The Relationship between PROP and Ethanol Preferences: An Evaluation of 4 Inbred Mouse Strains

Theresa L. White[1,2], Laura V. Dishaw[1], Paul R. Sheehe[2,3] and Steven L. Youngentob[2,3]

[1]Department of Psychology, Le Moyne College, Syracuse, NY 13214, USA, [2]Department of Neuroscience and Physiology, SUNY Upstate Medical University, Syracuse, NY 13210, USA and [3]SUNY Developmental Exposure Alcohol Research Center, Binghamton University, Binghamton, NY 13902, USA

Correspondence to be sent to: Steven L. Youngentob, Department of Neuroscience and Physiology, SUNY Upstate Medical University, 750 East Adams Street, Syracuse, NY 13210, USA. e-mail: youngens@upstate.edu

Abstract

Ethanol's taste attributes undoubtedly contribute to the development of drug preference. Ethanol's taste is both sweet and bitter. Taster status for bitter 6-*n*-propylthiouracil (PROP) has been proposed as a genetic marker for alcoholism; however, human results are conflicting. We collected preference scores for both tastants in 4 mouse strains selected on the basis of previously reported taste preference, with the generally accepted idea that inbred mice show minimal within-strain variation. Eighty-eight male mice (22 per strain) participated. The strains were as follows: C57BL/6J, ethanol preferring; BALB/cJ, ethanol avoiding; SWR/J, PROP avoiding; and C3HeB/FeJ, PROP neutral. Using a brief-access (1-min trials) 2-bottle preference test, we assessed the taste response of each strain to PROP and ethanol on separate days. Although PROP avoiding versus neutral mice could be segregated into significantly different populations, this was not the case for ethanol avoiding versus preferring mice, and all strains showed high variability. On average, only BALB/cJ, SWR/J, and C3HeB/FeJ mice conformed to their literature-reported preferences; nonetheless, there were a substantial number of discordant animals. C57BL/6J did not conform to previous results, indicating that they are ethanol preferring. Finally, we did not observe a significant relationship between PROP and ethanol preferences across strains. The high variability per strain and the number of animals in disagreement with their respective literature-reported preference raise concerns regarding their utility for investigations underlying mechanisms of taste-mediated ingestive responses. Absent postingestive consequences, the brief-access results suggest a possible degree of previously masked polymorphisms in taste preferences or a more recent drift in underlying genetic factors. The absence of a relationship between PROP and ethanol indicates that the bitter quality in ethanol may be more highly related to other bitter compounds that are mediated by different genetic influences.

Key words: alcoholism, brief-access preference testing, ethanol preference, inbred mouse strains, PROP preference

Introduction

Alcoholism is a disorder that affects millions of people worldwide. In the United States, the lifetime risk for developing alcohol dependence is approximately 15% in the general population (American Psychiatric Association 2000). It is estimated that 40–60% of the risk for the development of alcoholism can be attributed to genetics (American Psychiatric Association 2000). Of the polygenic factors, variations in the genetically determined chemosensory attributes of ethanol's taste undoubtedly play a role in the development of ethanol preference and potentially, subsequent intake (e.g., Lanier et al. 2005).

The taste qualities associated with ethanol are both sweet and bitter (Wilson et al. 1973; Scinska et al. 2000; Mattes and

DiMeglio 2001; Lemon et al. 2004; Blednov et al. 2007; Blizard 2007). With particular regard to this latter component quality, evaluation of taste sensitivity to 6-*n*-propylthiouracil (PROP, Barnicot et al. 1951; Bartoshuk et al. 1994) has shown that individuals vary widely in their ability to perceive bitterness and that this variability is genetically determined. Indeed, bitter sensitivity lies along a wide continuum that ranges from the inability to perceive PROP (i.e., tastes like water) to what is termed a ''super taster'' (i.e., intense bitter perception, Duffy et al. 2004). Because of these genetically determined differences in bitter taste, recent research has attempted to evaluate whether such variations can serve as a genetic marker for a variety of food preferences

(Drenowski and Rock 1995; Kaminski et al. 2000), most notably ethanol. The studies investigating the relationship between PROP taster status and ethanol preference, in humans, however, have produced conflicting results (Pelchat and Danowski 1992; Kranzler et al. 1996, 1998; DiCarlo and Powers 1998; Duffy et al. 2004; Lanier et al. 2005). Some studies support the relationship between taster status and ethanol intake, indicating that nontasters perceive the taste of ethanol to have fewer negative and more pleasant taste sensations (Duffy et al. 2004) and that taster status may serve as a genetic marker for alcoholism (DiCarlo and Powers 1998). By contrast, other studies demonstrate that there is no strong evidence for a connection between taster status and ethanol preference (Kranzler et al. 1996, 1998). One possible explanation for these contradictory findings is, perhaps, the myriad of social and psychological factors (e.g., Cloninger 1987; Brook et al. 1988; Smith 1994; Pitkänen et al. 2005; Schuckit and Smith 2006) that often confound human investigations into specific contributing factors of ethanol preference. Alternatively, the genetics underlying the perception of other bitter compounds (Hansen et al. 2006) may play a more prominent role.

