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# The effects of cocaine, alcohol and cocaine/alcohol combinations in conditioned taste aversion learning

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**Abstract**

We have recently reported that alcohol attenuates cocaine place preferences. Although the basis for this effect is unknown, alcohol may attenuate cocaine reward by potentiating its aversive effects. To examine this possibility, these experiments assessed the effects of alcohol on cocaine-induced taste aversions under conditions similar to those that resulted in attenuated place preferences. Specifically, Experiments 1 and 2 assessed the effects of alcohol (0.5 g/kg) on taste aversions induced by 20, 30 and 40 mg/kg cocaine. Experiment 3 examined the role of intertrial interval in the effects of alcohol (0.5 g/kg) on cocaine (30 mg/kg) taste aversions. In Experiments 1 and 2, cocaine was effective at conditioning aversions. Alcohol produced no measurable effect. Combining cocaine and alcohol produced no greater aversion than cocaine alone (and, in fact, weakened aversions at the lowest dose of cocaine). In Experiment 3, varying the intertrial interval from 3 days (as in the case of Experiments 1 and 2) to 1 day (a procedure identical to that in which alcohol attenuated cocaine place preferences) resulted in significant alcohol- and cocaine-induced taste aversions. Nonetheless, alcohol remained ineffective in potentiating cocaine aversions. Thus, under these conditions alcohol does not potentiate cocaine's aversiveness. These results were discussed in terms of their implication for the effects of alcohol on cocaine-induced place preferences. Further, the effects of alcohol on place preferences conditioned by cocaine were discussed in relation to other assessments of the effects of alcohol on the affective properties of cocaine and the implications of these interactions for alcohol and cocaine co-use.


*Keywords:* Alcohol; Aversion; Cocaine; Rats; Sprague–Dawley; Drug interaction

## 1. Introduction

Alcohol consumption commonly co-occurs with cocaine use (Caetano and Schafer, 1996; Caetano and Weisner, 1995; Carroll et al., 1993; Grant and Harford, 1990; Heil et al., 2001; Higgins et al., 1994). For instance, the prevalence of cocaine and alcohol co-use has been reported to be as high as 85% in the general population (see Grant and Harford, 1990) and 62% in a treatment-seeking population (Caetano and Schafer, 1996; Caetano and Weisner, 1995; Carroll et al., 1993; Heil et al., 2001; Higgins et al., 1994). Although it remains unknown why individuals use this combination at such high rates, many have suggested that alcohol possesses the ability to modulate the affective (e.g., rewarding, aversive, anxiogenic) properties of cocaine in a manner that increases the likelihood of their co-use (see Farré et al., 1993; Knackstedt and Ettenberg, 2005; Lewis and June, 1994; Magura and Rosenblum, 2000; McCance-Katz et al., 1998; see also Moolten and Kornetsky, 1990). Specifically, alcohol may either increase cocaine's rewarding properties (see Farré et al., 1993; Lewis and June, 1994; McCance-Katz et al., 1998; see also Moolten and Kornetsky, 1990) and/or decrease its aversive (including anxiogenic) effects (Knackstedt and Ettenberg, 2005; Magura and Rosenblum, 2000; McCance-Katz et al., 2005).

We have recently reported that alcohol modulates cocaine's rewarding properties within the place conditioning design (see Busse et al., 2004; Busse and Riley, 2002). In particular, cocaine-induced place preferences were significantly *attenuated* when animals were conditioned with a combination of 0.5 g/kg alcohol and 20, 30 or 40 mg/kg cocaine (see Busse et al., 2004; Busse and

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Riley, 2002). Although the bases for the attenuation of cocaine-induced place preferences by alcohol remains unknown, e.g., whether such interactions reflect pharmacokinetic and/or pharmacodynamic changes (see Horowitz et al., 1997; McCance-Katz et al., 2005; Pan and Hedaya, 1999), it is possible that, under some conditions, alcohol may increase both the rewarding (see Farré et al., 1993; Lewis and June, 1994; McCance-Katz et al., 1998; see also Moolten and Kornetsky, 1990) and aversive effects of cocaine (Etkind et al., 1998; Grakalic and Riley, 2002). Under these conditions, e.g., high doses of cocaine, the potentiation of cocaine's aversive effects may mask or outweigh any potentiation that occurs to its rewarding effects. Interestingly, Le Pen et al. (1998) offered a similar interpretation of their findings that place preferences induced by 20 mg/kg cocaine were attenuated by pretreatment with the dopamine uptake inhibitor, GBR12783. Specifically, they attributed the attenuation by GBR12783 to a masking of cocaine rewarding properties by its potentiation of cocaine's aversive effects.

