

Self-administration behavior is maintained by the psychoactive ingredient of marijuana in squirrel monkeys

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Many attempts to obtain reliable self-administration behavior by laboratory animals with delta-9-tetrahydrocannabinol (THC), the psychoactive ingredient in marijuana, have been unsuccessful^{1–5}. Because self-administration behavior has been demonstrated in laboratory animals for almost all other psychoactive drugs abused by humans⁶, as well as for nicotine, the psychoactive ingredient in tobacco⁷, these studies would seem to indicate that marijuana has less potential for abuse. Here we show persistent intravenous self-administration behavior by monkeys for doses of THC lower than doses used in previous studies, but comparable to doses in marijuana smoke inhaled by humans.

Most previous studies of THC self-administration^{1–5} used intravenous unit doses higher than those calculated from clinical studies. Moreover, due to the lipophilic nature of THC, its

very low solubility in water, and the high THC doses usually used, many studies were done using THC in suspension. Here we used low, clinically relevant doses of THC in a clear solution that rapidly distributed THC to the brain after intravenous administration⁸. THC was dissolved in a vehicle containing 0.4–1.0% Tween-80 and 0.4–1.0% ethanol in saline (using a modification of previous procedures^{9,10}). Before this study began, squirrel monkeys (*Saimiri sciureus*) with venous catheters had been trained to press a lever for intravenous (i.v.) injections of cocaine¹¹. During daily sessions from Monday to Friday, monkeys sat in a chair in an isolation chamber, and injections (0.2 ml in 0.2 s) were administered from a pump outside the chamber¹¹. A green stimulus light was turned on at the start of each session; 10 presses of a lever turned off the green light and produced a 2-s red light paired with injection of 30 µg/kg cocaine (a 10-response, fixed-ratio schedule of i.v. drug injection, FR10). There was a one-minute 'time out' period after each injection, during which the chamber was kept dark and lever presses had no consequences. Each self-administration session lasted one hour.

At the start of this study, saline was substituted for cocaine injections for five sessions. Responding declined within a few sessions, and monkeys self-administered only one to four injections per session (Fig. 1a). When 2 µg/kg injections of THC were substituted for saline, responding immediately increased and stabilized within a week, and approximately 30 injections of THC were self-administered per session. Substitution of vehicle for THC caused a significant progressive decline in the number of injections delivered per session ($F_{5,15} = 7.683$, $p < 0.001$, one-way ANOVA for repeated measures) and in the rates of responding ($F_{5,15} = 3.118$, $p < 0.05$, data not shown). Replacement of vehicle injections with 4 µg/kg injections of THC resulted in immediate recovery of drug-taking behavior. When the THC dose was varied, an inverted U-shaped dose–response curve was obtained (Fig. 1b and c). THC maintained significantly higher numbers of injections per session ($F_{4,12} = 8.239$, $p = 0.002$) and rates of responding ($F_{4,12} = 4.702$, $p = 0.016$) than vehicle, with maximal responding at 2 and 4 µg/kg THC per injection.

For human subjects smoking a marijuana cigarette containing 15 mg of THC, actual THC intake¹² is about 3 mg. Assuming that humans take 10 to 15 puffs per cigarette, each puff contains 200 to 300 µg of THC, or 2.9 to 4.3 µg/kg THC (for an average

