METABOLISM AND DISPOSITION OF Δ⁹-TETRAHYDROCANNABINOL IN MAN

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Δ⁹-Tetrahydrocannabinol (Δ⁹-THC) has been shown to be the active component of marihuana and hashish (17, 25–27). The Δ⁹-THC content of these materials varies depending upon the source of the *Cannabis* plant. Good quality marihuana of Mexican origin contains about 1.5 % of THC, whereas hashish (the resin obtained from the flowering tops) contains approximately 5 to 10 times more THC. Preparations from other geographic regions, *e.g.*, Jamaica or Vietnam, contain more Δ⁹-THC. Previously, Δ⁹-THC utilized for research purposes had been extracted from the crude plant. However, this compound has recently been synthesized chemically (29), and synthesized radiolabeled material[3] is now available. This has made possible studies of the disposition and metabolism of Δ⁹-THC in animals (1–3, 7, 16, 18, 22, 28, 34, 35) and man (23, 24).

Disposition of Δ⁹-THC. Initially, studies in man were conducted on normal volunteers who professed no previous exposure to *Cannabis* and who had no history of recent administration of other drugs or medications. The subjects were given 0.5 mg of ¹⁴C-Δ⁹-THC (specific activity of 17.5 μc/mg; labeled in the two and four positions on the benzene ring) prepared under aseptic conditions in absolute alcohol for parenteral injection. The radioactive drug was slowly injected over an interval of 1 min into the tubing of a rapidly flowing intravenous infusion of 5 % dextrose in water. Blood samples were drawn at various times thereafter and analyzed for Δ⁹-THC, total radioactivity and ether-extractable radioactivity. The results from three non-users are shown in figure 1. After the intravenous administration of Δ⁹-THC, the plasma levels of unchanged ¹⁴C-Δ⁹-THC, total radioactivity and the radioactivity in an ether extract at first declined rapidly during the first few hours, presumably as a consequence of the redistribution and uptake into tissues. After equilibration the plasma levels of all three fractions declined more slowly. The Δ⁹-THC persisted in plasma for at least 3 days. Metabolites of Δ⁹-THC, as depicted by total radioactivity and ether-extractable radioactivity, were present in plasma within 10 min after the intravenous administration of the drug and also persisted for at least 3 days. Chromatography of a heptane extract of plasma pooled from various time periods

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[3] The ¹⁴C-Δ⁹-THC has been synthesized by Dr. Monroe E. Wall (Research Triangle Institute, North Carolina) and made available through Drs. John A. Scigliano and Monique C. Braude of the National Institute of Mental Health.
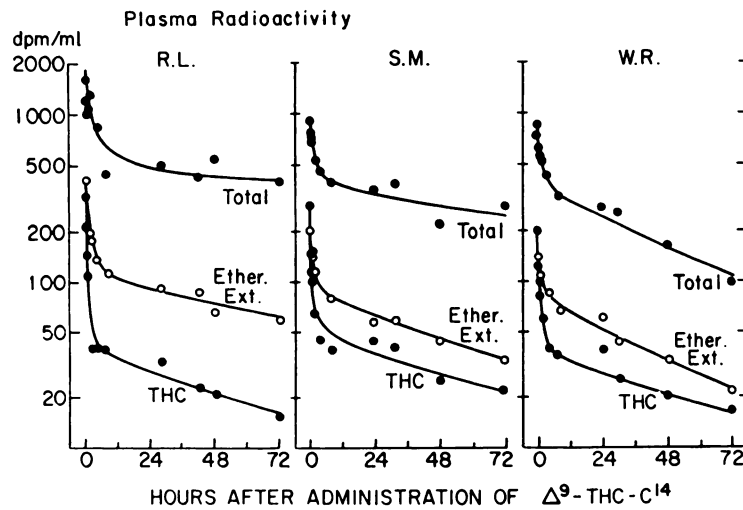
Fig. 1. Plasma levels of Δ^9-tetrahydrocannabinol (Δ^9-THC), total radioactivity and ether-extractable radioactivity in non-users after the intravenous injection of ^{14}C-Δ^9-THC. Three normal volunteers were given 0.5 mg of ^{14}C-Δ^9-THC in 1 ml of ethanol. The radioactive solution was injected during a 1-min interval into the tubing of a rapidly flowing intravenous infusion of 5% dextrose in water. Blood samples were drawn in heparinized syringes from the opposite arm at various times. Plasma was assayed for Δ^9-THC, total radioactivity and ether-extractable radioactivity by liquid scintillation spectrometry.

confirmed the presence of Δ^9-THC in plasma at all times examined (fig. 2). The presence of 11-hydroxytetrahydrocannabinol in plasma was also confirmed by chromatography (fig. 2). 11-Hydroxy-tetrahydrocannabinol has been demonstrated to be as active or even more active than Δ^9-THC in certain animal tests (7, 34) and could possibly be the active form of Δ^9-THC *in vivo*.

