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Cannabinoid precipitated withdrawal by the selective cannabinoid receptor antagonist, SR 141716A

Mario D. Aceto [a,*], Susan M. Scates [a], John A. Lowe [b], Billy R. Martin [a]

[a] *Department of Pharmacology and Toxicology, Virginia Commonwealth University, Richmond, VA 23298-0613, USA*
[b] *Pfizer Central Research, Pfizer Inc., Groton, CT 06340, USA*

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Abstract

Precipitated withdrawal in rats chronically exposed to Δ^9-tetrahydrocannabinol, the major psychoactive principle of the marijuana plant, was unequivocally demonstrated for the first time using a selective antagonist, SR 141716A (*N*-(piperidin-1-yl)-5-(4-chlorophenyl)-1(2,4-dichloro-phenyl)-4-methyl-1*H*-pyrazole carboxamide · HCl). This demonstration should provide a powerful stimulus for the systematic study of dependency on the psychoactive cannabinoids.

Keywords: Δ^9-Tetrahydrocannabinol; Precipitated withdrawal; SR 141716A, (*N*-(piperidin-1-yl)-5-(4-chlorophenyl)-1(2,4-dichloro-phenyl)-4-methyl-1*H*-pyrazole carboxamide · HCl)

Recently, Rinaldi-Carmona and co-workers (Rinaldi-Carmona et al., 1994) reported that SR 141716A (*N*-(piperidin-1-yl)-5-(4-chlorophenyl)-1(2,4-dichlorophenyl)-4-methyl-1*H*-pyrazole carboxamide · HCl) bound with high affinity to the central cannabinoid receptor and acted as an antagonist versus a variety of pharmacological and behavioral cannabinoid effects in rodents. The availability of such a tool provided a unique opportunity to study and demonstrate dependence on Δ^9-tetrahydrocannabinol.

We used the continuous intraperitoneal infusion method because of the great flexibility regarding daily adjustment of dose and because physical dependence develops more rapidly when tissues or receptors are exposed continuously to a substance. Unanesthetized male Sprague-Dawley rats previously fitted with intraperitoneal cannulae (Teiger, 1974) were infused continuously with selected dose regimens of Δ^9-tetrahydrocannabinol, or appropriate controls. At the end of 4 days of continuous exposure to Δ^9-tetrahydrocannabinol, the rats were challenged i.p. with a dose of 10 mg/kg of SR 141716A. The dose of SR 141716A used in these studies was shown to antagonize several marijuana-like effects in mice and rats (Rinaldi-Carmona et al., 1994).

Two experiments were conducted, the second to confirm the results of the first and to extend the dose range. In both experiments, the rats were randomly allocated to treatment groups and were likewise assigned to a numbered cage on a rack to prevent bias. The Δ^9-tetrahydrocannabinol dose regimens were either 0.5, 2.5 or 12.5 mg/kg on day 1. Each starting dose was doubled each day throughout the infusion. The control vehicle was emulphor/ethanol/sterile saline (1 : 1 : 18). A trained observer scored the behavior. These studies were approved by the Institutional Animal Care and Use Committee.

A marked change in the Δ^9-tetrahydrocannabinol-infused animals was evident approximately 10 min after the injection of SR 141716A, and these effects subsided within the hour. The behavioral signs included head shakes, facial tremors, tongue rolling, biting, wet-dog shakes, eyelid ptosis, facial rubbing, paw treading, retropulsion, immobility, ear twitch, chewing, licking, stretching and arched back. The signs facial rubbing and wet-dog shakes were quantified. A gauge of the intensity of this behavioral activation is depicted in Fig. 1 for wet-dog shakes. The plateau in the dose-response curve probably reflects an upper limit of responsive-

* Corresponding author. Department of Pharmacology and Toxicology, Virginia Commonwealth University, MCV Campus, Box 980613, Richmond, VA 23298-0613, USA. Tel. (804) 828-8397, fax (804) 828-2117.