Although it is possible that alcohol's attenuation of cocaine-induced place preferences is a function of an increase in cocaine's aversive effects, there are several difficulties with this interpretation. For example, the attenuation of cocaine-induced place preferences by alcohol (as well as by other drugs, see above) may actually reflect a decrease in cocaine reward rather than a potentiation of its aversive effects (see Gaiardi et al., 1998). Such an effect would also be reflected in a change in the ability of cocaine to induce a place preference. Further, much of the evidence suggesting that alcohol potentiates the aversive effects of cocaine (Etkind et al., 1998; Grakalic and Riley, 2002) do so under different parametric conditions (e.g., route of administration, sex and strain of subject, intertrial interval) than those assessing the effects of alcohol on cocaine-induced place preferences (Busse et al., 2004; Busse and Riley, 2002). These parametric variables have all been shown to be significant factors in aversion learning with cocaine (Elkins et al., 2003; Ferrari et al., 1991; Glowa et al., 1994; Grabus et al., 2004; Grigson and Freet, 2000; van Haaren and Hughes, 1990; see Riley and Freeman, 2004). As such, it remains unknown whether the conditions under which alcohol attenuates cocaine-induced conditioned place preferences also potentiate cocaine's aversiveness. The present series of experiments tested this more directly by examining the ability of alcohol to potentiate cocaine-induced taste aversions under conditions similar to those in which alcohol attenuates cocaine-induced place preferences. Specifically, Experiments 1 and 2 examined the effects of alcohol on conditioned taste aversions induced by a variety of doses of cocaine in male Sprague–Dawley rats injected with cocaine intraperitoneally. Experiment 3 examined the contribution of intertrial interval in mediating the effects of alcohol on cocaine-induced taste aversions.

## 2. General methods

### 2.1. Subjects

Male Sprague–Dawley rats (Harlan Sprague Dawley Laboratories), weighing approximately 250 to 350 g at the start of each experiment, were housed in separate hanging wire cages in a room maintained on a 12 L : 12 D light cycle (lights on at 0800 hours) and at an ambient temperature of 23 °C. Food and water were available ad libitum except where noted. Animals were handled daily beginning 2 weeks prior to the start of each experiment in order to limit any effects of handling stress during conditioning and testing. Procedures recommended by the Guide for the Care and Use of Laboratory Animals (National Research Council, 1996), the Guidelines for the Care and Use of Mammals in Neuroscience and Behavioral Research (National Research Council, 2003) and the Institutional Animal Care and Use Committee at American University were followed at all times.

### 2.2. Drugs

Cocaine hydrochloride (generously supplied by the National Institute on Drug Abuse) was dissolved in distilled water and was injected intraperitoneally (IP) in a concentration of 10 mg/ml (cocaine doses are expressed as the salt). Ethyl alcohol was prepared in a 15% solution with distilled water (*v*/*v*) and was also injected IP. Cocaine and alcohol were administered as separate injections. Vehicle injections were distilled water and were matched in number and volume to the injections of cocaine and alcohol. Saccharin (0.1% sodium saccharin, Sigma Chemical Co., St. Louis, MO) was prepared as a 1 g/l solution in tap water.

### 2.3. Procedure

Phase I: Habituation. Following 23-h water deprivation, subjects were given 20-min access to water (presented in graduated 50-ml Nalgene tubes). This procedure was repeated daily until all subjects were approaching and drinking from the tube within 2 s of its presentation.

Phase II: Conditioning. On Day 1 of this phase, all subjects were given 20-min access to a novel saccharin solution. Immediately following saccharin access, subjects were rank ordered on saccharin consumption and assigned to their respective groups (i.e., either a vehicle, cocaine-only, alcohol-only or cocaine/alcohol treatment group; group designation differs for each experiment). All injections were given within 10 min of removal of the saccharin bottles.

The following 3 days (or 1 day, as in the case of Experiment 3) were water-recovery sessions wherein all subjects were given 20-min access to water. No injections were given following water access on these days. This alternating procedure of conditioning/water recovery was repeated until all subjects received four complete cycles. On the day following the last cycle, all subjects were given 20-min access to saccharin in a Final Aversion Test. No injections followed this access.