Studies identical to those performed in non-users of marihuana were repeated on subjects who had a history of chronic marihuana usage, defined for our purposes as smoking marihuana approximately once daily for at least 1 year immediately before the investigation. They had no history of habitual ingestion of other drugs or medications during the past year and, in general, did not appear to differ from the normal volunteers except for their history of exposure to *Cannabis*. The subjects smoked marihuana 12 to 14 hr before the intravenous administration of the Δ^9-THC and did not smoke during the remainder of the study. As in studies with non-smokers, plasma levels of Δ^9-THC, total radioactivity and ether-extractable radioactivity declined more rapidly during the early time periods than during the later times (fig. 3). The plasma half-life of Δ^9-THC was significantly shorter in chronic *Cannabis* users than in normal volunteers, both at the early time periods (table 1) and the later time periods (table 2). To determine whether this was the result of different patterns of distribution and tissue binding in the two groups of subjects, apparent volumes of distribution were calculated. There appeared to be no significant difference in the volume of distribution between the users and non-users for Δ^9-THC and total radioactivity,

indicating a similar distribution of the ^{14}C-Δ^9-THC in both groups (table 3). This suggests that the difference in plasma half-lives was due to different rates of metabolism.

Excretion of Δ^9-*THC.* In man, Δ^9-THC was extensively metabolized to more polar compounds which were excreted in the urine and feces. Urinary excretion
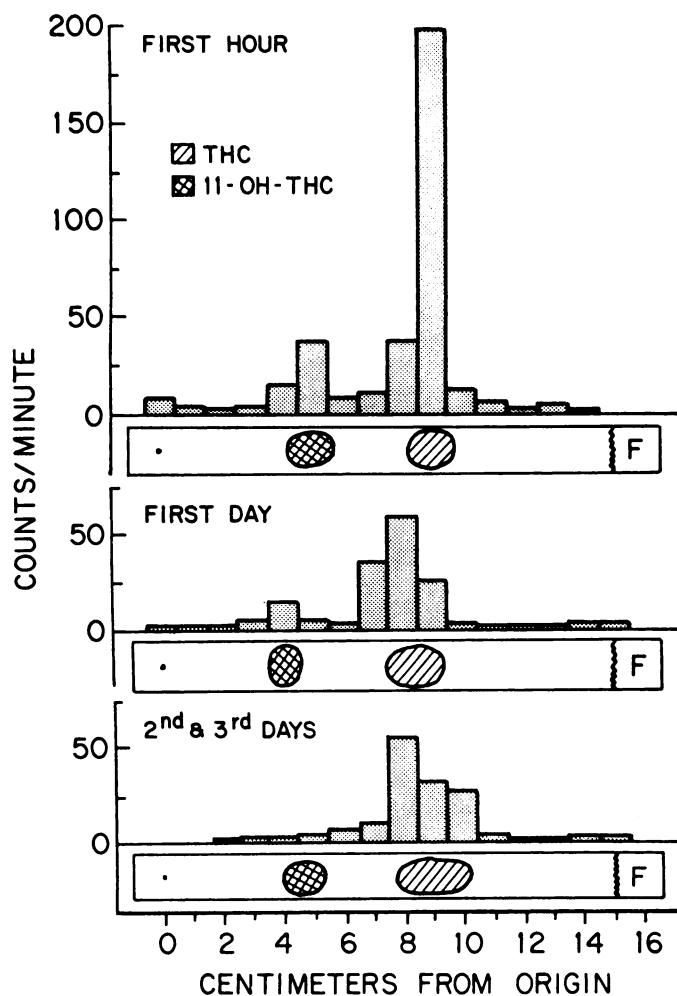

