Published in final edited form as:
Dev Psychobiol. 2014 July ; 56(5): 943–954. doi:10.1002/dev.21171.

AGE-DEPENDENT MDPV-INDUCED TASTE AVERSIONS AND THERMOREGULATION IN ADOLESCENT AND ADULT RATS

Andrew P. Merluzzi[1], **Zachary E. Hurwitz**[1], **Maria A. Briscione**[1], **Jennifer L. Cobuzzi**[1], **Bradley Wetzell**[1], **Kenner C. Rice**[2], and **Anthony L. Riley**[1]

[1]Psychopharmacology Laboratory Department of Psychology American University Washington, DC 20016

[2]Chemical Biology Research Branch National Institute on Drug Abuse Bethesda, MD 20892

Abstract

Adolescent rats are more sensitive to the rewarding and less sensitive to the aversive properties of various drugs of abuse than their adult counterparts. Given a nationwide increase in use of “bath salts,” the present experiment employed the conditioned taste aversion procedure to assess the aversive effects of 3,4-methylenedioxypyrovalerone (MDPV; 0, 1.0, 1.8 or 3.2 mg/kg), a common constituent in “bath salts,” in adult and adolescent rats. As similar drugs induce thermoregulatory changes in rats, temperature was recorded following MDPV administration to assess if thermoregulatory changes were related to taste aversion conditioning. Both age groups acquired taste aversions, although these aversions were weaker and developed at a slower rate in the adolescent subjects. Adolescents increased and adults decreased body temperature following MDPV administration with no correlation to aversions. The relative insensitivity of adolescents to the aversive effects of MDPV suggests that MDPV may confer an increased risk in this population.

Keywords

MDPV; bath salts; taste aversions; body temperature

1. INTRODUCTION

Use and abuse of “bath salts,” a new group of designer drugs composed primarily of synthetic cathinones, has been increasing in recent years (Bronstein, Spyker, Cantilena, Green, Rumack, & Dart, 2011). Poison control centers across the United States have reported a dramatic increase in the number of calls relating to these substances, with 0 calls in 2009 to 302 in 2010 and 2,237 in 2011 (NDIC, 2011). Further, anecdotal reports describe symptoms of paranoid psychotic behavior, agitation, hallucinations and delirium following use of “bath salts” (Penders, 2012). It is interesting to note that while the synthetic compounds found in “bath salts” are routinely changing in an effort to circumvent laws on

Please address all correspondence to: Andrew P. Merluzzi Psychopharmacology Laboratory Department of Psychology American University 4400 Mass. Ave., NW Washington, DC 20016 Phone: (202) 885-1721 Fax: (202) 885-1081 andrewmerluzzi@gmail.com.

The authors declare no conflict of interest.

banned substances (USDEA, 2011), there seem to be several common components, e.g., mephedrone, methylone and 3,4-methylenedioxypyrovalerone (MDPV), the latter of which has been a popular constituent (Airuehia, 2012). MDPV is a potent dopamine (DA) and norepinephrine (NE) reuptake inhibitor that increases extracellular concentrations of these neurotransmitters in the mesolimbic pathway and has increased potency and selectivity for the catecholamines as compared with cocaine (Baumann, Partilla, Lehner, Thorndike, Hoffman, & Holy, 2012). By these mechanisms, MDPV induces its euphoric and hallucinogenic effects (Ross, Reisfield, Watson, Chronister, & Goldberger, 2012).

Although there have been many anecdotal reports documenting both the physical and behavioral effects of MDPV (McClean, Anspikian, & Tsuang, 2012; Mugele, Nañagas, & Tormoehlen, 2012; Airuehia, 2012), as well as papers describing patterns and trends in its use (Olives, Orozco, & Stellpflug, 2012), there are relatively few controlled studies examining the drug in rodents (or in humans). In one of the first reports documenting the behavioral effects of MDPV, adult Sprague-Dawley rats were able to acquire and maintain intravenous self-administration (SA) of 0.05, 0.1 or 0.2 mg/kg per infusion MDPV, doses which were also successful in lowering intracranial self -stimulation (ICSS) thresholds (Watterson, Kufahl, Nemirovsky, Sewalia, Grabenauer, Thomas, Marusich, Wegner, & Olive, 2012). In an assessment of its discriminative stimulus properties in rats, MDPV has been reported to substitute for cocaine and methamphetamine (Forster, Taylor, & Gatch, 2012; for similar research in mice, see Fantegrossi, Gannon, Zimmerman, & Rice, 2012). Related work with rats has shown that MDPV is capable of maintaining self-administration up to 5.8 mg/kg/h in adult male Wistar rats and also induces stereotypies at doses higher than 1.5 mg/kg (Aarde, Huang, Creehan, Dickerson, & Taffe, 2013). MDPV has also been shown to induce locomotor activation at low doses while suppressing locomotor activation and inducing stereotypies at high doses, effects similar to those of d-methamphetamine (Huang, Aarde, Angrish, Houseknecht, Dickerson, & Taffe, 2012). These data, in conjunction with anecdotal reports of human abuse, demonstrate that MDPV produces a number of behavioral effects similar to other CNS stimulants.

Although reports on the vulnerability to drugs of abuse commonly focus on reward, there is also another affective property of the drug, i.e., its aversive effect, which has been shown to impact drug taking and may also contribute to abuse vulnerability (Riley, 2011). Specifically, drug use is thought to be a function of the relative balance between its rewarding and aversive effects, with the aversive effects serving to limit drug intake (Riley, 2011). Conditioned taste aversion (CTA) learning is one procedure that has been used to assess the aversive effects of a number of drugs of abuse (Freeman & Riley, 2008), including morphine (Sherman, Pickman, Rice, Liebeskind, & Holman, 1980), cocaine (Goudie, Dickins, & Thornton, 1978), THC (Edwin, 1975) and ethanol (Eckardt, 1976). Notably, MDPV has not been examined for its ability to induce taste aversions, despite the upsurge of use and reports of negative side effects in recent years, as well as the fact that aversions are induced by compounds with similar pharmacological mechanisms (for cocaine see Goudie et al., 1978; for MDMA and *d*-amphetamine see Lin, Atrens, Christie, Jackson, & McGregor, 1993; for methamphetamine see Martin & Ellinwood, 1973). Accordingly, in the present study, MDPV was tested for its ability to induce aversions. This assessment was

made in both adult (Experiment 1) and adolescent (Experiment 2) rats given that for a variety of drugs of abuse aversions are dependent upon age with adolescent animals generally displaying weaker aversions than adults (for reviews, see Doremus-Fitzwater, Varlinskaya, & Spear, 2010; Spear, 2013). Assessing such age differences in aversion learning with MDPV will provide information regarding abuse liability in a population that may be more vulnerable to use and abuse due to a reduction in aversive protectant effects. It is interesting in this context that the recent increase in use of “bath salts” is among teenagers (NDIC, 2011).

In the present series of experiments, body temperature was also assessed to further characterize MDPV's effects in adolescent and adult rats and to determine if temperature changes were related to taste aversion conditioning. Changes in body temperature are common with psychostimulant administration (for MDMA see Dafters & Lynch, 1998; for methamphetamine see Fukumura, Cappon, Pu, Broening, & Vorhees, 1998; for cocaine see Cappon, Morford, & Vorhees, 1998) and have recently been reported for MDPV (see Fantegrossi et. al., 2012). Fantegrossi and colleagues reported that ambient temperature differentially impacted thermoregulatory responses in mice treated with 3, 10 or 30 mg/kg. Specifically, hyperthermic effects outside of the normal circadian range were observed in mice maintained at an ambient temperature of 28°C, but not at a temperature of 22°C (though no significant effects were reported given the large variability of the measure and small group sizes). Interestingly, a more recent study found no temperature effects in adult male Wistar rats following MDPV administration (Aarde et al., 2013), although it is unclear if it induces temperature changes in adolescent animals. Given the reported temperature effects following acute MDPV administration in mice and the suggestion that thermoregulatory effects may underlie aversions induced by ethanol (Cunningham, Niehus, & Bachtold, 2006), the present series of experiments examined taste aversions and temperature changes induced by MDPV administration in adolescent and adult rats.

