Life Sciences, Vol. 58, No. 25, pp. PL 365-372, 1996

0024-3205/96 $15.00 + .00

ELSEVIER

PII S0024-3205(96)00237-8

PHARMACOLOGY LETTERS
Accelerated Communication

GENETIC DIFFERENCES IN Δ^9-TETRAHYDROCANNABINOL-INDUCED FACILITATION OF BRAIN STIMULATION REWARD AS MEASURED BY A RATE-FREQUENCY CURVE-SHIFT ELECTRICAL BRAIN STIMULATION PARADIGM IN THREE DIFFERENT RAT STRAINS

Marino Lepore[1], Xinhe Liu[1], Virginia Savage[1], Daniel Matalon[1] and Eliot L. Gardner[1,2]

Division of Neuropsychopharmacology, Departments of [1]Psychiatry and [2]Neuroscience, Albert Einstein College of Medicine, New York, NY 10461-1602, U.S.A.

(Submitted September 6, 1995; accepted September 25, 1995; received in final form April 15, 1996)

Abstract. Lewis, Fischer 344, and Sprague-Dawley rats were implanted with electrodes in the medial forebrain bundle and trained to lever press for brain stimulation reward using a rate-frequency curve-shift electrical brain stimulation paradigm based on a series of 16 pulse frequencies ranging from 25 to 141 Hz in descending order. Once reward thresholds were stable, rats were given 1.0 mg/kg Δ^9-tetrahydrocannabinol (Δ^9-THC), the psychoactive constituent in marijuana and hashish, or vehicle, by intraperitoneal injection. Lewis rats showed the most pronounced Δ^9-THC-induced enhancement of brain reward functions. Sprague-Dawley rats showed an enhancement of brain reward functions that was approximately half that seen in Lewis rats. Brain reward functions in Fischer 344 rats were unaffected by Δ^9-THC at the dose tested. These results are consistent with previous work showing Lewis rats to be highly sensitive to the rewarding properties of a variety of drugs of abuse, including opiates, cocaine, and alcohol, while Fischer 344 rats are relatively less sensitive. They extend such previous findings to cannabinoids, and further suggest that genetic variations to other cannabinoid effects may also exist.

Key Words: **Δ^9-tetrahydrocannabinol, cannabinoids, genetic differences, brain stimulation reward**

Introduction

We have previously reported that Δ^9-tetrahydrocannabinol (Δ^9-THC), the psychoactive constituent in marijuana and hashish, enhances dopamine levels in reward-relevant limbic and forebrain dopaminergic loci, and that this effect is more pronounced in Lewis rats than in Fischer 344 rats (1,2). Recently, attention has focused on genetic differences in vulnerability to abused drugs. Differences in neurotransmitter synthesis, transport, and release, as well as receptor, second messenger, and immediate early gene function have been documented in the reward-related dopamine neurons in the Lewis as compared to the Fischer 344 strain (3,4). These differences have been proposed to constitute the neurobiological substrate of the phenotypic vulnerability to drugs of abuse shown by Lewis as compared to Fischer 344 rats (3,4). As shown by many previous studies, the essential neuropharmacological commonality amongst drugs of abuse (with the possible

Corresponding author: Dr. Eliot L. Gardner, Program in Brain and Behavior, Division of Neuropsychopharmacology, Forchheimer Building G-49, Albert Einstein College of Medicine, 1300 Morris Park Avenue, New York, NY 10461-1602, USA; Telephone: 718-430-3737; FAX: 718-430-8772; E-mail: gardner@aecom.yu.edu.

exception of the LSD-like hallucinogens) is their ability to acutely enhance brain reward functions, presumably deriving their euphorigenic properties and abuse potential therefrom (5). We previously showed that this brain reward enhancing property extends to Δ^9-THC, using a titrating threshold electrical brain stimulation procedure (6). In those experiments, we observed a brain reward enhancing threshold shift after Δ^9-THC administration in Lewis rats, but did not methodically test other rat strains (6). In the present study, we examined whether strain-specific differences in Δ^9-THC-induced enhancement of brain reward functions would be seen, testing three different strains of laboratory rat. We used the rate-frequency curve-shift electrical brain stimulation paradigm (7,8), another quantitative brain reward threshold measurement paradigm that is methodologically different from the titrating threshold brain stimulation procedure, but which was designed to achieve a similar goal - measurement of brain reward thresholds independent of motor effects, and capable of discriminating the separate neurobiological substrates of motivation and reward that may be differentially affected by neuropharmacological manipulations (9-11).

