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ORIGINAL INVESTIGATION

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Differential development of behavioral tolerance and the subsequent hedonic effects of alcohol in AA and ANA rats

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Abstract *Rationale:* There at least two ways in which tolerance development to alcohol's behavioral effects could interact with its subsequent intake: 1) tolerance to alcohol's reward or reinforcing effects per se could lead to increased consumption, and 2) tolerance to alcohol's aversive effects could unmask alcohol's rewarding effects. These two mechanisms may differentially interact with preexisting genetic traits underlying alcoholism. *Objectives:* Alcohol's subjective attributes were assessed in selectively bred AA and ANA rats after the development of tolerance to alcohol's behaviorally disruptive effects on lever-press performance. *Methods:* Rats were trained to press a lever under an FR30 schedule of food presentations. Group-dependent differential access to intoxicated practice, using a typical pre-post drug administration design, was utilized to promote the development of alcohol tolerance in only the group receiving intoxicated practice sessions. Subsequently, rats were trained to associate alcohol with unique place and taste stimuli in order to assess the relative changes in the approach towards, or avoidance of alcohol-related cues in each group. *Results:* Groups of AA and ANA rats given access to intoxicated practice demonstrated tolerance development. These groups subsequently conditioned place preferences and failed to develop conditioned taste aversions to alcohol. Passive alcohol exposure in the ANA rats set the occasion for the development of a place preference and delayed taste conditioning. AA rats exposed to passive alcohol exposure failed to condition place preferences and developed rapid taste aversions. Saline control rats failed to develop tolerance or place preferences but did condition a robust alcohol-induced taste aversion. *Conclusions:* AA and ANA rats differ in their behavioral and pharmacokinetic response to chronic alcohol exposure. Compensatory responses interacting with approach-avoidance behaviors appear to be learned during intoxicated practice in the AA rats and during both intoxicated practice and passive exposure in the ANA rat line.

Key words AA and ANA rats · Associative learning · Conditioned place preference · Conditioned taste · Aversion · Behavioral tolerance · Alcohol

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Introduction

Three pairs of rat lines have been genetically selected for high versus low ethanol (EtOH) preference in two-bottle choice procedures: the ALKO AA (alcohol-accepting) and ANA (alcohol non-accepting) lines (Ericksson 1969; Eriksson and Narhi 1973); the P (preferring) and NP (non-preferring) lines (Li et al. 1981); and the HAD (high alcohol drinking) and LAD (low alcohol drinking) lines (Li et al. 1988). In addition to the differences in voluntary EtOH consumption, these rat lines present behavioral differences as well as differences in the activity of a number of neurotransmitter systems, both in the presence and absence of EtOH (George et al. 1990; Sinclair and Li 1990; Krimmer and Schechter 1991; Gianoulakis et al. 1992; Krimmer 1992; Schechter 1992; Dyr et al. 1993; Gordon et al. 1993; Hyytia 1993; Paivarinta and Korpi 1993; Nurmi et al. 1994; Ritz et al. 1994).

In a set of studies, Lê and Kiianmaa (1988) suggested that the AA rats develop a greater degree of tolerance to the depressive effects of alcohol than the ANA rats. Lê and Kiianmaa (1988) also demonstrated differential rates of acute alcohol tolerance development in AA and ANA rats for both hypothermia and sleep-induction. Low to moder-

ate doses of alcohol usually stimulate locomotor activity in AA rats but not the ANA rats (Hilakivi et al. 1984). The differential behavioral or autonomic response to environmental stressors, which has been characterized as a measure of "emotionality" or affective responsiveness, has been reported in the AA and ANA selectively bred lines as well (Sinclair et al. 1987, 1989; Sinclair and Li 1990; Korpi et al. 1988). These data suggest that the line of rats selectively bred for decreased alcohol consumption, the ANA line, are more reactive to aversive stimuli leading to an aversion to the strong flavor of alcohol or the interoceptive cues associated with alcohol consumption.

The current series of experiments were designed to assess the changes in the interoceptive properties of alcohol induced by the development of behavioral tolerance. The behavioral tasks most often used to assess drug-induced motivational parameters associated with their hedonic valence are based on the degree of approach-avoidance behaviors expressed by an animal. The repeated pairing of specific exteroceptive (place conditioning) or interoceptive (taste conditioning) stimuli may lead to a conditioned preference (approach towards) or aversion (avoidance of) the drug-paired stimuli (Young 1959; Goudie 1979; White et al. 1987). We have previously reported that the development of tolerance to alcohol's rate-decreasing effects was associated with a degree of tolerance development to alcohol's aversive effects of alcohol, as measured by subsequent taste and place conditioning in Sprague-Dawley rats (Gauvin and Holloway 1992a, 1992b). The current studies were designed to extend these findings to the genetically bred AA and ANA rat lines.