An animal-based approach offers the possibility of a high level of experimental and genetic control for evaluating the relationship between bitter perception and ethanol preference. Prior investigations have independently identified differences among inbred mouse strains for PROP sensitivity (e.g., Nelson et al. 2003) and ethanol preference (e.g., Belknap et al. 1993). However, the testing procedures used to establish particular taste preferences have not been equivalent across all tastants and studies. More specifically, the research has typically used variations of the more traditional "extended-access" 2-bottle preference testing paradigm (Belknap et al. 1993; Harder and Whitney 1998) to determine specific strain preferences for a particular tastant. This testing procedure, although easily applied, allows an animal access to the tastant of interest for extended time periods (e.g., hours or even days). As such, in these studies it is difficult to separate preferences in consumption due to taste perceptions, from those resulting from potential nontaste postingestive consequences of a substance (which can, over time, influence apparent taste preference results; Davis et al. 1975; Spector 2003; Glendinning, Bloom, et al. 2005). Accordingly, it is possible that previously published reports (using the extended-access 2-bottle preference test) of ethanol preferring or avoiding inbred mouse strains (e.g., Belknap et al. 1993) may not accurately reflect the taste preferences of these strains. Rather, they may reflect preferences acquired as a result of a differential responsiveness to the postabsorptive properties of the drug or its interaction with taste perception. Recent findings of high variability in individual animal ethanol preference scores within the C57BL/6J strain may support this possible concern and suggest that this strain may not be as strongly ethanol preferring as previously reported (Little et al. 1999; O'Callaghan et al. 2002). In sim-

ilar fashion, nontaste-related postabsorptive consequences are also a concern in prior evaluations of PROP sensitivity. The effects of stimulus toxicity found in many bitter compounds may directly affect the results of extended-access 2-bottle tests. In addition, because PROP is a commonly used treatment for hyperthyroidism (Waseem et al. 1998), exposure to this chemical over extended periods could potentially induce hypothyroidism in test subjects, thereby influencing preference results.

In order to study the potential relationship between bitter (i.e., PROP) and ethanol preference, we obviated the foregoing concerns by uniformly applying brief-access 2-bottle testing methodology (Smith et al. 2001; Boughter et al. 2002; Glendinning, Chyou, et al. 2005) to an evaluation of both tastants in 4 genetically different strains of mice. Each strain was selected based on its literature-reported taste preference (i.e., C57BL/6J, ethanol preferring; BALB/cJ, ethanol avoiding; C3HeB/FeJ, PROP neutral; and SWR/J, PROP avoiding), using the extended-access 2-bottle procedure (e.g., Belknap et al. 1993; Nelson et al. 2003). In so doing, the goals of the present research were 2-fold: 1) to determine whether taste preferences within each mouse strain for a particular tastant are consistent with the previously reported literature when tested with a procedure that effectively eliminates nontaste postingestive effects and 2) to determine whether, and to what extent, there was a predictive relationship between a particular animal's bitter (i.e., PROP) and ethanol preferences.

Method

Subjects

A total of 88-male mice from 4 different strains (22 per strain) participated in the study. All mice were 6–10 weeks old and were purchased from Jackson Laboratories (Bar Harbor, ME). Each strain was chosen for either its ethanol preference or PROP sensitivity based upon previous reports in the literature (C57BL/6J, ethanol preferring and BALB/cJ, ethanol avoiding [Belknap et al. 1993]; SWR/J, PROP avoiding; and C3HeB/FeJ, PROP neutral [Nelson et al. 2003]). Testing occurred in 11 cohorts of mice, with each cohort consisting of 2 mice from each of the 4 strains (8 mice per cohort). During experimentation, mice were housed individually in standard ventilated micro-isolator cages in a temperature and humidity controlled vivarium. The animals were kept on a 12-h light–dark cycle, and all testing took place during the light portion of the cycle.

Testing apparatus

Taste preferences were determined using a computer-monitored 2-bottle preference apparatus, similar to the one used by Glendinning, Chyou, et al. (2005). This device consisted of a standard shoebox mouse cage ($28.5 \times 17.5 \times 12.5$ cm) that had been painted black. Two vertical slits (3×28 mm)

were cut side-by-side, approximately 3.25 cm apart into one of the 17.5×12.5 cm walls of the cage. Two stainless steel sipper tubes that were monitored with contact lickometers were placed 2 mm outside the vertical slits (one sipper tube each). A grounded 8×12–cm stainless steel plate was positioned inside the cage and adjoined to the wall containing the sipper tubes. Contact between the sipper tubes and the stainless steel floor was detected by separate touch-sensitive circuits.

