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European Journal of Pharmacology 398 (2000) 251–258

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# Effects of Δ9-tetrahydrocannabinol, (*R*)-methanandamide, SR 141716, and *d*-amphetamine before and during daily Δ9-tetrahydrocannabinol dosing

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Received 11 April 2000; accepted 14 April 2000

**Abstract**

We examined the effects of Δ9-tetrahydrocannabinol (Δ9-THC), (*R*)-(+)-arachidonyl-1′-hydroxy-2′-propylamide ((*R*)-methanandamide, AM 356), SR 141716, and *d*-amphetamine on fixed-ratio (FR) responding maintained by food in rats before and during daily dosing with Δ9-THC. Rats responded under a FR 10 schedule of food reinforcement. Cumulative dose–response curves for the various drugs were determined before and during daily Δ9-THC administration. All four drugs dose-dependently decreased responding both before and during daily dosing with Δ9-THC (18 mg/kg/day). The dose–response curves for both Δ9-THC and (*R*)-methanandamide were shifted to the right with daily dosing with Δ9-THC, indicating tolerance to the effects of Δ9-THC and cross-tolerance to the effects of (*R*)-methanandamide. The doses of *d*-amphetamine examined produced similar effects both before and during daily dosing with Δ9-THC. The effects of SR 141716 were not consistently altered by daily Δ9-THC administration. These results indicate that tolerance develops to the effects of Δ9-THC, when Δ9-THC is administered repeatedly. These results also indicate that cross-tolerance to (*R*)-methanandamide develops with repeated Δ9-THC administration. 

*Keywords:* Δ9-Tetrahydrocannabinol; (*R*)-Methanandamide; Anandamide; SR 141716; Amphetamine; Operant behavior; (Rat); Tolerance; Cross-tolerance; Dependence

## 1. Introduction

In this study, we examined the effects of Δ9-tetrahydrocannabinol (Δ9-THC), (*R*)-(+)-arachidonyl-1′-hydroxy-2′-propylamide ((*R*)-methanandamide), SR 141716, and amphetamine on operant behavior before and during a period of daily Δ9-THC administration. Our motivation for this was twofold. First, we were interested in examining whether cross-tolerance develops to the rate-decreasing effects of (*R*)-methanandamide with daily Δ9-THC administration. Second, we were interested in examining whether sensitization to the rate-decreasing effects of SR 141716 develops with daily THC administration. We included amphetamine as a control for any non-specific effects of daily Δ9-THC administration.

We were interested in examining whether the rate-decreasing effects of Δ9-THC and (*R*)-methanandamide on operant behavior were due to similar pharmacological mechanisms because of our observations with these two cannabinoids using a drug discrimination procedure. We observed that (*R*)-methanandamide, a stable analog (Abadji et al., 1994; Khanolkar et al., 1996) of anandamide, a putative endogenous cannabinoid (Hanus et al., 1993; Mechoulam et al., 1994), occasions Δ9-THC appropriate responding in rats trained with lower doses of Δ9-THC (1.8–3.0 mg/kg), but does not reliably occasion Δ9-THC appropriate responding in rats trained with a high dose of Δ9-THC (5.6 mg/kg; Järbe et al., 1998, 2000). One interpretation of these results is that Δ9-THC is a higher

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0014-2999/00/$ - see front matter 
PII: S0014-2999(00)00318-6

efficacy cannabinoid than (*R*)-methanandamide. This interpretation would be consistent with similar results with opioids using drug discrimination procedures (Young, 1991), and would be consistent with studies on the N-type calcium channel showing that anandamide is a partial agonist (Mackie et al., 1993). However, other interpretations of these results are possible.

The doses of (*R*)-methanandamide that could be tested in the drug discrimination procedure were limited by the rate-decreasing effects of (*R*)-methanandamide. If these rate-decreasing effects of (*R*)-methanandamide are not produced by the same pharmacological mechanism as the rate-decreasing effects of Δ9-THC, then the testing of higher doses of (*R*)-methanandamide would be prevented because of these non-cannabinoid rate-decreasing effects of (*R*)-methanandamide. Thus, if higher doses of (*R*)-methanandamide could be tested, then (*R*)-methanandamide might have completely occasioned Δ9-THC appropriate responding in the high-dose Δ9-THC training condition as well. Alternatively, these rate-decreasing effects of (*R*)-methanandamide could be mediated by the same pharmacological mechanism as for Δ9-THC, but these rate-decreasing effects could be due to different pharmacological mechanisms than the Δ9-THC-like discriminative effects of cannabinoids.