Materials and methods

This experiment was conducted in three phases. During phase I rats were trained in a single lever food-motivated lever-press response. Groups of rats were differentially exposed to either alcohol or saline in a typical pre-post design (Chen 1968) during a period of chronic drug administration to examine the development of behavioral tolerance to the rate-disruptive effects of alcohol on lever-press responding. Phases II and III were conducted to examine the functional role that differential alcohol exposure had on the development of shifts in alcohol's aversive and/or rewarding effects as approach-avoidance measurements using taste and place conditioning. To reduce order-effects in phases II and III, half the rats of each group received taste conditioning first and place conditioning second. The other half of the rats received the training in reverse order. The research protocols had the prior approval of the University of Oklahoma Health Sciences Center's animal care and use committee. The principles of laboratory animal care (NIH publication #85-23, revised 1985) were strictly adhered to during the course of these studies.

Animals

Twenty-four AA (Alko-Alcohol) and 24 ANA (Alko Non-Alcohol) male rats were generously donated by K. Kiianmaa [Research Laboratories, State Alcohol Company (Alko Ltd), Helsinki, Finland]. The animals and colony rooms were maintained by technicians and veterinarians from the AAALAC-accredited Department of Animal Resources at the University of Oklahoma Health Sciences Center. Animals were initially given free access to both food and water and allowed 1 month to accommodate to the new environment. Subsequently, rats were food-deprived and maintained at 85% of their free-feeding weights by supplemental feeding to the food acquired during daily experimental sessions during phase I (behavioral tolerance development) of the study. During phases II and III (taste and place conditioning) of the study rats were placed back on the free access to food (see below). Target weights were allowed to increase by 10–15 g per month during phase I of the study to allow for normal growth. The body weights were unregulated during phases II and III.

Apparatus

Phase I

Twelve rodent chambers were each equipped with a single lever, stimulus lamp, and pellet dispenser; each chamber was housed within a sound attenuating enclosure cabinet (Lafayette Instruments, Lafayette, Ind., USA). All behavioral/experimental contingencies and data collection in phase I were accomplished by PROMAL-based Commodore 64C microcomputer systems (American Neuroscience Research Foundation, Yukon, Oklahoma, USA) interfaced with the six experimental chambers (Rayfield Instruments, Waitsfield, N.H., USA).

Phases II and III

The place conditioning apparatus consisted of two main conditioning compartments connected to each other by a third compartment in a straight alleyway configuration. Detailed descriptions of these chambers have been previously described (Gauvin et al. 1994a). The number of compartment entries, time in each compartment, and general activity of the subjects were assessed and recorded by sets of infrared photobeams located near the floors of each compartment and linked by a photobeam controller (DIG-723, Med. Associates, Inc., East Fairfield, Vt., USA) to a Commodore 64C microcomputer system. The microcomputer system controlled the experimental contingencies and recorded all measures from four sets of conditioning chambers simultaneously (American Neuroscience Research Foundation).

Behavioral tasks

After the 1-month acclimation period and the reduction to the 85% free-feeding weights, training procedures were implemented. Rats were shaped by successive approximation to press the lever for food pellets (45 mg; Bioserv Inc., Frenchtown, N.J., USA) during a three-cycle training session. The multi-cycle sessions were identical to those previously used by this laboratory (Gauvin et al. 1994b). All sessions began with an IP injection of saline and a 15-min timeout period (no food reinforcement and stimulus and house lights off); each timeout period was followed by a 10-min time-in period (schedule-controlled food reinforcement with stimulus and house lights on). Over successive training sessions, the number of lever-press responses required to produce a single food pellet was raised from an initial FR1 to FR30. Training continued with saline injections administered at the beginning of each timeout period until rates of responding across all three cycles for 5 consecutive days varied by less than ±10%. Once stable lever-press performance was achieved, rats were run in five cycle sessions three times per week until rates of responding were stable across all five cycles preceded by saline injections. These latter sessions were instituted to accommodate the rats to the five-cycle dose-response test sessions.

EtOH dose-effect curve procedures

Once stable performance was achieved across the three- and five-cycle training sessions, cumulative dose-effect tests were conduct-

ed using a five-cycle cumulative dosing procedure. Baseline stability performance was determined by IP injections of saline alone in a volume equivalent to that of the corresponding EtOH dose that would be administered in subsequent EtOH cumulative dosing test sessions. The day after this five-cycle saline test session was completed, an initial dose-effect curve was determined in each subject using a cumulative injection regimen at the beginning of each session and each timeout period. Saline and four sequential EtOH doses (10% w/v solution in 0.9% saline) were administered by IP injections. The EtOH doses were 0.25, 0.5, 0.5, and 0.75 g/kg, which provided cumulative doses of 0.25, 0.75, 1.25, and 2.0 g/kg.

Once these initial dose-effect functions were obtained, rats from each selectively bred line were subdivided into three treatment groups that were equivalent in their: (1) saline baseline rates-of-responding, (2) ED_{50} for EtOH-induced response rate suppression, and (3) body weights. The three subgroups in each rat line subsequently received differential EtOH exposure during a 30-day chronic EtOH period. A second dose-effect function was generated at the end of the chronic dosing regimen, the day following a five-cycle saline test session which was used as a baseline of drug-free rates-of-responding.