Methods

Animals and surgical preparation: Adult male Lewis (LEW), Fischer 344 (F344), and Sprague Dawley (SD) rats (Charles River, MA) were anesthetized with pentobarbital (55 mg/kg) and a monopolar stainless steel electrode (Plastics One, VA), aimed at the medial forebrain bundle at the level of the lateral hypothalamus, was chronically implanted in the brain of each rat using standard surgical and stereotaxic techniques. The electrode was insulated to within 0.5 mm of the tip, and a stainless steel wire wrapped around several stainless steel skull screws served as a current return. The implant stereotaxic coordinates (12) were -2.56 mm posterior to Bregma, $\pm$ 1.9 mm from the midline, and -8.6 mm from the skull surface. Correct placement of electrodes was confirmed by standard histology following completion of experiments. Rats were allowed at least 1 week recovery before testing.

Apparatus: Animals were tested in a conventional operant chamber modified for delivery of brain stimulation and equipped with a retractable lever on which the rat pressed to obtain stimulation. Brain stimulation was delivered by an electrically isolated constant current stimulator. Trains of 0.1 msec cathodal pulses, of 250 msec duration, were delivered contingent on a single lever press. A PC-AT microcomputer monitored the rats' responses, recorded data, and set appropriate current and pulse frequencies (see below). The microcomputer-driven brain stimulation apparatus was essentially identical to that previously described by Gallistel and colleagues (8).

Drugs: Δ^9-THC (National Institute on Drug Abuse) dissolved in 100% ethanol (20 mg/ml) was prepared for injection by evaporating the Δ^9-THC-ethanol solution under a stream of nitrogen gas. A 0.2 ml volume of 100% ethanol was then added to the concentrated Δ^9-THC, and this Δ^9-THC-ethanol solution was suspended in 45% 2-hydroxypropyl-β-cyclodextrin (β-CD) at a concentration of 1.0 mg/ml. This final solution was then used for Δ^9-THC injections. A 45% β-CD solution containing 0.2 ml of 100% ethanol was used for vehicle injections. All Δ^9-THC or vehicle solutions were injected intraperitoneally in a volume of 1.0 ml/kg body weight. All Δ^9-THC injections were given at a dose of 1.0 mg/kg, which we previously found optimal for enhancing electrical brain stimulation reward functions (2,6), for enhancing dopamine levels in reward-relevant forebrain dopaminergic loci (1,2), and for producing Δ^9-THC-induced conditioned place preference (13).

Procedure: Animals were screened for electrical brain-stimulation behavior using 250 msec trains of 150 Hz stimulation, with the current set at 200 μA. Once animals acquired self-stimulation behavior, the current was adjusted to produce moderate rates of responding (45-60 responses/30

sec). Animals were then trained using a rate-frequency procedure (8). Animals were trained to lever press for a series of 16 different pulse frequencies, ranging from 25 to 141 Hz, presented to the animal in descending order. At each separate pulse frequency, animals responded for two 30-second time periods ("bins"), following which the pulse frequency was decreased by 0.05 log units. Following each 30-second bin, the lever was retracted for 10 seconds. The two 30-second "bins" at each frequency were tested consecutively (rather than the whole range of frequencies being tested sequentially twice). Animals were trained on this rate-frequency procedure until reward thresholds were stable. Two commonly used measures of reward threshold were used: M_{50} - the pulse frequency at which the animals responded at half-maximum (7,14,15); and Θ_0 - the frequency at which animals failed to respond for rewarding stimulation (7,14), a reliable neurophysiological and psychophysical index of stimulation efficacy (7,14,16). Reward thresholds were considered stable when M_{50} and Θ_0 were within 0.01 log units of their respective previous values for three consecutive days. Of the larger number of animals surgically implanted with stimulation electrodes in the desired brain locus and trained on the rate-frequency brain stimulation reward procedure, 5 animals in each group reached this level of reward threshold stability. When this stability was reached, animals were injected with the vehicle solution and tested in the brain reward procedure. The next day, animals were injected with Δ^9-THC and tested in the brain reward procedure. On each test day, testing began 20 minutes after vehicle or Δ^9-THC injection.