### 2.4. Statistical analysis

Differences in absolute saccharin consumption were assessed using a repeated measures ANOVA with the between-group factor of Group and the within-subjects factor

of Trial. All determinations of statistical significance were made at $p<.05$. Tukey's HSD post-hoc tests were used to confirm any main effect of Group or Trial or any significant Group × Trial interaction.

## 3. Experiment 1

Experiment 1 assessed the effects of alcohol (0.5 g/kg) on taste aversions induced by 20 mg/kg cocaine. Specifically, following habituation and initial saccharin presentation (Trial 1), 34 subjects were rank ordered on saccharin consumption and assigned to receive either two injections of vehicle (Group V–V; $n=8$), 20 mg/kg cocaine and vehicle (Group C–V; $n=8$), 0.5 g/kg alcohol and vehicle (Group V–A; $n=8$) or the cocaine/alcohol combination (Group C–A; $n=10$). The doses of cocaine and alcohol were based on Busse and Riley (2002), wherein place preferences induced by 20 mg/kg cocaine were attenuated by 0.5 g/kg alcohol (but see Busse et al., 2004).

## 4. Results: experiment 1

The overall 4 × 5 repeated measures ANOVA indicated a significant main effect for Group and Trial [$F(3, 30)=3.285$, $p=0.03$; $F(4, 120)=18.352$, $p<0.0001$, respectively], as well as a Group × Trial interaction [$F(12, 120)=2.032$, $p=0.03$; see Fig. 1]. Post-hoc analysis established that overall saccharin consumption was greater on Trials 2, 3 and 4 and on the Final Aversion Test than it was on Trial 1 ($ps<0.05$). Animals injected with cocaine alone (i.e., Group C–V), however, consumed significantly less saccharin across trials than animals in Groups V–V, V–A and C–A ($ps<0.05$). More specifically, subjects in Group C–V consumed less saccharin than Group V–V on Trials 3 and 4 and Groups V–A and C–A on Trial 4. The fact that subjects injected with cocaine alone drank less than those injected with the cocaine/alcohol combination suggests that alcohol *attenuated* the aversions induced by cocaine.

## 5. Discussion

As compared to vehicle controls (Group V–V), 20 mg/kg cocaine (Group C–V) induced a weak aversion. Animals conditioned with 0.5 g/kg alcohol alone (Group V–A), on the other hand, displayed no measurable aversion, an effect consistent with other work demonstrating that this dose of alcohol does not induce aversions in either the place preference or taste aversion design (see Busse et al., 2004; Busse and Riley, 2002; Etkind et al., 1998; Fidler et al., 2004; Grakalic and Riley, 2002). Interestingly, animals that underwent taste aversion conditioning with the combination of 20 mg/kg cocaine and 0.5 g/kg alcohol never differed from Group V–V, but did differ from Group C–V on Trial 4, perhaps suggestive of a weakening in cocaine's ability to condition a taste aversion by alcohol.

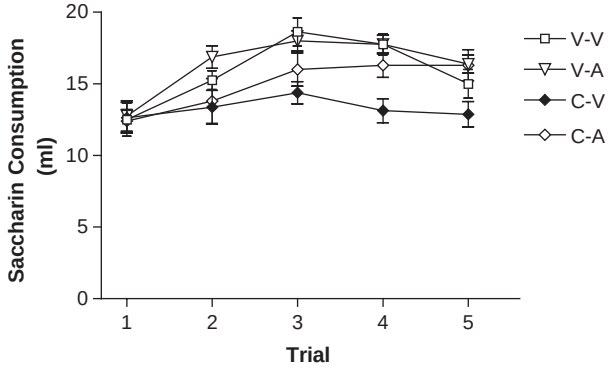


Fig. 1. Mean (±SEM) absolute saccharin consumption in ml across trials for animals conditioned with vehicle (Group V–V), 0.5 g/kg alcohol (Group V–A), 20 mg/kg cocaine (Group C–V) or the cocaine/alcohol combination (Group C–A).