Place conditioning task

After dose-effect curves generated in the conditioning task had demonstrated alcohol tolerance, a 1-week washout period was imposed prior to subsequent conditioning. On a 3-day cycle, each rat was intubated with either 2 g/kg alcohol (20% w/v alcohol in tap water) or tap water (gavaged using an 18-g feeding tube; Harvard Bioscience) and immediately placed into the conditioning apparatus for 30 min. Each alcohol-environment pairing session was followed by a day off to insure that any residual effects of the alcohol administrations did not carry over into the water-environment pairing sessions. The order of testing was randomly assigned for each subgroup. After two sets of conditioning trials (6 days), each rat was retested for side preference, as described above. Each rat received a total of eight stimulus/environment pairing sessions (four alcohol, four water) and two conditioning test sessions (T1 and T2).

Taste conditioning task

After a 1-week washout period, in which all rats were allowed free access to food and water, rats were place on a short-term water deprivation schedule. Rats were allowed access to tap water in the home cage for 15 min (1030–1045 hours) in the morning and for 60 min (1500–1600 hours) in the afternoon in calibrated drinking tubes. This procedure was continued for 3 days, until the animals were drinking approximately 10 ml water during the morning access session. On day 4, rats were presented with a 0.1% w/v solution of sodium saccharin in place of water in the morning drinking session. Immediately after 15-min access to the saccharin solution, each rat was injected with 1.5 g/kg alcohol (10% w/v alcohol in sterile saline) and the total volume of saccharin consumed was recorded. They were allowed access to tap water for 60 min during the afternoon period. On day 7, the amount of the saccharin solution was recorded and the experiment terminated.

Tail blood alcohol concentrations

Blood alcohol concentrations (BACs) were quantified using a gas chromatographic headspace sampling technique (for details see Gauvin et al. 1994b).

Drugs

Ethyl alcohol USP (190 proof) was purchased from US Industrial Chemicals Company (Houston, Tex., USA) and diluted in normal sterile saline to 10% w/v for all injections, and diluted in normal tap water to 20% w/v for the intubations used in the place conditioning assay.

Data analysis

The individual ED_{50} scores for response rate suppression in the FR30 behavioral task was calculated by linear regression analysis (least squares procedure) of the response rate data from individual dose-effect curves. The rates-of-responding were expressed as a percentage of control rates-of-responding during each of the five cycles of saline injections during test sessions conducted the day immediately preceding the alcohol dose-effect function tests. The group comparisons for the behavioral dose-effect curves were analyzed using an independent group [strain: AA versus ANA (2)×treatment: Pre, Post, Sal (3)], repeated measures [dose (4)×time (2)] mixed factor analysis of variance (ANOVA) with a posteriori comparisons for individual dose and group comparisons using Duncan's multiple range tests. For the taste and place conditioning tasks, group comparisons for changes in the total volume of saccharin consumed during the 15-min morning access period were analyzed using an independent group repeated measures mixed-factor ANOVA. In the place conditioning task, each rat received a "preference score" which was calculated from test session data. The total time of the 30-min undrugged, free-access test sessions spent in the non-preferred compartment (bias strategy) were compared after habituation and the two conditioning test sessions using an independent group repeated measures mixed-factor ANOVA. Blood alcohol levels were expressed in mg/dl and compared across groups by similar repeated measures ANOVAs, as well. All data were analyzed using a personal computer based statistical analysis package [Complete Statistical Systems (CSS); Statistica, Tulsa, Oklahoma, USA].

Results

Table 1 shows the group mean blood alcohol curves (BACs) for the initial dose effect functions generated in the AA and ANA rats. There were significant differences in the blood alcohol levels between the AA and ANA rats for all four tested doses of alcohol [Main group, $F(1,44)=16.83$, $P<0.001$; Main time, $F(3,132)=694.3$, $P<0.001$; all post-hoc $P<0.05$]. The AA rats had significantly lower blood alcohol concentrations 15 min after each administered dose. As can be seen in the behavioral dose-effect curves (Fig. 1, Fig. 2), the lower BACs in the AA rat lines were related to higher rates-of-responding during the initial dose-effect functions generated in the behavioral task for each subgroup [open symbols; Main group, $F(1,46)=4.53$,

Table 1 Differential blood alcohol levels in 24 ALKO-AA and 24 ALKO-ANA rats. Cumulative dosing procedure was used to assess BACs corresponding to the "time-in" periods of the operant sessions associated with the behavioral dose-effect functions

Cumulative EtOH dose (g/kg)	Blood alcohol levels±1 SEM (mg/dl)	
	AA rats	ANA rats
0.25	22.76±1.85	30.69±1.76
0.75	40.98±1.93	53.13±1.66
1.25	72.67±3.31	85.62±2.53
2.00	124.31±4.25	145.28±5.4