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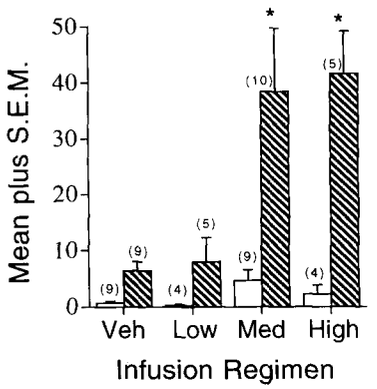

Fig. 1. Depiction of the data designated wet-dog shakes following challenge with the cannabinoid antagonist in Δ^9-tetrahydrocannabinol-dependent rats. The starting doses for the low, medium (Med), and high Δ^9-tetrahydrocannabinol dose regimens were 0.5, 2.5, and 12.5 mg/kg per day, respectively. Each starting dose was doubled each day for a total of 4 days. The sign was quantified during a 1-h observation period after SR 141716A. The data are expressed as means plus S.E.M. The number of subjects per treatment regimen is indicated in parentheses. Open and hatched bars represent wet-dog shakes elicited by rats challenged with vehicle and SR 141716A, respectively. * Significantly different ($P < 0.05$) from vehicle control.

ness due to the numerous competing behaviors. Analysis of variance of the data indicated overall statistical significance for facial rubbing ($F = 11.25$, $P < 0.0001$) and wet-dog shakes ($F = 6.40$, $P < 0.0001$). Post-hoc comparisons using the Bonferroni/Dunn procedure for each sign revealed significant differences ($P < 0.05$) in response when compared to vehicle controls. All the control groups for these signs, either before or after challenge with SR 141716A, were not statistically different. The other signs were observed and scored once when they occurred. Statistical analysis (Bonferroni/Dunn procedure) of the body weight data revealed no significant differences among the treatment groups; they all gained weight as did the vehicle controls during the course of the experiments.

Kaymakcalan et al. (1977) examined the behavior of rats following cessation of chronic administration of Δ^9-tetrahydrocannabinol and some of the signs were the same as those noted by us in the present study. Importantly, facial rubbing was not mentioned and wet-dog shakes were seen rarely. Other workers (Verberne et al., 1980) found that challenge with serotonin biogenic amine reuptake inhibitors in rats chronically treated with Δ^9-tetrahydrocannabinol elicited withdrawal signs. Again, wet-dog shakes were rarely observed. In addition, since the amine reuptake inhibitors were given 24 h after abrupt withdrawal of Δ^9-tetrahydrocannabinol, the syndrome cannot be said to have been precipitated.

The availability of a functional experimental model and selective cannabinoid antagonist provide a powerful impetus for the systematic study of the effect of chronic exposure to cannabinoids. Characterization of the behavioral expressions provides leads regarding the underlying neural substrates and role of the recently described cannabinoid receptors.

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References

Kaymakcalan, K., I.H. Ayhan and F.C. Tulunay, 1977, Naloxone-induced or postwithdrawal abstinence signs in Δ^9-tetrahydrocannabinol-tolerant rats, Psychopharmacology 55, 243.

Rinaldi-Carmona, M., F. Barth, M. Héaulme, D. Shire, B. Calandra, C. Congy, S. Martinez, J. Maruani, G. Néliat, D. Caput, P. Ferrara, P. Soubrié, J.C. Brelière and G. Le Fur, 1994, SR141716A, a potent and selective antagonist of the brain cannabinoid receptor, FEBS Lett. 350, 240.

Teiger, D.G., 1974, Induction of physical dependence on morphine, codeine, and meperidine in the rat by continuous infusion, J. Pharmacol. Exp. Ther. 190, 408.

Verberne, A.J.M., D.A. Taylor and M.R. Fennessy, 1980, Withdrawal-like behaviour induced by inhibitors of biogenic amine reuptake in rats treated chronically with Δ^9-tetrahydrocannabinol, Psychopharmacology 68, 261.