Sigmoid curve-fitting of rate-frequency brain stimulation reward functions: Each rate-frequency brain stimulation reward function generated by a given animal over a given descending series of pulse frequencies was mathematically fitted, by iterative computer programs derived from the Gauss-Newton algorithm for nonlinear regression, to three different sigmoid curve-fitting mathematical growth models that appear to accurately fit rate-frequency brain stimulation reward functions (14). The three curve-fitting models (14) used were the Gompertz model ($Y'=ae^{-e^{(b-cX)}}$), the logistic model ($Y'=a/[1+e^{(b-cX)}]$), and the Weibull function ($Y'=a[1-e^{-(bX)^c}]$); where Y' is the rate of response (number of lever presses for rewarding brain stimulation per unit of time), X is the pulse frequency, and a, b, and c are parameters approximated from each empirical rate-frequency data curve (a representing the asymptotic response rate value, b relating to the intercept of the rate-frequency curve with the Y axis, and c representing the rate at which Y increments). From each curve-fitting model, a solution for M_{50} and Θ_0 was obtained. Thus, for each rate-frequency brain stimulation reward function generated by a given animal over a given descending series of pulse frequencies, three solutions for M_{50} and Θ_0 were obtained. The three solutions for M_{50} were averaged and the three solutions for Θ_0 were averaged, to produce a mean M_{50} and a mean Θ_0 for each rate-frequency brain stimulation reward function generated by a given animal over a given descending series of pulse frequencies. The mean M_{50} values and mean Θ_0 values were then subjected to statistical analyses.

Statistical analyses: Two-way analyses of variance were performed on maximum response rates (Y_{max}), mean M_{50} values, and mean Θ_0 values under baseline, vehicle, and drug conditions (17). Post-hoc comparisons were made using Tukey's t-test (17).

Results

Figure 1 shows the mean rate-frequency brain stimulation reward functions in the medial forebrain bundle as affected by systemic injections of 1.0 mg/kg Δ^9-THC in the three different rat strains. Table 1 gives the mean Y_{max}, M_{50}, and Θ_0 values ($\pm$ S.E.M.) generated by the three rat strains under vehicle and Δ^9-THC conditions.

A two-way analysis of variance (ANOVA) with repeated measures was performed on the maximum

response rate (Y_{max}) per 30 sec bin. No differences were found in Y_{max} between the three strains of rats ($F_{(2,8)}=2.39$, $p>0.05$). Furthermore, Δ^9-THC administration did not produce any reliable differences in Y_{max} ($F_{(2,8)}=2.15$, $p>0.05$), and no significant interaction between rat strain and drug treatment was found with respect to Δ^9-THC effects on Y_{max} ($F_{(4,16)}=0.961$, $p>0.05$). These results indicate that the maximum number of responses emitted within a 30 second period were not dependent on either rat strain or Δ^9-THC administration.

A two-way ANOVA with repeated measures performed on the mean Θ_0 values revealed a highly significant effect of Δ^9-THC ($F_{(2,8)}=12.92$, $p<0.01$), indicating that reward thresholds (as measured by Θ_0) were significantly lowered following Δ^9-THC administration. A significant interaction between rat strain and drug treatment was also observed with respect to Θ_0 ($F_{(4,16)}=28.97$, $p<0.0001$). Post hoc analyses using Tukey's t-test revealed that no significant differences existed in baseline Θ_0 threshold measures between rat strains (all p values >0.05), and that Δ^9-THC did not affect Θ_0 threshold measures in the F344 rat strain (all p values >0.05). In the LEW rat strain, Δ^9-THC significantly lowered the Θ_0 threshold measure when compared to vehicle administration ($t=9.18$, $p<0.001$) or baseline ($t=9.08$, $p<0.001$). Δ^9-THC also significantly lowered Θ_0 threshold measures when compared to vehicle ($t=6.56$, $p<0.001$) or baseline ($t=6.96$, $p<0.001$) in the SD rat strain. The difference between Δ^9-THC's effects on Θ_0 reward thresholds in the LEW strain and in the F344 strain was highly significant ($t=14.94$, $p<0.001$). Further, the Δ^9-THC-induced lowering of brain reward Θ_0 thresholds was significantly greater in the LEW strain than in the SD strain ($t=3.41$, $p<0.05$).