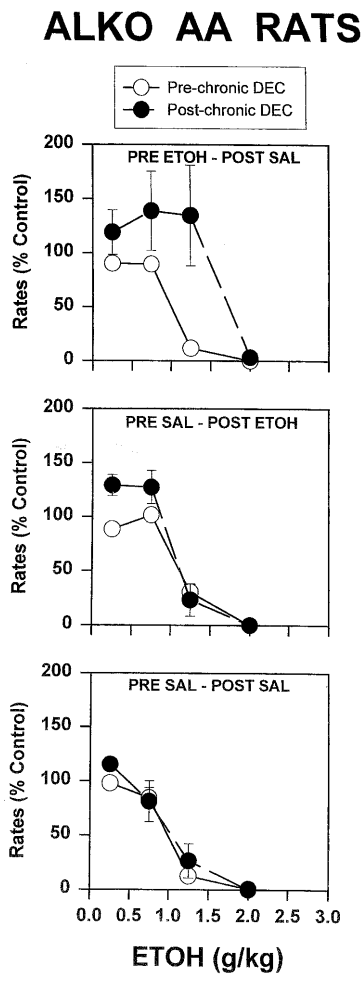

Fig. 1 The effects of cumulative doses of alcohol on the rates-of-responding, expressed as a percentage of saline baseline control rates, in three groups of ALKO-AA rats before (*open circles*) and after (*closed circles*) a 30-day period of chronic alcohol administrations. The rates-of-responding were expressed as a percentage of control rates-of-responding during each of the five cycles of saline injections during test sessions conducted the day immediately preceding the alcohol dose-effect function tests. Each point represents the mean (±SEM) of eight rats. The *top panel* represents the data from rats given alcohol intoxicated practice during the chronic exposure period (pre-session injections). The *middle panel* represents the data from alcohol control rats, which received equivalent post-session alcohol injections, without intoxicated practice. The *lower panel* represents the data from saline control rats

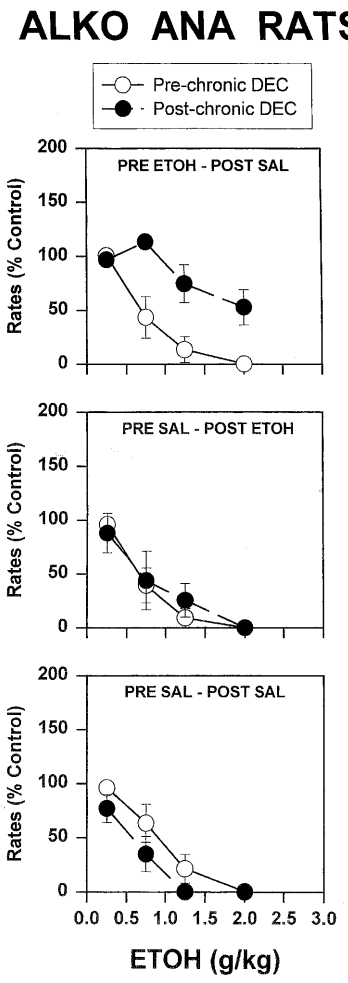

Fig. 2 The effects of cumulative doses of alcohol on the rates-of-responding, expressed as a percentage of saline baseline control rates, in three groups of ALKO-ANA rats before (*open circles*) and after (*closed circles*) a 30-day period of chronic alcohol administrations. Details as in text and Fig. 1

$P<0.05$; Main cycles, $F(3,138)=130.41$, $P<0.001$; Main group×Cycle interactive effects, $F(3,138)=8.08$, $P<0.001$]. When these initial dose-effect curves and BACs were completed, the AA and ANA rats were subdivided into groups in a typical pre-post drug administration design. Within each strain, the subgroups of rats did not differ from each other on either the ED_{50} for ethanol's rate-suppressing effects on performance or baseline body weights (all group *t*-tests non-significant). The initial behavioral ED_{50}s for the AA rat line (expressed in g/kg) were: Pre-EtOH, Post-Sal: 1.03±0.09; Pre-Sal, Post-EtOH: 1.1±0.12; Pre-Sal, Post-Sal: 1.02±0.11. For the ANA line, the initial behavioral ED_{50}s were: Pre-EtOH, Post-Sal: 0.78±0.15; Pre-Sal, Post-EtOH: 0.74±0.13; Pre-Sal, Post-Sal: 0.88±0.14.

As seen in Fig. 1 and Fig. 2, the 1 month chronic dosing regimen selectively produced behavioral tolerance, defined as a shift-to-the-right in the dose-effect curves, in the AA and ANA rats exposed to intoxicated practice, only [Groups Pre-EtOH, Post-Sal; filled circles, top panels; Fig. 1, AA rats: Main treatment effects, $F(2,21)=4.61$, $P<0.01$; Main dose effects, $F(3,63)=48.7$, $P<0.001$; Main DEC effects, $F(1,21)=27.6$, $P<0.001$; Treatment×Dose interactive effects, $F(6,63)=2.97$, $P<0.05$; Dose×DEC, $F(3,63)=3.9$, $P<0.01$; Treatent×Dose×DEC interactive effects, $F(6,63)=2.74$, $P<0.05$; Fig. 2, ANA rats: Main treatment, $F(2,21)=8.66$, $P<0.001$; Main dose, $F(3,63)=38.28$, $P<0.001$; and Main DEC effects, $F(1,21)=27.59$, $P<0.001$; Treatment×Dose interactive effects, $F(6,63)=3.42$, $P<0.01$; Dose×DEC interactive effects, $F(3,63)=3.17$, $P<0.05$]. Those rats in both selected lines which received equal alcohol exposure but no intoxicated practice failed to show any significant right-