FIG. 2. Radiohistogram of thin-layer chromatography of Δ^9-THC extracted from plasma at various times. R.L. represents a typical subject. Aliquots of plasma from the 1st hr, from the remainder of the 1st day and from the 2nd and 3rd days were pooled separately and extracted with four volumes of a solution of heptane containing 1.5% isoamyl alcohol. The extract was evaporated to dryness *in vacuo*, redissolved in a small volume of ethanol, applied to an Eastman silica-gel chromatogram sheet and developed in a hexane:acetone (3:1) system. Authentic Δ^9-THC and 11-hydroxy-Δ^8-THC were cochromatographed with the heptane extract. The sheet was cut into 1-cm strips from the origin to the solvent front and placed in vials containing scintillation solution. Radioactivity was determined by liquid scintillation spectrometry.
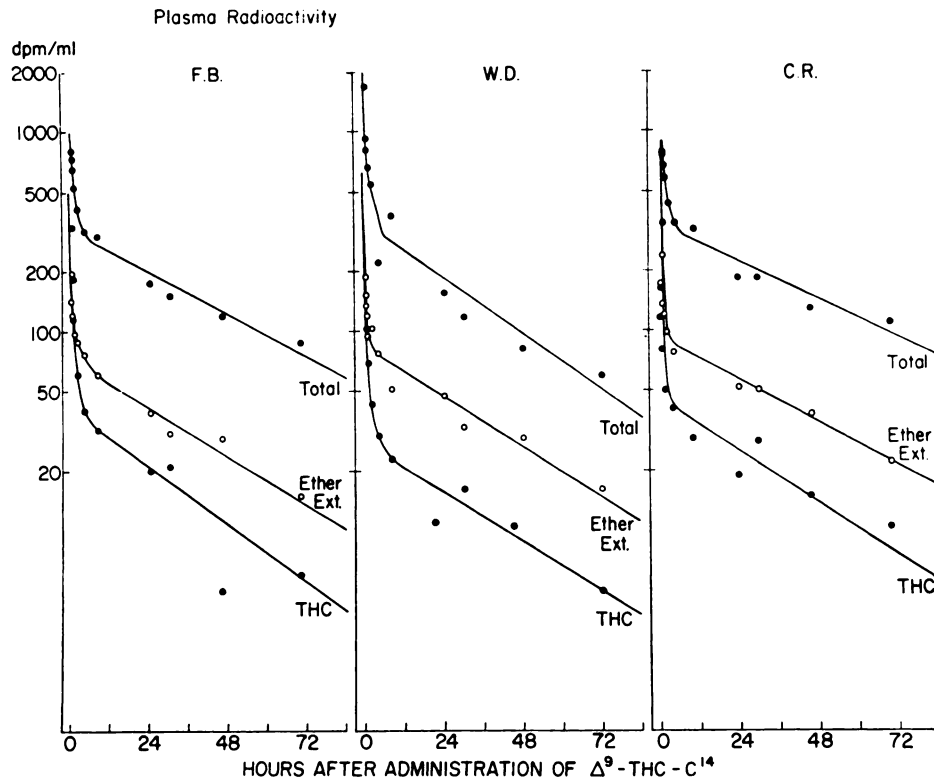
Fig. 3. Plasma levels of Δ^9-THC, total radioactivity and ether-extractable radioactivity in chronic *Cannabis* users after the intravenous injection of ^{14}C-Δ^9-THC. Three long-term *Cannabis* users were given a slow intravenous infusion of 0.5 mg of ^{14}C-Δ^9-THC in 1 ml of ethanol. The radioactive solution was injected during an interval of 1 min into the tubing of a rapidly flowing intravenous infusion of 5% dextrose in water. Blood samples were drawn in heparinized syringes from the opposite arm at various times. Plasma was assayed for Δ^9-THC, total radioactivity and ether-extractable radioactivity by liquid scintillation spectrometry.