Tastants

Three tastants were used in this study. The first, Polycose (Abbott Laboratories, Columbus, OH), is an easily digestible glucose polymer that mice generally find preferable to water (Glendinning et al. 2002). A 0.16-M aqueous solution served as a training stimulus. The experimental testing stimuli consisted of a 0.003-M PROP solution (Aldrich Chemical Company, Milwaukee, WI) and a 5% (v/v) ethanol solution (Pharmco, Brookfield, CT). These concentrations were chosen because they represented moderate levels of the stimuli that have been previously shown to indicate significant strain differences in the respective mice (Belknap et al. 1993; Nelson et al. 2003).

Brief-access training procedure

Prior to testing, mice were trained over a 3-day period to access fluid and sample from both sipper tubes in the testing apparatus (Glendinning, Chyou, et al. 2005). In order to access fluid, the mice were trained to stand on the stainless steel plate and protrude their tongue through one of the 2 slits, in order to make contact with the tip of a sipper tube. In doing so, completion of the touch circuit resulted in the delivery of approximately 0.005 cc of fluid (either tastant or water). During training sessions, mice were given a choice between 2 sipper tubes, one containing tap water whereas the other contained a Polycose solution (Glendinning, Chyou, et al. 2005). The position of the sipper tube containing the tastant was counterbalanced across both training trials and sessions.

Each training session consisted of a 5-min habituation period followed by 2 trials that were separated by a 5-min interval. Beginning with the first lick to either sipper tube (i.e., trial initiation), the number of licks to each tube was recorded separately for each of the 2 trials per session. The duration of each pair of trials during the training sessions progressively decreased on days 1, 2, and 3 to 5, 2, and 1 min in length, respectively. The timing for the final day of training was identical to the parameters required for testing (5-min habituation period, two 1-min trials, and a 5-min intertrial interval).

In order to provide sufficient motivation for the behavioral task, the mice were water deprived for 22.5 h prior to each training session. Because the mice received an insignificant amount of their daily fluid needs during the training sessions, each animal was provided with 1-h ad libitum food and water access immediately following training sessions 1 and 2. Following training session 3, a 24-h period of ad libitum food and water access was provided.

Brief-access testing procedure

Each mouse participated in 2 testing sessions, one for each of the 2 tastants, PROP and ethanol. Following the posttraining 24-h ad libitum access period, the mice were placed on a restricted access protocol and then tested. That is, a 23.5-h food and water restriction period preceded the first testing session (either PROP or ethanol) with each mouse having access to a total of 2 ml of tap water and 1 g of food pellets. After the first testing session, animals were again given a 24-h period of ad libitum access to food and water which, in turn, was again followed by a second period of limited access prior to the second testing session. This approach of interposing a limited access period with ad libitum access was utilized in order to minimize the potential contribution of caloric demand on subsequent preference testing without disturbing the previously learned sampling behavior.

Each testing session incorporated a 5-min habituation period that was followed by two 1-min brief-access trials with a 5-min intertrial interval. Trial initiation and data collection were identical to the training period. Consequently, mice had two 1-min trials with access to sipper tubes containing water and tastant that was balanced for tastant location. Only one tastant was evaluated on any particular testing day. The order of tastant testing (i.e., PROP and ethanol) and sipper tube location (tastant vs. tap water) were randomized and balanced across testing sessions, trials, within and across mouse strain, within a cohort, and across cohorts.

Preference measure

Each animal's "measure of preference" (MP) both for ethanol and for PROP, compared with water, was calculated by first subtracting the number of licks to water (across the two 1-min trials) from the number of licks to the specific tastant evaluated (across trials; i.e., tastant − water) and then dividing that value by the total number of licks emitted during the testing session (tastant + water). As a result, MP is a measure that ranges from −1 to +1, with the values standardized to indicate a preference for the tastant when positive, of a preference for water when negative, and of no preference when the value is 0.