TABLE 1.
Mean Y_{max}, M_{50}, and Θ_0 Values (± S.E.M.) Generated by Three Rat Strains after Injection with Vehicle or Δ^9-THC

		LEW	SD	F344
Y_{max}	VEHICLE	41.5 ± 2.8	32.4 ± 4.3	38.4 ± 6.3
	Δ^9-THC	39.9 ± 3.1	32.2 ± 4.0	36.2 ± 7.8
M_{50}	VEHICLE	81.5 ± 5.3	74.4 ± 2.5	79.6 ± 4.6
	Δ^9-THC	72.1 ± 3.7	73.8 ± 2.3	80.1 ± 4.4
Θ_0	VEHICLE	67.9 ± 4.5	67.7 ± 2.3	72.6 ± 4.1
	Δ^9-THC	60.5 ± 4.7	63.2 ± 2.8	75.1 ± 4.1

A two-way ANOVA with repeated measures performed on the mean M_{50} values indicated that Δ^9-THC significantly affected M_{50} threshold measures ($F_{(2,8)}=10.20$, $p<0.01$), and that there was a significant interaction between rat strain and M_{50} thresholds ($F_{(4,16)}=6.23$, $p<0.01$). Post hoc tests using Tukey's t-test revealed that no significant differences in baseline M_{50} threshold measures existed between the three rat strains (all p values >0.05). Further, Δ^9-THC did not lower M_{50} values in either the F344 or SD rat strains (all p values >0.05). Δ^9-THC lowered M_{50} brain reward threshold values in only the LEW rat strain when compared to baseline ($t=5.26$, $p<0.01$) or vehicle ($t=5.47$, $p<0.01$). Further, M_{50} values were significantly lower following Δ^9-THC administration in the LEW strain rats as compared to the F344 rats ($t=11.54$, $p<0.001$) and in the LEW strain rats as compared to the SD rats ($t=7.96$, $p<0.001$).