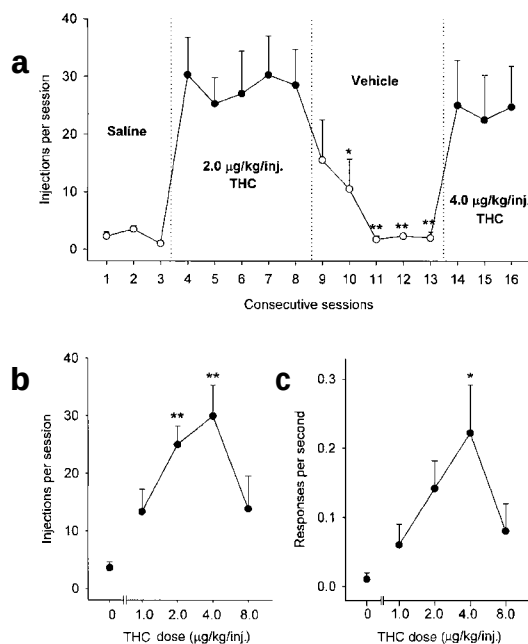
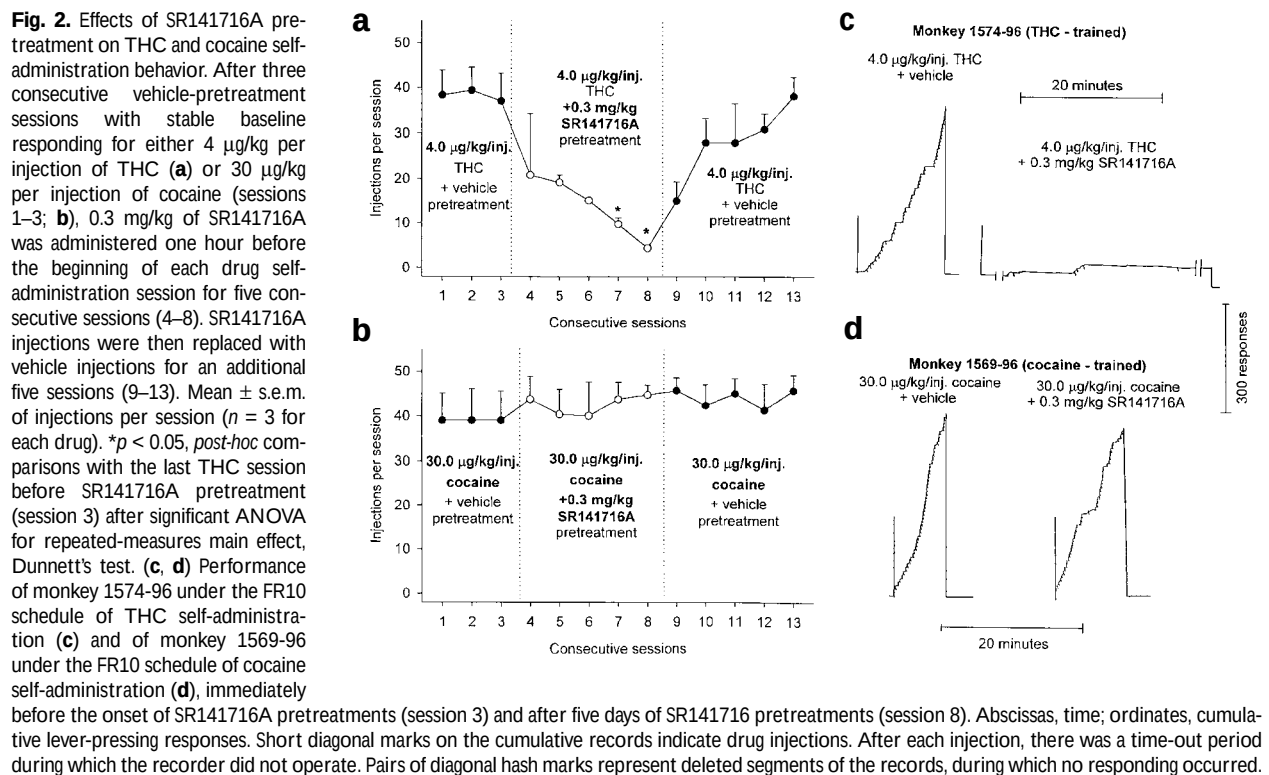


Fig. 1. Initial acquisition of THC self-administration behavior, and effects of varying injection dose of THC in squirrel monkeys. **(a)** Mean $\pm$ s.e.m. of injections per session ($n = 4$ monkeys). Following saline extinction (sessions 1–3 are the last three sessions of saline extinction), monkeys self-administered THC (2 µg/kg per injection) for 5 sessions (4–8). Self-administration behavior was then extinguished by replacing injections of THC with injections of its vehicle for an additional five sessions (9–13). When vehicle injections were replaced with 4 µg/kg injections of THC, self-administration behavior immediately recovered (sessions 14–16). * $p < 0.05$; ** $p < 0.01$; *post-hoc* comparisons with the last THC session before vehicle extinction (session 8), after significant ANOVA for repeated-measures main effect, Dunnett's test. Subsequently, a THC dose–response curve was established. Both 2 and 4 µg/kg per injection doses of THC were retested along with two additional doses (1 and 8 µg/kg per injection). Each dose of THC was tested for five consecutive sessions with five intervening vehicle-extinction sessions between each dosage condition. Number of injections per session **(b)** and overall rate of responding in the presence of the green light signaling THC availability **(c)** are presented as a function of injection dose of THC (mean $\pm$ s.e.m. of the last three sessions within the five sessions in which each dose was tested; $n = 4$). * $p < 0.05$; ** $p < 0.01$; *post-hoc* comparisons with vehicle conditions after significant ANOVA for repeated-measures main effect, Dunnett's test.



body weight of 70 kg). This is in perfect agreement with the 2 to 4 μ g/kg injection doses that maintain THC self-administration in squirrel monkeys.