Results

As outlined above, a total of 88 mice from 4 different strains participated in the study. Further, each of these strains was selected on the basis of literature reports of their particular tastant consumption preference, with the generally accepted idea that mice from inbred strains show minimal within-strain variation (Taft et al. 2006). Consequently, SWR/J (PROP avoiding) and BALB/cJ (ethanol avoiding) were predicted to prefer water (i.e., a negative MP value) for

the reason that they should avoid their respective tastant. By contrast, C57BL/6J (ethanol preferring) and C3HeB/FeJ (PROP neutral) strains were predicted to either prefer the tastant to that of water (i.e., a positive MP value for the C57BL/6J) or, at the very least, have a neutral preference (i.e., MP ≈0 for the C3HeB/FeJ). As illustrated in Figure 1A, the MP values for the 2 strains of mice obtained for their PROP perception (namely, C3HeB/FeJ and SWR/J) could be segregated into distinctly different populations [$t(42)$ = 2.79, P = 0.008]. By contrast, this was not the case for the mice obtained for their ethanol preferences (i.e., C57BL/6J and BALB/cJ; Figure 1C) [$t(42)$ = 1.43, P = 0.16], despite some tendency toward segregation around neutral MP values. In considering these results, it is noteworthy that the distributions of MP values relative to the tastant for which the strains were not selected (based on the literature) were unambiguously overlapping (Figure 1B,D). More specifically, Figure 1B illustrates the complete overlap of the MP distribution values to PROP for the strains selected for their

ethanol preferences, whereas Figure 1D shows a similar picture of the MP distribution values to ethanol for strains selected for their PROP preferences.

With further regard to the individual mouse strains specifically chosen for a particular tastant, Figure 1A,C also highlights the high variability among these animals; so much so there was a substantial amount of overlap of individual MP values even between the significantly different mouse strains selected for their PROP preference (Figure 1A). To be sure, the data suggest that although the 2 strains of mice (as a group) selected for their PROP preference could be discriminated, observing an individual animal's tastant preference would not necessarily be prognostic of its strain.

Concordance with literature-reported preference

Although the upper panels in Figure 1 (A and C) illustrate the differences in preferences between the respective strains chosen for a particular tastant, they do not provide a formal
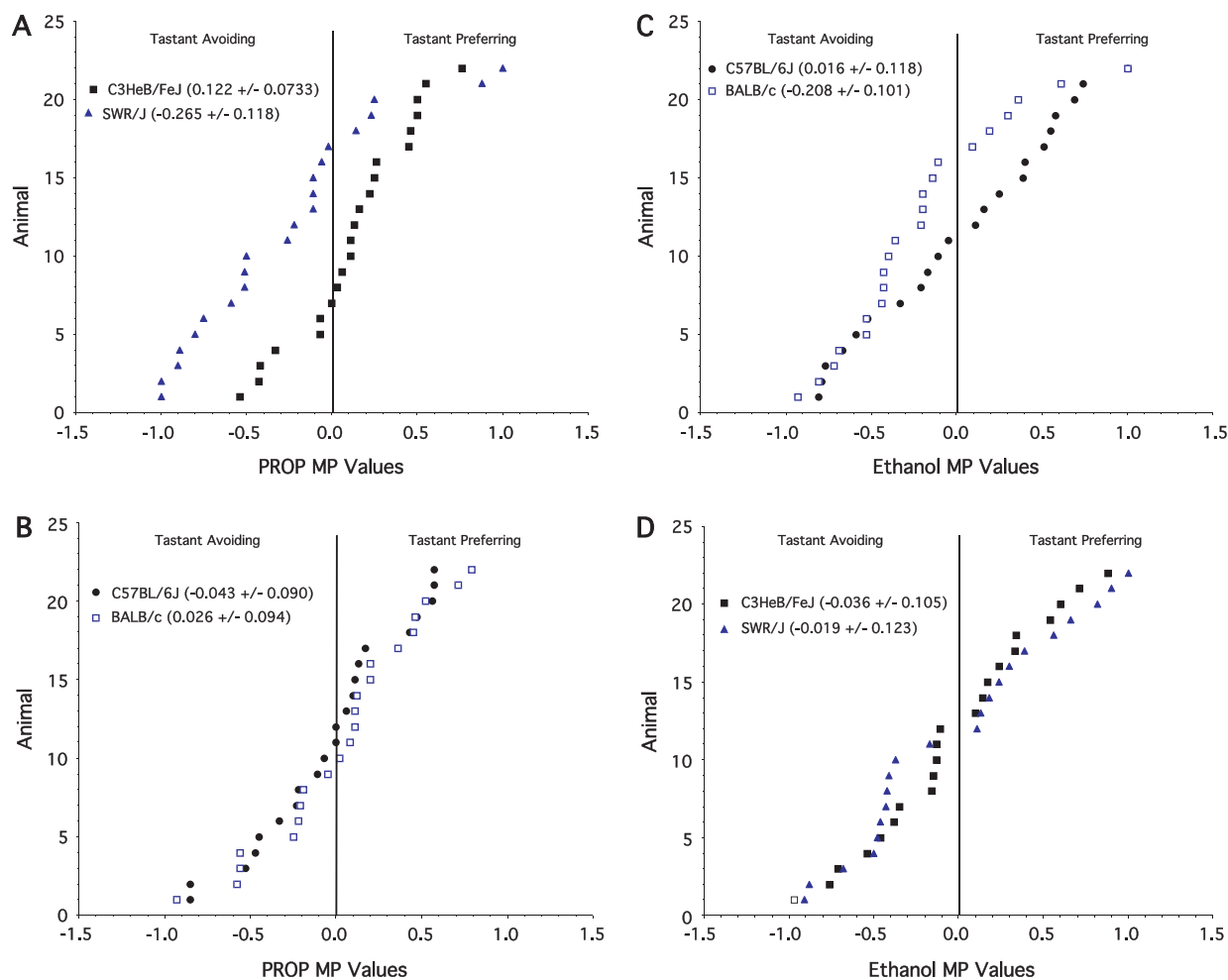

