CHAPTER 3

Marihuana

D.R. Compton, L.S. Harris, A.H. Lichtman, and B.R. Martin

A. Introduction

Among the drugs of abuse which are regulated under the United States Controlled Substances Act and the International Conventions, none has created more intense public debate and controversy than marihuana. Marihuana is one of many names given to the leaves and flowering tops of the plant *Cannabis sativa*. The plant grows in all temperate regions of this planet and has been used commercially as a source of fiber and oil. Wherever the plant grows people have learned to ingest the material for its introxicating effects. The usual routes of administration are by mouth or smoking. It has been estimated that, worldwide, more than one hundred million individuals are regular users of the plant material. However, accurate data on this situation are not available. In the United States, use data have been collected on a regular basis using two large surveys, the National Household Survey on Drug Abuse (Substance Abuse and Mental Health Services Administration 1994) and the Monitoring the Future Survey (Johnston et al. 1994) which covers eighth, tenth, and twelfth grade students in public and private schools. Figure 1 presents the data over time for lifetime, annual, 30 day, and daily use of marihuana among high school seniors in the United States (Johnston et al. 1994). As can be seen, use peaked from 1978 to 1980 and had been declining slowly up to 1992. From 1992 to 1993 there was a significant increase in all use categories. Even more disturbing was the fact that very similar trend data were reported for eighth and tenth graders. This was matched in 1992 and 1993 by a decrease in the reported "perceived risk" from use of the drug.

Data from the Household Survey revealed similar findings among those 12–17 years old. It is estimated that in 1993, 600 000 individuals in this age group used marihuana weekly. There was also an increase in reporting that "obtaining marihuana is fairly or very easy." These trends should give us early warning of increased future public health problems. This is especially true when one looks at the increasing concentrations of Δ^9-tetrahydrocannabinol (Δ^9-THC), the psychoactive principal found in confiscated samples of the plant material. This is illustrated in Fig. 2. During the years of peak use in the United States (1976–1980) the average Δ^9-THC content of confiscated cannabis was about 1.5%. There was a steady increase

C. R. Schuster et al. (eds.), *Pharmacological Aspects of Drug Dependence*

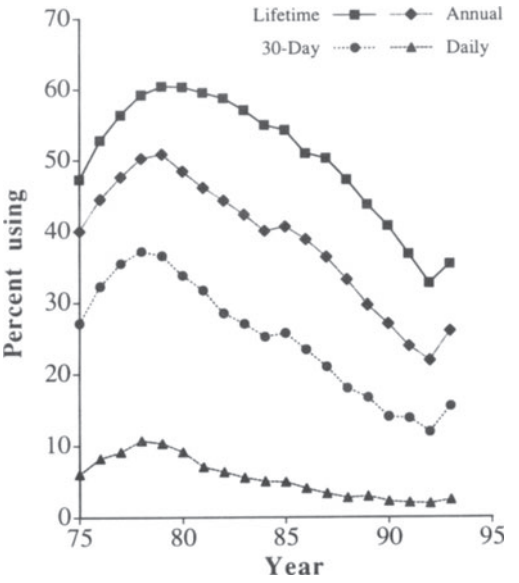

Fig. 1. Prevalence of marihuana use in high school seniors in the U.S. Incidence is defined as having used marihuana at least once during their lifetime, the last year, or the last 30 days or by daily use in the last 30 days

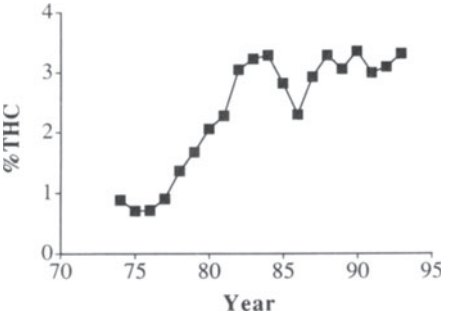

Fig. 2. Δ^9-Tetrahydrocannabinol (Δ^9-*THC*) content in confiscated marihuana in the U.S. The data are expressed as an average of the percentage in individual samples

to the mid-1980s, when it stabilized at 3.0%–3.5%, double that of earlier years (ElSohly and Ross 1994). It should be noted, however, that seized samples of buds and sinsemilla (flowering tops of the female *Cannabis* plant) have considerably higher concentrations of Δ^9-THC. Indeed, concentrations of Δ^9-THC as high as 20%–30% have been reported in individual samples. Thus, marihuana with a very high concentration of psychoactive Δ^9-THC is

regularly appearing on the street in the United States. We have every reason to believe that a similar situation exists internationally.

The general structure of the active principle of cannabis was elucidated in the 1940s. Indeed, very potent psychoactive cannabinoids were synthesized by ADAMS and colleagues. However, it remained for MECHOULAM and colleagues, in the 1960s, to first isolate in pure form and identify (−)-*trans*-Δ^9-tetrahydrocannabinol (Δ^9-THC) as the molecule primarily responsible for the psychoactive properties of the plant material. For decades the pharmacological activity of the cannabinoids was attributed to some nonspecific mechanism usually associated with its lipid solubility and disruption of cell membranes. A large body of structure-activity data and the demonstration of stereoselectivity gradually led to the postulation of more specific mechanisms. In recent years there has been an explosive advance in our knowledge of cannabis action. Specific binding sites have been demonstrated, receptors cloned and sequenced, purported endogenous ligands isolated and identified and, most recently, a competitive antagonist has been reported. This review will provide a relatively extensive and critical examination of these recent findings and will attempt to put them in context with our previous knowledge of the fascinating properties of this natural product.

B. Cellular and Molecular Effects

I. Neurochemistry

1. Effects on Neurotransmitters

a) Traditional Monoamine and Cholinergic Systems

Cannabinoids affect a wide variety of neurotransmitter pathways in the central nervous system and several of these share common second messenger systems, thus providing potential common sites for biochemical interactions mediated by the cannabinoid receptor. That cannabinoids can potentiate the actions of norepinephrine or acetylcholine by altering their receptors or second messenger systems has been the subject of numerous reviews (DEWEY 1986; PERTWEE 1990, 1992). However, neither adrenergic, dopaminergic, serotonergic or cholinergic agents (agonist or antagonist) bind directly to the CB_1 receptor (HOWLETT et al. 1992). The CB_1 receptor is the cannabinoid receptor primarily located in brain neuronal tissue, discussed in detail below.

Δ^9-THC decreases the release of acetylcholine from frog nerve via a presynaptic action, which was proposed to occur due to a decrease in the influx of calcium into nerve terminals (KUMBARACI and NASTUK 1980). Cannabinoids either suppress (NIEMI 1979), enhance (TURKANIS and KARLER 1986), or produce biphasic effects on neuronal transmission (TRAMPOSCH et al. 1981). Cannabinoids reportedly interact synergistically with cholinergic

agonists in the production of catalepsy, tremor, circling, salivation, lacrimation, hypothermia and drinking (PERTWEE 1990).

b) Dopamine and the Reward System

Cannabinoids have been shown to enhance the formation of dopamine (DA) and reported to stimulate the release of DA from rat corpus striatum, nucleus accumbens and medial prefrontal cortex. Enkephalinergic neurons synapse upon DA neurons in the nucleus accumbens, the site proposed to modulate the reward system for all addicting drugs. Thus, drugs that alter opioid activity in this region alter the release of DA, which may in turn underlie the rewarding properties of the drugs. Theoretically, naloxone should attenuate the rewarding properties of all addicting drugs. The interaction of cannabinoids with opioids in reward mechanisms in the brain has recently been reviewed (GARDNER 1992; GARDNER and LOWINSON 1991). Data indicate that cannabinoids interact with opioids allosterically, either presynaptically on the enkephalinergic neuron or on the opioid receptor of the DA neuron, to produce the reinforcing effects. This research endeavor has been particularly provocative in that traditionally this reward system has been closely linked to agents with strong reinforcing properties, such as morphine and cocaine. The reinforcing properties of cannabinoids have been more difficult to characterize and quantitate. Hence, these findings provide an additional avenue of pursuing the etiology of marihuana self-administration and placing it in the context of agents with high abuse potential. Thus, opioid/cannabinoid interactions may play an important role in the subjective effects of the cannabinoids, in addition to other pharmacological effects, as also discussed below.

Dopaminergic regulation of cannabinoid receptor mRNA levels in rat caudate putamem has been observed (MAILLEUX and VANDERHAEGHEN 1993a). Furthermore, recent desciptions of the interactions between the DA system and the endogenous cannabinoid, anandamide (discussed below), provide further credence for the existence of an interrelationship between the dopaminergic and cannabinoid systems (CHEN et al. 1993; GARDNER and LOWINSON 1991; NAVARRO et al. 1993; RODRIGUEZ DE FONSECA et al. 1992b).

c) Amino Acid Transmitters

A variety of cannabinoid-mediated effects have been attributed to modulation of amino neurotransmitter systems (PERTWEE 1990, 1992). Cannabinoids have been reported to enhance the turnover of γ-aminobutyric acid (GABA). Interpretation of the actions of cannabinoids on amino acid neurotransmitter synthesis has not been straightforward, because there is evidence that they inhibit as well as stimulate neurotransmitter reuptake. Evidence also suggests that cannabinoids can potentiate the actions of GABA by altering receptors or second messenger systems. Cannabinoids reportedly interact with GABA agonists in the production of catalepsy,

excitement, hypothermia and antinociception (Pertwee 1990). Recently, it has been reported that anandamide acts in a fashion similar to that of other cannabinoids to enhance GABAergic transmission (Wickens and Pertwee 1993).

d) Opioid Interactions

The cannabinoids produce effects which have much in common with the opioids, such as antinociception, hypothermia, catalepsy (in rats), cross-tolerance to morphine, and attenuation of naloxone-precipitated withdrawal from opiates. With regard to withdrawal symptoms, the interaction of the cannabinoids with the opiates is very ambiguous. As early as 1942, opioid withdrawal in humans was reported to be attenuated by marihuana administration (Adams 1942). Conversely, the irreversible μ antagonist chlornaltrexamine has been shown to decrease tolerance to Δ^9-THC (Tulunay et al. 1981). Blockade by cannabinoids of naloxone-precipitated withdrawal (jumping in opioid-tolerant animals) has been summarized (Martin 1986; Pertwee 1992). However, in morphine-tolerant mice, a series of both psychoactive and nonpsychoactive cannabinoids prevented withdrawal jumping (Bhargava 1976). Similarly, in morphine-tolerant rats, Δ^9-THC and cannabinol (CBN), but not cannabidiol (CBD, a marihuana constituent lacking pscyhoactive properties), attenuated withdrawal scores of a variety of behaviors (Chesher and Jackson 1985). While these findings should not be ignored, they raise the question whether this is a specific action of cannabinoids on opioid withdrawal and strongly suggest that the cannabinoid receptor is not involved. In a morphine-tolerant guinea-pig ileum model, Δ^9-THC attenuated withdrawal both in vivo and in vitro (using an ileum preparation). The in vitro effects of Δ^9-THC are presumably due to a decrease in acetylcholine release. Blockade of naloxone-precipitated withdrawal jumping has been shown to occur when cannabinoids are administered up to 24 h prior to the precipitated event, and specificity suggested by the fact that cannabinoids failed to alter the withdrawal (jumping behavior) following chronic amphetamine treatment (Bhargava 1978). However, it is unclear why Δ^9-THC was effective 24 h prior, while other cannabinoids were only effective for up to a 2 h pretreatment period. In morphine-tolerant rats, Δ^9-THC attenuated naloxone-precipitated "wet dog" shakes and defecation, but failed to attenuate any other behaviors associated with opioid withdrawal (Hine et al. 1975). Attenuation of morphine withdrawal signs by Δ^9-THC, nantradol, and nabilone have also been observed in the dog (Gilbert 1981). The mechanism by which cannabinoids attenuate opioid withdrawal in humans or in other animal species is unclear. However, cannabinoid-induced blockade of the release of various mediators of opioid withdrawal, such as acetylcholine and norepinephrine, has been proposed. Another possibility includes interactions of the cannabinoids and opioids with a common, possibly non-CB_1 related, second messenger system.

As in withdrawal studies, investigations of the cross-tolerance between the cannabinoids and opioids have also resulted in ambiguity. Although symmetrical cross tolerance between the opioids and cannabinoids have been shown in some studies (Hine 1985; Kaymakcalan and Deneau 1972), it was not observed in other studies measuring analgesia and/or heart rate. Cross-tolerance was not observed in pigeons (McMillan and Dewey 1972; McMillan et al. 1971), or in rats in nonanalgesic evaluations (Newman et al. 1974), or in shock avoidance measures (Newman et al. 1974). Asymmetrical cross-tolerance was observed whereby, in Δ^9-THC-tolerant mice, tolerance to the hypothermic effect of morphine was observed, though this was not so for antinociception (Bloom and Dewey 1978). In morphine-tolerant mice, cross-tolerance to the antinociceptive effects of Δ^9-THC has been demonstrated, but there was no cross-tolerance to the hypothermic effect of Δ^9-THC. Similar asymmetric cross-tolerance was observed using measures of motor activity in rats (Tulunay et al. 1982).