of radioactivity was greatest during the initial 24 hr and then gradually tapered off during successive days. Similarly biliary excretion, reflected about 1 day later in the fecal excretion, was greatest during the first few days. Radioactive metabolites of Δ^9-THC were excreted for more than 7 days. The fecal route of excretion in man was more prominent than the renal route; however, both played a significant role (fig. 4). About 40 % of the total radioactivity was recovered from the feces of both non-users and chronic users during the 7-day collection period. However, radioactivity excreted in the urine accounted for about 30 % in chronic users and 22 % in non-users. Man appears to be intermediate between the rabbit, who excretes the major portion of radioactive metabolites of Δ^9-THC in the urine (2), and the rat, who excretes very little in the urine (1, 22). Significantly more radioactivity was excreted in the marihuana smokers' urine than in the non-smokers' urine (fig. 4). These findings also suggest that chronic users of mari-

TABLE 1

Fate of ¹⁴C-Δ⁹-tetrahydrocannabinol in chronic cannabis users and non-users during initial phases

Non-smoking Subjects	(A) $t_{1/2}$	(B) $t_{1/2}$	Chronic Smoking Subjects	(A) $t_{1/2}$	(B) $t_{1/2}$
	min	min		min	min
R. L.	16	120	W. D.	8	98
S. M.	16	142	F. B.	13	117
W. R.	11	146	C. R.	10	123
Mean	14.33 ± 1.66*	136 ± 8.08	Mean	10.33 ± 1.45*	112 ± 7.5

Values represent mean ± standard error. Plasma levels of ¹⁴C-Δ⁹-tetrahydrocannabinol (from 10-min to 4-hr samples) were plotted on semilogarithmic paper. This resulted in a biphasic curve, which was divided into its components (designated A and B) and half-lives for each phase calculated.

* $P < .01$.

TABLE 2

Fate of ¹⁴C-Δ⁹-tetrahydrocannabinol (Δ⁹-THC), total radioactivity and ether-extractable radioactivity in chronic cannabis users and cannabis non-users

	Non-Smokers				Chronic Smokers		
Subjects	Plasma half-lives (hr)			Subjects	Plasma half-lives (hr)		
	Total	Ether	THC		Total	Ether	THC
R. L.	78	67	49	W. D.	26	28	29
S. M.	82	53	66	F. B.	34	30	24
W. R.	42	42	52	C. R.	40	34	29
Mean	67 ± 13	54 ± 7*	56 ± 5†		33 ± 4	31 ± 2*	27 ± 2†

Plasma levels of ¹⁴C-Δ⁹-THC, total radioactivity, and ether-extractable radioactivity from 10 min to 72 hr were plotted on semilogarithmic paper. The half-lives were calculated from that phase representing metabolism (the slow phase).

* $P < .05$ (significant).

† $P < .001$ (highly significant).

TABLE 3

Apparent volumes of distribution of total radioactivity and Δ⁹-tetrahydrocannabinol (Δ⁹-THC)

	Non-Smokers			Chronic Smokers	
Subjects	Total Radioactivity	THC	Subjects	Total Radioactivity	THC
	liters	liters		liters	liters
R. L.	34	497	W. D.	58	742
S. M.	60	516	F. B.	63	498
W. R.	50	439	C. R.	60	453
Mean	48 ± 7.5	484 ± 23.3		60 ± 1.5	564 ± 90

Values represent mean ± standard error.

huana metabolize Δ^9-THC more rapidly to polar metabolites which are capable of being excreted in the urine.