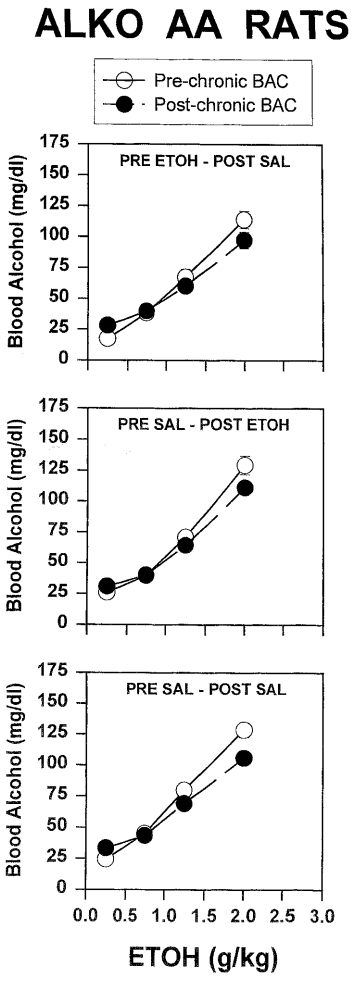

Fig. 3 The effects of cumulative doses of alcohol on the blood alcohol concentrations (*BACs*), expressed in mg alcohol per dl blood, in three groups of ALKO-AA rats before (*open circles*) and after (*closed circles*) a 30-day period of chronic alcohol administration. Each point represents the mean (±SEM) of eight rats. The *top panel* represents the data from rats given alcohol intoxicated practice during the chronic exposure period (pre-session injections). The *middle panel* represents the data from alcohol control rats, which received equivalent post-session alcohol injections, without intoxicated practice. The *lower panel* represents the data from saline control rats. The BACs correspond to the "time-in" or active period of operant responding associated with the behavioral dose-effect curves (see Fig. 1)

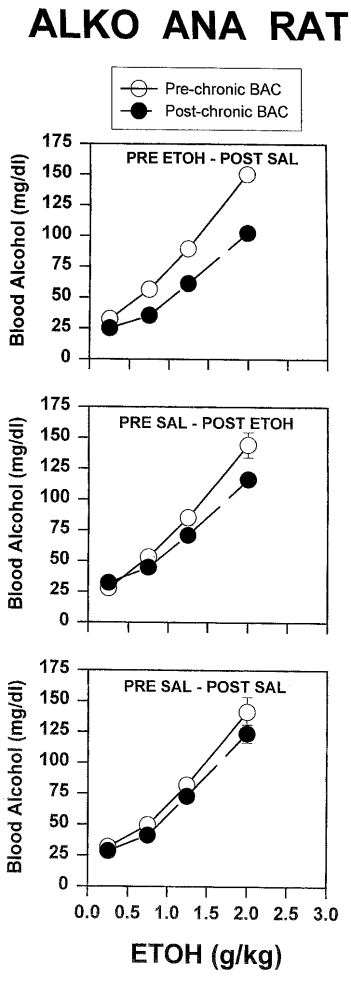

Fig. 4 The effects of cumulative doses of alcohol on the blood alcohol concentrations (*BACs*), expressed in mg alcohol per dl blood, in three groups of ALKO-ANA rats before (*open circles*) and after (*closed circles*) a 30-day period of chronic alcohol administrations. Each point represents the mean (±SEM) of eight rats. For details see Fig. 4. The BACs correspond to the "time-in" or active period of operant responding associated with the behavioral dose-effect curves (see Fig. 2)

ward shifts in the behavioral dose-effect curves (middle panels, filled circles). Saline control groups of both AA and ANA rats failed to show any significant shifts in behavioral dose-effect curves (bottom panels).

Figure 3 and Fig. 4 show the pre- and post-chronic BACs associated with each cycle of the behavioral dose-effect curves. Across all subgroups, the AA rat line did not show any change in alcohol levels during the time-in periods associated with the behavioral task. On the other hand, the ANA rat lines showed a significant reduction in BACs in the subgroups of rats receiving chronic alcohol exposure. Significantly lower blood levels were found at the 0.75, 1.25 and 2.0 g/kg doses in the subgroup of ANA rats receiving intoxicated practice (groups Pre-EtOH, Post-SAL; all Duncan's test $P<0.05$). The subgroup of ANA rats which received identical alcohol exposure to the first subgroup but not given access to intoxicated practice (groups Pre-Sal, Post-EtOH) had lower BACs for only the highest test dose (2.0 g/kg) during the post-chronic dose-effect tests. The subgroup of ANA rats that received pre- and post-session saline injections did not show significant changes in the BACs.