Similarities between the opioids and cannabinoids include their antinociceptive properties. Although there were early reports of cannabinoid-induced antinociceptive properties (Bloom and Dewey 1978; Sofia et al. 1973), it became clear that route of administration plays a critical role in the expression of this pharmacological property (Martin 1985a). Early results on the antinociceptive effects of the cannabinoids following injection into spinal sites (Gilbert 1981; Yaksh 1981), which has recently been investigated more extensively (Welch 1993; Welch and Stevens 1992), point to the participation of spinal sites in this cannabinoid action. The antinociceptive effect of intrathecal (i.t.) CP-55,940, a potent cannabinoid (Johnson and Melvin 1986), was attenuated in spinal-transected rats, indicating that cannabinoid-induced antinociception was mediated at both spinal and supraspinal sites (Lichtman and Martin 1991). However, Δ^9-THC (i.t.) produced the same degree of antinociception in mice that were spinal-transected as those that had the spinal cord intact (Smith and Martin 1992), suggesting that the effects of i.t. administered Δ^9-THC in mice are spinally mediated. The possibility remains that central components may also play a role following administration by other routes.

In vivo, the opioid antagonist naloxone has been shown to attenuate the antinociceptive effects of 11-OH-Δ^9-THC (Wilson and May 1975). However, many investigators have shown that naloxone fails to block the effects of various parenterally administered cannabinoids (Chesher et al. 1973; Chesher and Jackson 1985; Martin 1985a; Sanders et al. 1979). Naloxone, administered i.t., subcutaneously (s.c.), or intracerebroventricular (i.c.v.) also failed to block the antinociception induced by a variety of i.t., i.c.v. or spinally administered cannabinoids (Gilbert 1981; Welch 1993; Welch and Stevens 1992; Yaksh 1981). However, the irreversible μ antagonist chlornaltrexamine was reported to attenuate the antinociceptive and hypothermic effects of Δ^9-THC (Tulunary et al. 1981).

It has been shown that the antinociceptive effect of Δ^9-THC and morphine are additive following intravenous (i.v.) administration, thus implying distinct mechanisms of action (Gennings et al. 1993). In vitro the effects of Δ^9-THC on adenylyl cyclase have been shown to be insensitive to naloxone blockade and additive with the decrease in adenylyl cyclase observed with morphine (Bidaut-Russell and Howlett 1988). In rat striatum, a potent cannabinoid agonist was not found to be additive with either morphine or the dopamine agonist LY 171555 in decreasing cAMP levels (Bidaut-Russell and Howlett 1991). Results of another study using opioids and cannabinoids, alone and in combination, indicate that the cannabinoids and opioids may alter cAMP levels via similar mechanisms. The common final pathway of cAMP modulation by cannabinoids and opioids may be the phosphorylation of similar proteins, such as synapsins I and II, which are involved in the release of neurotransmitters (Childers et al. 1992). The binding of μ and δ opioids has been shown to be displaced by the cannabinoids in brain, but only at high concentrations (Vaysse et al. 1987), whereas δ opioid binding is not displaced by cannabinoids (Devane et al. 1986). The cannabinoid receptor was shown to be dense in the striatum (Herkenham et al. 1990), which is an area also associated with a dense population of opioid receptors (Yaksh et al. 1988). It is intriguing that, despite the data suggesting independent mechanisms of action, the effects of morphine have been found to be enhanced by orally administered Δ^6- and Δ^9-THC (Mechoulam et al. 1984).

Intrathecal administration of several cannabinoids leads to synergism with i.t. and i.c.v. administered morphine in the production of antinociception in mice (Smith and Martin 1992; Welch and Stevens 1992). Although pretreatment with morphine ehnanced the effects of Δ^9-THC, pretreatment of the mice with naloxone (s.c. or i.t.) failed to block the antinociceptive effects of the cannabinoids indicating that the cannabinoid-induced antinociception does not occur via interactions with the μ opioid receptor. Pretreatment of mice with Δ^9-THC significantly enhanced the potency of i.t. administerred morphine. Parallel shifts in morphine dose-response curves were produced not only with Δ^9-THC, but also with 11-OH-Δ^9-THC, Δ^8-THC and levonantradol, but interestingly not by CP-55,940. Thus, the antinociceptive effects of i.t. administered morphine are enhanced by the pretreatment with some, but not all, cannabinoids active at the CB_1 receptor (Welch and Stevens 1992).

Recently, the blockade of cannabinoid antinociception by the κ opioid antagonist, nor-binaltorphimine (nor-BNI) has been reported (Welch 1993). Antinociception produced by Δ^9-THC and Δ^8-THC (i.v., ED_{80} doses) was blocked by the κ antagonist nor-BNI, and the dose-effect curve for Δ^9-THC was shifted to the right in a parallel fashion. Specificity was suggested by the fact that the δ antagonist ICI 174,864 (i.t.) was without effect. Though Δ^9-THC activity was additive to that of a κ agonist (U50,488H), it

was unexpected that Δ^9-THC would produce a parallel 37-fold shift to the left in the dose-effect curve of a δ agonist (DPDPE). The AD50 values for nor-BNI vs i.t. administered Δ^9-THC, Δ^8-THC, levonantradol, and CP 55,940 ranged from 1.1 to 4.5 μg/mouse. Interestingly, the effects of i.v. CP-55,940 were blocked by i.v. nor-BNI, but not i.t. or i.c.v. nor-BNI, suggesting a locus of action for nor-BNI vs i.v. cannabinoids outside the central nervous system. Selectivity is indicated by the fact that nor-BNI blocks the antinociceptive effects of i.t. Δ^9-THC, without altering responses of catalepsy, hypothermia, or hypoactivity (SMITH et al. 1993). However, the inability of naloxone to block cannabinoid-induced antinociception raises questions as to the involvement of opioid receptors in the nor-BNI effects, since all κ opioid antinociceptive effects described can be blocked by naloxone (despite the need for high doses of antagonist). In addition κ opioid binding has been shown to remain unaltered by cannabinoids (VAYSSE et al. 1987), and neither nor-BNI nor the κ antagonist U50,448H bind to the CB_1 receptor (WELCH 1993). Thus, the nature of the nor-BNI effect is unclear, though it may be related to its ability to block the antinociceptive effects of ketoralac, which acts via a reduction in the synthesis of prostaglandins. (UPHOUSE et al. 1993). The interaction of nor-BNI with prostanoid formation has not been evaluated but might provide an alternative mechanism by which nor-BNI blocks the cannabinoid antinociception.

To summarize, the interactions between opioids and cannabinoids most likely involve a combination of indirect interactions mediated through numerous neurotransmitter systems as well as direct interactions between endogenous cannabinoid and opioid systems. At present, the relationship between these two systems under normal physiological circumstances is unclear. However, recruitment of either system through either pathological pain or opioid withdrawal can be manipulated by the other system. Past efforts to meld these two drug classes into a single entity represents an oversimplification and a misunderstanding of their biochemical and cellular actions.

2. Receptors

a) Pharmacological Characteristics

The structure-activity relationship (SAR) for cannabinoids has been reviewed extensively elsewhere (RAZDAN 1986), so only a brief statement of some aspects of the structural requirements for cannabinoid activity are presented. These and other data suggested the existence of specific cannabinoid receptors before identification by ligand binding and confirmation via cloning techniques.

Enantioselectivity is an important criterion for drug-receptor interactions because enantiomers share the same physicochemical characteristics. Initial

studies with Δ^9-THC failed to demonstrate complete enantioselectivity. However, almost complete enantioselectivity (Mechoulam et al. 1988) can be achieved when highly pure enantiomers are obtained, as demonstrated pharmacologically with 11-OH-Δ^8-THC-dimethylheptyl (Little et al. 1989). It had generally been assumed that an intact three-ring structure, based upon Δ^9-THC, was essential for activity since CBD (a bicyclic structure) lacks psychoactivity (Razdan 1986). However, a bicyclic derivative of 9-*nor*-9β-hydroxyhexahydrocannabinol, which also had a dimethylheptyl side chain (rather than the traditional pentyl group) at the C3 position (Melvin et al. 1984), proved to have a pharmacological profile similar to that of Δ^9-THC, though much more potent. This synthetic strategy led to the development of CP 55,940 which proved to be 4–25 times more potent than Δ^9-THC depending upon the pharmacological measure (Little et al. 1988). CP-55,940 and related novel bi- and tricyclic analogs have subsequently been referred to as nonclassical cannabinoids. CP-55,940 was radiolabeled in an attempt to discover a cannabinoid binding site (Devane et al. 1988).

The systematic approach taken in the development of cannabinoid antinociceptive agents (Johnson and Melvin 1986) helped define many of the structural determinants of cannabinoid action and produced extremely potent agonists. Some of these nonclassical analogs are as much as 700 times more potent than Δ^9-THC (Little et al. 1988). Other investigators prepared 11-OH-Δ^8-THC-DMH (Mechoulam et al. 1988), which also proved to be several hundred times more potent than Δ^8-THC in several behavioral evaluations (Little et al. 1989), as well as 11-OH-Δ^9-THC-DMH, which exhibited similarly high potency (Martin et al. 1991). In addition, a hexahydro-analog of the 11-OH-THC-DMH has proven to be potent and useful in ligand binding assays (Devane et al. 1992a).

Attempts to develop nonulcerogenic nonsteroidal anti-inflammatory drugs led unexpectedly to the discovery of yet another class of cannabinoid compounds, the aminoalkylindole (AAI) drugs, which are structurally distinct from both the traditional and nonclassical cannabinoids, yet bind to the cannabinoid receptor (Kuster et al. 1993) and exhibit cannabinoid behavioral effects (Compton et al. 1992a). WIN-55,212-2 (the prototypic AAI cannabinoid) was one of a series of analogs whose antinociceptive properties could not be explained by inhibition of either cyclooxygenase or by opioid mechanisms. Results in both radiolabeled CP-55,940 and WIN 55,212 ligand binding assays indicate similar rank potencies and suggest identical binding sites (Pacheco et al. 1991). That the AAI analogs share a common pharmacological profile with Δ^9-THC is indicated by the fact that the (+)-enantiomer and several related analogs exhibited ED_{50} values in the range of those of Δ^9-THC for producing hypoactivity, antinociception, hypothermia and ring immobility in mice. Additionally, they generalized from the Δ^9-THC cue in the rat drug discrimination paradigm despite considerable response rate suppression (Compton et al. 1992a). The (−)-isomer was inactive up to the highest dose tested.

b) Ligand Binding and Biochemical Characteristics

Convincing evidence for a receptor binding site for the cannabinoids did not emerge until the late 1980s. The highly lipophilic nature of Δ^8- and Δ^9-THC produced a large degree of nonspecific and nonsaturable binding which, coupled with their relatively low receptor affinities, provides the most likely explanation for failure of earlier investigators to characterize a cannabinoid binding site in brain (HARRIS et al. 1978; ROTH and WILLIAMS 1979), though they were able to demonstrate high-affinity, saturable binding of [^{3}H]Δ^8-THC to hepatoma cells in culture. Attempts to circumvent lipophilicity problems with the hydrophilic cannabinoid [^{3}H]5′-trimethylammonium-Δ^8-THC were also unsuccessful in that this ligand labeled a site which interacted both with pharmacologically active and inactive cannabinoids and which was later identified as a myelin basic protein (NYE et al. 1984, 1985). However, radiolabeling the potent bicyclic analog CP-55,940 proved to be a successful strategy for characterizing a cannabinoid binding site in brain homogenates (DEVANE et al. 1988).

Studies with CP-55,940 were the first to provide convincing evidence that a cannabinoid receptor existed. In rat brain cortical membranes, reported K_D values for CP 55,940 range from 0.13 to 5 nM and B_{max} values on the order of 0.9–3.3 pmol/mg protein (COMPTON et al. 1993; DEVANE et al. 1988; WESTLAKE et al. 1991). A limited series of analogs exhibited an excellent correlation between antinociceptive potency and affinity for this binding site (DEVANE et al. 1988). Subsequently, this correlation was extended to include a large number of cannabinoids and several behavioral measures (COMPTON et al. 1993). A high degree of correlation was found between the K_I values and in vivo potency in the mouse for depression of spontaneous locomotor activity, and for production of antinociception, hypothermia, and catalepsy. Similarly high correlations were demonstrated between binding affinity and in vivo potency in both the rat drug discrimination model and for psychotomimetic activity in humans. Thus, these studies suggest that the requirements for activation of the cannabinoid receptor are similar across different species and that this receptor is sufficient to mediate many of the known pharmacological effects of cannabinoids. This binding site has also been characterized with [^{3}H]1-OH-hexahydrocannabinol-DMH (DEVANE et al. 1992a), [^{3}H]11-OH-Δ^9-THC-DMH (THOMAS et al. 1992), and [^{3}H]WIN-55,212-2, and the findings are consistent with those reported for [^{3}H]CP-55,940.