Metabolism of Δ^9-*THC*. Studies were performed to investigate the metabolic products of Δ^9-THC in man. Urine contained essentially no unchanged Δ^9-THC and only a small quantity of 11-hydroxy-tetrahydrocannabinol. Enzymatic hydrolysis with glusulase (a mixture of β-glucuronidase and sulfatase) increased the proportion of 11-hydroxy-tetrahydrocannabinol; however, the major fraction of the urinary radioactivity consisted of more polar compounds. The effect of pH on the extraction of urinary radioactivity was examined in an attempt to characterize the metabolites of Δ^9-THC (table 4). Urine was adjusted to various pH's and extracted with several organic solvents. The percentage of extracted radioactivity increased as the pH of the urine was decreased, *i.e.*, became more
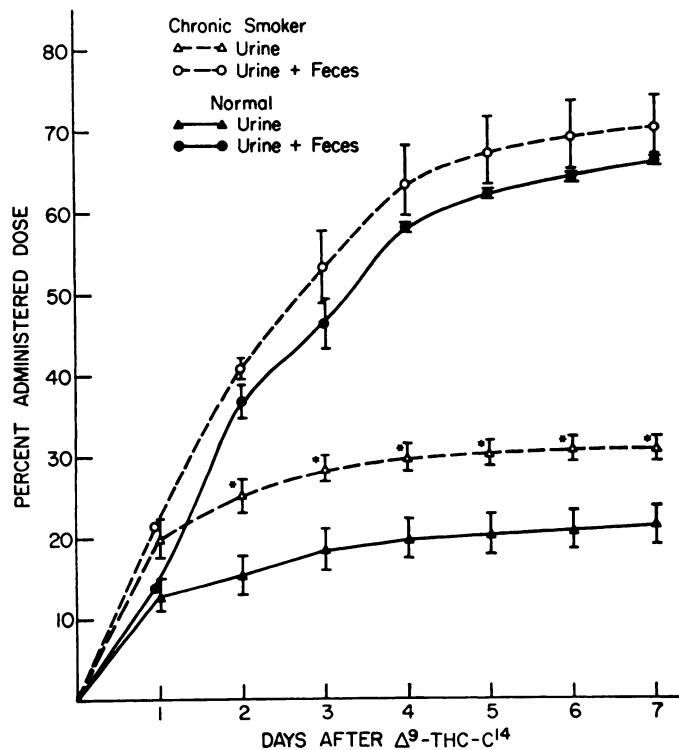
Fig. 4. Comparison of the cumulative excretion of radioactivity in chronic marihuana users and non-users after the intravenous injection of ^{14}C-Δ^9-THC. Three chronic users and three non-users were studied. Urine and feces were collected for at least 8 days after the intravenous administration of ^{14}C-Δ^9-THC. Urine and feces were frozen until analyzed. The feces were suspended in three volumes of methanol and vigorously shaken for 10 min on a mechanical shaker. The material was centrifuged, and an aliquot of the methanol extract was assayed for total radioactivity. Urine was assayed directly for total radioactivity by liquid scintillation spectrometry. Fecal radioactivity is represented by the difference between total radioactivity and urinary radioactivity. Significant differences are indicated (*).

acid. The largest percentage increase occurred in the ether-extraction step. We are presently attempting to identify the major urinary metabolites. The radioactivity present in feces did not show any difference with respect to the effect of pH on its extraction properties (table 5). 11-Hydroxy-tetrahydrocannabinol was found to be a metabolite of Δ^9-THC in feces. In several subjects this appeared to account for a considerable quantity of the fecal radioactivity (about 20 %).

Weil et al. (37), and Jones and Stone (21) have reported that after smoking marihuana the effects are maximum within 15 min, diminished between 30 min to 1 hr and largely dissipated by 3 hr. The plasma levels of Δ^9-THC and its metabolites seen after the intravenous injection of ^{14}C-Δ^9-THC suggest that the effects are terminated by redistribution and metabolism of the drug after the high initial levels. The presence of 11-hydroxy-tetrahydrocannabinol at the early times would be consistent with the hypothesis that 11-hydroxy-tetrahydrocannabinol is an active metabolite of Δ^9-THC (7, 16, 23, 25, 34). The more rapid disappearance of Δ^9-THC in chronic Cannabis smokers indicates that repeated exposure to marihuana can induce enzymes which are responsible for its metabolism. The classical work of Conney and Burns (4, 5, 8–11) and Remmer (30–32) demonstrated that certain drugs and polycyclic hydrocarbons can induce enzymes

TABLE 4

Effect of pH on the extraction of radioactivity from urine

	Percentage of Radioactivity Extracted			
	pH 6.5	pH 5.0	pH 4.0	pH 3.0
Heptane	1.3	3.3	5.2	8.0
Ether	6.6	34.7	45.5	61.2
Ethyl acetate	13.2	16.3	20.7	18.1
Total extracted	21.1	54.3	71.4	87.3
Residual	78.9	45.6	28.5	12.7

Values represent mean of four subjects (two users and two non-users) determined in duplicate.

TABLE 5

Effect of pH on the extraction of radioactivity from feces

	Percent Radioactivity Extracted	
	pH 6.5	pH 5.0
Heptane	66.5	65.3
Ether	16.1	18.6
Ethyl acetate	5.0	4.5
Total extracted	87.6	88.4
Residual	12.4	11.6

Values represent mean of four subjects (two users and two non-users) determined in duplicate.

which metabolize other drugs. Several studies in animals and in man have demonstrated that after long-term administration many drugs can induce their own metabolism (6, 14, 15, 20, 33). It is not unlikely that a similar effect occurs with chronic marihuana usage.