It is quite surprising that, in the present context, alcohol appeared to weaken cocaine's aversiveness, especially since most evidence, to date, suggests that alcohol potentiates the aversive effects of cocaine within this design (Etkind et al., 1998; Grakalic and Riley, 2002). Still, such an effect is consistent with others examining the interaction between alcohol and cocaine's anxiogenic properties (see Knackstedt and Ettenberg, 2005; Magura and Rosenblum, 2000). In particular, Knackstedt and Ettenberg (2005) trained animals with intravenous (IV) cocaine to run to a goal box at the end of a runway apparatus. Interestingly, animals displayed approach–avoidance behaviors (a suggested index of anxiety) before entering the goal box. The administration of alcohol after training sessions, however, reduced the number of retreats animals displayed on these tests. Such an effect was interpreted as a weakening of the negative consequences of cocaine by alcohol (see also McCance-Katz et al., 2005). If this is the case, then it is possible that the weakening of cocaine-induced taste aversions by alcohol is truly a reflection of a decrease in cocaine's aversiveness.

A second interpretation of these data, however, is that alcohol decreased the rewarding properties of cocaine. In fact, it has recently been postulated that the taste "aversion" design reflects changes in the rewarding properties of drugs rather than their aversive effects. In particular, Grigson and her coworkers have suggested that decreases in fluid consumption that occur when saccharin is paired with cocaine reflect a devaluation of the saccharin solution in comparison (or an anticipation of) cocaine (see Grigson and Twining, 2002; Grigson and Freet, 2000). That is, animals lower their consumption of the rewarding saccharin solution in anticipation of the subsequent rewarding cocaine experience. If this were the case, then alcohol's attenuation of cocaine-induced taste aversions would be more reflective of a decrease in the rewarding properties of cocaine then a decrease in its aversiveness. As noted, such an interpretation would be consistent with the place conditioning work presented by Busse and Riley (2002) and Busse et al. (2004).

Independent of these suggested bases for the results of Experiment 1, it is clear that the conditions under which these animals were tested did not support a potentiation in cocaine-induced taste aversions by alcohol. Although it is not known why alcohol did not potentiate cocaine-induced aversions, it is possible that the aversive effects of 20 mg/kg cocaine were weak and not subject to potentiation (for a dose–response comparison for cocaine CTAs, see Busse et al., 2005b; Ferrari

et al., 1991). Interestingly, prior work from this lab and others has reported that 20 mg/kg cocaine when administered intraperitoneally produces a weak aversion (see also Fig. 1). It is important to note in this context that although Busse and Riley (2002) reported that 0.5 g/kg alcohol attenuated place preferences conditioned by 20 mg/kg cocaine, this effect was not evident in a subsequent experiment by this group (see Busse et al., 2004). In fact, Busse et al. (2004) only found an attenuation in cocaine-induced place preferences when combining alcohol with higher cocaine doses (i.e., 30 and 40 mg/kg). As such, under these conditions, 20 mg/kg cocaine may be near threshold for detecting any potentiation in its aversiveness by alcohol.

Given this possibility, the purpose of Experiment 2 was to assess the effects of 0.5 g/kg alcohol on taste aversions induced by 30 and 40 mg/kg cocaine. Specifically, following habituation and initial saccharin exposure (Trial 1), 48 subjects were rank ordered on saccharin consumption and assigned to receive either two injections of vehicle (Group V–V; $n=8$), 20 or 30 mg/kg cocaine and vehicle (Groups C30–V; $n=8$; C40–V; $n=8$), 0.5 g/kg alcohol and vehicle (Group V–A; $n=8$) or the respective cocaine/alcohol combinations (Groups C30–A; $n=8$; C40–A; $n=8$). In addition to being well above the threshold for inducing taste aversions (see Busse et al., 2005b; Ferrari et al., 1991; Grakalic and Riley, 2002), these doses of cocaine induce place preferences that are robustly attenuated by 0.5 g/kg alcohol (see Busse et al., 2004).

## 6. Results: experiment 2

The overall $6\times5$ repeated measures ANOVA indicated a significant main effect for Group and Trial [$F(5, 35)=6.891$, $p=0.0001$; $F(4, 140)=24.948$, $p<0.0001$, respectively], as well as a significant Group × Trial interaction [$F(20, 140)=5.351$, $p=0.03$; see Fig. 2]. Tukey's HSD post-hoc analyses established that overall saccharin consumption was less on Trials 2, 3, 4 and on the Final Aversion Test, as compared to Trial 1 ($ps<0.05$). Similar to Experiment 1, alcohol, when administered alone, produced no decrease in saccharin consumption compared to vehicle ($p<0.05$). Animals administered cocaine, alone or in combination with alcohol (Groups C30–V, C40–V, C30–A and C40–A), displayed a significant reduction in saccharin intake across trials compared to subjects administered vehicle (Group V–V) or alcohol alone (Group V–A; $ps<0.05$). There were no differences in overall saccharin consumption between animals administered cocaine alone (Groups C30–V and C40–V) and their respective cocaine/alcohol counterparts (Groups C30–A and C40–A).