Autoradiographic studies of the cannabinoid receptor have shown a heterogeneous distribution in brain that is conserved throughout a variety of mammalian species, including humans, with most of the sites in the basal ganglia, hippocampus and cerebellum (HERKENHAM et al. 1990, 1991b). Binding sites are also abundant in the cerebral cortex and striatum. It is interesting to speculate that these sites correlate with some of the pharmacological effects of marihuana, for example, cognitive impairment

(hippocampus and cortex), ataxia (cerebellum), catalepsy (basal ganglia), hypothermic and endocrine effects (hypothalamus), and even relatively low toxicity (paucity of receptors in the brainstem). Similar results have been obtained in studies conducted with [^{3}H]WIN 55,212 (Jansen et al. 1992) and [^{3}H]11-OH-Δ^9-THC-DMH (Thomas et al. 1992).

With regard to the existence of cannabinoid receptors in peripheral tissues, an examination of [^{3}H]CP 55,940 binding in all major organs of the rat resulted in detectable binding only in the immune system (Lynn and Herkenham 1994). Binding was detected in B lymphocyte-enriched areas (marginal zone of the spleen, cortex of the lymph nodes and nodular corona of Peyer's patches) but not in T lymphocyte-enriched areas (thymus and periarteriolar lymphatic sheaths of the spleen) and macrophage-enriched areas (lung and liver). Cannabinoid receptor binding in mouse spleen was consistent with Δ^9-THC inhibition of forskolin-stimulated cAMP accumulation in this tissue (Kaminski et al. 1992). Enantioselective immune modulation was observed with CP-55,940 and 11-OH-Δ^8-THC-DMH. In both cases, the (−) enantiomer demonstrated greater immunoinhibitory potency than the (+) isomer. Scatchard analysis of [^{3}H]CP 55,940 binding suggested a single binding site with a K_D of 910 pM and a B_{max} of approximately 1000 receptors/spleen cell. It is unclear why other sites were not found since cannabinoids apparently directly inhibit neuronal activity in peripheral sites (Kumbaraci and Nastuk 1980) as well as directly affect various smooth muscle preparations (e.g., vas deferens, ileum). A likely explanation is the lack of highly selective ligands for receptor subtypes.

Further validation of a receptor is often derived from manipulation of the endogenous system. One reaction of neuronal systems to the continued presence of agonist is receptor down-regulation. Temporally, in most systems, this process follows desensitization and is characterized by a loss of ligand binding at cell membrane receptors. Chronic exposure to Δ^9-THC results in the development of tolerance to the behavioral effects of Δ^9-THC (Dewey 1986). In mice, tolerance has been shown to occur for most Δ^9-THC-induced behaviors (Compton et al. 1990a). Long-term exposure to Δ^9-THC (90 days) apparently does not irreversibly alter the cannabinoid receptor, since 60 days after cessation of the treatment the receptor affinity and number were unaltered (Westlake et al. 1991). Down-regulation of receptor density has been observed in discrete brain regions of animals tolerant to Δ^9-THC (Oviedo et al. 1993; Rodriguez de Fonseca et al. 1994) but not in whole brain homogenates (Abood et al. 1993).

c) Receptor Cloning

Although SAR and receptor binding provide a compelling argument for existence of a cannabinoid receptor, the cloning of the protein provided definitive evidence. Homology screening with an oligonucleotide probe based on the structure of a G-protein coupled receptor (substance K) led to the

isolation of a unique clone from a rat brain library (MATSUDA et al. 1990). Subsequently, data from autoradiographic studies indicated that the distribution of the mRNA for the clone closely corresponded to that of the cannabinoid receptor. Thus, a ligand for this "orphan receptor" was eventually identified following the screening of many candidate ligands (opioids, neurotensin, angiotensin, substance P, neuropeptide Y, and others) when cannabinoids were found to act via this molecule. CP-55,940 and Δ^9-THC and other psychoactive cannabinoids (but not CBD and CBN) were found to inhibit adenylyl cyclase in cells transfected with the clone.

After the discovery of the rat cannabinoid receptor sequence, a human cannabinoid receptor cDNA was identified (GÉRARD et al. 1990). The nucleic acid sequences of these two clones were 90% identical, while the respective receptor proteins were 98% identical at the amino acid level. The human clone was expressed in COS cells and specific binding was demonstrated with [^{3}H]CP-55,940 (GÉRARD et al. 1990). The message for this receptor was also detected in the brains of the dog, rat, and guinea pig, but not found in dog stomach, spleen, kidney, liver, heart, or lung. Interestingly, the message was also identified in human testis, with a trace amount present in dog testis. The discrepancies between these results and those from receptor binding (LYNN and HERKENHAM 1994) include the failure to detect mRNA in the spleen (an organ which exhibits receptor binding) and the failure to detect binding in testis (an organ which contains message).

A peripheral receptor has been identified that is structurally distinct from the brain receptor (MUNRO et al. 1993). This receptor is expressed in macrophages in the marginal zone of the spleen and exhibits 44% homology with the receptor identified in brain tissues (MATSUDA et al. 1990), though this value rises to 68% in the transmembrane domains. Since multiple receptor subtypes exist, a consistent receptor nomenclature was adopted. The receptor nomenclature committee of IUPHAR recommended that the cannabinoid receptor be abbreviated as CB with a numerical subscript assigned according to order of discovery. Thus, the receptor isolated initially in brain tissue (MATSUDA et al. 1990) is designated CB_1, while that identified in the spleen (MUNRO et al. 1993) is designated CB_2. Though only a limited number of cannabinoids were evaluated, based upon binding properties it was concluded that the CB_2 receptor was indeed cannabinoid. The cloning of this receptor is consistent with the findings of others (KAMINSKI et al. 1992) showing that the spleen contains a cannabinoid binding site as well as the requisite mRNA.

The sequence of the cannabinoid receptors falls into the growing category of G-protein coupled receptors, which share structural and functional homologies. Signal transduction of ligand-receptor binding occurs via GTP-binding and G-proteins. Despite the fact that three sites of glycosylation are predicted from the structure of the receptor, biochemical studies indicate that the cannabinoid receptor need not be glycosylated to decrease cAMP (HOWLETT et al. 1990b). Structurally all are predicted to possess seven

transmembrane domains based upon the amino acid sequence. When the CB_1 receptor amino acid sequence was compared with that of 38 other G-protein coupled receptors, the cannabinoid receptor was found to be in a novel subgroup along with the ACTH and melanocortin receptors (Mountjoy et al. 1992). This subset of receptors: (1) lacks proline residues in the fourth and/or fifth membrane domains (where existence in G-protein coupled receptors is though to participate in forming a binding "pocket" by introduction of a bend into the linear nature of the α-helical structure); (2) lacks one or both of the cysteine residues (disrupting disulfide bond formation between the first and second extracellular loops); and (3) possesses amino acid residue homology of between 32% and 39%. Though the cannabinoid receptor shares 20% homology with the δ opioid receptor (Evans et al. 1992), assigning relevance to this degree of homology is difficult. Since the opioid receptor belongs to a class of peptide-responsive receptors, it is conceivable that the cannabinoid receptor is also responsive to an (as yet unidentified) endogenous peptide. Regardless, knowledge of the conserved amino acids between receptors that bind different ligands provides suitable target amino acids for site-directed mutagenesis studies.

Molecular techniques also provide the means to examine the expression of mRNA. Cannabinoid receptor binding and mRNA levels were examined in whole brain homogenates prepared from mice that had been treated chronically with Δ^9-THC (Abood et al. 1993). No alterations in cannabinoid receptor mRNA or protein levels were found in whole brain homogenates, though the chronic treatment was sufficient to induce a 27-fold tolerance in one behavioral assay. However, it is possible that alterations might occur within distinct brain regions (Oviedo et al. 1993), and such changes would not be apparent in whole brain preparations (Abood et al. 1993).

Cell lines transfected with the rat and human cannabinoid receptor clones have been investigated for their binding and signal transduction properties (Felder et al. 1992). The affinity of [^{3}H]CP 55,940 was similar to other preparations. The number of sites in the cell line expressing the human cannabinoid receptor was comparable to that of rat cerebellum, while the expression of the rat receptor was lower. Interestingly, cannabinoid receptor-mediated inhibition of cAMP accumulation was significantly reduced in the cell line overexpressing the human receptor. The rank order of potency of 16 cannabinoids evaluated for both receptor affinity and adenylyl cyclase inhibition proved to be nearly indentical to that in an earlier report of receptor binding in rat brain and multiple behavioral effects (Little and Martin 1991).

d) Molecular Modeling

Characterization of the interaction between the receptor and the ligand is crucial for understanding receptor activation, developing selective agonists, understanding antagonist actions, and distinguishing receptor subtypes. Two

molecular modeling approaches can be useful in this regard. The first strategy involves modeling the receptor itself, which is quite difficult and therefore has received relatively little attention. The second strategy involves developing a pharmacophore which describes the three-dimensional steric and electrostatic properties of an agonist. Though the discovery of the AAI cannabinoids underscores the limitations of the empirical approach to drug design, one technique used to evaluate the structural determinants for ligand binding and biological activity is computer-assisted molecular modeling. Studies have focused on the role of the phenolic hydroxyl in possible ligand receptor interactions (Reggio et al. 1990; Semus and Martin 1990) and the importance of the C9 position substituent and the spatial orientation of the associated ring (Reggio et al. 1991, 1993). Use of comparative molecular field analysis to analyze pharmacological and binding data has produced a three-dimensional pharmacophore of the electrostatic and steric forces of cannabinoids capable of quantitating the variations in the potencies of a wide variety of cannabinoids (Thomas et al. 1991). Steric repulsion "behind" the ring associated with C9 and the double bond of Δ^9-THC was associated with decreased binding affinity and pharmacological potency. The steric bulk of a side chain (located at C3 of the phenolic ring) can be extended by adding up to a total of seven carbons, which increases affinity and potency. This model possessed reasonable predictive capabilities and accommodated the AAI cannabinoids. However, considerable refinement is needed before the emergence of a predictive model for either receptor subtypes or selective ligands. In general, these models have provided descriptive models of SARs without divulging new insights.

e) Cannabinoid Antagonist

The search for a cannabinoid antagonist has been the topic of a previous review (Martin et al. 1987). Historically, lack of a cannabinoid antagonist has hindered research progress, since antagonists have generally played major roles in the characterization of many receptor systems. Numerous marihuana constituents, along with weakly active or inactive cannabinoid analogs, have been evaluated for potential antagonist properties with relatively little success. Although there have been some intriguing observations with CBD, there is no convincing evidence it is a specific antagonist. One report indicates that 11-nor-Δ^9-THC-carboxylic acid is capable of antagonizing the cataleptic effects produced by Δ^9-THC (Burstein et al. 1987). However, this observation has not been replicated in other laboratories. Though some drugs can (sometimes partially) attenuate some of the effects of Δ^9-THC, most alterations produced by such agents apparently simply represent the net effect of drugs possessing opposite effects (e.g., amphetamine stimulation plus cannabinoid inhibition of motor activity) rather than specific antagonism.

Agents which bind irreversibly to receptors have proven to be useful in developing antagonists for several classes of drugs. Reports thus far indicate

that nitrogen mustards CBD and Δ^8-THC lack agonist and antagonist effects (Compton et al. 1990b; Little et al. 1987). By contrast, photoactivation of azido-analogs of Δ^8-THC results in irreversible binding to the cannabinoid receptor in vitro (Burstein et al. 1991; Charalambous et al. 1992), and 5′-azido-Δ^8-THC exhibited potent in vivo effects. Although photoactivatable analogs do not provide a means for producing antagonism in vivo, they suggest that a reactive group at the terminal position of the side chain may cause irreversible binding, which could produce delayed antagonism.

Receptor-specific cannabinoid antagonists have been reported in the AAI class of drugs (Ward et al. 1991). One AAI antagonist was capable of producing a rightward shift in the in vitro dose-response curve of various agonists. However, the antagonist was most effective against AAI agonists (shifts of 20-fold or more) but much less effective (approximately fivefold shift) against natural and synthetic cannabinoids. Additionally, the drug exhibited only moderate to weak affinity for the cannabinoid receptor and was not capable of blocking the effects of cannabinoids under in vivo conditions (Compton et al. 1992a).