In order to assess the relative changes in the behavioral responses to alcohol during chronic drug exposure and the resulting metabolic tolerance which seemed to have selectively developed in the ANA rat lines, the rates-of-responding were plotted as a function of the blood alcohol levels. Figure 5 and Fig. 6 show the changes in the rates-of-responding as a function of BACs for each of the six subgroups. These comparisons show that the subgroup of

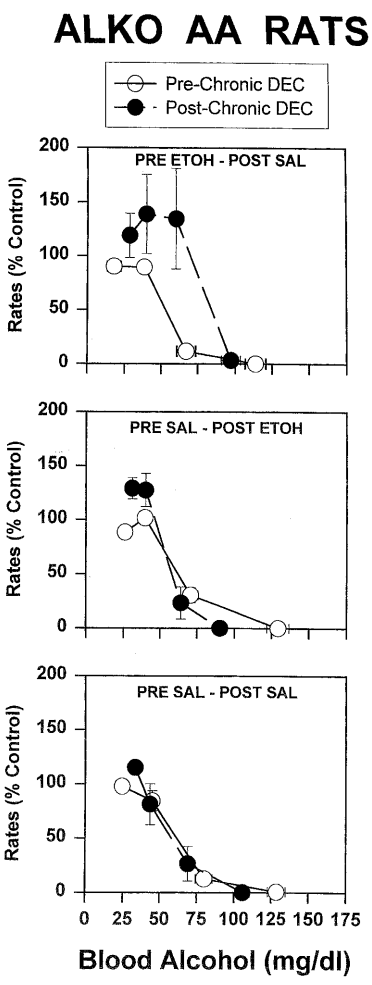

Fig. 5 The rates of operant responding during the cumulative dose-effect tests, expressed as a percentage of saline baseline control rates, plotted as a function of blood alcohol concentrations, expressed in mg/dl, in three groups of ALKO-AA rats before (*open circles*) and after (*closed circles*) a 30-day period of chronic alcohol administrations. Each point represents the mean (±SEM) of eight rats. The *top panel* represents the data from rats given alcohol intoxicated practice during the chronic exposure period (pre-session injections). The *middle panel* represents the data from alcohol control rats, which received equivalent post-session alcohol injections, without intoxicated practice. The *lower panel* represents the data from saline control rats

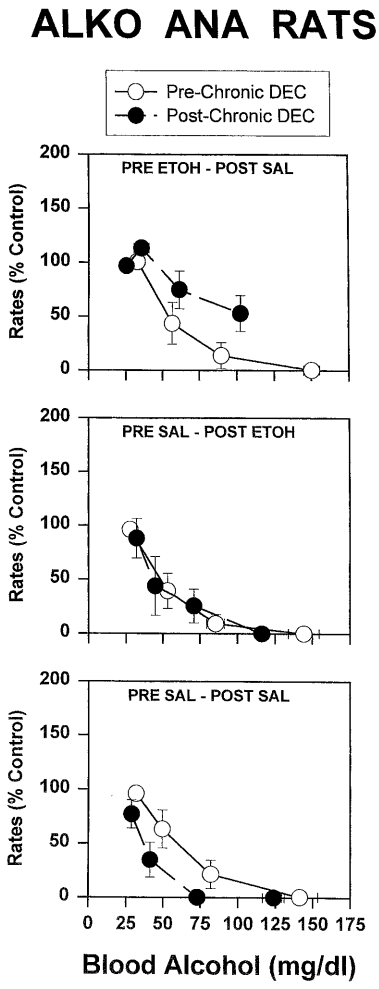

Fig. 6 The rates of operant responding during the cumulative dose-effect tests, expressed as a percentage of saline baseline control rates, are plotted as a function of blood alcohol concentrations, expressed in mg/dl, in three groups of ALKO-ANA rats before (*open circles*) and after (*closed circles*) a 30-day period of chronic alcohol administrations. Each point represents the mean (±SEM) of eight rats. The *top panel* represents the data from rats given alcohol intoxicated practice during the chronic exposure period (pre-session injections). The *middle panel* represents the data from alcohol control rats, which received equivalent post-session alcohol injections, without intoxicated practice. The *lower panel* represents the data from saline control rats

rats given access to intoxicated practice (groups Pre-EtOH, Post-Sal) of both the AA and ANA lines selectively developed tolerance. While the AA rat lines seemed to have developed sensitivity to the rate-increasing effects of the low to intermediate test doses of alcohol, the AA line did not develop tolerance to the rate-decreasing effects of the 2.0 g/kg test dose. On the other hand, the ANA rat lines did not appear to be sensitive to the locomotor stimulating effects of the low to intermediate doses of alcohol, but did develop a greater degree of tolerance to the rate-depressing effects of alcohol on behavioral performance when compared to their AA cohorts.

Figure 7 show the results of the conditioned place preference assays which were completed after phase I. Alcohol-induced place preferences were conditioned in the AA (top panel) and ANA (bottom panel) rat lines. The rats from both selectively bred lines which were given access to intoxicated practice in the behavioral task during phase I developed the strongest alcohol-induced place preferences (Pre-ETOH, Post-SAL, $P<0.001$). Passive drug exposure without intoxicated practice in the behavioral task (groups Pre-SAL, Post-ETOH) did not set the occasion for the subsequent development of alcohol-induced place preferences in the AA rat line (top panel) but this same exposure produced a significant shift in preference scores in the ANA rat line (bottom panel). Alcohol failed to induce conditioned place preferences or aversions in either of the control groups of rats (Pre-Sal, Post-SAL).