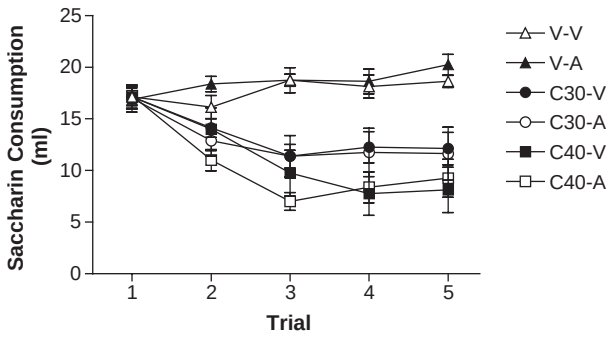


Fig. 2. Mean (±SEM) absolute saccharin consumption in ml across trials for animals conditioned with vehicle (Group V–V), 0.5 g/kg alcohol (Group V–A), 30 mg/kg cocaine (Group C30–V), 40 mg/kg cocaine (Group C40–V) or the respective cocaine/alcohol combination (Groups C30–A and C40–A).

In the context of the Group × Trial interaction, Group C40–A significantly reduced saccharin intake by Trial 2, as compared to vehicle controls (Group V–V) and animals conditioned with alcohol alone (Group V–A; $ps<0.05$; see Fig. 2). By Trial 3, all cocaine and cocaine/alcohol groups reduced saccharin consumption relative to Groups V–V and V–A ($ps<0.05$). However, these groups did not differ from each other. On Trial 4, saccharin intake in Groups C40–V and C40–A remained significantly reduced below that in animals in Groups V–V and V–A ($ps<0.05$), while all other comparisons failed to reach statistical significance. By the Final Aversion Test, all cocaine and cocaine/alcohol groups displayed a significant reduction in saccharin intake below that of animals administered alcohol alone ($ps<0.05$), while only Groups C40–V and C40–A differed from vehicle controls ($ps<0.05$). Thus, if any potentiation occurred with the cocaine/alcohol combination, it was weak and limited to Trial 2.

## 7. Discussion

Compared to vehicle controls (Group V–V), subjects injected with 30 and 40 mg/kg cocaine (Groups C30–V and C40–V, respectively) displayed a modest decrease in saccharin consumption, an effect consistent with other reports on the aversiveness of cocaine at these doses and by this route (Busse et al., 2005b). Similar to Experiment 1, however, no decrease was evident in animals administered 0.5 g/kg alcohol (Group V–A). Interestingly, as compared to their cocaine-only counterparts, the combinations of cocaine and alcohol (Groups C30–A and C40–A) produced similar decreases in saccharin consumption across most trials, although a faster acquisition of aversions was noted in Group C40–A. That this difference was only evident on Trial 2 suggests that if any potentiation occurred, it was weak and short-lived. Thus, the results of Experiment 2 present no significant evidence that, under these experimental conditions, alcohol potentiates the aversive properties of cocaine.

One additional variable that may impact the likelihood of alcohol potentiating the aversive effects of cocaine is the interval between successive conditioning trials. Specifically, in Experiments 1 and 2 animals were given access to the saccharin solution and injected with a drug (or drug combination) once every 4 days. Although this procedure is common in work on conditioned aversion learning (see Riley and Freeman, 2004; see also www.CTAlearning.com) and similar to the procedure used in the initial demonstrations of alcohol's potentiation of cocaine-induced taste aversions (see Etkind et al., 1998; Grakalic and Riley, 2002), it is different than that which was employed by Busse and Riley (2002) and Busse et al. (2004) in their assessment of the effects of alcohol on

cocaine-induced place preferences. In these latter assessments, the pairing of the place conditioning chamber with the drug (or drug combination) was given every other day. Such a difference may be important given that the aversive effects of drugs have been reported to vary with this parameter (Domjan, 1980). The purpose of Experiment 3, therefore, was to investigate the effects of 0.5 g/kg alcohol on taste aversions induced by 30 mg/kg cocaine when the pairing of saccharin and cocaine occurred every other day. Specifically, following habituation and initial saccharin exposure, 40 subjects were rank ordered on initial saccharin consumption and assigned to receive either two injections of the vehicle (Group V–V; $n=8$), 30 mg/kg cocaine and vehicle (Groups C–V; $n=12$), 0.5 g/kg alcohol and vehicle (Group V–A; $n=8$) or the cocaine/alcohol combination (Groups C–A; $n=12$). Conditioning occurred every other day for a total of four conditioning trials.