However, a novel chemical structure (typifying a fourth subclass of cannabinoid structures besides the traditional, nonclassical, and AAI subclasses) has been described as a truly specific competitive cannabinoid receptor antagonist (Rinaldi-Carmona et al. 1994). This analog (SR141716A) is most closely related in structure to the AAI class of cannabinoids (both possessing a nitrogen-containing, five-member, heterocyclic ring), but instead of being a carboxy-aryl-substituted indole, like the AAI analogs, it is a carboxyamide-substituted pyrazole with phenyl ring substituents. Though data strongly suggest that the analog is the first specific competitive cannabinoid receptor antagonist to be effective in vivo, it has only been shown that SR141716A blocks the in vivo effects of WIN-55,212 (an AAI cannabinoid). The ability of this drug to block the effects of other cannabinoids must be demonstrated in light of the data presented on the AAI antagonists. Additionally, only antagonism of WIN-55,212-mediated hypothermia has been evaluated in terms of time course of action and specificity with respect to noncannabinoid hypothermic agents.

3. Second Messenger and Other Transduction Mechanisms

a) Adenylyl Cyclase

The role of cannabinoids in the modulation of cAMP levels in cell culture and in homogenates of brain regions has been widely demonstrated. Δ^9-THC decreased epinephrine-and prostaglandin-stimulated levels of cAMP in fibroblasts (Kelly and Butcher 1973) and decreased cAMP levels in the *Tetrahymena* (Zimmerman et al. 1981) and in nonstimulated rat heart homogenates (Li and Ng 1984). However, the effect in the fibrolast preparation was biphasic and a function of incubation time (Kelly and Butcher 1979), so pharmacological relevance was unclear. In contrast, the effects of

cannabinoids on membrane fluidity in the liver and heart may alter coupling of glucagon to the G_s-protein leading to activation of adenylyl cyclase by glucagon in the liver and isoproterenol in the heart (HILLARD et al. 1990). This effect was shown to be enantioselective, so it has been proposed that the receptor associated with the cannabinoids may utilize a phospholipid as part of the recognition site.

The findings that cannabinoids inhibited forskolin-stimulated adenylyl cyclase preceded the characterization of the receptor (HOWLETT 1985; HOWLETT and FLEMING 1984). In neuroblastoma (N18TG2) or neuroblastoma X glioma (NG108-15) cell lines, cannabinoid-induced inhibition of cAMP formation has been consistent, reproducible, and independent of interaction with prostanoid, opioid, muscarinic, or adrenergic systems (HOWLETT 1984, 1985; HOWLETT and FLEMING 1984). It also was not blocked by antagonists of other classical neurotransmitters (BIDAUT-RUSSELL and HOWLETT 1991; DEVANE et al. 1986; HOWLETT et al. 1992). Cannabinoid-induced inhibition of cAMP formation in NG108-15 cells was rapid and reversible (DILL and HOWLETT 1988l; HOWLETT 1985), occurred at low cannabinoid concentrations (HOWLETT et al. 1986), and was consistent with the SARs established for the cannabinoids (HOWLETT 1987; HOWLETT et al. 1990b; HOWLETT and FLEMING 1984). The ability of cannabinoid analogs to inhibit adenylyl cyclase correlated well with their potency in several pharmacological assays, suggesting a cause-effect relationship (HOWLETT et al. 1990a).

Cannabinoids were reported to interact with a ribosylated membrane protein identified as the G_i-protein (HOWLETT et al. 1986). This result was subsequently reinforced by the finding that pertussis toxin attenuated cannabinoid effects on adenylyl cyclase (HOWLETT et al. 1988). Monovalent cations are recognized for their modulatory role in G-protein/receptor coupling (e.g., sodium) which is generally required for optimal inhibition of adenylyl cyclase by G_i/G_o-coupled receptors. In contrast, cannabinoid (and $GABA_B$) agonists inhibited adenylyl cyclase in a sodium-independent fashion in the cerebellum, but in a sodium-dependent fashion in the striatum (PACHECO et al. 1994). This differential effect was not due to either the receptor or the effector, so it is possible that different G-proteins are involved in these two brain regions.

Biochemical tolerance has been useful in studying receptor-regulated adenylyl cyclase activity. The cellular response to an agonist declines reversibly after continued exposure to drug. Exposure of N18TG2 neuroblastoma cells to Δ^9-THC attenuated cannabinoid-inhibited adenylyl cyclase activity without affecting cell morphology or growth (DILL and HOWLETT 1988). Cells pretreated for 24 h with 1 μM Δ^9-THC showed unaltered levels of basal cAMP, secretin-stimulated cAMP, and carbachol-inhibited cAMP, but Δ^9-THC produced only a 17% decrease (cf. 35% in controls) of cAMP accumulation. Thus, the desensitization was specific for the cannabinoid receptor-mediated response. The desensitization process was time- and dose-dependent and reversible.

In contrast to cell culture data, however, the modulation of forskolin-stimulated cAMP levels in mouse brain synaptosomes was not identical for all cannabinoids; Δ^9-THC and Δ^8-THC produced biphasic effects, while nonclassical cannabinoids only produced inhibition (Little and Martin 1991). Additionally, levonantradol (which produces pronounced cannabinoid effects) did not alter cAMP levels, while CP-56,667 (the largely inactive enantiomer of CP-55,940) inhibited cAMP, which suggested little correlation between pharmacological activity and modulation of adenylyl cyclase. Inhibition of adenylyl cyclase activity was also observed for AAI cannabinoids in rat brain membranes (Pacheco et al. 1991).

Though in vitro studies are relatively consistent and in vivo studies have suggested that cannabinoid administration to rodents altered cAMP accumulation in brain, the effects were modest and frequently difficult to reproduce (Martin et al. 1994). The initial work in rodent brain indicates that the levels of cAMP in the brain are altered in a biphasic manner by cannabinoids in the mouse. The intraperitoneal (i.p.) administration of Δ^9-THC has been shown to increase cAMP in whole brain and brain regions, while higher doses decrease cAMP levels (Dolby and Kleinsmith 1974). These effects have been proposed to be correlated with the initial stimulatory effects of low doses of the cannabinoids, whereas the depressant effects were with the higher doses of the cannabinoids (Dolby and Kleinsmith 1977). Similarly, cannabinoids increased cAMP levels in other preparations, but the effect did not correlate well with the psychoactive potency of the cannabinoids (Hillard and Bloom 1983). In contrast, another study found that i.v. administration of cannabinoids did not alter cAMP concentrations in five brain regions of the mouse, while Δ^8-THC increased cAMP in only one brain region (Askew and Ho 1974).

Despite evidence suggesting cannabinoid receptor/adenylyl cyclase association, it has been difficult to establish which pharmacological effects are mediated through this pathway. Most efforts have concentrated on demonstrating a role for adenylyl cyclase in cannabinoid-induced antinociception (Howlett et al. 1988). However, the affinity of cannabinoids at the CP-55,940 binding site and potency at inhibiting adenylyl cyclase (Devane et al. 1988) have been shown to be similar in rank order to the production of not only antinociception, but also hypothermia, spontaneous activity and catalepsy by the cannabinoids (Compton et al. 1993). Yet, pertussis toxin abolished the antinociceptive effects of cannabinoids, and i.t. administration of both forskolin and chloro-cAMP attenuated the antinociceptive effects of Δ^9-THC (Welch et al. 1994). These results support a role for adenylyl cyclase in the actions of cannabinoids since these agents either elevate or mimic cAMP. It should be noted that these agents did not completely abolish the cannabinoid effects and that actions other than adenylyl cyclase may be implicated by pertussis toxin.

b) Calcium Ion Channels

There has been reasonable evidence supporing a role for cannabinoid modulation of neurotransmitter release (Dewey 1986). Calcium is the likely mediator of this action given its well characterized role in neurotransmitter release. Cannabinoids act to decrease the release of acetylcholine by decreasing the influx of presynaptic calcium (Kumbaraci and Nastuk 1980). Additionally, the effects of pertussis toxin on cannabinoid response could as easily be attributed to G-protein-linked ion channels as to adenylyl cyclase activity. Cannabinoids are known to decrease calcium uptake to several brain regions (Harris and Stokes 1982), although this effect does not correlate with psychoactivity. Direct measurement of the effects of cannabinoids on free intracellular calcium in brain tissue (using intracellular calcium indicators) has shown that depolarization-induced rises in intracellular calcium are attenuated by Δ^9-THC, though at micromolar concentrations (Martin et al. 1989). These concentrations are similar to those required for the alteration of neuronal transmission (Kumbaraci and Nastuk 1980), but higher than those required to block calcium uptake (Harris et al. Stokes 1982). In contrast, others (Okada et al. 1992) have reported that Δ^9-THC did not perturb calcium levels in rat brain. Yet, in mouse thymocytes Δ^9-THC has been shown to decrease concanavalin A-stimulated levels of free intracellular calcium by both inhibition of calcium influx and inhibition of intracellular mobilization of calcium. These authors proposed that such changes in calcium may explain the immune suppression observed with the cannabinoids (Yebra et al. 1992).

Very low concentrations of Δ^9-THC (0.1 n*M*) have been shown to enhance potassium-stimulated rises in intracellular calcium, while intermediate concentrations (1–50 n*M*) block potassium-stimulated rises in intracellular calcium. Electrophysiological studies in neuroblastoma cells indicated that 1–100 n*M* concentrations of several cannabinoids inhibited an Ω conotoxin-sensitive, high voltage-activated calcium channel. This effect was blocked by the administration of pertussis toxin and was independent of the formation of cAMP. Since the L-type calcium channel blocker nitrendipine failed to alter the cannabinoid effect, it was concluded that cannabinoids apparently interact with an N-type calcium channel. Such an effect would lead to a decrease in the release of neurotransmitters (Mackie and Hille 1992). Results from a similar study revealed that cannabinoids inhibit I_{Ca} current in neuroblastoma cells, but the effect was not dose-related suggesting lack of a receptor-mediated event. However, it was pertussis toxin- and Ω conotoxin-sensitive (Caulfield and Brown 1992). Thus, while cannabinoids have been shown to alter intracellular calcium, the role of the cannabinoid receptor in these events has been questioned. Additionally, in CHO cells cannabinoids were shown to induce a nonspecific release of intracellular calcium. Both the active (−)- and inactive (+)-enantiomers of the potent cannabinoid 11-OH-Δ^8-THC-DMH were able to release calcium in non-transfected and CB_1 transfected cells (Felder et al. 1992).

The interaction of the adenylyl cyclase system with intracellular calcium has also been documented (Brostrom et al. 1978). cAMP has been shown to produce rises in free intracellular calcium in synaptosomes (Okada et al. 1989; Olson and Welch 1991). Such interactions may lead to cellular events responsible for the release of neurotransmitters, such as the phosphorylation of calcium channels which increases calcium conductance (Reuter 1983). Thus, the modulation of intraceullar calcium by cannabinoids is possible either via their interaction with adenylyl cyclase or by a mechanism independent of the formation of cAMP.

Although in vitro studies indicate a role for calcium in the effects of the cannabinoids, in vivo administration of various calcium channel modulators to mice has yielded results which indicate a lack of involvement of calcium directly in the antinociceptive effects of i.t. administered cannabinoids, whereas calcium modulation of i.c.v. administered cannabinoids is observed (Welch et al. 1994). The antinociceptive effects of the cannabinoids (i.t.) were not altered directly by the administration of calcium or by other modulators such as nimodipine, verapamil, Ω conotoxin, thapsigargin, BAYK 8644, or ryanodine. In addition, cannabinoids administered i.t. were blocked by the calcium-gated potassium channel blocker, apamin, but not by blockers of any other potassium channels. These data indicate that the antinociceptive effects of the cannabinoids in the spinal cord are not mediated by calcium channels. Unlike the i.t. situation, the i.c.v. administration of cannabinoids results in antinociception which is blocked by i.c.v. administration of calcium. In addition, thapsigargin (i.c.v.) blocks the effects of Δ^9-THC. Thus calcium modulation appears to play a role in the antinociceptive effects of cannabinoids in the brain. Apamin (i.c.v. or i.t.) fails to block the antinociceptive effects of i.c.v. administerred cannabinoids. Thus, the modulation of potassium channels by the cannabinoids may differ in the brain and in the spinal cord.

One other possible mechanism by which cannabinoids may decrease calcium entry is via interaction with the receptor-operated calcium channels stimulated by NMDA. Blockade of the NMDA-stimulated calcium channel has been described for (+)-11-OH-Δ^8-THC-DMH, an analog which is devoid of psychoactive effects of cannabimimetic properties in rodents (Feigenbaum et al. 1989).

c) Prostaglandins and Other Systems

Compelling evidence for the involvement of other second messenger systems in the pharmacological effects of cannabinoids does not exist, though there are many research avenues still open because cannabinoids appear to have some effect on almost any selected system or biochemical pathway (Martin 1986; Mellors 1979). One example is the effects of cannabinoids on cellular ATPases (Martin 1986; Pertwee 1988). Generally, the cannabinoids inhibit both cellular Na^+/K^+ ATPase and Mg^{2+}/Ca^{2+} ATPase. Thus, the effects of cannabinoids on calcium may, in part, be due to the alteration of Ca^{2+}

ATPase. activity. The effects of the cannabinoids on cellular energy via Na^+/K^+ ATPases disruption may explain the inhibition of neurotransmitter uptake. Most investigators have concluded that effects of the cannabinoids in ATPases may result from membrane perturbation or fluidization by the cannabinoids.