There is evidence that the effects of anandamide and Δ9-THC are not identical. For instance, intrathecal (i.t.) Δ9-THC induced analgesia, but not i.t. anandamide-induced analgesia, is antagonized by nor-BNI in the rat (Welch et al., 1995). Similarly, mice tolerant to the hypothermic effects of Δ9-THC are not cross-tolerant to the hypothermic effects of anandamide (Pertwee et al., 1993). Thus, there are differences between the pharmacological effects of anandamide and Δ9-THC, and presumably also differences between (*R*)-methanandamide and Δ9-THC. In order to more fully understand whether the failure of (*R*)-methanandamide to occasion complete Δ9-THC (5.6 mg/kg), appropriate responding in a drug discrimination paradigm represents (i) non-cannabinoid effects of (*R*)-methanandamide, (ii) differing efficacies and/or (iii) differing sites of cannabinoid rate-decreasing and Δ9-THC-like discriminative effects, we examined the extent to which daily Δ9-THC administration produces cross-tolerance to the rate-decreasing effects of (*R*)-methanandamide. In other studies, tolerance to the effects of either i.t. Δ9-THC induced analgesia or the twitch inhibiting effects of Δ9-THC in the mouse vas deferens has been shown to confer cross-tolerance to the effects of anandamide (Welch et al., 1995; Pertwee et al., 1993) and in the case of the mouse vas deferens, cross-tolerance to (*R*)-methanandamide also has been reported (Pertwee and Griffin, 1995). Conversely, repeated anandamide administration produces cross-tolerance to the analgesic effects of Δ9-THC (Fride, 1995; Welch, 1997).

We were also interested in how the effects of SR 141716 changed with daily Δ9-THC administration for a different reason. Physical dependence upon cannabinoids has not always been easy to demonstrate. A similar situation had also existed with benzodiazepines. When the benzodiazepine antagonist flumazenil became available, physical dependence could more easily be demonstrated using precipitated withdrawal procedures (Lukas and Griffiths, 1982). In addition to the observable signs of drug withdrawal, benzodiazepine withdrawal (Lamb and Griffiths, 1984) and precipitated withdrawal from either benzodiazepines (Lucki and Kucnarik, 1990; Takada et al., 1989) or opioids (Gellert and Sparber, 1977) is accompanied by decreases in the rate of operant responding. Beardsley et al. (1986) showed that withdrawal from Δ9-THC would disrupt operant behavior in the monkey, and that this disruption is reversed by the readministration of Δ9-THC. In this experiment, we examined whether the rate-decreasing effects of the cannabinoid antagonist SR 141716 were enhanced during daily Δ9-THC administration in a manner similar to that seen with opioids and benzodiazepines.

## 2. Materials and methods

### *2.1. Subjects*

Eight male Sprague–Dawley rats (Taconic Farms, Germantown, NY), weighing between 275 and 300 g upon arrival to the laboratory, were used. These rats had previously participated in an experiment that examined the effects of ethanol (0.2–1.6 g/kg) and fluvoxamine (0.1–30.0 mg/kg) on food-maintained behavior (Lamb and Järbe, 1997). A period of about 3 months separated drug administration for the two experiments. Rats were individually housed in polycarbonate cages. The animals had unlimited access to water, but access to food was limited to 12–15 g/day (on Fridays, rats received 45 g of food for the weekend). This food allotment was provided shortly following experimental sessions, maintaining bodyweights at 285–355 g during the course of the experiment, and resulted in adequate motivation. The vivarium had a 12-h light/12-h dark illumination cycle, and rats were trained and tested during the light phase.

### *2.2. Apparatus*

Experimental sessions were conducted in operant chambers containing three levers. Only the left lever was utilized in these experiments. Behavioral contingencies were controlled using Med-PC (Med-Associates, St. Albans, VT).

### *2.3. Procedures*

Experimental sessions were conducted 5 days a week and consisted of four 5-min periods of access to food under a Fixed–Ratio (FR) 10 schedule of reinforcement.

Each 5-min period of food availability was preceded by a 15-min time-out period in which responding had no programmed consequences. During each 5-min period of food availability, every tenth response on the left lever when the stimulus light above the lever was lighted (FR 10) resulted in the delivery of two 45 mg Noyes (Formula A) food pellets and the beginning of a 10-s time-out period. During this 10-s time-out period, the stimulus light above the lever was turned off, the house light was illuminated, the white noise generator turned on, and responses had no programmed consequences. Following this 10-s time-out period, the FR 10 schedule was again in effect, the house-light and white noise turned off, and the stimulus light above the lever turned on, unless the 5-min period of food availability had expired. If the 5-min period of food availability had expired, then a 15-min time-out period occurred during which all lights were out and no white noise was provided, or if the fourth food availability period was just completed the experimental session ended. An identical 15-min time-out period also preceded the first period of food availability.