## 8. Results: experiment 3

The overall $4\times5$ repeated measures ANOVA indicated a significant main effect for Group and Trial [$F(3, 36)=12.533$, $p<0.0001$; $F(4, 144)=25.493$, $p<0.0001$, respectively], as well as a significant Group × Trial interaction [$F(12, 144)=2.032$, $p<0.0001$; see Fig. 3]. Specifically, although post-hoc analysis established that overall saccharin consumption was greater on Trial 2 than it was on Trial 1 ($p<0.05$), animals consumed less saccharin on the Final Aversion Test than they did on Trial 2 ($p<0.05$). In the context of the main effect for Group, post-hoc analyses confirmed that animals conditioned with 30 mg/kg cocaine (Group C–V), 0.5 g/kg alcohol (Group V–A) and the cocaine/alcohol combination (Group C–A) consumed less overall saccharin than vehicle controls (Group V–V; $ps<0.05$). In addition, Group C–A also consumed significantly less saccharin than animals in Group V–A ($p<0.05$).

Upon examination of the Group × Trial interaction, post-hoc analysis indicated that animals in Groups C–V and C–A consumed less saccharin than Group V–V on Trials 2, 3 and 4 ($ps<0.05$). In addition Group V–A drank less saccharin than Group V–V on Trial 3 and 4 and on the Final Aversion Test, but more than Group C–V and C–A on Trial 4 and on the Final Aversion Test ($ps<0.05$). Groups C–V and C–A did not differ in saccharin consumption at any point in conditioning. Thus, these data indicate that the combination of cocaine and alcohol did not produce greater aversions than cocaine alone, despite alcohol (i.e., 0.5 g/kg) producing an aversion on its own.

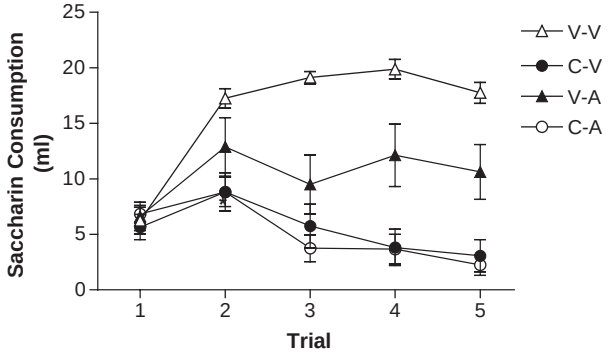


Fig. 3. Mean (±SEM) absolute saccharin consumption in ml across trials (with a 1 day intertrial interval) for animals conditioned with vehicle (Group V–V), 0.5 g/kg alcohol (Group V–A), 30 mg/kg cocaine (Group C–V) or the cocaine/alcohol combination (Group C–A).

## 9. Discussion

Compared to vehicle controls (Group V–V), animals injected with 30 mg/kg cocaine (Group C–V) displayed a modest decrease in saccharin consumption by Trial 2. This reduction was significantly different than that which occurred in animals conditioned with alcohol alone (Group V–A). Interestingly, and unlike the previous experiments, animals administered 0.5 g/kg alcohol (Group V–A) every other day also displayed a reduction in saccharin consumption as compared to Group V–V (an effect likely due to the change in the intertrial interval during conditioning; see Domjan, 1980). Similar to animals conditioned with cocaine alone (Group C–V), animals injected with the drug combination (Group C–A) displayed a significant reduction in saccharin consumption from vehicle controls (Group V–V). This reduction, however, never differed from that in animals administered cocaine alone. It is important to note that initial saccharin consumption on Trial 1 was low, relative to Experiments 1 and 2. It is, therefore, possible that no potentiation was evident because subsequent reductions in saccharin consumption were not possible (due to the initially low level of consumption at baseline). Following the final aversion test, all subjects were given four extinction trials in which saccharin was given with no subsequent injections. There was no difference in the rate and degree of extinction between animals given cocaine and those given the cocaine/alcohol combination (data not shown). Thus, the failure to see any difference between the cocaine and cocaine/alcohol groups was not likely a function of an inability to see any differences due to the "basement" effects during acquisition. Thus, despite that fact that alcohol produced aversions on its own, no potentiation in the aversiveness of cocaine was evident when animals underwent aversion conditioning with the combination of cocaine and alcohol every other day.