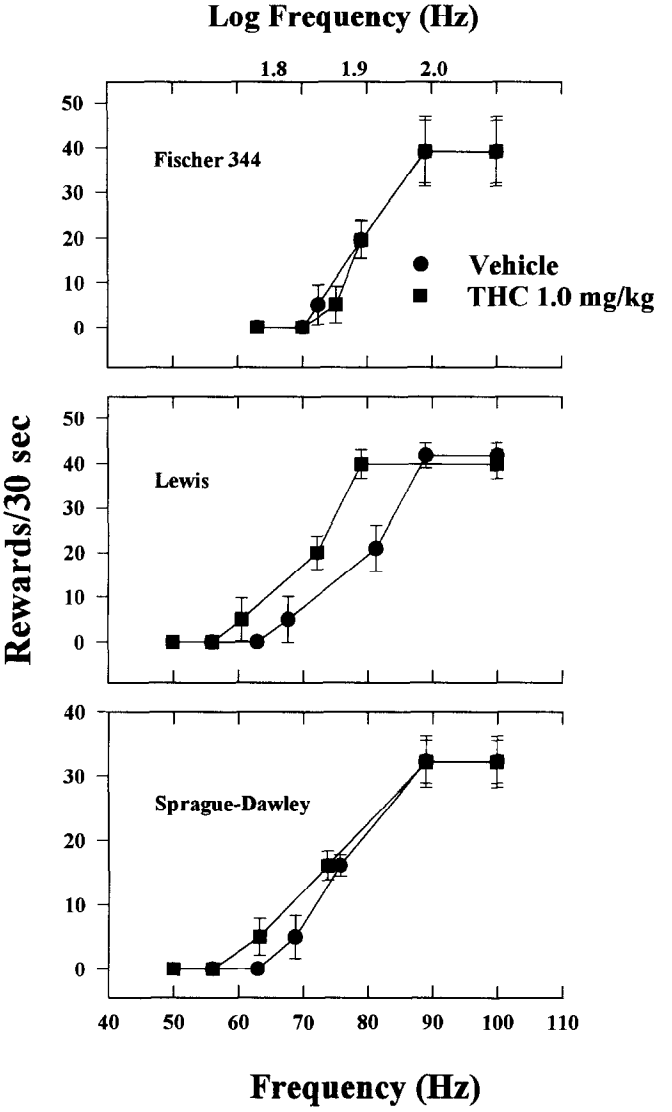

Fig. 1

Mean rate-frequency brain stimulation reward functions in the medial forebrain bundle as affected by systemic injections of 1.0 mg/kg Δ^9-tetrahydrocannabinol (Δ^9-THC) in three different rat strains. In Fischer 344 rats, Δ^9-THC did not significantly affect the brain stimulation reward function curve. In Lewis rats, Δ^9-THC significantly shifted the reward function curve to the left (i.e., enhanced brain reward), as indicated by significant leftward shifts in both M_{50} and Θ_0 points (see text). In Sprague-Dawley rats, Δ^9-THC enhanced brain stimulation reward only in terms of a significant leftward shift in the Θ_0 point (see text). Although animals were tested at 16 different pulse frequencies from 25 to 141 Hz, only the central portion of each mean rate-frequency brain stimulation reward function is depicted in this figure, as rewards/30 sec were asymptotic for all animals below 50 Hz and above 100 Hz.

Discussion

For many drugs of abuse, genetic differences influence drug preference, propensity for drug self-administration, and ease of drug-conditioning to previously neutral environmental cues (3,4,18, 19,22). These genetic differences presumably work through a neurobiological substrate to produce the observed phenotypic differences at the behavioral pharmacology level (3,5). Since the essential neuropharmacological commonality amongst abused drugs of different classes appears to be their ability to enhance brain reward functions (5,20), it seems reasonable to hypothesize the existence of genetic differences in the effects of abuse-prone drugs on the neurobiological substrate(s) encoding and subserving brain reward functions. The present findings of clear genetic differences in the effects of Δ^9-THC, the psychoactive constituent of marijuana and hashish, on brain reward thresholds measured directly by the rate-frequency curve-shift method, appears to support such an hypothesis. It is very interesting that rats of the inbred strain (LEW) shown in the present study to be most vulnerable to Δ^9-THC, in terms of reward-threshold shifts in the medial forebrain bundle neurons carrying reward-related neural signals from the ventral tegmental area to the nucleus accumbens (5,21), also: a) learn opiate or cocaine self-administration more readily, b) work harder for opiate or cocaine self-administration, c) place-condition for opiates or cocaine more readily, and d) show high ethanol preference, all in comparison to rats of other strains (3,4,18,19, 22). This suggests that LEW rats may have a generalized vulnerability to the rewarding effects of abused drugs, and that this vulnerability may derive from an augmented sensitivity to abused drugs of the brain-reward neural circuit of the ventral tegmental area - medial forebrain bundle - nucleus accumbens axis. At the same time, the relative *lack* of sensitivity of F344 rats to Δ^9-THC shown here should not be over-interpreted. First, only one dose of Δ^9-THC was tested, and the presently-observed failure of F344 rats to respond to Δ^9-THC with shifts in brain reward functions may reflect *relative*, rather than total, lack of sensitivity of brain reward mechanisms in F344 rats to Δ^9-THC. At higher Δ^9-THC doses, F344 rats might show shifts in brain reward functions similar to those in LEW and/or SD rats. In fact, in one of our prior studies of Δ^9-THC-induced enhancement of dopamine levels in reward-relevant forebrain loci (1), a strain difference in Δ^9-THC-induced effects likely to reflect a *relative*, rather than total, difference in response to Δ^9-THC between strains *was* seen (i.e., the difference between strains could be partially overridden by increased dose of Δ^9-THC). Second, the mean rate-frequency brain stimulation reward functions generated in the present study by F344 rats under vehicle conditions are reasonably similar to those generated by LEW and SD rats under vehicle conditions (figure 1). Thus, the reward system in the F344 strain would appear to function at least approximately as well (basally) as in the LEW and SD strains. So, the present strain differences would appear to reflect differences in responsiveness to drug rather than differences in basal brain reward functions. Also to be considered is the possibility that the presently-observed strain differences in response to Δ^9-THC could reflect pharmacokinetic, rather than pharmacodynamic, differences. We previously raised this possibility for our findings of strain differences in enhancement of dopamine levels in reward-relevant forebrain loci by Δ^9-THC (1), although there is *no* evidence for strain differences in Δ^9-THC absorption, distribution, biotransformation, and/or excretion. In fact, in the present study, essentially *identical* degrees of sedation and responsiveness to environmental stimuli were observed in rats of the three strains while under the influence of Δ^9-THC. Therefore, we are inclined to the belief that the present results reflect genetic differences in dopaminergic neuronal sensitivity and/or dopaminergic regulatory responsiveness to Δ^9-THC, as we have previously suggested (1,2).