### 2.4. Drugs

#### 2.4.1. Cumulative dosing procedure

Drug doses were administered in a cumulative fashion on test days (Fridays). The lowest dose in a dose–response curve would be administered at the beginning of the time-out period preceding the first period of food availability. Following this period of food availability at the beginning of the second 15-min time-out period, the second lowest dose in the dose–response curve was administered. This sequence continued until all four doses were administered, permitting the determination of a four point dose–response curve in a single session. Doses are reported as the cumulative dose given, i.e., the dose reported is the sum of the dose given and the doses that were given before it. For example, if at the beginning of the first time out period, a dose of 1 mg/kg was given, a dose of 2 mg/kg would be given at the beginning of the second time-out and the reported dose would be 3 mg/kg. Dose–response curves were determined at weekly intervals on Fridays as noted above.

#### 2.4.2. Daily Δ9-THC administration

During the period of daily Δ9-THC administration, Δ9-THC was given immediately before each experimental session (i.e., at the beginning of the first 15 min time-out period) and at mid-day on the weekends with no subsequent operant sessions. On the 2 days immediately preceding the beginning of daily Δ9-THC administration, vehicle was administered before the experimental sessions. On the subsequent 10 days, 10 mg/kg Δ9-THC was administered. Subsequently, the daily dose was increased to 18 mg/kg. Following 16 days on this dosing schedule, redeterminations of the various dose–response curves began. When the D-amphetamine and SR 141716 dose–response curves were determined during daily Δ9-THC administration, 18 mg/kg Δ9-THC was administered following rather than before the experimental session. When the Δ9-THC and (*R*)-methanandamide dose–response curves were determined during daily Δ9-THC administration, the daily dose of 18 mg/kg Δ9-THC was omitted.

#### 2.4.3. Drug preparations

Δ9-THC, dissolved in ethanol, was provided by NIDA and stored at −20°C until used. (*R*)-Methanandamide was synthesized according to Abadji et al. (1994) and sent to the site of behavioral evaluation in helium capped vials. Upon arrival, (*R*)-methanandamide was also dissolved in ethanol, appropriate amounts withdrawn, the ethanol evaporated under a stream of nitrogen, the residue then dissolved in a solution of propylene glycol and Tween-80, and stored at −20°C. Shortly before being used, the solute was diluted with normal (0.9%) saline after the solute had been sonicated for 20–30 min. This procedure was followed for preparing suspensions of Δ9-THC as well. SR 141716 (a gift from Sanofi Recherché, France) was dissolved in a propylene glycol/Tween-80 mixture before being diluted with saline. All cannabinoid related drugs were administered i.p. in volumes ranging between 1 and 10 ml/kg. Doses of SR 141716 less than 10 mg/kg were given in a volume of 1 ml/kg. Doses of Δ9-THC or (*R*)-methanandamide less than 10 mg/kg were given in a volume of 2 ml/kg. Doses of 10–18 mg/kg Δ9-THC, and 10 mg/kg (*R*)-methanandamide or SR 141716 were given in a volume of 3 ml/kg. Doses of 30 mg/kg (*R*)-methanandamide or SR 141716 were given in a volume of 5 ml/kg. Higher doses of Δ9-THC or (*R*)-methanandamide were given in volumes of 6–10 ml/kg. Suspensions were used within 0.5 h after saline was added to the drug/propylene glycol/Tween-80 mixture. *d*-Amphetamine sulfate was purchased from Sigma (St. Louis, MO), dissolved in saline and administered i.p. in a volume of 1 ml/kg. All doses are expressed in the forms indicated above.

### 2.5. Statistics

The $ED_{50}$s and their 95% confidence limits were calculated according to Tallarida and Murray (1987). Means, standard deviations, paired *t*-tests, confidence limits and analysis of variance (ANOVA) were calculated using SYSTAT (version 5.2.1; Systat, Evanston, IL), run on a Macintosh Quadra 700; $\alpha = 0.05$.