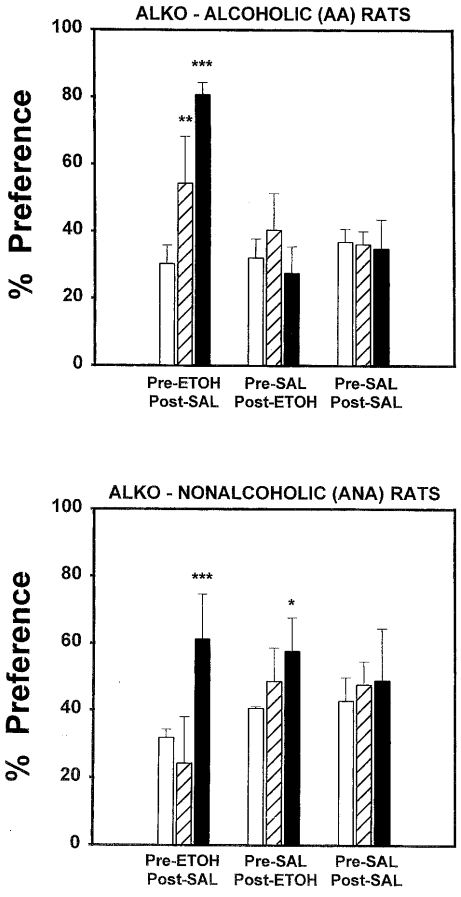

Fig. 7 Place conditioning induced by intragastric administration of alcohol. Group mean (±SEM) preference scores of ALKO-AA rats (*top panel*) and ALKO-ANA rats (*bottom panel*) expressed as a function of treatment conditions in the phase I operant sessions. The total time of a 30-min session spent in the non-preferred compartment divided by the total time spent in both conditioning compartments is expressed as a percentage, and is used as a measure of side preference. Place approach infers a positive hedonic valence for the unique environmental cues of the compartment. Initial side preferences/bias (*open bars*) were used to determine which of two compartments would be paired with alcohol administrations. After double alcohol-water pairings, rats were retested for side preference (T1 *hatched bars*, T2 *solid bars*). **P*<0.05, ***P*<0.01, ****P*<0.001

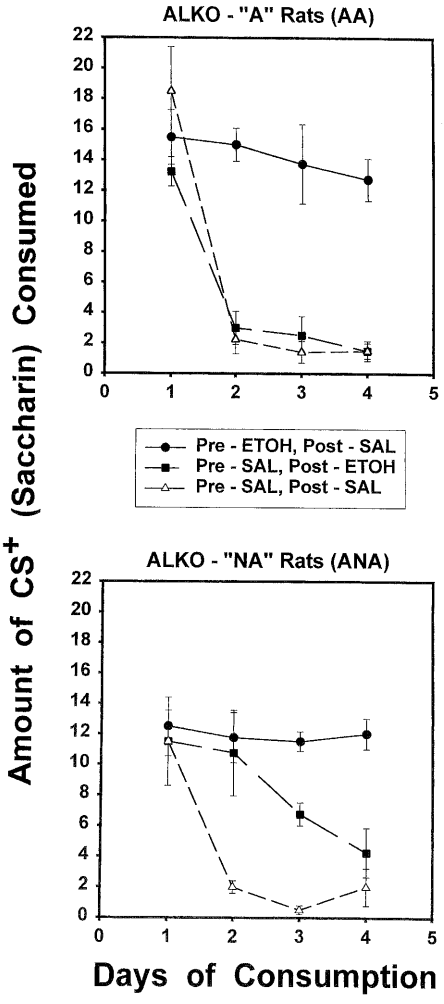

Fig. 8 Taste conditioning induced by 1.5 g/kg IP administered alcohol in ALKO-AA (*top panel*) and ALKO-ANA rats (*bottom panel*). Mean (±SEM) saccharin consumption (expressed in ml) over 4 consecutive days of taste-aversion conditioning are shown for each of three groups of rats of both selectively bred lines which had been previously trained in an FR30 food-reinforced operant task. Taste aversion, defined as a significant decline in voluntary saccharin consumption over the conditioning trials, infers a negative hedonic valence for the interoceptive alcohol cues associated with the alcohol injections

To assess the relative contribution of behavioral tolerance development on the aversive attributes of alcohol the results of the conditioned taste aversion assays are shown in Fig. 8. The subgroups of both the AA (top panel) and ANA lines (bottom panel) given access to intoxicated practice in the behavioral sessions failed to develop alcohol-induced conditioned taste aversions (groups Pre-EtOH, Post-Sal: closed circles). Passive drug exposure in the behavioral task for the AA rat line failed to confer tolerance to the aversive attributes of alcohol as measured by the taste aversion assay (groups Pre-Sal, Post-EtOH, top panel); however, this same passive alcohol exposure did retard the development of the conditioned taste aversion in the ANA rat lines (closed squares, bottom panel). Control groups of both AA and ANA rat lines developed robust alcohol-induced conditioned taste aversions (open triangles).