In several biological systems it has been shown that activation of receptors coupled to the formation of cyclic nucleotides results in a decrease in phosphoinositides (NISHIZUKA 1983, 1984). Δ^9-THC decreases the formation of *myo*-inositol trisphosphate (IP_3) in pancreatic islets (CHAUDRY et al. 1988). It is possible that the cannabinoids alter intracellular calcium and thus neuronal transmission by IP_3 formation. IP_3 formation has been shown to enhance the release of calcium from the endoplasmic reticulum, an organelle partially responsible for the buffering of intracellular calcium levels (NISHIZUKA 1983, 1984). However, there is no evidence that the effects of Δ^9-THC in brain or spinal cord are mediated through IP_3, though involvement in peripheral (e.g., cardiac) effects is still uncertain. While the binding of the cannabinoids within the cerebellum colocalized with that of forskolin, protein kinase C distribution was not localized in a similar pattern (HERKENHAM et al. 1991a). These studies support a role for cAMP rather than IP_3 in the actions of cannabinoids in the cerebellum. However, the interaction of the cannabinoids with IP_3 in other brain and spinal cord regions is not precluded.

Since, in the pituitary, cGMP enhances the formation of inositol phosphates (NAOR 1990), a possible interrelationship between IP_3 and cGMP formation has been hypothesized. Also, it has been shown that levonantradol (but not its inactive enantiomer dextronantradol) decreases basal and isoniazid-induced increases in cGMP in the cerebellum, possibly via an interaction with GABA (KOE et al. 1985; LEADER et al. 1981). In most systems the role of cGMP is unclear, although cGMP produces antinociceptive effects when injected into the brain of mice (VOCCI et al. 1978); therefore, it is possible the cannabinoids alter either IP_3 or cGMP formation in the production of antinociception. Though cGMP has also been linked to nitric oxide formation in the cerebellum, and cannabinoid mechanism of action pursued intensely in this brain region, there are no reports on the interaction of cannabinoids with nitric oxide.

Previous studies have suggested a role for cannabinoid agonists in arachidonic acid release and membrane phospholipid turnover. Δ^9-THC released arachidonic acid from mouse peritoneal cells and S49 cells (AUDETTE et al. 1991), and this effect was attenuated by pertussis toxin or cholera toxin. Thus, the release of arachidonic acid would appear to involve the G_i-protein. However, cell lines transfected with cannabinoid receptor have been evaluated recently for possible signal transduction systems (FELDER et al. 1992). Though CP-55,940 was able to release [^{3}H]arachidonic acid (at concentrations greater than $100\,\mu M$), it also did so in nontransfected CHO cells. Additionally, the inactive (+)-enantiomer of the potent cannabinoid

agonist 11-OH-Δ^8-THC-DMH was also able to stimulate [^{3}H]arachidonic acid release. These data indicated lack of involvement of the cannabinoid receptor.

The role of prostaglandins in the activity of cannabinoids is an area of research that has been previously reviewed (Burstein 1992; Martin 1986). Though anandamide, the endogenous ligand for the cannabinoid receptor, has been shown to be an ethanolamide derivative of arachadonic acid (Devane et al. 1992b), the relationship of this product to others in the arachadonic acid cascade remains questionable. Several behavioral effects, in particular cataleptic and antinociceptive effects, of the cannabinoids have been proposed to be related to the formation of prostaglandins. In vitro cannabinoids have been shown to produce diverse effects on prostaglandin synthesis. Both inhibition (Burstein et al. 1973; Burstein and Raz 1972; Burstein et al. 1974; Reichman et al. 1987; Spronck et al. 1978) and stimulation of prostaglandin formation (Burstein and Hunter 1981; Burstein et al. 1982, 1985; White and Tansik 1980) have been observed and are blocked by aspirin and mepacrine. Δ^9-THC has been shown to inhibit prostaglandin (PG)E_1 formation (Howes and Osgood 1976) in rat brain. However, PGE_1 and Δ^9-THC act synergistically in the production of antinociception as well as cataleptic, anticonvulsant and sedative effects (Bhattacharya et al. 1980). Blockers of prostaglandin formation, such as aspirin and indomethacin, have been shown to modify the antinociceptive, cataleptic, and hypotensive effects of Δ^9-THC in rodents, supporting the notion that cannabinoids may increase the formation of prostaglandins. (Burstein et al. 1982; Dalterio et al. 1981; Fairbairn and Pickens 1979, 1980; Jorapur et al. 1985). Similar findings have been reported to occur in humans (Perez-Reyes et al. 1991), in whom some behavioral effects of the cannabinoids have been shown to be attenuated by indomethacin. Mice immunized against PGE_2 had reduced cataleptic effects (Burstein et al. 1989; Hunter et al. 1991). Since antibodies presumably could not enter the central nervous system, the effect was thought to be primarily peripheral. These results were in agreement with those indicating a rise in levels of PGE_2 and $PGF_{2\alpha}$ following administration of Δ^9-THC (Bhattacharya 1986). Since the binding of PGE_2 was decreased following Δ^9-THC administration, it appeared there were increased levels of the prostaglandin which decreased its binding (Hunter et al. 1991). In contrast, Δ^9-THC has also been shown to inhibit the release of $PGF_{2\alpha}$ (Raffel et al. 1976) in rat brain.

4. Integration of Systems

a) Endogenous Cannabinoid System

Attempts to identify an endogenous ligand have resulted in the isolation of anandamide, an arachidonic acid derivative, from porcine brain which bound with high affinity to the cannabinoid receptor (Devane et al. 1992b).

Anandamide specifically bound to membranes from cells transfected with the cannabinoid receptor, but not to membranes from control nontransfected cells (Vogel et al. 1993).

Additionally, anandamide inhibited forskolin-stimulated adenylyl cyclase in transfected cells (but not in control nontransfected cells), an effect which was blocked by pretreatment with pertussis toxin (Vogel et al. 1993). Inhibition of adenylyl cyclase activity by anandamide in CHO cells expressing the human cannabinoid receptor was also observed and also blocked by pertussis toxin (Felder et al. 1993). N-type calcium channels were inhibited by anandamide in N-18 neuroblastoma cells. Additionally, inhibition of N-type calcium channels was voltage-dependent and *N*-ethylmaleimide sensitive (Mackie et al. 1993).

Anandamide was also shown to inhibit electrically stimulated contractions of mouse vas deferens much in the same fashion as Δ^9-THC. These effects were mediated via presynaptic actions on cholinergic neurons. Anandamide also reversed the stimulation of the miniature endplate potential firing frequency in the frog neuromuscular junction that was induced by hypertonic gluconate (Van der Kloot 1994). Since the protein kinase A inhibitor Rp-cAMPS also blocks this stimulatory effect, it is possible the anandamide effect was mediated via protein kinase A. However, the increase in frequency produced by Sp-cAMPS (a protein kinase activator) was not attenuated by anandamide. Thus, anandamide inhibits the gluconate effect without altering protein kinase A activity, though apparently via calcium effects (see above).

Anandamide has also been reported to produce effects in the rat on the hypothalamic-pituitary-adrenal axis similar to those produced by Δ^9-THC (Weidenfeld et al. 1994). Anandamide (i.c.v.) decreased CRF-41 levels in the median eminence and increased serum ACTH and corticosterone levels. These findings are consistent with the observations that cannabinoids exhibit anxiogenic properties (Onaivi et al. 1990).

Preliminary studies in mice also indicated that anandamide shares some of the behavioral and other pharmacological effects of Δ^9-THC (Fride and Mechoulam 1993). Other investigators (Crawley et al. 1993) also found a similar reduction in spontaneous activity and body temperature in mice treated with anandamide. However, more detailed studies (Smith et al. 1994) show that though anandamide and Δ^9-THC are very similar, there are also distinct differences. Of minor importance is the relatively short duration of action of anandamide, and the weak potency (anandamide is 4- to 20-fold less potent than Δ^9-THC). Interestingly, anandamide is largely inactive following i.p. administration, with the exception of the ability to produce profound sedative effects. Also, the antinociceptive properties of anandamide suggest a divergence from mechanisms for the production of other effects. The time course for anandamide-mediated antinociception is significantly longer than other effects and (unlike Δ^9-THC) is insensitive to administration of nor-BNI (see opiate interactions above).

Anandamide could function as an endogenous neurotransmitter or neuromodulator, since synthetic and metabolic pathways exist (Deutsch and Chin 1993). Synthesis was demonstrated by incubating arachidonic acid and ethanolamine in the presence of rat brain homogenate. Anandamide was also synthesized in bovine brain fortified with arachidonate and ethanolamide (Devane and Axelrod 1994), with the level of synthesis being greatest in the hippocampus, intermediate (twofold lower) in the thalamus, striatum or frontal cortex, and lowest (five- to sixfold less) in the cerebellum, an area with the greatest receptor density. Based upon the fact that anandamide synthesis is enzyme CoA- and ATP-independent, it was proposed that synthesis occurred via a novel eicosanoid pathway (Kruszka and Gross 1994). Anandamide was readily taken up by neuroblastoma or glioma cells and rapidly degraded by an amidase which can be blocked by phenylmethylsulfonyl fluoride (PMSF), a nonspecific peptidase and esterase inhibitor (Deutsch and Chin 1993). The degradative enzyme resides in the membranes (Deutsch and Chin 1993), which is corroborated by the fact that degradation occurs within ligand binding assays (Adams et al., in press; Childers et al. 1994). Anandamide was also degraded by brain, liver, kidney and lung tissue, but not heart or muscle. There is also evidence that the metabolism of anandamide can be blocked by trifluoromethyl ketone, α-keto-ester and α-keto-amide analogs of anandamide by acting as transition state inhibitors (Koutek et al. 1994). Though separate enzymes appeared to be responsible for synthesis and degradation, since PMSF did not block synthesis (Deutsch and Chin 1993), others have found that PMSF did inhibit synthesis (Devane and Axelrod 1994). Therefore, the question of multiple enzymes for anandamide synthesis and metabolism remains unanswered.

Anandamide may not be the only endogenous cannabinoid. A family of anandamides (similar structurally and physicochemically) may exist, since other endogenous unsaturated fatty acid ethanolamides (homo-γ-linolenylethanolamide and docosatetraenylethanolamide) have been isolated and also bind to the cannabinoid receptor (Hanus et al. 1993; Mechoulam et al. 1994). Additionally, unlike the anandamides or glycerol derivatives, a more hydrophilic endogenous substance was described (Evans et al. 1994), which could be released from neurons in a calcium-dependent fashion.

The last decade of progress in the cannabinoid field now supports the postulate that a cannabinoid neurochemical system exists. However, its role in the brain and its relationship to other neurochemical systems remains to be elucidated. Without direct evidence for a primary functional role, it would seem that the cannabinoid system is largely neuromodulatory, which is supported by the fact (Rinaldi-Carmona et al. 1994) that a putative cannabinoid antagonist (SR 141716A) administered alone appears to be devoid of typical cannabinoid effects in various rodent models (temperature, nociception, catalepsy, forced motor activity).

b) Spinal and Peripheral Cannabinoid Receptors

[^{3}H]CP-55,940 binds to the substantia gelatinosa of the spinal cord (HERKENHAM et al. 1990) at a level approximately 10% of that found in the substantia nigra, where maximal cannabinoid binding occurs. The substantia gelatinosa is responsible for the processing of pain transmission (YAKSH et al. 1988). Though the density of cannabinoid receptors is low relative to that of the brain, it is still much higher than that of substance P, which is a recognized transmitter involved in pain processing, in the dorsal horn of the spinal cord (IVERFELDT et al. 1988). Additionally, the substantia gelatinosa is also the principle location of the opioid receptors in the dorsal horn (GAMSE et al. 1979). The colocalization of these two systems may be critical to the synergism observed following i.t. administration of inactive doses of cannabinoids and active doses of morphine (i.t. or i.c.v.) in the production of antinociception in mice (SMITH and MARTIN 1992; WELCH and STEVENS 1992). Though parallel shifts in the morphine dose-response curve were produced by pretreatment with several cannabinoids, it is not clear that the response is mediated by a known cannabinoid receptor, since CP-55,940 was inactive in these procedures. However, it is possible that an as-yet-unidentified receptor might exist in spinal tissue, although describing the binding to this site would apparently require use of a radioligand other than CP-55,940.