2. EXPERIMENT 1: ADULTS: METHODS

2.1 Subjects

Thirty-three experimentally naïve male Sprague-Dawley rats (Harlan Laboratories; Indianapolis, IN) arrived at the facility on postnatal day (PND) 21, weighing approximately 40 g. Animals were delivered to the animal facility at this age to permit the control of their environment during maturation (housing conditions, light and dark cycle, etc.). Food and water was available *ad libitum* unless noted otherwise. Procedures recommended by the National Research Council (1996), the Committee on Guidelines for the Care and Use of Animals in Neuroscience and Behavioral Research (2003) and the Institutional Animal Care and Use Committee at American University were followed at all times.

2.2 Apparatus

Upon arrival to the animal colony, subjects were initially handled and then group-housed (3 rats per bin) in polycarbonate bins (23 × 44 × 21 cm) with maple woodchip bedding. All subjects were maintained on a 12:12 light-dark cycle (lights on at 0800h) and at an ambient temperature of 22.5°C ± 1.5. All conditioning and testing occurred during the light phase of

the light-dark cycle. During adaptation and conditioning, animals were transferred to individual hanging wire-mesh (24.3 × 19 × 18 cm) test cages located in another room, but were returned to their group-housing bins afterwards (see details below).

2.3 Drugs and Solutions

3,4-Methylenedioxypyrovalerone hydrochloride (synthesized at the Chemical Biology Research Branch of the National Institute on Drug Abuse) was dissolved in sterile isotonic saline (0.9%) at a concentration of 1 mg/ml and was subsequently filtered through a 0.2 μm filter to remove any contaminants before being administered intraperitoneally (IP) at a dose of 1.0, 1.8 or 3.2 mg/kg. Sterile isotonic saline was also filtered before being administered to vehicle controls equivolume to the highest dose of MDPV administered (3.2 mg/kg). Volume of the injection was manipulated in favor of concentration given the influence concentration has on the absorption/distribution of the drug. Sodium saccharin (0.1%; Sigma) was prepared daily as a 1g/l solution in tap water.

2.4 Procedure

2.4.1 Conditioned Taste Aversion and Temperature Analyses—Phase I: Adaptation. Subjects were brought into the laboratory on PND 21 and were maintained on *ad libitum* food and water until PND 77. On this day, subjects were handled and weighed and temperature probes were implanted. Specifically, the injection site was aseptically cleaned with alcohol and the temperature transponders (Bio Medic Data Systems, Seaford Delaware; Model # IPTT-300) were rapidly inserted subcutaneously into each animal's left flank with a hypodermic needle. For the next 7 days (PND 77-83), *ad libitum* water consumption was recorded and the temperature transponders were checked daily to assess placement by palpating the injection site and for proper function by attempting to record the temperatures. On the following day (PND 84), each animal's available water was reduced to 50% of the previous day's measurement to encourage consumption of water in the individual test cages to take place on the subsequent day. On this day, subjects were removed from their group-housed bins, scanned for body temperature, weighed and placed into individual test cages. Once completed, the test cages were wheeled to the designated testing room where subjects were given 45-min access to tap water in graduated 50-ml Nalgene tubes. After 45 min, the bottles were removed, consumption was recorded and subjects remained in the hanging cages for an additional 20 min before being returned to their group-housed bins and given *ad libitum* water for the next 22.5 h. On the next day, the amount of water available for each bin was again reduced to 50% (as described above) with the exception that individual test cage consumption was also factored into the previous 22.5 h of consumption. On the subsequent day, subjects were again scanned, weighed and placed into the test cages and given 45-min access to tap water before being returned to their group-housed home cages with *ad libitum* water for the next 22.5 h. This twoday cycle (50% on day one, test cage-access followed by *ad libitum* access on day two) was repeated a total of four times at which point consumption was stable in all subjects.

Phase II: Conditioning. Following the final adaptation cycle, animals were given access to water for 1.5 h and bottles were then removed from the bins completely for 21 h before undergoing saccharin conditioning in the test cages. On this conditioning trial, animals'

temperatures were recorded during weighing and handling (1000h) before being placed in the test cages. The initial scan during handling was to ensure that the probe was functioning, and these data were not considered in any statistical analyses. Subjects were then given 45-min access to a novel saccharin solution (1 g/l) in the test cages after which they remained for an additional 20 min. At this point, subjects (independent of their group-housed bin) were assigned to one of four groups such that saccharin intake was comparable among groups. Based on these group assignments, animals were scanned for body temperature and injected with either MDPV (1.0, 1.8 or 3.2 mg/kg IP) or vehicle 20 min following saccharin access. Animals were then immediately returned to their group-housed bins and given *ad libitum* water for the next 22.5 h. This procedure yielded Groups 0 (n = 9), 1.0 (n = 8), 1.8 (n = 8) and 3.2 (n = 8) for which the number indicates the dose of MDPV administered. In addition to the scan prior to drug administration, animals were scanned 30, 60, 90 and 120 min post-injection. For each temperature recording, the probe was scanned twice and the two measurements averaged, with the two measurements never differing by more than 0.9°C. The temperature data were uploaded to a spreadsheet from the Bio Medic Data Systems scanner. On the next day, subjects in each bin were completely water deprived and this two-day cycle (deprivation on day one; saccharin access, injection, temperature scan time-course and 22.5 h ad libitum recovery on day two) was repeated four times.

Phase III: Two-bottle test. On the day following the final conditioning cycle, subjects were again transferred to test cages where two 50-ml Nalgene tubes (one containing tap water; the other containing the 0.1% sodium saccharin solution) were affixed to the cage for 45 min and consumption of both solutions was recorded. Placement of the bottle was counterbalanced across subjects to prevent positioning effects. After the 45 min access, bottles were removed, consumption recorded and animals were left in the cages for an additional 60 min.

2.4.2 Statistical Analyses—Saccharin consumption (ml) on the four conditioning trials was analyzed using a 4 (Dose) × 4 (Trial) mixed model ANOVA. In the presence of significant interactions, one-way ANOVAs with Tukey's HSD *post hocs* were used to assess differences between dose groups on each trial. One-way ANOVAs with Tukey's HSD *post hoc* analyses were employed to evaluate differences in the percent saccharin consumed and total fluid consumed between the different dose groups on the two-bottle test. Statistical analyses of body temperature were based on the mean of two serial scans per animal. A 4 (Dose) × 5 (Days) × 5 (Interval) mixed model ANOVA was used to evaluate differences in temperature between dose groups across intervals and days (adaptation and four conditioning trials). In the presence of significant three-way interactions, a 4 (Dose) × 5 (Interval) ANOVA was used to assess differences in temperature between dose groups across intervals for each day. In the presence of significant interactions, one-way ANOVAs with Tukey's HSD *post hoc* analyses were used to evaluate differences between dose groups at each interval. All statistical analyses were based on significance level of $\alpha = 0.05$ with the exception of the age comparisons for two-bottle test data where $\alpha = 0.0125$ due to Bonferroni corrections.

3. EXPERIMENT 2: ADOLESCENTS: METHODS

All procedures were matched to Experiment 1 with the following exceptions; 33 experimentally naïve animals arrived at the laboratory on PND 21 and had the temperature transponders implanted upon arrival; adaptation began on PND 28; only 2 days of water access in the test cages were employed prior to conditioning that began on PND 32; prior to water and/or saccharin access during adaptation, conditioning, and the two-bottle test, subjects were deprived of water for a full 24 h to ensure drinking. The distribution of animals between drug groups was identical to Experiment 1 (n=8 for all drug groups, n=9 for vehicle) as was the ambient room temperature.

4. RESULTS

4.1 Experiment 1: Adults

4.1.1 Acquisition—The 4 × 4 mixed-model ANOVA on saccharin consumption (ml) during conditioning revealed significant effects of Dose [$F(3, 29) = 14.263$, $p < 0.05$] and Trial [$F(3, 87) = 4.937$, $p < 0.05$] as well as a significant Dose × Trial interaction [$F(9, 87) = 11.850$, $p < 0.05$] (see Figure 1A). A subsequent one-way ANOVA indicated significant differences in consumption between groups on Trials 2 [$F(3, 32) = 9.231$, $p < 0.05$], 3 [$F(3, 32) = 18.748$, $p < 0.05$] and 4 [$F(3, 32) = 25.383$, $p < 0.05$]. Tukey's *post hoc* analysis revealed that on Trials 2 and 3 Groups 1.8 and 3.2 drank significantly less saccharin than Groups 0 and 1.0. By Trial 4, all MDPV-treated subjects drank significantly less saccharin than Group 0, and Groups 1.8 and 3.2 drank significantly less saccharin than Group 1.0.