Discussion

There were significant differences in the blood alcohol concentrations between the AA and ANA rat lines in the present study. Eriksson (1969; Eriksson CJP 1981), Eriksson and Narhi (1973) and Eriksson and Rusi (1981) have previously reported a line difference in body weights between the AA and ANA rats which may have been a contributing factor to these BAC differences. Prior to the present set of studies an initial kinetics function was determined in both lines of rats the second week af-

ter arrival in the United States (data not shown). Each rat received IP administration of 2 g/kg alcohol and tail blood samples were collected. The kinetics functions demonstrated equivalent slopes with parallel lines [$t(18)=0.885$, NS]. At the time of these tests, the group mean body weight for the ANA rats was 332±8.78 g while the AA group mean body weight was 284±5.9 g. The differential BACs shown in the present paper were most likely due to differences in absorption-rates since, in this initial kinetics test, the BACs for the AA rats peaked 1 h after injection and the BAC of the ANA rats peaked 15 min after the administration. The differential rates-of-absorption were probably attributable to the differential body-weights previously reported.

A unique finding of the present study was that the shift-to-the-right in the alcohol dose-response curve in the AA rat line resulted from a strain-selective increase in the rates-of-responding during low to intermediate test doses of alcohol. The ANA rat line did not show this sensitivity to the rate-increasing effects of alcohol. Instead, the ANA rat lines seemed to develop tolerance to the rate-decreasing effects of intermediate to high doses of alcohol. Hilakivi et al (1984) have provided previous evidence to show that the AA rats appear to be more sensitive to the locomotor stimulating properties of alcohol than the ANA rats. In the present study, when the rates-of-responding between these two rat lines were compared with the blood alcohol concentrations during the behavioral sessions the resulting shifts in the behavioral dose-effect curves could not be attributed to the baseline strain differences in the blood alcohol levels. These data highlight the need to include BAC measurements in all studies evaluating tolerance development (Lê et al. 1992).

Kiianmaa et al. (1991) have suggested that AA rats develop a greater degree of tolerance to the depressive effects of alcohol than the ANA rats. The current data suggest that this tolerance development may reflect the development of sensitivity to the locomotor stimulating effects of alcohol. Kilbey and Sannerud (1984) have previously suggested that the development of sensitization to the locomotor effects of stimulants may be the best predictor of the degree or magnitude of tolerance development. Differential sensitivity to the locomotor stimulating effects of drugs in AA rats have been recently shown to generalize to both morphine and cocaine (Honkanen et al. 1999). The data from the present set of studies support these findings. These data also suggest that the learned compensatory strategy developed by the AA and ANA rat lines in response to the reduction of reinforcer deliveries during the chronic drug exposure period and the development of sensitivity to the locomotor stimulating effects of drugs, in general, may be the selectively bred trait which distinguishes these two rat lines in the development of behavioral tolerance.

On any occasion that a drug is given, its effects on behavior are crucially dependent on the specific behavioral and pharmacological parameters that characterize both the present circumstances of the investigation and the past history of the experimental subject (Blackman 1989). The questions of what is being learned during the development of tolerance to the behaviorally disruptive effects of alcohol and whether these learned events may significantly interact with alcohol's continued abuse has interest to both researcher and clinician. The functional significance of the development of alcohol tolerance in the present study is reflected in the subsequent assessment of the approach (rewarding) and avoidance (aversive) attributes of alcohol, as measured by taste and place conditioning. The pre-session administration of alcohol to both AA and ANA rats, which set the occasion for intoxicated practice and the development of behavioral tolerance to alcohol in the behavioral task, was associated with a shift in the hedonic valence of alcohol. This selective shift in the hedonic continuum was demonstrated by the conditioning of a place preference for the unique environmental cues associated with subsequent experimenter-administered alcohol. Rats from the AA selected line, which received equal alcohol exposures through post-session alcohol administrations but not intoxicated practice, failed to condition these place preferences. Interestingly, this passive alcohol exposure from post-session alcohol injections in the ANA rat line also set the occasion for the subsequent development of a place preference. However, the magnitude of this preference was less than the group of ANA rats that were given access to intoxicated practice. Saline control rats from both selected lines did not show any place conditioning to subsequent alcohol administrations. The development of place preferences in the group of ANA rats which received passive drug exposure is a unique characteristic which differentiates this group from the similarly treated rats of the AA rat lines.

Saline control groups from both rat lines developed a rapid and robust taste aversion to saccharin with alcohol conditioning. Rats from both the AA and ANA rat lines with the greatest tolerance development in the behavioral task (groups Pre-EtOH, Post-SAL) failed subsequently to condition this taste aversion to saccharin. Interestingly, the AA and ANA rats receiving post-session alcohol injections in the behavioral task demonstrated a different response to subsequent alcohol taste conditioning. The development of the taste aversion was retarded in the ANA post-session alcohol group – a phenomenon that was not demonstrated in the post-session AA rat group. The exact neural and operational mechanisms associated with the differential responses to intoxicated performance and to passive drug exposure in these two selected rat lines may underlie the differential voluntary alcohol consumption in drug-naive rats which identifies these lines as animal models of human alcohol abuse.

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