## 3. Results

### 3.1. Vehicle injections

The open circles in Fig. 1A show the mean rate of responding following sequential vehicle injections. The

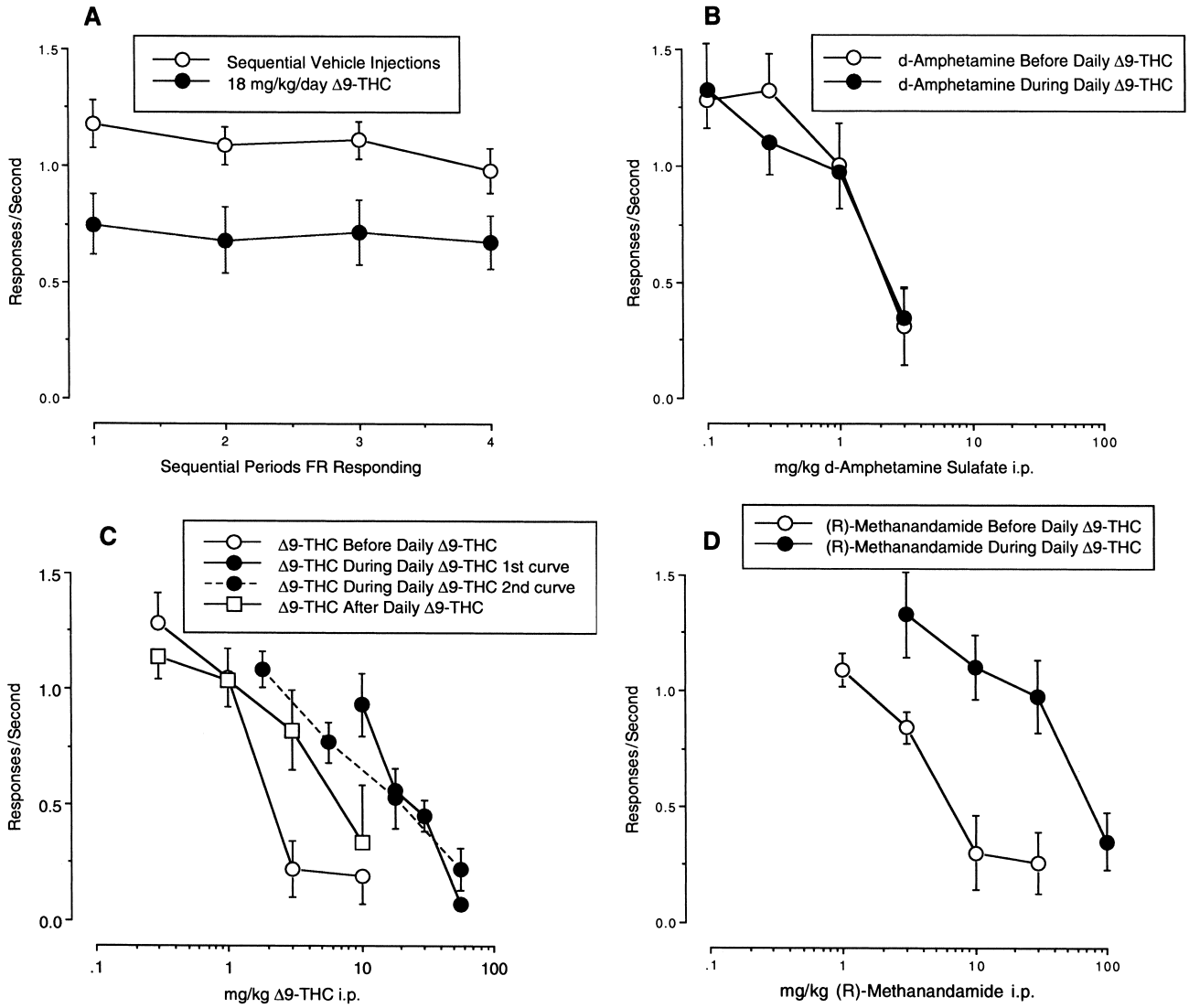


Fig. 1. The upper-left (A) graph shows mean (±S.E.M.) response rate in responses per second during four sequential periods of FR responding on the days before determinations of the effects of the various drugs either following four sequential vehicle injections (open circles) in the period before daily Δ9-THC administration or following a single injection of 18 mg/kg Δ9-THC (closed circles) during the period of daily Δ9-THC administration. The upper-right hand (B) graph shows the mean response rate following various doses of D-amphetamine both before (open circles) and during (closed circles) daily Δ9-THC administration (18 mg/kg/day, i.p.) with drug dose plotted on a log scale. The lower-left hand (C) graph shows the effects of various doses of Δ9-THC on response rate both before (open circles), during (closed circles; solid line represents the first determination of the Δ9-THC dose–response curve and the dashed line represents the second determination of the Δ9-THC dose–response curve) and 3 weeks following the discontinuation of daily Δ9-THC administration (open squares). The lower-right hand (D) graph shows the effects of various doses of (*R*)-methanandamide on response rate both before (open circles) and during (closed circles) daily Δ9-THC administration.

bars represent the standard error of the mean. These error bars for the vehicle were calculated using the standard deviation based upon all the determinations, but with a standard error value calculated with a reduced *N* equal to 8, so as to provide a conservative estimate for comparisons with the other mean values. As can be seen, responding occurred at a rate of about 1 response/s during each of these sequential sessions using volumes of vehicle corresponding to those used for the following dose–response curves. As can also be seen, all the means are within the standard error of the other means, and there are no significant differences between the means.