The CB_2 receptor (discussed above) is structurally distinct from the brain CB_1 receptor (MUNRO et al. 1993) and has not been found in brain. The primary distinction between CB_1 and CB_2, besides anatomical location and primary structure, appears to be their affinity for CBN relative to that for Δ^9-THC. A review of the (brain CB_1) binding literature indicates that no single study has generated displacement data on both CBD and CBN to allow proper comparison to the Δ^9-THC value. Additionally, K_I values for all three of these analogs vary considerably between studies. These facts underscore the necessity of further characterization of CB_2 before concluding that its binding profile is distinguishable from that of CB_1. The functional role these receptors may play in the immune system is uncertain. However, the potential discovery of endogenous cannabinoids from peripheral tissue (see above) may suggest that existence of peripheral cannabinoid neuromodulatory systems. Regardless, the existence of the CB_2 receptor suggests the possibility that yet other subtypes may exist.

c) Cardiovascular Mechanisms

The effects of cannabinoids on the vascular system appear to be mediated by altered autonomic control of both the heart and blood vessels (ADAMS et al. 1976; BENOWITZ et al. 1979; JANDHYALA and BUCKLEY 1977), and indeed the cannabinoids possess some anticholinergic properties which may contribute to this response (DREW and MILLER 1974; GASCON and PERES 1973; LAYMAN 1971; ROSELL et al. 1976, 1979). Effects on heart rate have been linked to

altered parasympathetic function of the vagus nerve (Benowitz et al. 1979; Hollister 1986). In the dog, several acute cardiovascular and autonomic effects of Δ^9-THC were not observed following chronic Δ^9-THC treatment, suggesting tolerance. However, prolonged use may also induce pharmacological properties and/or mechanisms of action which may not exist in acute exposure (Jandhyala and Buckley 1977) and might mask previously observed events. Since some (but not all) cardiovascular effects were observed with nonpsychoactive drugs (Adams et al. 1977), the molecular mechanisms involved with these particular effects are probably not related to activation of CB_1 receptors in the brain. Additionally, since not all cardiovascular effects appear to be mediated by central and autonomic systems, some of the effects of cannabinoids would appear to be mediated by peripheral mechanisms. This last postulate is tentative, but the discovery of at least one peripheral receptor and one potential peripheral endogenous cannabinoid provide indirect support for this contention. However, cannabinoid receptors have not been detected in cardiac or vascular tissues, but it is possible such hypothetical sites represent a new subtype of receptor which is less sensitive to CP-55,940 and therefore not bound under currently used conditions or ligand concentrations, or that effects are mediated by an intermediate substance produced elsewhere in the periphery.

C. General Pharmacology

I. Pharmacokinetics

1. Absorption and Distribution

Δ^9-THC is absorbed rapidly and efficiently via the inhalation route. Detectable amounts of Δ^9-THC (7–18 ng/ml) have been measured following a single puff of marihuana smoke by individuals, and during a multiple puff session peak Δ^9-THC concentrations developed prior to the termination of smoking (Huestis et al. 1992b; Perez-Reyes et al. 1981). Despite considerable intersubject variability, experienced individuals developed peak Δ^9-THC concentrations in excess of 100 ng/ml after smoking marihuana cigarettes (THC content 1.32 to 2.54%) (Cocchetto et al. 1981; Huestis et al. 1992b; Lemberger et al. 1972b; Ohlsson et al. 1980; Perez-Reyes et al. 1982). The initial increase in Δ^9-THC blood concentrations during smoking is followed by rapid redistribution to tissues. Subsequent release back into the circulation occurs slowly, which produces a prolonged elimination half-life.

Oral ingestion of Δ^9-THC or marihuana leads to the production of similar pharmacological effects as smoking, although substantial differences exist in the rate of onset of effects and in the amounts of cannabinoids appearing in blood. Following oral dosing with 15–20 mg of Δ^9-THC there

was a gradual increase in blood levels of Δ^9-THC over a period of 4–6 h (WALL et al. 1983). Peak concentrations of Δ^9-THC were in the 10–15 ng/ml range, while concurrent 11-hydroxy-Δ^9-THC concentrations were in the range of 1–6 ng/ml. 11-*nor*-9-carboxy-Δ^9-THC concentrations were increased approximately twofold over those observed following intravenous dosing of Δ^9-THC.

Distribution of Δ^9-THC begins of occur immediately upon absorption. Mean peak Δ^9-THC concentrations declined by 50% approximately 10 min after the plateau was reached following smoking. Subsequently, concentrations declined much more slowly, but remained detectable for at least 4 h. Much longer detection times for Δ^9-THC have been reported, particularly in studies in which sensitive analytical methodologies were utilized. Concentrations of deuterium-labeled Δ^9-THC in plasma of chronic marihuana users were detected for 13 days by GC/MS techniques (JOHANSSON et al. 1988).

2. Metabolism and Excretion

Following the rapid redistribution of Δ^9-THC to body tissues there is a slow release from these tissues back to the circulatory system, which results in a prolonged elimination half-life. Δ^9-THC is metabolized in humans by a variety of oxidative routes which first produce hydroxylated metabolites, followed by conversion to carboxylic acids, and subsequent excretion as conjugates. The metabolite, 11-hydroxy-Δ^9-THC, is active (LEMBERGER et al. 1972a); however, it is formed in trace amounts when marihuana is smoked, though greater amounts may be formed following oral ingestion. About 50% of a dose of Δ^9-THC is excreted in feces and 15% is excreted in urine over a period of several days (WALL et al. 1983). The primary metabolite excreted in urine is conjugated 11-*nor*-9-carboxy-Δ^9-THC. Blood concentrations of Δ^9-THC peak prior to drug-induced effects. The discrepany between time course effects and cannabinoid blood concentrations, which was first raised almost 50 years ago (LOEWE 1946), remains unsolved.

Marihuana plant material cooked in brownies and consumed by male volunteers was studied to evaluate oral absorption (CONE et al. 1988). Subjects scored significantly higher on behavioral measures after consumption of brownies containing Δ^9-THC than placebo; however, the effects were slow to appear and were variable. Urinalysis indicated that substantial amounts of 11-*nor*-9-carboxy-Δ^9-THC were excreted in urine over a period of 3–14 days.

The metabolism of Δ^9-THC to 11-hydroxy-Δ^9-THC and to 11-*nor*-9-carboxy-Δ^9-THC occurs rapidly with peak blood concentrations of 11-hydroxy-Δ^9-THC appearing shortly after peak Δ^9-THC concentrations following either intravenous or oral administration (HUESTIS et al. 1992a; WALL et al. 1983). Peak 11-nor-9-carboxy-Δ^9-THC concentrations appear later (1–2 h) and decline slowly thereafter.

Due to complex distribution and elimination phases, a number of kinetic models have been proposed to describe plasma Δ^9-THC data. Blood levels

for Δ^9-THC during the first 6 h after smoking have been adequately described by a triexponential function (Barnett et al. 1982). Disposition of Δ^9-THC was described empirically as being represented by a two-compartment model with first order input from smoking. Others have utilized two- and four-compartment models to describe the disposition of Δ^9-THC administered intravenously. Half-life estimates for plasma Δ^9-THC range from 18 h to 4 days. Use of very sensitive assays usually results in longer half-lives and less variable Δ^9-THC clearance from blood that ranged from 650 to 1000 ml/min. Cannabinoids are excreted via bile and reabsorbed from the gastrointestinal tract, which likely contributes to their long half-life. Oral bioavailability of Δ^9-THC appears to be lower (6%–19%) than Δ^9-THC from smoked marihuana (14%–27%). Although several factors contribute to bioavailability, the experience of the smoker appears to play a key role. Subjects inhaling smoke from 4.5% Δ^9-THC marihuana cigarettes had a mean area-under-the-curve plasma concentration almost twice as high as that of subjects smoking 1.3% Δ^9-THC cigarettes (Perez-Reyes 1985). The expected AUC ratio based on the relative potency of the two cigarettes was 3.6:1, whereas the observed AUC ratio was 1.8:1. This discrepancy led to speculation that smokers could sense the rate of appearance and intensity of their "high" and would titrate their intake accordingly. Heavy marihuana users smoked more efficiently (23%–27% bioavailability) than light smokers (10%–14% bioavailability) leading to the conclusion that the experienced smokers utilized a more adept smoking technique e.g., deeper inhalations (Ohlsson et al. 1985). In studies involving drug administration by intravenous infusion of Δ^9-THC and by smoked marihuana (Lindgren et al. 1981; Ohlsson et al. 1982), there was a trend for heavy users to exhibit lower plasma concentrations than light users, but the differences were not statistically significant.

3. Relationship of THC Levels to Effects

Subjects begin to report behavioral effects after a single puff of marihuana smoke and these effects culminate at a time similar to or somewhat delayed with respect to blood Δ^9-THC concentrations (Huestis et al. 1992b). The delay between peak blood concentrations and peak drug effects is likely related to delays in penetration of the central nervous system and to subsequent redistribution of Δ^9-THC following rapid uptake by adipose tissues. The delay has been characterized as a counter-clockwise hysteresis between Δ^9-THC blood concentrations and drug (Barnett et al. 1982, 1985; Cone and Huestis 1993). Prior to equilibrium, plasma concentrations increase rapidly while effects develop more slowly. Consequently, at early times after smoking marihuana, plasma concentrations are high while effects are low; whereas at later times, plasma concentrations may be low while effects become highly prominent. This time discordance between blood concentrations of Δ^9-THC and effects has led to conclusions that no meaningful relationships exist between blood concentration and effect (Mason and McBay 1985; McBay 1986).

II. Effects on Organ Systems

1. Brain

a) Electroencephalogram

Alterations in EEG recordings are found in both humans and animals, but interpretation of such data is difficult. It has been suggested that the subcortical spike activity might be related to motor manifestations of marihuana use (Rosenkrantz 1983). In animals, the areas most sensitive to the effects of the cannabinoids were the hippocampus, amygdala, and septal areas. Identical measures are not available in humans.

b) Cerebral Blood Flow and Glucose Metabolism

Normally cerebral blood flow (CBF) and cerebral metabolic rate (CMR) are closely coupled with brain activity. Drug-induced changes in CBF or CMR are likely to be representative of a change in brain function (Mathew and Wilson 1993). Relatively little has been described concerning the effects of Δ^9-THC on these cerebral parameters (Mathew and Wilson 1992). Acute Δ^9-THC generally increases CBF (Mathew and Wilson 1992). A maximal, bilateral increase in CBF was observed 30 min following marihuana smoking, with greater increases observed in the frontal region and right hemisphere, though increases in both hemispheres correlated well with the degree of intoxication (Mathew et al. 1992). This correlation suggests that stimulation, rather than an inhibition of neuronal activity, is principally responsible for the observed effects. A decrease in CBF observed in inexperienced marihuana smokers has been attributed, in part, to the anxiety response sometimes observed in first time users. Increased global CBF has been reported in animals receiving Δ^9-THC as well as an increase in cerebral blood velocity (related to increased capillary perfusion). Also, decreased CBF was observed in chronic heavy abusers, but no alterations were observed under chronic conditions of moderate or mild marihuana abuse. This attenuation could be interpreted as development of tolerance with the emergence of an exaggerated compensatory mechanism (Mathew and Wilson 1992; Volkow and Fowler 1993) or possibly of the unmasking of inhibitory actions once tolerance has developed to the stimulatory effects of Δ^9-THC.

Acute Δ^9-THC generally increases the CMR of glucose (Mathew and Wilson 1992). However, Δ^9-THC has been shown to produce a biphasic response in limbic regions of animals (Margulies and Hammer 1991). Effects on CMR in humans may be limited to specific areas of the brain such as the cerebellum and prefrontal cortex (Volkow and Fowler 1993).

2. Immune System

a) Lymphoid Tissues

A thorough, and still relevant, review of the effects of marihuana and cannabinoids on the immune system is available (Munson and Fehr 1983). Though not definitive, the alteration of lymphoid organ weight is often considered an index of nonspecific immunosuppression. Δ^9-THC produced a reduction in thymus weight in monkeys, focal hemorrhages in rats (with no alteration in weight), and decreases in weight and cellularity in the mouse (Munson and Fehr 1983). Since the thymus provides immunocompetent lymphocytes to the secondary lymphoid organs, it seems plausible that marihuana negatively affects maturation of these cells in the developing individual (Pross et al. 1992a). However, no data are available concerning thymic changes in humans.

There are no consistent effects observed on the spleen following marihuana or cannabinoid administration to various animal species (Munson and Fehr 1983). The most consistent results appear to be in the mouse model, in which treatments of 8 days or less induce a hypocellularity concomitant with organ weight loss. One study indicates that administration of the nonpsychoactive cannabinoids CBD and CBN produced a decrease in the white pulp of the spleen, suggesting a reduction in lymphocytes. Other results indicated that Δ^9-THC could either enhance or suppress aspects of the immune response, depending on the specific immune stimulants used and the specific parameter of immunity measured (Pross et al. 1992c) as well as the age of the animal (Nakano et al. 1993). However, no data are available concerning splenic changes in humans.