4.1.2 Two-Bottle Test—A one-way ANOVA on the percent saccharin consumed on the two-bottle test revealed significant differences among groups [$F(3, 32) = 42.607$, $p < 0.05$] (see Figure 1B). Specifically, all MDPV-treated subjects drank a significantly lower percent of saccharin than Group 0, and Groups 1.8 and 3.2 drank a significantly lower percent of saccharin than Group 1.0. Additionally, a one-way ANOVA on total fluid consumed (data not shown) revealed significant differences among groups [$F(3, 32) = 3.675$, $p < 0.05$] such that Group 3.2 drank significantly less than Group 0.

4.1.3 Temperature Assessment—The 4 × 5 × 5 mixed model ANOVA on body temperature revealed significant effects of Day [$F(4, 580) = 19.765$, $p < 0.05$], Dose [$F(3, 145) = 16.542$, $p < 0.05$] and Interval [$F(4, 145) = 30.856$, $p < 0.05$] as well as significant Day × Dose [$F(12, 580) = 6.241$, $p < 0.05$], Day × Interval [$F(16, 580) = 7.10$, $p < 0.05$], Dose × Interval [$F(12, 145) = 2.485$, $p < 0.05$] and Day × Dose × Interval [$F(48, 580) = 1.417$, $p < 0.05$] interactions.

In relation to the significant three-way interaction, a 4 × 5 mixed model ANOVA on the last day of water adaptation indicated a significant effect of Interval [$F(4, 116) = 28.134$, $p < 0.05$] (data not shown). On the initial conditioning trial (Figure 2A), there was a significant effect of Interval [$F(4, 116) = 15.161$, $p < 0.05$]. On the second conditioning trial (Figure 2B), there was significant main effect of Interval [$F(4, 116) = 33.083$, $p < 0.05$] and Dose [$F(3, 29) = 6.229$, $p < 0.05$] as well as a significant Interval × Dose interaction [$F(12, 116) = 4.040$, $p < 0.05$]. A one-way ANOVA on Interval with Tukey's *post hoc* analyses revealed

that Groups 1.0 and 1.8 had higher temperatures than Group 0 at 60 and 90 min post-injection, with the additional effect that all drug-treated groups had higher body temperatures than Group 0 at 120 min post-injection. On the third conditioning trial (Figure 2C), there were significant main effects of Interval [$F(4, 116) = 38.791$, $p < 0.05$] and Dose [$F(3, 29) = 6.286$, $p < 0.05$] as well as a significant Interval × Dose interaction [$F(12, 116) = 4.455$, $p < 0.05$]. A one-way ANOVA on Interval with Tukey's *post hoc* analyses revealed that Groups 1.0 and 1.8 had higher temperatures than Group 0, with Group 1.0 exhibiting higher temperatures compared to Group 3.2 at 90 min post-injection. At 120 min post-injection, all drug-treated groups had higher body temperatures than Group 0. On the final trial (Figure 2D), there were significant main effects of Interval [$F(4, 116) = 32.233$, $p < 0.05$] and Dose [$F(3, 29) = 6.951$, $p < 0.05$] as well as a significant Interval × Dose interaction [$F(12, 116) = 4.203$, $p < 0.05$]. A one-way ANOVA on Interval with Tukey's *post hoc* analyses revealed that Groups 1.0 and 1.8 exhibited higher temperatures than Group 0 at 60 min, while at 90 and 120 min post-injection all drug-treated groups had increased body temperatures relative to Group 0.

4.2 Experiment 2: Adolescents

4.2.1 Acquisition—The 4 × 4 mixed model ANOVA on saccharin consumption (ml) during conditioning revealed significant effects of Dose [$F(3, 29) = 7.160$, $p < 0.05$] and Trial [$F(3, 87) = 11.216$, $p < 0.05$] as well as a significant Dose × Trial interaction [$F(9, 87) = 3.514$, $p < 0.05$] (Figure 3A). A subsequent one-way ANOVA indicated significant differences in consumption between the groups on Trials 3 [$F(3, 32) = 10.202$, $p < 0.05$] and 4 [$F(3, 32) = 6.794$, $p < 0.05$]. Tukey's *post hoc* analysis revealed that on Trials 3 and 4 all MDPV-treated groups differed from vehicle.

4.2.2 Two-Bottle Test—A one-way ANOVA on the percent saccharin consumed on the two-bottle test revealed significant differences among groups [$F(3, 32) = 9.432$, $p < 0.05$] (Figure 3B). Specifically, Groups 1.8 and 3.2 drank a significantly lower percentage of saccharin than Group 0 with Group 1.8 drinking less than Group 1.0. Additionally, a one-way ANOVA on total fluid consumed revealed significant differences among groups [$F(3, 32) = 4.208$, $p < 0.05$] with Groups 1.0 and 3.2 drinking significantly less than Group 0.

4.2.3 Temperature Assessment—The temperature probe of one subject in Group 0 failed to function after the first conditioning day. All data from this one subject were removed from temperature assessments leaving an $n = 8$. The 4 × 5 × 5 mixed model ANOVA on body temperature yielded significant effects of Day [$F(4, 560) = 6.367$, $p < 0.05$], Dose [$F(3, 140) = 11.351$, $p < 0.05$] and Interval [$F(4, 140) = 12.712$, $p < 0.05$] as well as significant Day × Interval [$F(16, 560) = 11.962$, $p < 0.05$], Dose × Interval [$F(12, 140) = 5.753$, $p < 0.05$] and Day × Dose × Interval [$F(48, 560) = 1.774$, $p < 0.05$] interactions.

The 4 × 5 ANOVA on the last day of water adaptation indicated a significant effect of Interval [$F(4, 112) = 75.350$, $p < 0.05$] (data not shown). On the initial conditioning trial (Figure 4A), there was a significant effect of Interval [$F(4, 112) = 12.226$, $p < 0.05$] as well as a significant Interval × Dose interaction [$F(12, 112) = 6.843$, $p < 0.05$]. A one-way

ANOVA on Interval with Tukey's *post hoc* analyses revealed that Group 1.0 exhibited higher body temperatures than Group 0 at the 0 min interval. At 30 min post-injection, Groups 1.8 and 3.2 had significantly lower temperatures than Group 0 with the added effect that Group 1.8 exhibited a significantly lower temperature than Group 1.0. At 60 min post-injection, Group 3.2 displayed lower temperatures than Group 1.0. On the second conditioning trial (Figure 4B), there was a significant effect of Interval [$F(4, 112) = 8.311$, $p < 0.05$] as well as a significant Interval × Dose interaction [$F(12, 112) = 7.421$, $p < 0.05$]. A one-way ANOVA on Interval with Tukey's *post hoc* analyses revealed that at the 0 min interval, Group 1.0 exhibited higher body temperatures than Group 0. At 30 min post-injection, Group 1.8 had a significantly lower temperature than Group 0. At 60 min post-injection, Groups 1.8 and 3.2 had significantly lower body temperatures compared to Group 1.0. At 90 min post-injection, Groups 1.0 and 3.2 had significantly higher body temperatures than Group 0. Finally, at 120 min post-injection, Group 3.2 exhibited significantly higher temperatures compared to Group 0. On the third conditioning trial (Figure 4C), there were significant effects of Interval [$F(4, 112) = 13.912$, $p < 0.05$] and Dose [$F(3, 28) = 3.217$, $p < 0.05$] as well as a significant Interval × Dose interaction [$F(12, 112) = 5.588$, $p < 0.05$]. A one-way ANOVA on Interval with Tukey's *post hoc* analyses revealed that at the 30 min interval, Groups 1.8 and 3.2 had significantly lower temperatures than Group 0 with the added effect that Group 1.8 exhibited a significantly lower temperature than Group 1.0. At 60 min post-injection Group 3.2 had significantly lower body temperatures compared to group 1.0, while at 120 min post-injection Group 3.2 had significantly higher body temperatures compared to Group 0. Additionally, at 90 min post-injection Groups 1.0 and 3.2 had significantly higher temperatures than Group 0. On the final conditioning trial, there was a significant effect of Interval [$F(4, 112) = 10.718$, $p < 0.05$] as well as a significant Interval × Dose interaction [$F(12, 112) = 3.009$, $p < 0.05$]. A one-way ANOVA on Interval with Tukey's *post hoc* analyses indicated that at 30 and 60 min post-injection Group 3.2 exhibited significantly lower temperatures compared to Groups 0 and 1.0.