We then compared THC and cocaine self-administration after treatment with SR141716A, a potent and selective antagonist of CB1 cannabinoid receptors¹³, which are thought to mediate most behavioral and neurochemical effects of THC^{9,14}. SR141716A (0.3 mg/kg, intramuscularly) or its vehicle was administered one hour before the experimental session for five consecutive sessions. SR141716A decreased the number of self-administered 4 μ g/kg THC injections to vehicle-control levels within a few sessions ($F_{5,10} = 3.908$, $p = 0.032$), and THC self-administration rapidly recovered when SR141716A treatment was stopped (Fig. 2a and c). In contrast, monkeys responding for cocaine under identical conditions, and with comparable rates and patterns of self-administration behavior (0.33 and 0.49 responses per second in monkeys self-administering THC or cocaine, respectively, during the last session before SR141716A pretreatment), showed no effect of pretreatment with the same dose of SR141716A ($F_{1,20} = 16.766$, $p = 0.015$, two-way ANOVA for repeated measures; Fig. 2b and d). Thus, the effect of SR141716A in suppressing THC self-administration did not seem to be due to any nonselective depressant effect on behavior, and the reinforcing effect of THC on self-administration behavior seemed to be due to its direct actions on CB1 cannabinoid receptors.

The active component of cannabis, THC, can act as a strong reinforcer of drug-taking behavior in an experimental animal, the squirrel monkey, as it does in humans. The THC self-administration behavior was comparable in intensity to that maintained by cocaine under identical conditions, and was obtained using a range of doses similar to those self-administered by humans smoking a single marijuana cigarette¹². These findings suggest

that marijuana has as much potential for abuse as other drugs of abuse, such as cocaine and heroin. The selective reduction in THC self-administration by the cannabinoid antagonist SR141716A indicates that this abuse potential is likely mediated by cannabinoid CB1 receptors in the brain.

ACKNOWLEDGEMENTS

Cocaine HCl, delta-9-tetrahydrocannabinol (THC) and SR141716A were obtained from NIDA, NIH. We thank Godfrey Redhi for technical assistance.

ACCEPTED 26 SEPTEMBER 2000

- Carney, J. M., Uwaydah, I. M. & Balster, R. L. *Pharmacol. Biochem. Behav.* 7, 357–364 (1977).
- Harris, R. T., Waters, W. & McLendon, D. *Psychopharmacologia* 37, 23–29 (1974).
- Kaymakcalan, S. *Bull. Narc.* 25, 39–47 (1973).
- Mansbach, R. S., Nicholson, K. L., Martin, B. R. & Balster, R. L. *Behav. Pharmacol.* 5, 219–225 (1994).
- Pickens, R., Thompson, T. & Muchow, D. C. *Bayer Symposium IV: Psychic Dependence* 78–86 (Springer, New York, 1973).
- Young, A. M. & Herling, S. in *Behavioral Analysis of Drug Dependence* (eds. Goldberg, S. R. & Stolerman, I.) 9–67 (Academic, Orlando, Florida, 1986).
- Goldberg, S. R., Spealman, R. D. & Goldberg, D. M. *Science* 214, 573–575 (1981).
- Mantilla-Plata, B. & Harbison, R. D. *Toxicol. Appl. Pharmacol.* 34, 292–300 (1975).
- Olsen, J. L. & Davis, K. H. *J. Pharm. Pharmacol.* 25, 344 (1973).
- Tanda, G., Pontieri, F. E. & Di Chiara, G. *Science* 276, 2048–2050 (1997).
- Spear, D. J., Muntaner, C., Goldberg, S. R. & Katz, J. L. *Pharmacol. Biochem. Behav.* 38, 411–416 (1991).
- Agurell, S. *et al. Pharmacol. Rev.* 38, 21–43 (1986).
- Rinaldi-Carmona, M. *et al. FEBS Lett.* 350, 240–244 (1994).
- Ledent, C. *et al. Science* 283, 401–404 (1999).