There are limited data on the effects of marihuana on lymph nodes (Munson and Fehr 1983). Despite the fact that proliferation of Ly2 (suppressor/cytotoxic) cells of splenic origin could be inhibited with low doses of Δ^9-THC, identical cells of lymph node origin were resistant to the suppressive effects, which illustrated the dependence of the immunomodulatory capability of Δ^9-THC on the organ source of lymphocytes (Pross et al. 1992b). No data are available concerning changes in human lymph nodes.

b) Immune System Cells

Δ^9-THC produces a reversible inhibition of macrophage extrinsic anti-herpes activity, while producing no effect on macrophage intrinsic activity (Cabral and Vasquez 1993). The suppressive effect of Δ^9-THC on extrinsic antiviral activity is reversible upon removal of the drug. Δ^9-THC did not alter virus uptake or replication within macrophage-like cells in culture. Other studies indicated that Δ^9-THC altered macrophage morphology, function, and motility. Studies in rodents indicated a potential decrease in motility, an increased ease of cytolysis, and variety of other, more minor, alterations (Munson and Fehr 1983). Δ^9-THC inhibited cell propagation and DNA

synthesis, though the magnitude of these effects was dependent upon the number of cells in the culture and the protein content in the culture medium (TANG et al. 1992). As the cell number increased, the Δ^9-THC effect decreased. *Legionella* grew much better in macrophages treated with low doses of Δ^9-THC, though there was no change in the number or viability of the macrophages. Thus, it is apparent that Δ^9-THC has the ability to enhance the growth of the intracellular opportunistic pathogen *Legionella* that grows in A/J mouse macrophages (ARATA et al. 1992). There do not appear to be consistent changes in the total number of leukocytes in humans (MUNSON and FEHR 1983). However, high in vitro concentrations of both psychoactive and nonpsychoactive cannabinoids exhibit some immunosuppressive activity on leukocytes (MOLNAR et al. 1987).

Natural killer (NK) cell activity was reduced following exposure to cannabinoids. Δ^9-THC did not inhibit the binding to target cells of either cloned NK cells or freshly isolated mouse spleen cells, though killing capacity was restricted. Therefore, Δ^9-THC appears to directly inhibit NK cell cytolytic activity at a postbinding stage (period following adhesion of NK cells to target cells) (KAWAKAMI et al. 1988). Δ^9-THC treatment resulted in a suppression of splenic NK activity (KLEIN et al. 1987). Further experiments suggested that the psychoactive cannabinoids suppress NK cell function by interacting directly with the killer cells and disrupting events postbinding and during the programming for lysis (KLEIN et al. 1987). Δ^9-THC applied in vitro was toxic to human peripheral blood lymphocytes at high concentrations, but at lower concentrations still produced an inhibitory effect on NK activity against a human tumor cell line (SPECTER et al. 1986).

c) B Cells and Antibody Formation

There is no consistent change in B cell number in humans following cannabis administration (MUNSON and FEHR 1983). The proliferation of B cells in response to mitogens (bacterial LPS) was reduced, but no changes were observed in monkeys following other (pokeweed) mitogen treatment. Similar effects have not been demonstrated in humans (MUNSON and FEHR 1983). In rodents, cannabinoids inhibit IgG and IgM antibody secretion, a B cell function (MUNSON and FEHR 1983). Treatment of monkeys also resulted in a decrease in IgG and IgM, but treatment for 6 months was required. There are no consistent changes in human basal antibody production.

d) T Cells

Functional measures of T cells include the in vitro measurement of stimulation (blastogenesis and secretion of cytokines) by mitogens and the ability to kill allogeneic cells (MUNSON and FEHR 1983). In vitro measures to mitogenic stimulation have proven inconsistent. Also, the ability of T_{killer} cells to destroy allogeneic mastocytoma cells was decreased by Δ^9-THC treatment (MUNSON and FEHR 1983). Exposure to Δ^9-THC also resulted in suppression

of concanavalin A-induced thymus cell proliferation, primarily evidenced in the single positive Ly2 (suppressor/cytotoxic) subpopulations (Pross et al. 1992a). Though Δ^9-THC was found to suppress mitogen-induced proliferation, it also enhanced anti-CD3 antibody-induced proliferation (Nakano et al. 1992). Δ^9-THC produced a suppression of Ly2 cell number following concanavalin A or phytohemagglutinin stimulation, but produced an increase of Ly2 cells following CD3 stimulation (Nakano et al. 1992; Pross et al. 1992c). However, it is clear that both age and organ source play a critical role in the generation of immunostimulation. This up-regulation of responsiveness was not seen in either lymph node cells of adult or young mice or in spleen cells of young mice, but was only observed on lymphocytes from adult spleens (Nakano et al. 1993; Pross et al. 1992b). Additionally, cotoxicity assays demonstrated that CTLs (cytotoxic T lymphocytes) from mice exposed to Δ^9-THC were deficient in anti-herpes virus (HSV1) cytolytic activity (Fischer-Stenger et al. 1992). However, in vivo Δ^9-THC treatment had little effect on the number of T lymphocytes expressing the Lyt-2 (cytotoxic cell) or L3T4 (helper cell) antigens. CTL from drug-treated mice were able to bind specifically to the HSV1-infected targets, but in vivo Δ^9-THC treatment affected CTL cytoplasmic polarization toward the virus-infected target cell, and granule reorientation toward the effector cell-target cell interface (following cell conjugation) occurred at a lower frequency. These results suggest that Δ^9-THC elicits dysfunction in CTL by altering effector cell-target postconjugation events (Fischer-Stenger et al. 1992). However, in humans the results are more variable.

e) Host Resistance

The immunomodulatory effects of marihuana include alterations in humoral, cell-mediated and innate immunity, and though most studies have shown immunosuppressive effects, there are reports that there may not be any deleterious effect or that some aspects of host immunity may be enhanced (Yahya and Watson 1987). Δ^9-THC or marihuana may reduced resistance to cancer growth and microbial pathogens in animals (Yahya and Watson 1987). In humans, studies have suggested potential links between episodes of marihuana abuse and increased infection by such organisms as those responsible for herpes and tuberculosis. These and similar anecdotal reports have not been corroborated (Munson and Fehr 1983). Studies conducted with appropriate control groups of humans have failed to indicate any decrease in resistance or any significant change in immunological responses related to T cell function. Evidence is generally supportive of some degree of immunosuppression only when in vitro studies are considered, and these have been flawed by the fact that most observations only occur at very high concentrations of Δ^9-THC (Hollister 1988). When experimental studies have been conducted to more closely mimic the actual clinical or human abuse situations, the evidence has been less compelling for immunosuppression, decreased host resistance, or increased infection (Hollister 1988).

3. Endocrine

a) Hypothalamic-Pituitary Hormones

In the rodent, acute administration of Δ^9-THC causes a decrease in the gonadotropins LH (luteinizing hormone) and FSH (follicular stimulating hormone). These effects appear to be mediated by disruption of the hypothalamic-pituitary system via alteration of dopaminergic, serotonergic, opioid and/or adrenergic controls of endocrine function (Fernández-Ruiz et al. 1992; Wenger et al. 1992). Decreased LH appears to be due to diminished release of LHRH (luteinizing hormone releasing hormone; also referred to as GnRH-gonadotropin releasing hormone), which was reported to accumulate in the hypothalamus (Rosenkrantz 1985), though some evidence suggests diminished biosynthesis of LHRH (Dewey 1986). One study suggests the effect of Δ^9-THC involves a mechanism which includes inhibitory actions within the preoptic-to-tuberal GnRH pathway (Tyrey 1992). Studies on the alteration of FSH levels in laboratory animals are somewhat inconclusive, though FSH levels have generally been found to be decreased (Rosenkrantz 1985). In humans, the acute alterations observed following marihuana or Δ^9-THC on gonadotropins are also somewhat unclear. Acute Δ^9-THC decreased LH, but did not change FSH, at the typically low doses abused by humans (Rosenkrantz 1985). Other evidence suggests either an increase, decrease, or no change in humans with either LH or FSH.

In rodents, the acute effect of marihuana or Δ^9-THC was reported to decrease prolactin (PRL) levels. Decreased PRL release in rodents appears to be due to diminished release of TRH (thyrotropin releasing hormone) (Rosenkrantz 1985), though it may also be altered by diminished GnRH. The reduction of PRL release following Δ^9-THC exposure, both in vivo and in vitro, might be elicited by a direct action of Δ^9-THC on the pituitary (Rodriguez de Fonseca et al. 1992a), though other data suggest that the initial site of action may not be in the region of the hypothalamus most intimately associated with pituitary function. Inhibition of the effects of Δ^9-THC on PRL (and ACTH, adrenocorticotropic hormone) by hypothalamic deafferentation suggests a more distant site for Δ^9-THC action (Tyrey 1984). In rats treated with estradiol, basal PRL levels were increased and a PRL surge occurred. However, estradiol stimulation of both basal and surge levels of PRL was significantly attenuated by concomitant Δ^9-THC treatment (Murphy et al. 1991). Lastly, though PRL was decreased in monkeys, in humans Δ^9-THC increased serum PRL levels following either oral or intravenous administration (Rosenkrantz 1985). No reports were found indicating whether lactation was altered in either humans of animals.

Growth hormone (GH) is generally reduced in both laboratory animals and humans (Rosenkrantz 1985). During a 9 h period following the acute

administration of Δ^9-THC to rats, the episodic secretion of GH was suppressed in terms of mean plasma level, peak height, and integrated peak amplitude analyses. Although the physiological mechanisms involved in this response were undetermined, the data indicated that Δ^9-THC can inhibit the hypothalamic-pituitary control of normal episodic GH secretion (Falkenstein and Holley 1992). Interestingly, the effect appears to be biphasic, since higher doses of Δ^9-THC could induce an increase in GH in rats. However, increases in human GH have not been reported (Rosenkrantz 1985).

b) Gonadal Hormones

The general effect of Δ^9-THC marihuana on the gonadal hormone testosterone is to decrease serum levels in rodents, monkeys, and humans (Rosenkrantz 1985). However, it is important to note that these changes in humans occur at high oral or moderate intravenous doses. Many studies using low doses of marihuana administered via the inhalation route showed no acute change in testosterone. Inhibition of plasma testosterone may be due to a direct effect on synthesis in Leydig cells (Burstein et al. 1978, 1979, 1980), although it appears that the Δ^9-THC-induced block of GnRH (gonadotropin releasing hormone) release results in lowered LH and FSH and subsequently reduced testosterone production by the Leydig cells of the testis (Harclerode 1984). Other results indicate that the nonpsychoactive cannabinoid CBD also suppresses hepatic testosterone oxidation at the 2α, 16α, and 17 positions through selective inhibition of a specific cytochrome P-450 in the adult male rat (Narimatsu et al. 1988). Additionally, smoked marihuana condensate, Δ^9-THC, and CBN have been found to inhibit specific binding of dihydrotestosterone to the androgen receptor, but did so with dissociation constants in the range of 210–580 n*M*. While it is difficult to interpret the meaning of low dissociation constants, some of the anti-androgenic effects associated with marihuana use may, at least in part, be due to inhibition at the receptor level (Purohit et al. 1980).