### *3.2. Dose–response curves before daily Δ9-THC*

As can be seen in Fig. 1C (open circles), administration of Δ9-THC produced dose-related decreases in response rate, with doses of Δ9-THC of 3 mg/kg or greater producing response-rate decreases below those produced by vehicle injections. (*R*)-Methanandamide produced similar effects with doses of 10 mg/kg or greater producing response-rate decreases below those produced by vehicle injections (Fig. 1D).

As can be seen Fig. 1B (open circles), the highest dose of *d*-amphetamine (3 mg/kg) produced response-rate de-

creases. There was also some tendency for the 0.3 mg/kg dose of *d*-amphetamine to increase responding as this point was slightly outside the upper bounds of the 95% confidence limits for the effects of drug vehicle.

SR 141716, as can be seen in the open circles of Fig. 2, produced dose-related decreases in response rate. These rate decreases were apparent even at the lowest dose examined (1 mg/kg), and became more pronounced with increasing doses of SR 141716 (paired *t*-tests between drug effects at each dose and appropriate vehicle controls, $t$'s $> 4.0$, $P$'s $< 0.0125$ for all four comparisons).

### 3.3. Effects of daily administration of Δ9-THC

As can be seen in Fig. 3, initial administration of 10 mg/kg Δ9-THC produced decreases in response rate that appeared to become even greater over the first few days of daily Δ9-THC administration. Subsequently, however, the effects of 10 mg/kg Δ9-THC lessened. When the dose of Δ9-THC was increased to 18 mg/kg, response rate showed a slight initial further decrease and then resumed an upward trend, but after a short while, seemed to plateau. Further evidence of this plateauing can be seen in Fig. 1A in which the closed circles represent the mean rate of responding following 18 mg/kg Δ9-THC prior to the experimental sessions preceding the determination of the dose–response curves in the second half of this experiment. As can be seen, rates of responding following 18 mg/kg Δ9-THC were lower (0.56–0.70 responses/s) during daily Δ9-THC administration than the rates of responding following vehicle administration before daily dosing with Δ9-THC began (1.01–1.09 responses/s). Thus, complete tolerance to the daily administered dose of Δ9-THC did not develop during the course of this experiment.

### 3.4. Dose–response curves during daily Δ9-THC

Daily administration of Δ9-THC (18 mg/kg) produced a rightward shift in the dose–response curves for Δ9-THC and for (*R*)-methanandamide as can be seen by comparing