Figure 1 A and C: Distributions of PROP **(A)** and ethanol **(C)** MP values for the tastant-selected mouse strains. B and D: Distributions of PROP **(B)** and ethanol **(D)** MP values for the tastant for which the mouse strain was not selected on the basis of previous testing literature. The vertical lines represent the point of neutral preference. The data in the parentheses of each strain legend represent the mean MP value and standard error of the mean for each strain, respectively.

evaluation of the extent to which each strain's mean MP value was in agreement with the prior literature. That is, regardless of whether 2 strains selected for opposing preferences for a tastant differed or not, it is possible that either one or both strains were not behaving according to prior literature reports. To evaluate this possibility, the MP values for the tastant for which each strain was selected were submitted to a t-test against a value of 0. In this respect, a significant t-test would indicate a nonneutral inclination for the tastant, and the direction of the t-value would indicate whether the strain avoided or preferred the tastant, thereby allowing an evaluation of literature concordance. The strains chosen to avoid their respective tastants behaved in a way that was demonstrably in keeping with the literature: SWR/J: $t(21) = -2.25$, nominal $P = 0.03$ (mean ± standard deviation [SD]: $0.265 ± 0.554$); BALB/cJ: $t(21) = -2.06$, nominal $P = 0.05$ (mean ± SD: $0.208 ± 0.477$). Note that the t-values were not only significantly different from 0 but also negative, which is the predicted direction for tastant-avoiding animals. In contrast to the tastant-avoiding strains, the mean MP values for the tastant-preferring (or at least neutral) animals were not different from 0: C3HeB/FeJ: $t(21) = 1.67$, $P = 0.11$ (mean ± SD: $0.122 ± 0.344$); C57BL/6J: $t(21) = 0.13$, $P = 0.90$ (mean ± SD: $0.016 ± 0.555$). In this regard, the C3HeB/FeJ animals, indeed, behaved in agreement with their literature-reported preference. That is, because these animals do not detect PROP, they should neither prefer nor avoid the tastant (Nelson et al. 2003). By contrast, the C57BL/6J mice showed no evidence of a preference behavior to ethanol that was consistent with reports in the literature.

Relationship between PROP and ethanol preferences

Using the brief-access testing protocol, the foregoing results indicated that, although highly variable in their preference scores, 3 of the 4 mouse strains produced tastant-specific MP values that were, on average, in keeping with the prior literature reports. Because each animal provided a preference measure for both PROP and ethanol, we used these data to examine whether there was evidence of an overall predictive relationship between the degree of preference for PROP and that for ethanol. To accomplish this, we calculated a Pearson's correlation coefficient using the individual animal MP values for both ethanol and PROP. The results of this analysis provided no evidence of a relationship between the preference values for the 2 tastants in the anticipated direction ($r = 0.011$, with a standard error of 0.11).

In examining a potential relationship between PROP and ethanol preference, using all 4 mouse strains and all preference values, it is possible that a potential relationship within an individual strain might have been obscured. To evaluate this, we performed a chi-square test of homogeneity using the z transforms of the individual strain data. This analysis provided no evidence of heterogeneity across strains [$\chi^2(3) = 0.94$, $P > 0.5$; Steel and Torrie 1980].

Discussion

In the present study, we used a brief-access 2-bottle taste preference paradigm (i.e., two 1-min trials; Glendinning, Chyou, et al. 2005) to measure the unconditioned lick response of 4 inbred mouse strains (BALB/cJ, C57BL/6J, SWR/J, and C3HeB/FeJ) to both ethanol versus water and PROP versus water. In doing so, this approach permitted us the ability to assess taste quality preferences (relative to water), while minimizing the potential influence of postingestive feedback or the effect of experience. Because each strain was initially chosen for a previously literature-evaluated tastant (either ethanol or PROP) and preference (either preferring, avoiding or neutral), as determined by long-term 2-bottle preference testing, the first goal of this study was to assess the prior literature results with the current brief-access paradigm. As highlighted qualitatively in Figure 1 and confirmed quantitatively, comparison of the strains indicated by the literature to avoid PROP (SWR/J) versus those indicated to be neutral (C3HeB/FeJ) demonstrated significant differences between their mean preferences values (MP). However, a difference was not observed between the strains noted to avoid ethanol (BALB/cJ) versus those noted to prefer the drug (C57BL/6J). Consequently, the brief-access paradigm only confirmed that the SWR/J and C3HeB/FeJ strains, on average, have differential preferences with respect to their literature-noted tastant. Nonetheless, the MP distributions for these 2 strains were in keeping with the more, general observation of wide variability within each of the 4 strains evaluated (Figure 1).