5. AGE COMPARISONS

Given that the adolescent and adult experiments were not run concurrently, the following age-difference analyses are exploratory in nature. Direct age comparisons were made on the acquisition of MDPV-induced taste aversions, but direct comparisons on body temperature were not performed given that the effects between adults and adolescents were in opposite directions (i.e., increased and decreased, respectively).

5.1 CTA Comparisons

Consumption for the drug-treated groups was transformed to a percent of the average consumption of Group 0 for each age group across each of the conditioning trials. For each trial, consumption in each age and dose group was calculated as a percent of the average absolute consumption of the vehicle-treated controls (Group 0) on that session. A 2 (Age) × 3 (Dose) × 4 (Trial) mixed-model ANOVA revealed significant main effects of Age [$F(1, 42) = 27.055$, $p < 0.05$], Dose [$F(2, 42) = 6.683$. $p < 0.05$] and Trial [$F(3, 126) = 70.582$, $p < 0.05$] as well as significant Trial × Age [$F(3, 126) = 10.923$, $p < 0.05$], Trial × Dose [$F(6, 126) = 3.885$, $p < 0.05$], Age × Dose [$F(2, 42) = 5.856$, $p < 0.05$] and a Trial × Age × Dose

[F (6, 126) = 3.035, $p < 0.05$] interactions. Subsequent one-way ANOVAs used to compare Age × Dose differences across conditioning trials revealed significant differences on Trials 2-4 with Tukey's *post hoc* analyses indicating that adolescents consumed a significantly higher percentage of saccharin than their adult counterparts at the doses of 1.8 and 3.2 mg/kg, reflective of the acquisition of weaker aversions in the adolescents at the two highest doses tested.

Additionally, Bonferroni-corrected independent sample t-tests (where $\alpha = 0.0125$) used to examine age differences in saccharin preference on the two-bottle test revealed that adolescents consumed a significantly higher percentage of saccharin relative to adults at 1.0 mg/kg [t (14) = 3.459, $p < 0.0125$], 1.8 mg/kg [t (14) = 3.955, $p < 0.0125$] and 3.2 mg/kg [t (14) = 2.906, $p < 0.0125$] with no differences at 0 mg/kg [t (14) = 4.909, $p > 0.0125$].

6. DISCUSSION

The present experiments sought to assess whether MDPV is capable of conditioning taste aversions and if such effects differ between adolescent and adult rats. As described, MDPV did induce taste aversions in both age groups, with adolescent subjects acquiring the aversions at a slower rate and to a lesser degree than their adult counterparts. Body temperatures revealed dose-dependent changes that were also age-dependent. Specifically, adults exhibited increased body temperatures while adolescents exhibited decreases compared to their own controls following acute exposure to MDPV, an effect not related to ambient temperature.

As with many drugs of abuse, aversions induced by MDPV were rapidly acquired (within several conditioning trials) and dose-dependent (for reviews, see Gamzu, Vincent, & Boff, 1985; Hunt & Amit, 1987; Verendeev & Riley, 2012), suggesting that, as indexed by the CTA procedure, MDPV has aversive effects. Like many other drugs of abuse, MDPV also has rewarding effects as measured in SA and ICSS designs. For example, rats self-administer MDPV (0.05, 0.1 and 0.2 mg/kg/infusion) and comparable doses of MDPV also lower ICSS thresholds (Aarde et al., 2013; Watterson et al., 2012). Interestingly, MDPV (at a dose effective in inducing aversions in the present assessment, i.e., 1 mg/kg), substitutes for cocaine and methamphetamine in a drug discrimination procedure (Forster et al., 2012). Additionally, methamphetamine and MDMA substitute with greater than 80% drug-appropriate responding in adult mice trained to discriminate 0.3 mg/kg MDPV from saline, (Fantegrossi et al., 2012), and MDPV produces increases in locomotor behavior similar to methamphetamine in rats (Huang et al., 2012). These reports suggest that MDPV has similar behavioral effects to that of other psychostimulants.

Given that drug use and abuse is a function of the relative balance of reward and aversion, characterizing these properties may be important to understanding their relative balance and MDPV's abuse potential. In this context, it is interesting that while MDPV induced aversions, adolescent subjects displayed significantly weaker aversions than adults. These results parallel a growing literature showing similar age-dependent aversive effects of a wide variety of drugs of abuse (for reviews, see Doremus-Fitzwater et al., 2010; Spear, 2013). In such assessments, taste aversions are generally induced at lower doses and are

acquired faster and to a greater degree in adult rats compared to their adolescent counterparts. Although little is known about the age differences in the rewarding effects of MDPV, the fact that it is less aversive in adolescent subjects suggests that this population may be vulnerable to its use and abuse.

Although the age differences in MDPV-induced aversions are clear, the basis for these differences remains unknown. It is possible that age-dependent differences in taste processing, learning and retention and stress reactivity could account for the differences in MDPV-induced aversions, although these factors have been shown to have little contribution (if any) in other assessments of aversion learning between adolescent and adults (see Anderson, Varlinskaya, & Spear, 2010 and Hurwitz, Merluzzi, & Riley, 2013 for a discussion of these issues). The fact that adolescents exhibit weaker taste aversions to a host of drugs of abuse may indicate some blunted sensitivity to drugs in general. However, studies using lithium chloride as the conditioning agent generally do not reveal age-dependent differences (Balcom, Coleman, & Norman, 1981; Guanowsky, Misanin, & Riccio, 1983; Klein, Mikulka, Domato, & Hallstead, 1977; Misanin, Guanowsky, & Riccio, 1983; Valliere, Misanin, & Hinderliter, 1988), indicating that adolescents and adults are comparably sensitive to the aversive effects of this compound. Further, a blunted response in learning the conditioning procedure in general also appears unlikely. Several reports have shown that adolescents acquire place preferences to several drugs of abuse more rapidly and at lower doses compared to their adult counterparts (Belluzzi, Lee, Oliff, & Leslie, 2004; Brielmaier, McDonald, & Smith, 2007; Vastola, Douglas, Varlinskaya, & Spear, 2002; though see Campbell, Wood, & Spear, 2000 for a report of no differences with morphine and cocaine), indicating that the age differences observed are procedurally-dependent and not indicative of a general blunted sensitivity in adolescents.

It is also possible that the adolescents and adults in the present experiment were differentially motivated to drink due to the effects of deprivation on developing animals. For example, adolescent and adult animals given access to fluid for only 20 min per day develop a significant difference in body weight loss (52% and 80% free-feeding weight, respectively), possibly causing greater motivation to drink saccharin in adolescent subjects (see Hurwitz et al., 2013). However, in a minimal deprivation procedure similar to the one used in the present experiment, adolescent and adult subjects weighed 95% and 94% of free-feeding, respectively, and exhibited similar results – that is, adolescents still acquired weaker aversions than their adult counterparts and did so on later trials. Further, in the present experiments, adolescents and adults in the vehicle-treated group both increased consumption of saccharin over trials and showed a similar preference for saccharin on the two-bottle test, indicating that for non-drug treated animals, the motivation to drink saccharin in the test cages was similar.