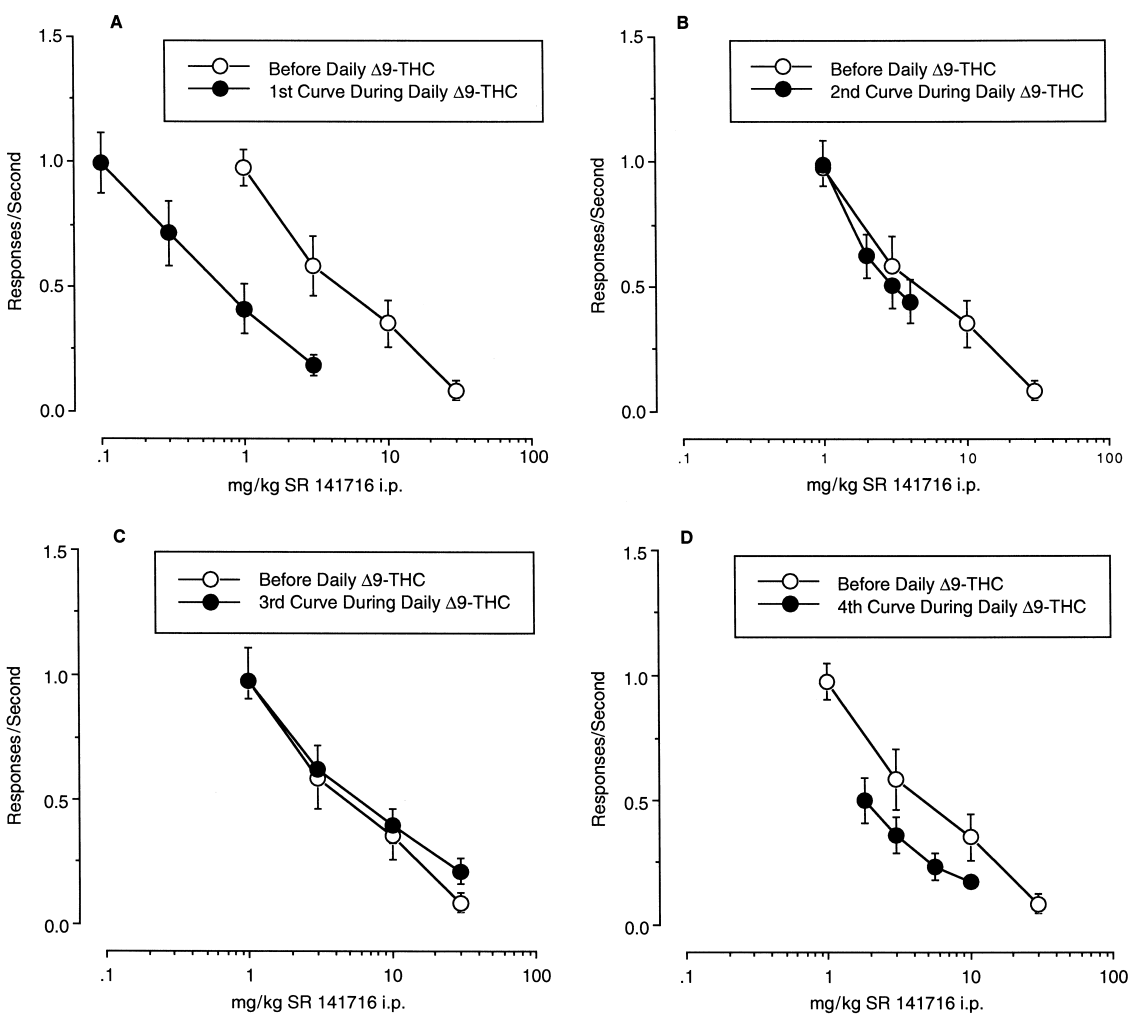


Fig. 2. The effects of various doses of SR 141716 plotted on a log scale on the mean (±S.E.M.) rate of responding in responses per second both before (open circles, same data in all four graphs) and upon four redeterminations (closed circles, upper-left 1st redetermination (A), upper-right 2nd (B), lower-left 3rd (C), and lower-right 4th (D)) during daily Δ9-THC administration (18 mg/kg/day) are shown.

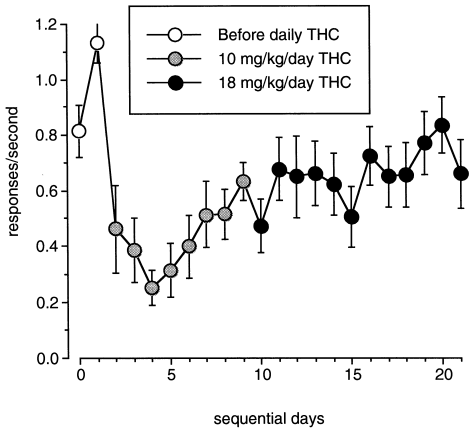


Fig. 3. The overall mean (±S.E.M.) rate of responding in responses per second is plotted across sequential experimental sessions. Days on which drug vehicle was injected before the experimental session are indicated by open circles. Days on which 10 mg/kg Δ9-THC (3 ml/kg) was injected before the experimental session are indicated by the hatched marked circles, and days on which 18 mg/kg Δ9-THC (5 ml/kg) was injected before the experimental session are indicated by the filled circles.

the curves with filled circles to those with open circles in Fig. 1C and D. The $ED_{50}$ for Δ9-THC before daily administration was 2.02 mg/kg (95% C.L., 1.54–2.66 mg/kg), while the $ED_{50}$ during daily administration was 15.61 mg/kg (9.26–26.30). Similarly, the (*R*)-methanandamide $ED_{50}$ was 5.77 mg/kg (3.97–8.36) before daily administration of Δ9-THC, but 26.93 mg/kg (14.99–48.37) during daily administration. Thus, daily administration of Δ9-THC produced significant rightward shifts in both the Δ9-THC and the (*R*)-methanandamide dose–response curves.

As can be seen, by comparing the two redeterminations of the Δ9-THC dose–response curve during daily Δ9-THC administration, one obtained early on during the period of daily Δ9-THC administration and the other near the end of daily Δ9-THC administration, the shift in the Δ9-THC dose–response curve was relatively stable. These shifts appear to be on the order of 10–30-fold for Δ9-THC and 3–10-fold for (*R*)-methanandamide.