The above not withstanding, we also examined whether the observed tastant-specific preferences for which each strain was selected was consistent with prior literature reports. In this regard, only the BALB/cJ, SWR/J, and C3HeB/FeJ strains, on average, conformed to their literature-predicted results. In other words, BALB/cJ avoided ethanol, SWR/J avoided PROP, and C3HeB/FeJ preferred PROP equally as well as water. However, even for these strains there were a substantial number of animals with discordant preferences (see Figure 1). By contrast, the C57BL/6J strain did not conform to the prediction that it is an ethanol-preferring strain. Certainly, roughly 50% of the animals in Figure 1C reside on either side of the neutral preference point (i.e., 0). Thus, the absence of an expected difference between the C57BL/6J and BALB/cJ strains resulted from a deviation by the C57BL/6J animals from an expected ethanol preference. Taken together, therefore, our results lend further support to the proposition that the C57BL/6J strain is either not as strongly ethanol preferring (Little et al. 1999) as previously indicated (York 1981; Belknap et al. 1993; Meliska et al. 1995; McMillen and Williams 1998; Middaugh et al. 1999) or rather that orosensory factors do not contribute to their preference for ethanol in long-term 2-bottle preference tests.

The general high variability and number of animals in disagreement with their respective literature prediction (across

the 4 strains) raises concerns regarding the utility of these animals for investigations of underlying mechanisms related to taste-mediated ingestive responses and suggests that other postingestive consequences may motivate ethanol intake in C57BL/6J mice (Rhodes et al. 2007).

In view of the above, it is worth considering possible factors that might account for the source of variance we observed within a mouse strain, the rather modest average preference scores, and the observation that the C57BL/6J strain did not conform to the earlier literature result. In this regard, our findings may be a function of the brief-access manner in which the animals were tested, intrinsic genetic change over time, or some interaction of the 2. In other words, we propose that, absent the potential for postingestive consequences, our approach more appropriately reflects the chemosensory (taste/odor) mediated quality preferences of these mouse strains than does the long-term access procedure. As such, the brief-access testing may have uncovered a substantial degree of polymorphisms or a more recent drift in the genetic factors underlying the response to the specific tastants for which each strain was chosen. This latter issue should not be underestimated. As recently noted by Taft et al. (2006), in spite of the best breeding efforts, inbred mouse strains can accumulate mutations over time that lead to increasing variation within a strain and phenotypic modification.

The second aim of the present study was to explore whether and to what degree there was a predictive relationship between preferences for PROP and ethanol. We hypothesized that a strong avoidance to PROP would be associated with low ethanol intake, whereas a neutral or positive preference would predict a relatively higher ethanol intake. Our results did not demonstrate a relationship between the 2 taste preferences. Thus, the more water a mouse chose to drink rather than PROP (presumably because they could detect its bitter taste) seemed to be unrelated to the amount of water they also tended to consume rather than ethanol.

One important factor that may have contributed to the lack of observed relationship is that the sensitivity to PROP is not the sole determining factor related to the bitterness of ethanol. Recent evidence from human twin studies indicated that the perception of bitterness might not be due to a single genetic factor (Hansen et al. 2006). More importantly, the genetic influences on the perception of PROP are mediated differently from that of other types of bitter compounds such as sucrose octa-acetate, caffeine and quinine, which appear to have a substantial amount of phenotypic covariation (Brasser et al. 2005; Hansen et al. 2006). If this, indeed, is the case for both mice as well as humans, then the present findings would indicate that the bitter quality in ethanol may be more highly related to the second of perhaps, at least, 2 sets of genetic influences. Such a proposition would be in keeping with the wide diversity of findings concerning the relationship between PROP and ethanol in studies using human subjects (Pelchat and Danowski 1992; Kranzler

et al. 1996, 1998; DiCarlo and Powers 1998; Duffy et al. 2004), as well as the present results.