With specific respect to Δ^9-THC and the psychoactive cannabinoids of which it is a constituent (e.g., marijuana), the present finding that Δ^9-THC significantly augments brain-stimulation reward as measured by the rate-frequency curve-shift quantitative electrophysiological threshold paradigm appears to confirm our previous findings that Δ^9-THC augments brain-stimulation reward as

measured by a titrating threshold brain stimulation procedure (2,6). We have also previously reported that Δ^9-THC dose-dependently enhances neurotransmitter (dopamine) efflux, as measured by both *in vivo* brain microdialysis and *in vivo* brain electrochemistry, in reward-relevant dopaminergic forebrain loci (1,2,23-25). We have further shown that this dopamine-enhancing action of Δ^9-THC occurs locally within the terminal fields of the ventral tegmental area - medial forebrain bundle - nucleus accumbens brain reward axis (26). In addition, we recently showed that Δ^9-THC produces a conditioned place preference similar to that produced by morphine and cocaine (13). These neuropharmacological and psychopharmacological actions of Δ^9-THC would appear to constitute a powerful neural substrate for marijuana's euphorigenic effects, and, therefore, for its abuse potential. Thus, marijuana - which for many years was felt to be an "anomalous" or "atypical" drug of abuse (i.e., was felt not to augment brain reward functions like other abuse-prone compounds) - may in fact not be "anomalous" or "atypical" at all.

The rate-frequency curve-shift brain stimulation reward paradigm used in the present study is believed to be an effective tool for testing the specificity of drug effects on brain reward (7,8,10, 11,15). A decrease in asymptotic performance (Y_{max}) is believed to indicate that the drug has interfered with the animal's capacity to perform the task, without affecting the rewarding potency of the stimulation (7,15). Conversely, lateral shifts in the rate-frequency curve (as measured by either the Θ_0 or M_{50} values) are believed to indicate that the drug has altered the rewarding potency *per se* of the brain stimulation, with leftward curve-shifts indicating enhanced reward potency and rightward curve-shifts indicating diminished reward potency (7,8,10,11,15). In the present study, the fact that, in the genetically vulnerable strains (especially the LEW), Δ^9-THC produced a leftward curve-shift without altering Y_{max} suggests that, at the dose used, Δ^9-THC has a specific effect on brain reward independent of performance or motivational effects.

As noted above, the titrating threshold electrical brain stimulation reward paradigm and the rate-frequency curve-shift brain stimulation reward paradigm are two different quantitative electrophysiological procedures for measuring brain reward thresholds independent of motor effects, and for discriminating the separate neurobiological substrates of motivation and reward that may be differentially affected by drugs. As also noted, we previously found that Δ^9-THC enhances brain reward functions as measured by the titrating threshold technique (2,6). The present finding that Δ^9-THC enhances brain reward functions as measured by the methodologically very different rate-frequency curve-shift brain reward paradigm thus appears to add additional validity to our initial findings with the titrating threshold procedure.

Finally, Δ^9-THC and other cannabinoids produce an extraordinarily wide range of pharmacologic actions - including analgesia, anti-emetic effects, anti-glaucoma effects, bronchodilation, anticonvulsant effects, and anti-ataxic effects (27,28). Given the present findings of significant genetic variation in Δ^9-THC's effect on brain stimulation reward, the possibility arises that significant genetic variations may also exist to other cannabinoid-induced pharmacologic actions.

Acknowledgements

This work was supported by National Institute on Drug Abuse research grant DA03622 to E.L.G. We thank the Research Technology Branch, NIDA, for the supplies of Δ^9-THC. M.L. is an Aaron Diamond Foundation Research Fellow. V.S. is an undergraduate research scholar of the Sue Golding Graduate Division, Albert Einstein College of Medicine. We thank Dr. Roger Brown of the National Institute on Drug Abuse, Dr. Billy R. Martin of the Medical College of Virginia, and Dr. Raphael Mechoulam of the Hebrew University of Jerusalem for their encouragement. We also thank Dr. T. Byram Karasu, Silverman Professor and Chairman, Department of Psychiatry, Albert

Einstein College of Medicine, for his support of this work.