In contrast, daily administration of Δ9-THC produced little change in the *d*-amphetamine dose–response curve as can be seen in Fig. 1B. All the points of the redetermination of the *d*-amphetamine dose–response curve are typically within one standard error of the original dose–response curve, with the exception of the 0.3 mg/kg dose, which is within two standard errors of the original point. The $ED_{50}$ for *d*-amphetamine before daily Δ9-THC administration was 2.05 mg/kg (1.34–3.15) and during daily Δ9-THC the $ED_{50}$ for *d*-amphetamine was 2.20 (1.42–3.40). Thus, the effects of *d*-amphetamine before and during daily Δ9-THC were quite similar.

The effects of daily Δ9-THC administration on the SR 141716 dose–response curve are either more complicated or less robust than with the other drugs examined. Fig. 2 shows the four redeterminations of the SR 141716 dose–response curve obtained during daily Δ9-THC administration (open circles before daily Δ9-THC, closed circles during daily 18 mg/kg Δ9-THC). As can be seen in Fig. 2A, the initial redetermination of the SR 141716 dose–response curve produced clear evidence of sensitization with the 3 mg/kg dose of SR 141716 now producing greater decreases in response rate than was the case before daily Δ9-THC administration (ANOVA with test as a factor and log-dose as a covariate resulted in $F > 12.0$ and $P < 0.001$ for test as a factor; paired *t*-test at 3 m/kg resulted in $t = 2.57$, $P < 0.05$). However, 1 week later, when the effects of four sequential 1 mg/kg injections of SR 141716 were determined (Fig. 2B), the effects were nearly identical to those initially found (same ANOVA resulted in $F < 0.5$, $P > 0.5$). The same can be said for the dose–response curve determined still one week later (Fig. 2C); this dose–response curve closely approximated the original dose–response curve (ANOVA $F < 1.0$, $P > 0.3$) and clearly differed from the curve first determined during daily Δ9-THC administration. The last SR 141716 dose–response curve was determined slightly more than 2 weeks later with the second Δ9-THC dose–response curve redetermined in between. This last SR 141716 dose–response curve was determined following administration of 18 mg/kg Δ9-THC 90 min before the beginning of food availability (see Tsou et al., 1995). This curve more closely approximates the original curve (Fig. 2D), but is difficult to interpret given the potential rate-decreasing effects of this dose of Δ9-THC alone.

### *3.5. Δ9-THC dose–response curve after daily Δ9-THC*

The results from when daily Δ9-THC administration was discontinued for 3 weeks and the Δ9-THC dose–response curve was redetermined is shown in Fig. 1C. This dose–response curve was mid-way between the curve determined before daily Δ9-THC administration and the curves determined during daily Δ9-THC administration, a finding congruent with a partial loss of tolerance to the rate-decreasing effects of Δ9-THC.

## 4. Discussion

Daily administration of Δ9-THC resulted in a rightward shift in the Δ9-THC dose–response curve. In other words, tolerance developed to the rate-decreasing effects of Δ9-THC on the operant behavior of the rat. This effect was replicable in so much as it was observed in both the Δ9-THC dose–response curves determined during daily Δ9-THC administration. This effect is also consistent with a number of previous reports showing that tolerance develops with repeated administration of Δ9-THC (e.g., McMillan et al., 1983). Furthermore, the failure to obtain complete tolerance under these conditions is consistent with other previous reports using similar conditions (e.g., Davis and Borgen, 1975; Beardsley and Martin, 2000).

Similarly, daily administration of Δ9-THC resulted in a rightward shift in the (*R*)-methanandamide dose–response curve. In other words, cross-tolerance developed to the rate-decreasing effect of (*R*)-methanandamide. This result is consistent with other studies showing tolerance to the effects of either i.t. Δ9-THC induced analgesia or the twitch inhibiting effects of Δ9-THC in the mouse vas deferens conferring cross-tolerance to the effects of anandamide (Welch et al., 1995; Pertwee et al., 1993), and in the case of the mouse vas deferens, cross-tolerance to (*R*)-methanandamide (Pertwee and Griffin, 1995). Conversely, repeated anandamide administration produces cross-tolerance to the analgesic effects of Δ9-THC (Fride, 1995; Welch, 1997). Our results are also consistent with previous studies showing cross-tolerance from the behavioral effects of Δ9-THC to other cannabinoids, such as Δ8-THC and 11-hydroxy-Δ9-THC, when Δ9-THC is administered chronically (McMillan et al., 1971; Kosersky et al., 1974).