Adolescents and adults also differed in the present assessments with respect to acclimatization in the animal facility (adolescents only had one week to adapt to the animal facility, adults had eight weeks), possibly confounding the taste aversion results. The adult rats were delivered to the facility on PND 21 to allow for the control of their developmental environment (e.g. housing conditions, light/dark cycle), whereas the adolescents were water deprived within one week of arriving at the facility. Though this difference in

acclimatization may have caused increased stress in the adolescent subjects, this is unlikely to account for the observed differences in taste aversion results. Studies assessing the effects of stress on taste aversion learning are mixed, with most illustrating that stress has no effect (Anderson, Hinderliter, & Misanin, 2006; Bourne, Calton, Gustavson, & Schachtman, 1992; Bowers, Amit, & Gringras, 1996; Misanin, Kaufhold, Paul, Hinderliter, & Anderson, 2006; Revusky & Reilly, 1989). The one study to test stress as a variable in adolescents and adults (isolation housing, restraint) found no evidence that stress induces age-dependent differences in ethanol-induced aversion learning (Anderson et al., 2010). The present data, along with the aforementioned reports on adolescent/adult CTA comparisons, implicate instead some developmental phenomenon that generalizes across many drugs and possibly reflects an overall insensitivity to the aversive properties of drugs for adolescents.

One possibility for this general insensitivity may be that adolescents and adults differ in their pharmacokinetic response to MDPV, and as such do not have comparable blood/brain levels following administration. In support of this possibility, adolescent mice have been reported to metabolize cocaine faster than their adult counterparts (McCarthy, Mannelli, Niculescu, Gingrich, Unterwald, & Ehrlich, 2004). This may also be true of MDPV in rats, although no research to date has investigated this possibility. Even so, unpublished data from our laboratory has shown that equal levels of morphine reach the brain in adolescent and adult LEW and F344 rats with similar taste aversion results (i.e. adolescents develop weaker taste aversions than adults in both strains). Therefore, while it is certainly possible adolescents and adults in the present experiment metabolized MDPV at different rates, it remains unknown how potential differences may relate to age-dependent differences in MDPV-induced taste aversions. At present, the most parsimonious explanation for age-dependent differences in aversion learning is that there is a difference in the aversive effects of MDPV in adolescents and adults.

A major difficulty with such a conclusion is the fact that little is known about the specific nature of the aversive effects of drugs in general, much less that of MDPV, which has only recently been investigated for its affective properties (Fantegrossi et al., 2012; Forster et al., 2012; Watterson et al., 2012; Aarde et al., 2013). A number of drug effects have been proposed as important in inducing aversions, including sickness, novelty, disruption of homeostasis and even reward (for recent reviews see Riley, 2011 and Verendeev & Riley, 2012). Although much has been speculated regarding the aversive properties of a host of compounds, including drugs of abuse, there is no consensus. One suggested mechanism proposed for ethanol-induced aversions is hypothermia (see Cunningham et al., 2006). For example, Cunningham and his colleagues have reported that rats that exhibit the strongest alcohol-induced aversions are those which have the greatest alcohol-induced decreases in core body temperature. Given that MDPV has been reported to affect body temperature in mice (Fantegrossi et al., 2012; though see Aarde et al., 2013 for no MDPV-induced temperature effects in rats), the present experiment assessed if MDPV induced temperature changes in adolescents and adults, if these changes differed in the two age groups and if these changes might be related to aversion learning during conditioning. In the present study, MDPV induced age-dependent changes in temperature. As described, adolescents generally decreased body temperature following MDPV. Interestingly, adolescent subjects at the lowest dose of MDPV (1 mg/kg) displayed elevated body temperature relative to control

subjects immediately prior to the injection on the first two conditioning trials but at no other time point for the remainder of conditioning. The basis for this difference is not clear, although it likely reflects individual variation unrelated to the drug, given that it was evident prior to drug exposure and because higher doses generally decreased body temperature in adolescent subjects. Overall, adolescents exhibited body temperature decreases and adults exhibited increases, effects that were unrelated to ambient temperature and independent of the degree of aversions acquired in either age cohort. The fact that both adolescents and adults acquired aversions (albeit to different degrees), yet differed directionally in terms of body temperature, argues that if temperature changes are related to MDPV-induced aversions this relationship is complicated and age-dependent. Importantly, on the final conditioning day, Pearson's correlations revealed no significant relationship between saccharin consumption and body temperature for either adults or adolescents at any time point post drug injection (all ps > .05). It is certainly possible that under different ambient temperatures, MDPV may have induced aversions that were more directly related to its effects on body temperature. As noted by Fantegrossi and colleagues (2012), ambient temperature has been shown to influence the degree of hyperthermia found in adult mice. Specifically, adult mice treated with 3, 10 and 30 mg/kg MDPV exhibited hyperthermia in a warm (28°C) but not cool (22°C) environment. Future research should assess how these two modulating factors interact by examining MDPV's thermoregulatory effects in adolescent and adult rats at varying ambient temperatures and establish if these effects are associated with strength of aversion conditioning.

The present experiments sought to determine whether MDPV, the primary constituent of “bath salts,” could induce conditioned taste aversions and whether these aversions varied by age. Given that the balance between the rewarding and aversive effects of a drug is thought to influence its abuse liability, the fact that adolescents exhibited weaker MDPV-induced aversions relative to adults suggests this population may be more vulnerable to its use and abuse (see Infurna & Spear, 1979; Schramm-Sapyta, Morris, & Kuhn, 2006; Schramm-Sapyta, Cha, Chaudhry, Wilson, Swartzwelder, & Kuhn, 2007; Anderson et al., 2010; Vetter-O'Hagen, Varlinskaya, & Spear, 2009; Shram, Funk, Li, & Lê, 2006 and Hurwitz et al., 2013 for similar findings with other drugs of abuse). This is especially concerning because MDPV use among teenagers has increased in recent years (NDIC, 2011). Although the age dependency in MDPV-induced aversions is clear, the specific mechanism underlying these effects remains unknown. It is increasingly important to investigate both the physiological and neurochemical mechanisms underlying reward and aversion to better understand use and abuse of MDPV and other drugs.

Acknowledgments

This research was supported by a grant from the Mellon Foundation to ALR, an American University Artists and Scholars Fellowship to APM and a Dean's Undergraduate Research Award to APM. A portion of this work was supported by the Intramural Research Programs of National Institute on Drug Abuse and National Institute on Alcohol Abuse and Alcoholism.

REFERENCES

Aarde SM, Huang PK, Creehan KM, Dickerson TJ, Taffe MA. The novel recreational drug 3, 4-methylenedioxypyrovalerone (MDPV) is a potent psychomotor stimulant: self-administration and locomotor activity in rats. Neuropharmacology. 2013; 71:130–140. [PubMed: 23597511]

Airuehia EW, Walker LY, Nittler J. A review of “bath salts”: Evolving designer drugs of abuse. Clinical Toxicology. 2012; 2(4)

Anderson MJ, Hinderliter CF, Misanin JR. The effects of chronic water deprivation on metabolic rate and long-trace taste-aversion conditioning in rats. Neurobiology of Learning and Memory. 2006; 85(3):199–205. [PubMed: 16321549]

Anderson RI, Varlinskaya EI, Spear LP. Ethanol-induced conditioned taste aversion in male Sprague-Dawley rats: Impact of age and stress. Alcoholism: Clinical and Experimental Research. 2010; 34(12):2106–2115.

Balcom FW, Coleman WR, Norman JL. Taste aversion learning and long-term retention in juvenile rats. Psychological Reports. 1981; 49(1):266. [PubMed: 7291421]

Baumann MH, Partilla JS, Lehner KR, Thorndike EB, Hoffman AF, Holy M, Rothman RB, Goldberg SR, Lupica CR, Sitte HH. Powerful cocaine-like actions of 3, 4-methylenedioxypyrovalerone (MDPV), a principal constituent of psychoactive ‘bath salts’ products. Neuropsychopharmacology. 2012; 38:552–562. [PubMed: 23072836]

Belluzzi JD, Lee AG, Oliff HS, Leslie FM. Age-dependent effects of nicotine on locomotor activity and conditioned place preference in rats. Psychopharmacology. 2004; 174(3):389–395. [PubMed: 14740150]

Bourne MJ, Calton JL, Gustavson KK, Schachtman TR. Effects of acute swim stress on LiCl- induced conditioned taste aversions. Physiology & Behavior. 1992; 51(6):1227–1234. [PubMed: 1322542]

Bowers W, Amit Z, Gringras M. Time-dependent exacerbation of amphetamine-induced taste aversions following exposure to footshock. Psychopharmacology. 1996; 125(1):43–49. [PubMed: 8724447]