References

1. J. CHEN, W. PAREDES, J.H. LOWINSON and E.L. GARDNER, Neurosci. Lett. 129 136-140 (1991).
2. E.L. GARDNER and J.H. LOWINSON, Pharmacol. Biochem. Behav. 40 571-580 (1991).
3. E.J. NESTLER, Sem. Neurosci. 5 369-376 (1993).
4. X. GUITART, D. BEITNER-JOHNSON, D.W. MARBY, T.A. KOSTEN and E.J. NESTLER, Synapse 12 242-253 (1992).
5. E.L. GARDNER, Substance Abuse: A Comprehensive Textbook, 2nd edn., J.H. Lowinson, P. Ruiz, R.B. Millman and J.G. Langrod (Eds), 70-99, Williams & Wilkins, Baltimore (1992).
6. E.L. GARDNER, W. PAREDES, D. SMITH, A. DONNER, C. MILLING, D. COHEN and D. MORRISON, Psychopharmacology 96 142-144 (1988).
7. E. MILIARESSIS, P.-P. ROMPRÉ, P. LAVIOLETTE, L. PHILIPPE and D. COULOMBE, Physiol. Behav. 37 85-91 (1986).
8. K.A. CAMPBELL, G. EVANS and C.R. GALLISTEL, Physiol. Behav. 35 395-403 (1985).
9. C.H. BIELAJEW and T. HARRIS, J. Psychiatr. Neurosci. 16 109-114 (1991).
10. J.M. LIEBMAN, Neurosci. Biobehav. Rev. 7 45-72 (1983).
11. E. MILIARESSIS and P.-P. ROMPRÉ, Behav. Neurosci. 101 827-831 (1987).
12. G. PAXINOS and C. WATSON, The Rat Brain in Stereotaxic Coordinates, Academic Press, New York (1982).
13. M. LEPORE, S.R. VOREL, J. LOWINSON and E.L. GARDNER, Life Sci. 56 2073-2080 (1995).
14. D. COULOMBE and E. MILIARESSIS, Behav. Neurosci. 101 209-214 (1987).
15. D.E. EDMONDS and C.R. GALLISTEL, J. Comp. Physiol. Psychol. 87 876-883 (1974).
16. E. MILIARESSIS, P.-P. ROMPRÉ and A. DURIVAGE, Brain Res. Bull. 8 693-701 (1982).
17. B.J. WINER, Statistical Principles in Experimental Design, 2dn edn., McGraw-Hill, New York (1971).
18. T. SUZUKI, F.R. GEORGE and R.A. MEISCH, J. Pharmacol. Exp. Ther. 245 164-170 (1988).
19. T.A. KOSTEN, M.J.D. MISERENDINO, S. CHI and E.J. NESTLER, J. Pharmacol. Exp. Ther. 269 137-144 (1994).
20. R.A. WISE, Pharmacol. Biochem. Behav. 13 [suppl.1] 213-223 (1980).
21. R.A. WISE and M.A. BOZARTH, Brain Res. Bull. 12 203-208 (1984).
22. F.R. GEORGE and S.R. GOLDBERG, Trends Pharmacol. Sci. 10 78-83 (1989).
23. J.M. NG CHEONG TON, G.A. GERHARDT, M. FRIEDEMANN, A.M. ETGEN, G.M. ROSE, N.S. SHARPLESS and E.L. GARDNER, Brain Res. 451 59-68 (1988).
24. J. CHEN, W. PAREDES, J. LI, D. SMITH, J. LOWINSON and E.L. GARDNER, Psychopharmacology 102 156-162 (1990).
25. J. CHEN, W. PAREDES, J.H. LOWINSON and E.L. GARDNER, Eur. J. Pharmacol. 190 259-262 (1990).
26. J. CHEN, R. MARMUR, A. PULLES, W. PAREDES and E.L. GARDNER, Brain Res. 621 65-70 (1993).
27. W. HALL, N. SOLOWIJ and J. LEMON, The Health and Psychological Consequences of Cannabis Use (National Drug Strategy Monograph Series No. 25), Australian Government Publishing Service, Canberra (1994).
28. R. MECHOULAM (Ed), Cannabinoids as Therapeutic Agents, CRC Press, Boca Raton, Florida (1986).