The observation of cross-tolerance from Δ9-THC to (*R*)-methanandamide in the present study is particularly interesting in light of our previous observation that (*R*)-methanandamide occasions Δ9-THC-appropriate responding in rats trained with a low dose of Δ9-THC, but not in rats trained with a high dose of Δ9-THC (Järbe et al., 1998, 2000). The doses of (*R*)-methanandamide that could be tested in these earlier studies were limited by the rate-decreasing effects of (*R*)-methanandamide. If (*R*)-methanandamide decreased responding by a different mechanism than Δ9-THC, then this difference rather than differences in the efficacy of the two drugs at occasioning Δ9-THC-appropriate responding might explain our earlier observations. Cross-tolerance from Δ9-THC to (*R*)-methanandamide makes this explanation of our previous observations less likely to be correct. Thus, this strengthens our suggestion that Δ9-THC and (*R*)-methanandamide differ in their efficacy to produce Δ9-THC-appropriate responding.

Daily administration of Δ9-THC did not result in any shifts in the dose–response curve of *d*-amphetamine. The effects of *d*-amphetamine determined before daily Δ9-THC administration were quite similar to those of *d*-amphetamine determined during daily Δ9-THC administration. This is consistent with a previous report that daily Δ9-THC administration does not alter the effects of *d*-amphetamine on operant behavior (McMillan et al., 1983). This replicates previous work, and serves as a negative control for this experiment; demonstrating that dose–response curves redetermined under these experimental conditions remain relatively stable.

This observation and the observation that the two Δ9-THC dose–response curves determined during daily Δ9-THC administration were quite similar becomes especially important given the effects observed with SR 141716. When the SR 141716 dose–response curve was initially redetermined during daily Δ9-THC administration, the expected leftward shift in the SR 141716 dose–response curve was observed. Such a leftward shit indicates sensitization to the effects of SR 141716. Recently, Beardsley and Martin (2000) reported that doses of SR141716 (3 and 9 mg/kg), that did not suppress behavior in rats that had not received chronic treatment with Δ9-THC, did suppress behavior in rats that had received chronic Δ9-THC (however, one should note that these doses of SR 141716 did suppress behavior in the present study). These effects are consistent with reported changes in the effects of narcotic antagonists, such as naloxone or naltrexone, with chronic opiate administration (e.g., Gellert and Sparber, 1977) and the observed changes in the effects of the benzodiazepine antagonist flumazenil with chronic benzodiazepine administration (Takada et al., 1989).

However, when the effects of SR 141716 were redetermined twice more at weekly intervals, this apparent sensitization disappeared. Similarly, when Beardsley and Martin (2000) administered SR141716 again on the day following their initial administration, SR 141716's rate suppressant effects were much reduced. Similar, but also poorly understood, reversals in the changes in the effects of narcotic antagonists or flumazenil have been observed when the effects of these drugs were redetermined in close temporal sequence (e.g., Huidobro et al., 1963; Lamb and Griffiths, 1985). When the temporal interval between SR 141716 was increased to 2 weeks between the 3rd and the 4th redetermination, a seemingly leftward shift in the SR 141716 dose–response curve occurred. This apparent shift is, however, difficult to interpret, because the 4th SR 141716 dose–response curve represents the effects of SR 141716 in combination with an acute dose of 18 mg/kg Δ9-THC, a dose of Δ9-THC that may have rate-decreasing effects when given alone.

In summary, this experiment provides evidence that (*R*)-methanandamide and by extension other fatty acid ethanolamides produce at least some of their behavioral effects through the same mechanism as other cannabinoids, such as Δ9-THC. In particular, conditions that produce tolerance to Δ9-THC resulted in cross-tolerance to (*R*)-methanandamide. These findings make it less likely that our earlier drug discrimination findings result from Δ9-THC and (*R*)-methanandamide decreasing FR responding by different mechanisms and thus, strengthen our earlier suggestion (Järbe et al., 1998, 2000) that (*R*)-methanandamide may have a lower efficacy than Δ9-THC at producing Δ9-THC-appropriate responding. One implication of which would be that fatty acid ethanolamides, such as (*R*)-methanandamide, might be less likely to produce Δ9-THC-like subjective effects in man.

## Acknowledgements

We thank NIDA for the supplies of Δ9-THC and Sanofi Recherché, France, for the supplies of SR 141716 used in

these experiments. We would also like to acknowledge the technical assistance of A. Dickstein and M. Harris. All procedures were approved by the Animal Care and Use Committee of Allegheny University of the Health Sciences. Preliminary results of this investigation were presented at the annual CPDD meeting, Nashville, TN, June 1997. Supported by NIDA grants DA 09064 and DA 00253 (Philadelphia), and DA 03801 and DA 00152 (Storrs).

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