Brielmaier JM, McDonald CG, Smith RF. Immediate and long-term behavioral effects of a single nicotine injection in adolescent and adult rats. Neurotoxicology and Teratology. 2007; 29(1):74–80. [PubMed: 17095188]

Bronstein AC, Spyker DA, Cantilena LR, Green JL, Rumack BH, Dart RC. 2010 annual report of the American Association of Control Centers' National Poison Data System (NPDS): 28th annual report. Clinical Toxicology. 2011; 49:910–941. [PubMed: 22165864]

Campbell JO, Wood RD, Spear LP. Cocaine and morphine-induced place conditioning in adolescent and adult rats. Physiology & Behavior. 2000; 68(4):487–493. [PubMed: 10713288]

Cappon GD, Morford LL, Vorhees CV. Enhancement of cocaine-induced hyperthermia fails to elicit neurotoxicity. Neurotoxicology and Teratology. 1998; 20(5):531–535. [PubMed: 9761591]

Cunningham CL, Niehus JS, Bachtold JF. Ambient temperature effects on taste aversion conditioned by ethanol: Contribution of ethanol-induced hypothermia. Alcoholism: Clinical and Experimental Research. 2006; 16(6):1117–1124.

Dafters RI, Lynch E. Persistent loss of thermoregulation in the rat induced by 3, 4-methylenedioxymethamphetamine (MDMA or “Ecstasy”) but not by fenfluramine. Psychopharmacology. 1998; 138(2):207–212. [PubMed: 9718291]

Doremus-Fitzwater TL, Varlinskaya EI, Spear LP. Motivational systems in adolescence: possible implications for age differences in substance abuse and other risk-taking behaviors. Brain and Cognition. 2010; 72(1):114–123. [PubMed: 19762139]

Eckardt MJ. Alcohol-induced conditioned taste aversion in rats; Effect of concentration and prior exposure to alcohol. Journal of Studies on Alcohol and Drugs. 1976; 37(3):334.

Edwin JK. Aversive effects of repeated injections of THC in rats. Psychological Reports. 1975; 37(3): 1051–1054. [PubMed: 1208721]

Fantegrossi WE, Gannon BM, Zimmerman SM, Rice KC. In vivo effects of abused ‘bath salt’ constituent 3, 4-methylenedioxypyrovalerone (MDPV) in mice: Drug discrimination, thermoregulation, and locomotor activity. Neuropsychopharmacology. 2012; 38:563–573. [PubMed: 23212455]

Forster, MJ.; Taylor, C.; Gatch, MB. “Bath salts”: Discriminative stimulus effects of several cathinone compounds.. Poster presented at the 74th annual meeting of the College on Problems of Drug Dependence (CPDD) in Palm Springs; CA. 2012.

Freeman, KB.; Riley, AL. The origins of conditioned taste aversion learning: A historical analysis.. In: Reilly, S.; Schachtman, TR., editors. Conditioned Taste Aversion: Neural and Behavioral Processes. Oxford University Press; New York: 2008. p. 9-33.

Fukumura M, Cappon GD, Pu C, Broening HW, Vorhees CV. A single dose model of methamphetamine-induced neurotoxicity in rats: Effects on neostriatal monoamines and glial fibrillary acidic protein. Brain Research. 1998; 806(1):1–7. [PubMed: 9739098]

Gamzu E, Vincent G, Boff E. A pharmacological perspective of drugs used in establishing conditioned food aversions. Annals of the New York Academy of Sciences. 1985; 443(1):231–249. [PubMed: 2861773]

Goudie A, Dickins D, Thornton E. Cocaine-induced conditioned taste aversions in rats. Pharmacology Biochemistry and Behavior. 1978; 8(6):757–761.

Guanowsky V, Misanin JR, Riccio DC. Retention of conditioned taste aversion in weanling, adult, and old-age rats. Behavioral and Neural Biology. 1983; 37(1):173–178. [PubMed: 6309133]

Huang PK, Aarde SM, Angrish D, Houseknecht KL, Dickerson TJ, Taffe MA. Contrasting effects of d-methamphetamine, 3,4-methylenedioxymethamphetamine, 3,4-methylenedioxypyrovalerone, and 4-methylmethcathinone on wheel activity in rats. Drug and Alcohol Dependence. 2012; 126(1-2):168–175. [PubMed: 22664136]

Hunt T, Amit Z. Conditioned taste aversion induced by self-administered drugs: Paradox revisited. Neuroscience & Biobehavioral Reviews. 1987; 11(1):107–130. [PubMed: 3554039]

Hurwitz ZE, Merluzzi AP, Riley AL. Age-dependent differences in morphine-induced taste aversions. Developmental Psychobiology. 2013; 55(4):415–428. [PubMed: 22585679]

Infurna RN, Spear LP. Developmental changes in amphetamine-induced taste aversions. Pharmacology Biochemistry and Behavior. 1979; 11(1):31–35.

Klein SB, Mikulka PJ, Domato GC, Hallstead C. Retention of internal experiences in juvenile and adult rats. Physiological Psychology. 1977; 5:63–66.

Lin HQ, Atrens DM, Christie MJ, Jackson DM, McGregor IS. Comparison of conditioned taste aversions produced by MDMA and d-amphetamine. Pharmacology Biochemistry and Behavior. 1993; 46(1):153–156.

Martin JC, Ellinwood EH. Conditioned aversion to a preferred solution following methamphetamine injections. Psychopharmacology. 1973; 29(3):253–261.

McCarthy LE, Mannelli P, Niculescu M, Gingrich K, Unterwald EM, Ehrlich ME. The distribution of cocaine in mice differs by age and strain. Neurotoxicology and Teratology. 2004; 26(6):839–848. [PubMed: 15451047]

McClean JM, Anspikian A, Tsuang JW. Bath salt use: A case report and review of the literature. Journal of Dual Diagnosis. 2012; 8(3):250–256.

Misanin JR, Guanowsky V, Riccio DC. The effect of CS-preexposure on conditioned taste aversion in young and adult rats. Physiology & Behavior. 1983; 30(6):859–862. [PubMed: 6310659]

Misanin JR, Kaufhold SE, Paul RL, Hinderliter CF, Anderson MJ. A time contraction effect of acute tail-pinch stress on the associative learning of rats. Behavioural Processes. 2006; 71(1):16–20. [PubMed: 16253440]

Mugele J, Nañagas KA, Tormoehlen LM. Serotonin syndrome associated with MDPV use: A case report. Annals of Emergency Medicine. 2012; 60(1):100–102. [PubMed: 22237165]

N.D.I.C.. Synthetic cathinones (bath salts): An emerging domestic threat. Department of Justice. , editor. Special Projects Unit, National Threat Analysis Branch; Johnstown, PA: 2011.

Olives TD, Orozco BS, Stellpflug SJ. Bath salts: The ivory wave of trouble. Western Journal of Emergency Medicine. 2012; 13(1):58–62. [PubMed: 22461923]

Penders TM. How to recognize a patient who's high on “bath salts”. The Journal of Family Practice. 2012; 61(4):210–212. [PubMed: 22482104]

Revusky S, Reilly S. Attenuation of conditioned taste aversions by external stressors. Pharmacology Biochemistry and Behavior. 1989; 33(1):219–226.

Riley A. The paradox of drug taking: The role of the aversive effects of drugs. Physiology & Behavior. 2011; 103(1):69–78. [PubMed: 21118698]

Ross EA, Reisfield GM, Watson MC, Chronister CW, Goldberger BA. Psychoactive “bath salts” intoxication with methylenedioxypyrovalerone. The American Journal of Medicine. 2012; 125(9): 854–858. [PubMed: 22682791]

Schramm-Sapyta NL, Cha YM, Chaudhry S, Wilson WA, Swartzwelder HS, Kuhn CM. Differential anxiogenic, aversive, and locomotor effects of THC in adolescent and adult rats. Psychopharmacology. 2007; 191(4):867–877. [PubMed: 17211649]

Schramm-Sapyta NL, Morris RW, Kuhn CM. Adolescent rats are protected from the conditioned aversive properties of cocaine and lithium chloride. Pharmacology Biochemistry and Behavior. 2006; 84(2):344–352.