Many investigators (12, 13, 19, 21, 36) reported that the psychological effects of Δ^9-THC are delayed after oral administration when compared to administration via inhalation. To study the reason for this delay, Δ^9-THC-^{14}C was administered orally in combination with a pharmacological dose of non-radiolabeled Δ^9-THC and blood samples taken at various times thereafter. Preliminary studies
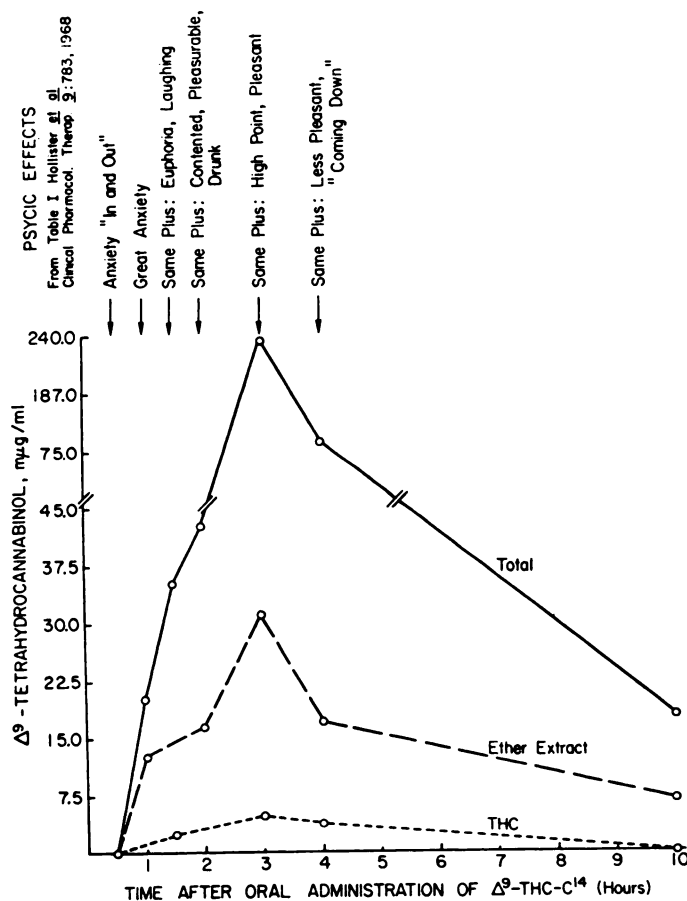
FIG. 5. Plasma levels of ^{14}C-Δ^9-THC, total radioactivity and ether-extractable radioactivity after the oral administration of 0.3 mg/kg of Δ^9-THC with 0.5 mg of ^{14}C-Δ^9-THC to a chronic *Cannabis* user. Blood samples were drawn at various times and plasma assayed for Δ^9-THC, total radioactivity and ether-extractable radioactivity by liquid scintillation spectrometry.

FIG. 5. (upper portion). Description of the time course for the psychic effects of Δ^9-THC after its oral administration (0.3 mg/kg). (Data extracted from L. E. Hollister, R. K. Richards and H. K. Gillespie, Clin. Pharmacol. Ther. 9: 783, 1968.

done in collaboration with Drs. James L. Weiss, August M. Watanabe and Philippe V. Cardon indicate that the plasma levels of Δ^9-THC, total radioactivity and ether-extractable radioactivity reach their maximum at 3 hr after ingestion of the drugs (fig. 5). The levels of Δ^9-THC and its metabolites in plasma correlated well with the psychic effects observed by Hollister et al. (19). After the oral administration of Δ^9-THC, very little unchanged Δ^9-THC was present in plasma whereas a large percentage of polar metabolites including 11-hydroxy-tetrahydrocannabinol were present. If this metabolite of Δ^9-THC were the active compound responsible for the effects of marihuana, then the alleged reverse tolerance to marihuana might be explained by the more rapid and extensive conversion of Δ^9-THC in chronic users of marihuana to the effective agent. Other possible explanations for this phenomenon could be due to an increased receptor sensitivity to Δ^9-THC, to a learned and heightened response to the effects of Δ^9-THC or to cumulative effects of repeated administration.

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