## 10. General discussion

Busse and his colleagues have recently suggested that alcohol's ability to modulate cocaine place preferences may be due to an alteration in the relative (and dose-dependent) balance of cocaine reward and aversion (Busse et al., 2004; Busse and Riley, 2002). Specifically, they suggest that when alcohol is co-administered with doses of cocaine high enough to support aversion learning (see Busse et al., 2005b; Etkind et al., 1998; Ferrari et al., 1991; Grakalic and Riley, 2002), increases in the aversive effects of cocaine mask or outweigh any change that occur to cocaine reward. The present experiments attempted to more directly examine this possibility by assessing the effects of alcohol on cocaine's aversive effects under conditions comparable to those in which the attenuation

was reported (i.e., male Sprague–Dawley rats injected intraperitoneally every other day) and in a design sensitive to the aversive effects of drugs, i.e., the CTA design (Hunt and Amit, 1987; Riley and Tuck, 1985).

Overall, the data from Experiments 1, 2 and 3 indicate that alcohol did *not* potentiate cocaine-induced taste aversions. This lack of potentiation was evident across a range of cocaine doses (20, 30 and 40 mg/kg) and under several different schedules of injections (i.e., every fourth day and every other day), although this latter condition did support the emergence of alcohol's aversive effects. The failure of alcohol to potentiate cocaine aversions is surprising given previous reports indicating such potentiation (Etkind et al., 1998; Grakalic and Riley, 2002). The differences between the results of the present series of studies and those previously reporting alcohol potentiation of cocaine aversions is likely due to parametric differences, e.g., route of administration and strain and sex of the subject, factors all of which have been shown to be important in mediating the aversive effects of cocaine (see Elkins et al., 2003; Ferrari et al., 1991; Glowa et al., 1994; Grabus et al., 2004; Grigson and Freet, 2000; van Haaren and Hughes, 1990).

In the context of the place conditioning work by Busse and Riley (2002; see also Busse et al. 2004), the results from these experiments do little to support the hypothesis that the attenuation in cocaine place preferences by alcohol is a result of its potentiation of the aversiveness of cocaine. Instead of increasing cocaine aversiveness and, in effect, outweighing any changes that occur to cocaine reward, alcohol may be directly reducing the rewarding effects of cocaine (independent of any specific changes in cocaine's aversive effects). Although untested in this report, there are several mechanisms that may account for alcohol's ability to reduce cocaine reward. For example, it has been reported that acute alcohol not only attenuates the increase in dopamine levels that occurs in the striatum following cocaine injections, but cocaine-induced motor activity as well (Dewey et al., 1997). This effect was attributed to GABAergic inhibition of dopamine activity. Similarly, other compounds that increase GABA levels reduce both the biochemical and behavioral effects of cocaine. Specifically, gamma vinyl-GABA (GVG, Vigabatrin), an irreversible inhibitor of GABA transaminase (an enzyme involved in the metabolism of GABA) reduces the cocaine-induced increase in dopamine levels in the nucleus accumbens in rats (Ashby et al., 1999; Gerasimov and Dewey, 1999; Morgan and Dewey, 1998) and primates (Dewey et al., 1998). Further, this compound blocks cocaine self-administration (Kushner et al., 1999) and cocaine-induced place preferences in the rat (Dewey et al., 1998). A similar effect on cocaine-induced place preferences was obtained by co-administering diazepam with cocaine (Meririnne et al., 1999). Thus, it possible that alcohol attenuates cocaine-induced place preferences by attenuating cocaine reward, an effect of decreasing dopamine levels, rather than by increasing its aversiveness.

A second possibility is that, in addition to affecting dopaminergic output via GABA, alcohol may also affect the ability of cocaine to condition a place preference by altering glutamate activity in the mesolimbic and mesocortical pathways. In fact, alcohol has been demonstrated to act as a glutamate antagonist at the NMDA receptor (Bienkowski et al., 1997; Krystal et al., 2003; Nagy, 2004; for a review see Kumari and Ticku, 2000). Such reductions in glutamate activity may inhibit cocaine's ability to condition a place preference (Harris and Aston-Jones, 2003; Nakagawa et al., 2005). Specifically, Harris and Aston-Jones (2003) demonstrated that microinjections of AP5 plus CNQX, glutamate antagonists, into the VTA blocked the development of cocaine-induced place preferences. A similar effect was reported by Nakagawa et al. (2005) wherein (*R*)-(−)-5-methyl-1-nicotinoyl-2-pyrazoline (MS-153), a glutamate transporter activator that reduces glutamate activity, attenuated the induction of cocaine place preferences. Thus, alcohol may attenuate the rewarding effects of cocaine by inhibiting glutamate activity.