Sherman JE, Pickman C, Rice A, Liebeskind JC, Holman EW. Rewarding and aversive effects of morphine: Temporal and pharmacological properties. Pharmacology Biochemistry and Behavior. 1980; 13(4):501–505.

Shram MJ, Funk D, Li Z, Lê AD. Periadolescent and adult rats respond differently in tests measuring the rewarding and aversive effects of nicotine. Psychopharmacology. 2006; 186(2):201–208. [PubMed: 16586088]

Spear LP. Adolescent neurodevelopment. Journal of Adolescent Health. 2013; 52(2):S7–S13. [PubMed: 23332574]

United States Drug Enforcement Agency (USDEA). Background, data and analysis of synthetic cathinones: Mephedrone (4-MMC), methylone (MDMC) and 3,4-methylenedioxypyrovalerone (MDPV). Department of Justice. , editor. Washington, DC: 2011.

Valliere WA, Misanin JR, Hinderliter CF. Age-related differences in the US preexposure effect on conditioned taste aversion in rats. Bulletin of the Psychonomic Society. 1988; 26(1):64–66.

Vastola BJ, Douglas LA, Varlinskaya EI, Spear LP. Nicotine-induced conditioned place preference in adolescent and adult rats. Physiology & Behavior. 2002; 77(1):107–114. [PubMed: 12213508]

Verendeev A, Riley AL. Conditioned taste aversion and drugs of abuse: History and interpretation. Neuroscience and Biobehavioral Reviews. 2012; 36(10):2193–2205. [PubMed: 22921283]

Vetter-O'Hagen C, Varlinskaya E, Spear L. Sex differences in ethanol intake and sensitivity to aversive effects during adolescence and adulthood. Alcohol and Alcoholism. 2009; 44(6):547–554. [PubMed: 19767625]

Watterson LR, Kufahl PR, Nemirovsky NE, Sewalia K, Grabenauer M, Thomas BF, Marusich JA, Wegner S, Olive MF. Potent rewarding and reinforcing effects of the synthetic cathinone 3, 4-methylenedioxypyrovalerone (MDPV). Addiction Biology. 2012 doi:10.1111/j.1369-1600.2012.00474.x.

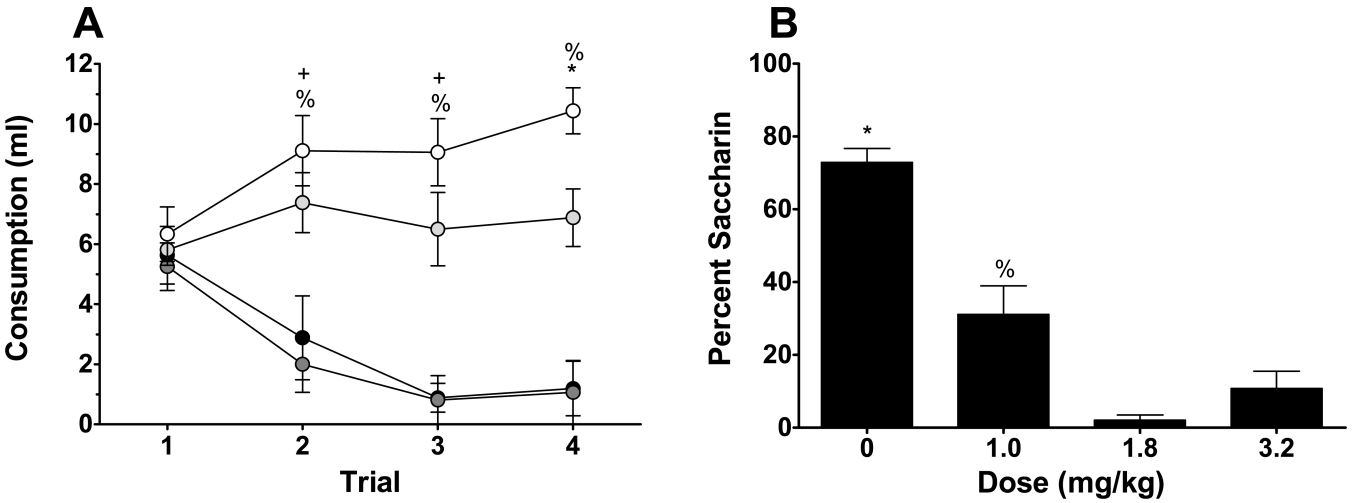

Figure 1.

Mean (± SEM) saccharin consumption (ml) by adults during acquisition (A) and mean (± SEM) percent saccharin consumed on the two-bottle test (B). $^{+}$denotes a significant difference between Group 0 and Groups 1.8 and 3.2. $^{\%}$denotes a significant difference between Group 1.0 and Groups 1.8 and 3.2. *denotes a significant difference between Group 0 and all drug treated-groups.

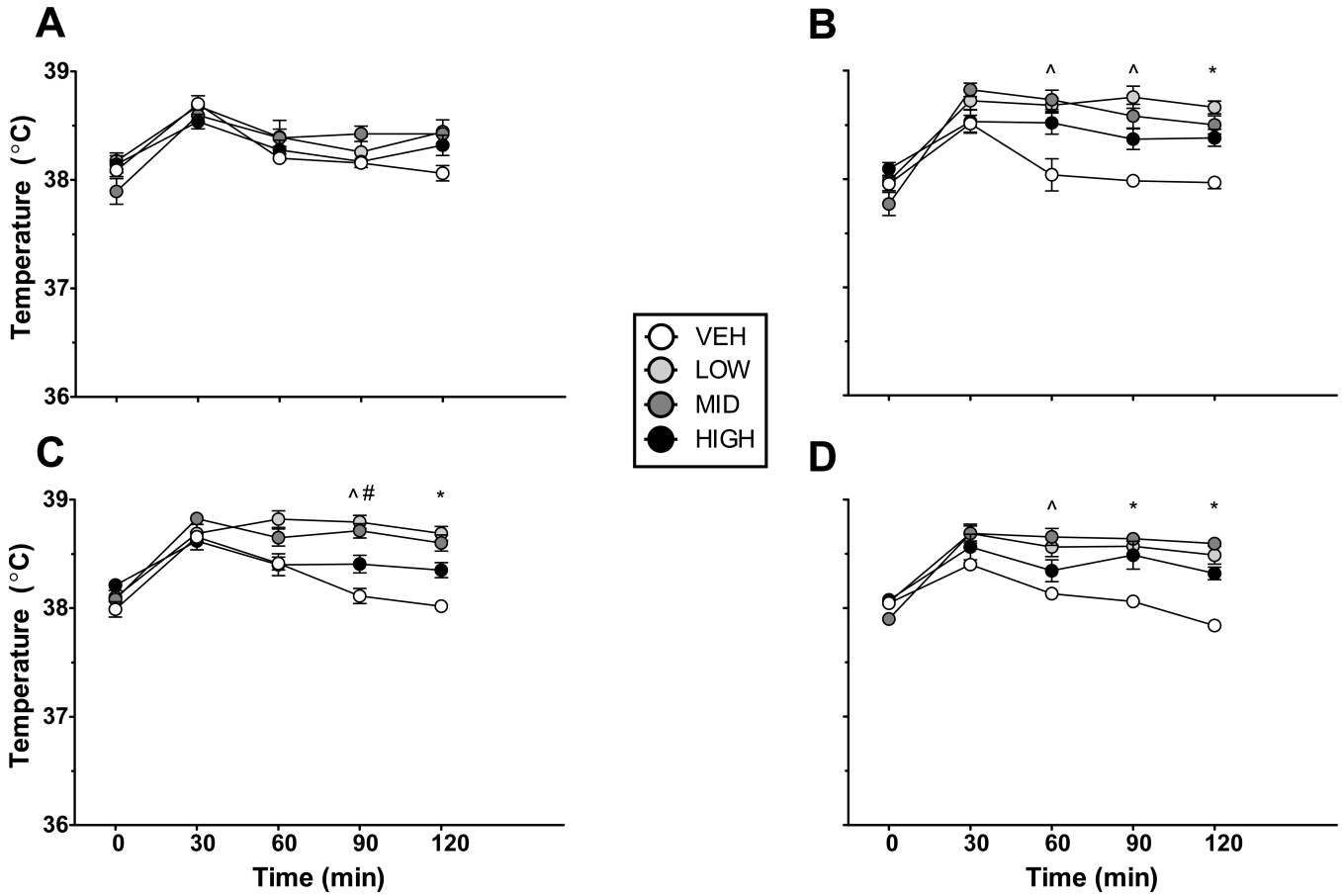

Figure 2.

