Numbers of mice correspond to those in Fig. 3.