The current data suggest that alcohol's attenuating effects on cocaine-induced place preferences are not a function of any changes in cocaine's aversiveness. It should be noted, however, that this analysis makes several assumptions. First, it assumes that the aversiveness of cocaine is comparably measured or detected in the taste and place aversion designs. There are several lines of evidence that suggest that this may not necessarily be the case. In fact, although both preparations are sensitive to the rewarding and aversive properties of drugs (Hunt and Amit, 1987; Riley and Simpson, 2001; Tzschentke, 1998; for a bibliography on place conditioning, see Schechter and Calcagnetti, 1993, 1998; for a bibliography on CTA, see www.CTAlearning.com), manipulations that result in a modulation in place conditioning do not necessarily produce parallel changes in taste aversion learning. For example, via lesioning of the medial prefrontal cortex, Isaac et al. (1989) converted cocaine-induced place preferences to place aversions with no corresponding change in cocaine-induced taste aversions. Similarly, Laviolette et al. (2002) reported that nicotine place preferences became place aversions after a lesion to the tegmental pedunculopontine nuclei, again with no change in nicotine-induced CTAs. Thus, the neural mechanisms mediating the affective properties of drugs in the place conditioning design may have little to no overlap with the neural mechanisms involved in CTA learning. Assessments of the changes in the aversiveness of cocaine in the place conditioning design may be more insightful to understanding the role of such changes in the attenuation of cocaine-induced place references by alcohol (see Cunningham et al., 2001).

A second assumption is that the conditioned taste aversion design is a sensitive and valid measure of drug aversion. As previously noted, it has recently been suggested that this design may instead be an index of the rewarding properties of drugs rather than a drug's aversiveness (see Grigson and Twining, 2002; Grigson and Freet, 2000; for a detailed discussion, see Foynes and Riley, 2004). That is, animals may avoid consuming a rewarding saccharin solution in anticipation of cocaine (anticipatory contrast). Therefore, if the attenuation in cocaine place preferences by alcohol presented by Busse and his colleagues (Busse and Riley, 2002; Busse et al., 2004) were a function of changes in the aversiveness of cocaine by alcohol, but the taste aversion design were more sensitive to changes in

drug reward, its use may not be appropriate in the context of this analysis.

Independent of these interpretational issues, the bases for the high rates of cocaine and alcohol co-use remain to be determined (see Introduction). Based on the place conditioning work of Busse and Riley (2002), the rewarding properties of cocaine appear to be weakened when this drug is combined with alcohol. Yet, others have shown that alcohol can alter the affective (rewarding and aversive) properties of cocaine in a manner that would predict an increased co-use of these compounds. For instance, Knackstedt and Ettenberg (2005) demonstrated a decrease in the anxiogenic properties of cocaine by alcohol (see also McCance-Katz et al., 2005). Further, others have shown that alcohol increases the rewarding properties of cocaine in both humans (see Farré et al., 1993; McCance-Katz et al., 1998) and animals (Lewis and June, 1994). Thus, what remains to be determined is why alcohol attenuates cocaine place preferences, an index of drug reward, and if (and under what conditions) this attenuation would be abated and/or reversed. Interestingly, it has been demonstrated that history with alcohol weakens alcohol's ability to potentiate cocaine-induced taste aversions (Grakalic and Riley, 2002). If the place conditioning design is a sensitive measure of both the rewarding and aversive properties of drugs (see Tzschentke, 1998; for a bibliography, see Schechter and Calcagnetti, 1993, 1998), and alcohol history affects one (or both) of these properties, then it is possible that the interaction between cocaine and alcohol within the place conditioning design would be affected by such a history (see Busse et al., 2005a). Under such conditions, alcohol may potentiate cocaine's rewarding effects.

In conclusion, under conditions similar to those employed by Busse and Riley (2002) and Busse et al. (2004), taste aversions induced by cocaine were not potentiated by alcohol. Although the basis for this failure and its implications for changes in apparent cocaine reward remain unknown, it is clear that continued investigation is needed to understand how, and under what conditions, alcohol alters the abuse liability of cocaine.

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