Mean (± SEM) body temperature (°C) of adults across the five intervals (0, 30, 60, 90, 120) over the four conditioning trials (Panels A-D). ^denotes a significant difference between Groups 1.0 and 1.8 and Group 0. *denotes a significant difference between all drug-treated groups and Group 0. #denotes a significant difference between Groups 1.0 and 3.2.

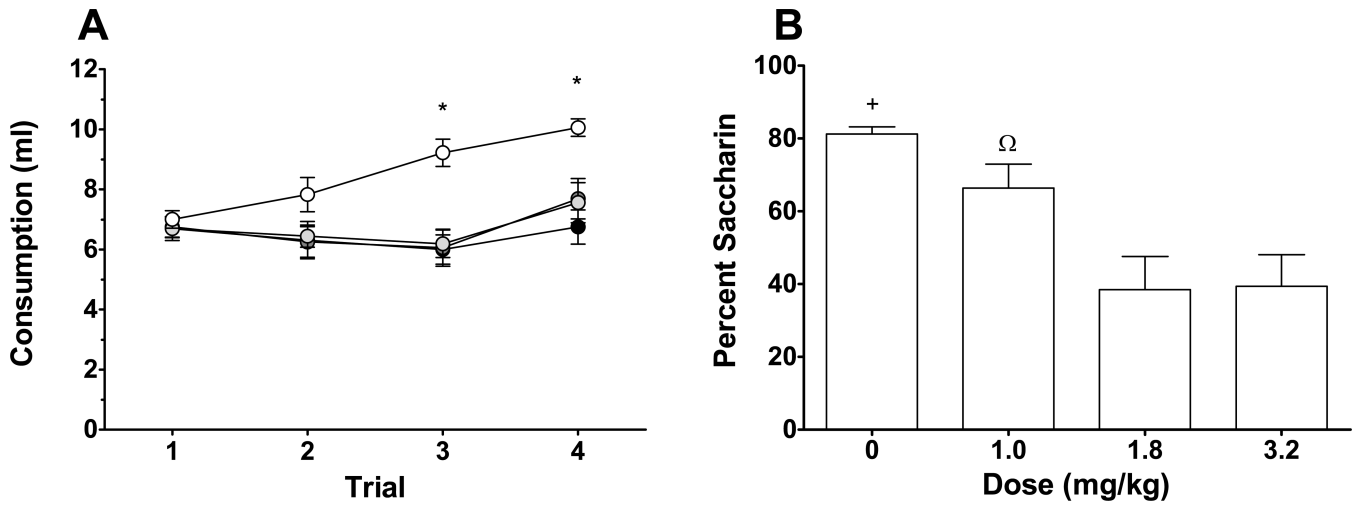

Figure 3.

Mean (± SEM) saccharin consumption (ml) by adolescents during acquisition (A) and mean (± SEM) percent saccharin consumed on the two-bottle test (B). *denotes a significant difference between Group 0 and all drug treated-groups. $^{+}$denotes a significant difference between Group 0 and Groups 1.8 and 3.2. $^{\Omega}$denotes a significant difference between Groups 1.0 and 1.8.

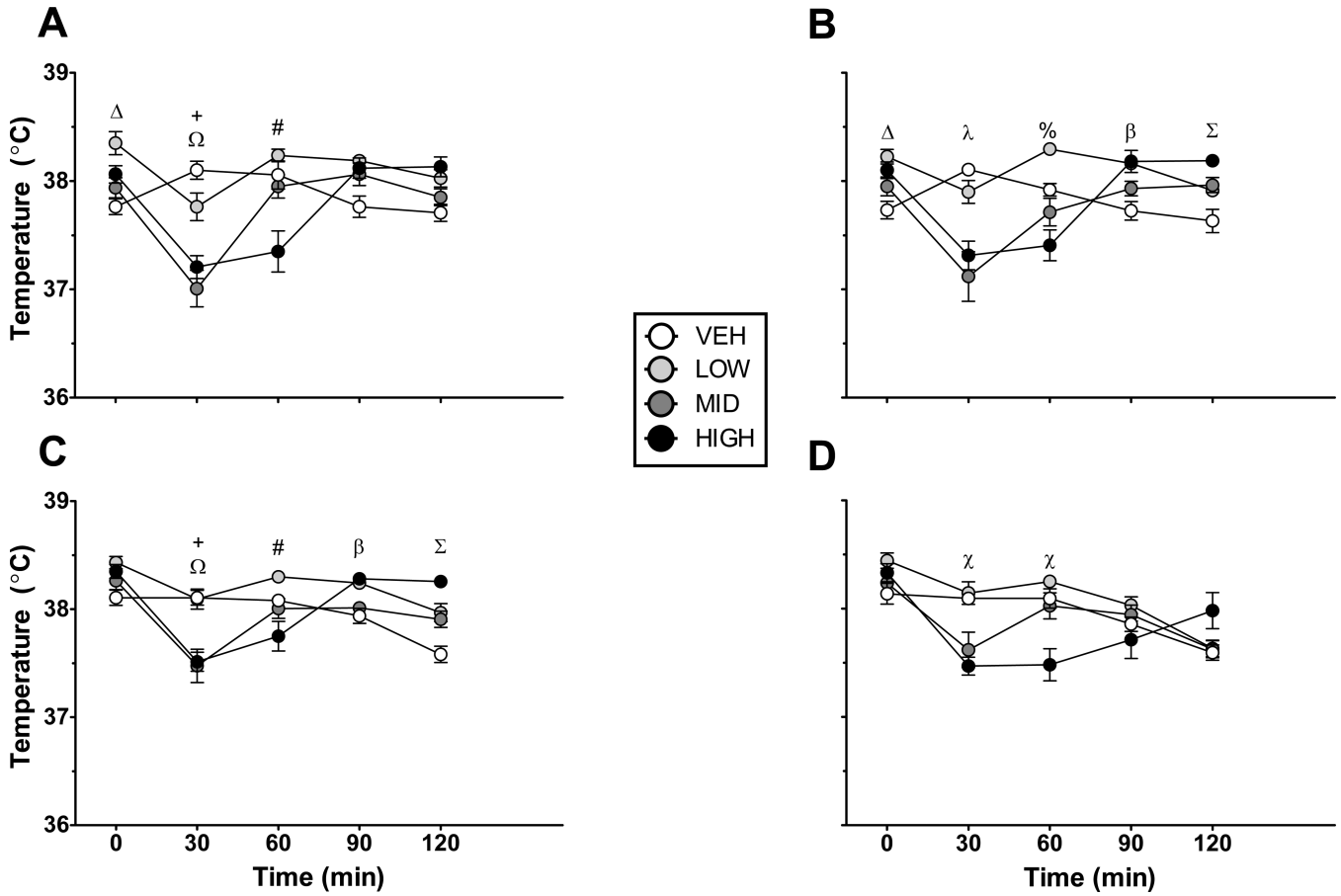

Figure 4.

Mean (± SEM) body temperature (°C) of adolescents across the five intervals (0, 30, 60, 90, 120) over the four conditioning trials (Panels A-D). $^{\Delta}$denotes a significant difference between Groups 0 and 1.0. $^{+}$denotes a significant difference between Group 0 and Groups 1.8 and 3.2. $^{\Omega}$denotes a significant difference between Groups 1.0 and 1.8. $^{\#}$denotes a significant difference between Groups 1.0 and 3.2. $^{\lambda}$denotes a significant difference between Groups 0 and 1.8. $^{\%}$denotes a significant difference between Group 1.0 and Groups 1.8 and 3.2. $^{\beta}$denotes a significant difference between Group 0 and Groups 1.0 and 3.2. $^{\Sigma}$denotes a significant difference between Group 0 and Group 3.2. ^{X}denotes a significant difference between Groups 0 and 1.0 and Group 3.2.