There is, of course, a caveat to the foregoing. Although there are similarities between rodent and human perceptions of the taste quality of bitter, there are also differences (Harder et al. 1996; Harder and Whitney 1998; Shi et al. 2003). Moreover, there are species-specific differences in the repertoire of bitter taste receptors (Shi et al. 2003). Nonetheless, the present data contribute to the literature in several ways. 1) The lack of a relationship between PROP and ethanol indicates that the bitter quality in ethanol may be more highly related to other bitter compounds that are mediated by different genetic influences. 2) The high variability per strain and the number of animals in disagreement with their reported long-term 2-bottle taste preference raise a note of caution regarding their utility for studying specific taste-mediated ingestive responses. 3) Our results confirm and extend upon recent findings that the C57BL/6J are not a strongly ethanol-preferring strain (Little et al. 1999; O'Callaghan et al. 2002). 4) Finally, in considering the question of the underlying basis for particular tastant preferences, our results speak to the importance of utilizing procedures that can distinguish between orosensory-mediated behaviors and those driven by postingestive consequences.

Funding

Acknowledgements

References

American Psychiatric Association. 2000. Diagnostic and statistical manual of mental disorders: dsm-iv. 4th ed. Washington (DC): American Psychiatric Association.

Barnicot NA, Harris H, Kalmus H. 1951. Taste thresholds of further eighteen compounds and their correlation with P.T.C. thresholds. Ann Eugen (Lond). 16:119–128.

Bartoshuk LM, Duffy VB, Miller IJ. 1994. PTC/PROP tasting: anatomy, psychophysics, and sex effects. Physiol Behav. 56(6):1165–1171.

Belknap JK, Crabbe JC, Young ER. 1993. Voluntary consumption of ethanol in 15 inbred mouse strains. Psychopharmacol. 112:503–510.

Blednov YA, Walker D, Martinez M, Levine M, Damak S, Margolskee RF. 2007. Perception of sweet taste is important for voluntary alcohol consumption in mice. Genes Brain Behav. doi: 10.1111/j.1601-183x.2007.00309.x. (OnlineEarly Articles).

Blizard DA. 2007. Sweet and bitter taste of ethanol in c57bl/6j and dba2/j mouse strains. Behav Genet 37(1):146–59.

Boughter JD Jr, St John SJ, Noel DT, Ndubuizu O, Smith DV. 2002. A brief access test for bitter taste in mice. Chem Senses. 27:133–142.

Brasser SM, Mozhui K, Smith DV. 2005. Differential covariation in taste responsiveness to bitter stimuli in rats. Chem Senses. 30(9):793–799.

Brook JS, Whiteman M, Nomura C, Gordon AS, Cohen P. 1988. Personality family and ecological influences on adolescent drug use: a developmental analysis. J Chem Depend Treat. 1:123–161.

Cloninger CR. 1987. Neurogenetic adaptive mechanisms in alcoholism. Science. 236:410–416.

Davis JD, Collins BJ, Levine MW. 1975. Peripheral drinking: control of gastro-intestinal filling as a negative feedback signal, a theoretical and experimental analysis. J Comp Physiol Psychol. 89:985–1002.

DiCarlo ST, Powers AS. 1998. Propylthiouracil tasting as a possible genetic association marker for two types of alcoholism. Physiol Behav. 64: 147–152.

Drenowski A, Rock CL. 1995. The influence of genetic taste markers on food acceptance. Am J Clin Nutr. 62:506–511.

Duffy VB, Peterson JM, Bartoshuk LM. 2004. Associations between taste genetics oral sensation and alcohol intake. Physiol Behav. 82:435–445.

Glendinning JI, Bloom LD, Onishi M, Zheng KH, Damak S, Margolskee RF, Spector AC. 2005. Contribution of α–Gusducin to taste-guided licking responses of mice. Chem Senses. 30:299–316.

Glendinning JI, Chyou S, Lin I, Onishi M, Patel P, Zheng KH. 2005. Initial licking responses of mice to sweeteners: effects of tas1r3 polymorphisms. Chem Senses. 30:601–614.

Glendinning JI, Gresdack J, Spector AC. 2002. A high-throughput screening procedure for identifying mice with aberrant taste and oromotor function. Chem Senses. 27:461–474.

Hansen JL, Reed DR, Wright MJ, Martin NG, Breslin PA. 2006. Heritability and genetic covariation of sensitivity to PROP, SOA, quinine HCl, and caffeine. Chem Senses. 31:403–413.

Harder DB, Gannon KS, Whitney, G. 1996. SW.B6-Soa(b) nontaster congenic strains completed and a sucrose octaacetate congenic quartet tested with other bitters. Chem Senses. 21:507–517.

Harder DB, Whitney G. 1998. A common polygenic basis for quinine and PROP avoidance in mice. Chem Senses. 23:327–332.

Kaminski LC, Henderson SA, Drewnowski A. 2000. Young women's food preferenced and taste responsiveness to 6-n-propylthiouracil. Physiol Behav. 68:691–697.

Kranzler HR, Moore PJ, Hesselbrock VM. 1996. No association of PROP taster status and paternal history of alcohol dependence. Alcohol Clin Exp Res. 20:1496–1500.

Kranzler HR, Skipsey K, Modesto-Lowe V. 1998. PROP taster status and parental history of alcohol dependence. Drug Alcohol Depend. 52:109–113.

Lanier SA, Hayes JE, Duffy VB. 2005. Sweet and bitter tastes of alcoholic beverages mediate alcohol intake in of-age undergraduates. Physiol Behav. 83:821–831.

Lemon CH, Brasser SM, Smith DV. 2004. Alcohol activates a sucrose-responsive gustatory neural pathway. J Neurophysiol. 92:536–544.

Little HJ, O'Callaghan MJ, Butterworth AR, Wilson J, Cole J, Watson WP. 1999. Low alcohol preference among the "high alcohol preference" C57BL/6J strain of mice; preference increased by saline injections. Psychopharmacol. 147:182–189.

Mattes RD, DiMeglio D. 2001. Ethanol perception and ingestion. Physiol Behav. 72:217–229.

McMillen BA, Williams HL. 1998. Role of taste and calories in the selection of ethanol by C57BL/6NHsd and Hsd:ICR mice. Alcohol. 15:193–198.

Meliska CJ, Bartke A, McGlacken G, Jensen RA. 1995. Ethanol, nicotine, amphetamine, and aspartame consumption and preferences in C57BL/6 and DBA/2 mice. Pharmacol Biochem Behav. 50:619–626.

Middaugh LD, Kelley BM, Bandy AE, McGroarty KK. 1999. Ethanol consumption by C57BL/6J/BL6 mice: influence of gender and procedural variables. Alcohol. 17:175–183.

Nelson TM, Munger SD, Boughter JD Jr. 2003. Taste sensitivities to PROP and PTC vary independently in mice. Chem Senses. 28:695–704.

O'Callaghan MJ, Croft AP, Watson WP, Brooks SP, Little HJ. 2002. Low alcohol preference among the "high alcohol preference" C57BL/6J/BL10 mice; factors affecting such preference. Pharmacol Biochem Behav. 72:475–481.

Pelchat ML, Danowski S. 1992. A possible genetic association between PROP-tasting and alcoholism. Physiol Behav. 51:1261–1266.

Pitkänen T, Lyyra A, Pulkkinen L. 2005. Age of onset of drinking and the use of alcohol in adulthood: a follow-up study from age 8-42 for females and males. Addiction. 100:652–661.

Rhodes JS, Ford MM, Yu CH, Brown LL, Finn DA, Garland T Jr, Crabbe JC. 2007. Mouse inbred strain differences in ethanol drinking to intoxication. Genes Brain Behav. 6(1):1–18.

Schuckit MA, Smith TL. 2006. An evaluation of the level of response to alcohol externalizing symptoms and depressive symptoms as predictors of alcoholism. J Stud Alcohol. 67:215–27.

Scinska A, Koros E, Habrat B, Kukwa A, Kostowski W, Bienkowski P. 2000. Bitter and sweet components of ethanol taste in humans. Drug Alcohol Depend. 60(2):199–206.

Shi P, Zhang J, Yang H, Zhang Y. 2003. Adaptive diversification of bitter taste receptor genes in mammalian evolution. Mol Biol Evol. 20:805–814.

Smith BK, Volaufova J, West DB. 2001. Increased flavor preference and lick activity for sucrose and corn oil in SWR/J vs. AKR/J mice. Am J Physiol Regul Integr Comp Physiol. 281:R596–R606.

Smith GT. 1994. Psychological expectancy as mediator of vulnerability to alcoholism. Ann NY Acad Sci. 708:165–171.

Spector AC. 2003. Psychophysical evaluation of taste function in non-human mammals. In: Doty RL, editor. Handbook of olfaction and gustation. 2nd ed. New York: Marcel Dekker. p. 861–879.

Steel RGD, Torrie JH. 1980. Principles and procedures of statistics: a biomedical approach. 2nd ed. New York: McGraw Hill Book Company. p. 280–281.

Taft RA, Davisson M, Wiles MV. 2006. Know thy mouse. Trends Genet. 22:649–653.

Waseem M, Seshadri KG, Kabadi UM. 1998. Successful outcome with methimazole and lithium combination therapy for propylthiouracil-induced hepatoxicity. Endocr Pract. 4:197–200.

Wilson CWM, O'Brien CO, MacAirt JG. 1973. The effect of metronidazole on the human taste threshold to alcohol. Addiction. 68:99–110.

York JL. 1981. Consumption of intoxicating beverages by rats and mice exhibiting high and low preferences for ethanol. Pharmacol Biochem Behav. 15:207–214.