Marihuana abuse during the time of established hormonal cycles may render human females anovulatory and produce delayed and smaller surges in estrogen and progesterone (Rosenkrantz 1985). However, data in female monkeys did not corroborate alterations in progesterone, though treatment for 1 year suggested a shortened luteal phase and either decreased or had no effect on serum estradiol and progesterone (Rosenkrantz 1985). Rat data suggest that Δ^9-THC is neither pro- nor antiestrogenic with respect to phase I responses (increased uterine macromolecular uptake within 6 h of estrogen administration), but in terms of phase II responses (hyperplasia and hypertrophy occurring 12–24 h following estrogen administration), Δ^9-THC was modestly pro-estrogenic in the progesterone-treated uterus, but was anti-estrogenic in the presence of estradiol. These estrogen agonistic/antagonistic effects of Δ^9-THC on uterine phase II responses did not adversely affect the process of implantation and decidualization (Paria et al. 1992). Δ^9-THC

antagonizes estradiol action on the anterior pituitary. Δ^9-THC also prevented the estradiol-induced increase in pituitary weight but had no effect on either the uterine or oviduct weight response (Paria et al. 1992).

c) Thyroid Hormones

Decreases in both T_3 (triiodothyronine) and T_4 (thyroxine) have been documented in rodents and appear to result from diminished TSH (thyroid stimulating hormone; thyrotropin) release. Diminished TSH levels appear to be due to the inhibition of its release. Additionally, there is a disruption of iodine uptake and release. In humans a similar disruption in iodine uptake and release was observed, but no change in levels of either T_3 or T_4. However, the subjects were chronic abusers (4–7 years) so the lack of an effect may be due to tolerance (Rosenkrantz 1985).

d) Glucocorticoid Hormones

Δ^9-THC can induce certain endocrine changes including stimulation of adrenocortical function (Dewey 1986). The effects of cannabinoids on glucocorticoid hormones and receptors (Eldridge and Landfield 1990; Rodriguez de Fonseca et al. 1991b), with special reference to the hippocampus, have been reviewed recently (Eldridge and Landfield 1992). In summary, cannabinoids stimulated adrenal corticosterone secretion either alone or in combination with physiological stressors (e.g., footshock) in laboratory animals. Δ^9-THC-induced increases in corticosterone appeared to be mediated by increased release of pituitary ACTH (Dewey 1986; Rosenkrantz 1985). Also, it has been suggested that normal functioning of the pituitary-adrenal axis requires a properly functioning hippocampus, which may suggest one region through which cannabinoid-mediated alterations could be induced (Eldridge and Landfield 1992). Additionally, Δ^9-THC appears to interact in a noncompetitive fashion with the type II corticosteroid-binding receptor in the hippocampus. Since Δ^9-THC was able to down-regulate this receptor in the hippocampus, it appears Δ^9-THC possesses at least partial agonist activity, suggesting that cannabinoids may disinhibit the negative feedback control of endogenous glucocorticoids (Eldridge and Landfield 1992). Additionally, corticosterone treatment appears to increase binding of the cannabinoid ligand CP-55,940 to the hippocampus but not the cerebellum (Eldridge and Landfield 1992). These findings suggest some specificity of the interaction between cannabinoids and glucocorticoids. Lastly, corticosterone was able to partially reverse the inhibition of binding of CP-55,490 induced by in vitro addition of GTP analogs (Eldridge and Landfield 1992). In humans, no effect on corticosterone level was produced by acute oral Δ^9-THC at doses pharmacologically relevant to human abuse patterns (Rosenkrantz 1985), nor under a variety of related clinical regimens. One of the most notable observations was that Δ^9-THC administration

induced aging-like degenerative changes in rat brain similar to that resulting from elevated corticosterone (Landfield et al. 1988).

It has now been demonstrated that an i.c.v. injection of anandamide (50–150 μg/rat) significantly increases serum levels of ACTH and corticosterone in a dose-dependent manner and causes pronounced depletion of CRF-41 in the median eminence (Weidenfeld et al. 1994). These data suggest that anandamide parallels Δ^9-THC in activating the hypothalamo-pituitary adrenal axis via mediation of a central mechanism which involves the secretion of CRF-41. It is of interest that the caudate-putamen of adrenalectonized rats contains 50% higher levels of mRNA for the cannabinoid receptor than the controls. This increase could be counteracted by dexamethasone (Mailleux and Vanderhaeghen 1993b). Taken together with the findings of Weidenfeld and colleagues, it seems possible that the corticoid and anandamide systems could be mutually regulatory.

e) Reproduction

The accumulation of many reproduction studies in both animals and humans have produced conflicting results and widely varying conclusions with time, which may be due in great part to a combination of differences in experimental design and interspecies differences in drug tolerance (Wenger et al. 1992). However, Δ^9-THC has been described as a reproductive toxicant in both humans and animals in various studies (Basloch 1983). In animal studies, Δ^9-THC produces adverse effects on gametogenesis (both oogenesis and spermatogenesis), on embryogenesis (organogenesis and fetal development), and upon postnatal development (Tuchmann-Duplessis 1993). Conclusions that marihuana may be linked to infertility have been proposed, in part, due to data indicating large reductions in sperm concentrations following administration of four to sixteen marihuana joints per week for a 4 week period (Buchanan and Davis 1984). However, as these reductions occurred in the absence of changes in FSH, LH, PRL, cortisol, and T_4 or testosterone, they would appear to be related to direct cellular effects rather than neuronal disruption. Other evidence suggesting alterations to the male reproductive tract include findings of oligospermia with Leydig and Sertoli cell dysfunction, though there did not appear to be an associated sterility (Rosenkrantz 1985). Evidence suggesting gynecomastia remains controversial (Rosenkrantz 1985). Besides altered spermatogenesis, there may also be alterations in sex organ physical characteristics (Bloch 1983; Rosenkrantz 1985), which appears to be the result of both direct actions on tissues and the indirect effect of reduced androgenic hormones. Though the effects of marihuana on fertility in men may involve the pituitary-hypothalamic axis, other effects seem to be produced via alteration of specific cells of the testis. Interestingly, the mechanism of action, at least on the Sertoli cells, appears to involve a pertussis toxin-independent pathway for the reduction of FSH-induced accumulation of cAMP (Heindel and Keith 1989), suggesting

cellular effects that are independent of the cannabinoid receptor described in brain tissue. Additional evidence that this response may not be mediated through the cannabinoid receptor includes the fact that Δ^9-THC does not alter forskolin stimulation of cAMP in the Sertoli cells and that cannabinoids devoid of psychoactivity also inhibit FSH-induced accumulation of cAMP (Heindel and Keith 1989).

4. Cardiovascular

a) Blood Pressure

The effects of cannabinoids of cardiovascular function have been reviewed (Dewey 1986; Hollister 1986; Tennant 1983). Generally, the hypotensive effect in animals appears to be centrally mediated, though some direct action upon the heart (Smiley et al. 1976), or the nerve terminals of the heart, appears likely (Gascon and Peres 1973; Jandhyala and Buckley 1977). Also, the effect of Δ^9-THC on animals can be biphasic, with an initial vasoconstrictive phase and an associated increase in vascular pressure, followed by a period of hypotension (Adams et al. 1976). One difference between animal and human data is that the effect in humans generally appears to have been smaller in magnitude, though this may be due to the use of relatively small doses as well as postural considerations in humans (Rosenkrantz 1985). The blood pressure response in humans (Hollister 1986; Lemberger et al. 1974; Malit et al. 1975; Weiss et al. 1972) was minimal (though orthostatic hypotension occurred), while a clear hypotensive effect was observed in animals (Cavero et al. 1973; Dewey et al. 1972; Pradhan 1984; Williams et al. 1973). However, no lasting effects on blood pressure have been described (Tennant 1983). Inhibited vascular reflexes and decreased peripheral resistance have been reported (Rosenkrantz 1983). Incidentally, the time course of the psychoactive effect closely parallels the time course of the reddening of the conjunctiva, which is due to local vasodilation (Hollister 1986).

b) Heart Rate and EKG

In animals, a bradycardia was the predominant effect observed following marihuana administration (Tennant 1983), although biphasic responses have been described following administration of sufficiently low doses of Δ^9-THC (Rosenkrantz 1983). Yet, in humans tachycardia was produced (Hollister 1986; Lemberger et al. 1974; Linton et al. 1975). Interestingly, the time course of the tachycardia in humans closely parallels the time course of the psychoactive effects (Hollister 1986). The tachycardia in humans appears to have been due to sympathetic stimulation in combination with parasympathetic inhibition (Hollister 1986; Rosenkrantz 1983). The net effect of marihuana on humans was to increase myocardial work load, myocardial oxygen demand, and to decrease oxygen delivery (Hollister

1986) which was also observed in animals (Smiley et al. 1976). Smoking marihuana may dispose individuals to heart problems such as angina (Aronow and Cassidy 1975). Despite this, there is no evidence for significant changes in EKG, and field studies failed to disclose any abnormalities in heavy abusers (Kochar and Hosko 1973; Tennant 1983).

5. Gastrointestinal

In some humans a significant degree of diarrhea and related gastrointestinal upset (vomiting, cramps) occurred following marihuana abuse (Tennant 1983). It was suggested that this may have been due to the fact that cannabinoids apparently decrease gastric acid secretion (Gahlinger 1984), which could make the smoker more susceptible to infection by gastrointestinal bacteria and thereby produce an aggravated diarrhea. However, the mechanism of action is unclear, but it is possible that cannabinoids interfered with one or more of the neuronal controls of gastric secretion. In animal models, low doses of intravenous psychoactive cannabinoids exerted an inhibitory effect on GI transit and motility and slowed the rate of gastric emptying and small intestinal transit (Shook and Burks 1989). Therefore, marihuana is capable of altering function of the GI tract, suggesting that low doses might produce mild constipation. However, it is possible that the humans suffering diarrhea and cramps ingested quantities larger than the relative amount evaluated in the animal nodels.

6. Renal

Renal toxicity has been observed following intravenous injection of cannabis extracts. Generally, renal insufficiency is not observed in humans, though there was at least one instance of such a report. Following consumption of a cannabis butter preparation an elderly man suffered constipation and urinary retention. The problem was sufficiently severe that the man required urethral catheterization. It was hypothesized that the mechanism of action might have been interference with peripheral cholinergic activity (Tennant 1983). However, Δ^9-THC has also been shown to possess an antihistaminergic activity on the rabbit kidney, which seems to be a competitive antagonism at the histamine H1-receptor (Turker et al. 1975). Also, evidence exists to suggest a Δ^9-THC-induced release of prostaglandin-like material from rabbit kidney (Kaymakcalan et al. 1975). The actions of cannabinoids on renal function has not received much attention in recent years.

III. Toxicity

1. Respiratory Effects

One effect of marihuana that is produced regardless of route of administration is the depression of the respiratory system, and this effect appears to be

mediated by central mechanisms. Lethality in animals could readily be demonstrated following acute drug administration (Rosenkrantz 1983) and appears to have been due to respiratory depression (Rosenkrantz 1983), which was preceded by dyspnea and apnea (Forney and Kiplinger 1971). These effects on respiration may have been due to an upward shift in the carbon dioxide set point as well as depression of the respiratory center in the medulla (Dewey 1986). However, marihuana-induced deaths in humans, in the absence of any other drugs, are an unheard of event. Thus, lethality data for marihuana in humans is primarily anecdotal. However, the lethality of Δ^9-THC administered by various routes and formulations in different species indicates large differences between oral and intravenous lethality (Rosenkrantz 1983). Lastly, there was unpublished evidence to suggest a role for cardiac arrest in the production of lethality following high dose cannabinoid administration, but whether this was a result of respiratory depression or other factors was unclear and unsubstantiated (Rosenkrantz 1983).

2. Psychotic Episodes

The suggestion that Δ^9-THC induces psychopathologies (Bartolucci et al. 1969; George 1970; Talbott and Teague 1969) has been examined and a listing of medical literature associating marihuana with mental illness compiled (Nahas 1993b; Nahas and Latour 1992). However, attempts to identify a "cannabis psychosis" have been unsuccessful (Dewey 1986; Hollister 1986; Taschner 1983; Thornicroft 1990), even in parts of the world where consumption of marihuana has previously been associated with admission to hospitals for psychiatric conditions (Chkili and Ktiouet 1993; Defer 1993).

The effects of marihuana on schizophrenic symptoms are widely recognized to be detrimental, yet approximately one-third of all schizophrenics continue to self-medicate with marihuana (Negrete 1993). Paranoid schizophrenics apparently recognize the worsening of symptomatology brought on by marihuana. Schizophrenics abusing marihuana have been reported to be more difficult to effectively treat, or their symptoms worsen even when appropriate neuroleptic levels were maintained (Knudsen and Vilmar 1984). Marihuana appears to consistently exacerbate the "positive" symptomatology of schizophrenia while producing inconsistent effects on "negative" symptoms. Patients who self-medicate with marihuana indicate their goal is to reduce negative symptoms.

The question of the causal relationship between abuse of marihuana and the development of schizophrenia has not been established, although some believe abuse leads to psychosis (Allebeck 1993; Negrete 1993). Those individuals abusing marihuana who also develop psychiatric problems suffer from rapid-onset schizophrenia and exhibit positive symptoms including auditory hallucinations and commenting voice (Allebeck 1993). Of those

schizophrenics that previously abused marihuana, almost 70% developed psychosis after more than 1 year of marihuana abuse. Though the mental abnormalities and related conditions attributed to cannabis abuse exist, it does not appear as though the psychosis can be distinguished from that either: (a) induced by other drugs of abuse or (b) found as endogenous schizophrenia (Taschner 1983). It is difficult to point to any one drug as the causative agent since these individuals are polydrug abusers. Proper studies have not been performed to determine the relative risk of development of psychiatric problems within marihuana abusers compared to nonabusers. However, the relative risk would actually appear to be small given the widespread abuse of marihuana.

3. Neurochemical and Histological Effects

The potential of neurochemical and histological damage produced by cannabinoids has been evaluated in both rats and monkeys. These results have been reviewed previously (Ali et al. 1991; Slikker et al. 1992). Generally, 7 months after a 1 year period of inhalation exposure of male rhesus monkeys, there was no evidence of neurochemical, histological or electron microscopic alterations in hippocampal volume, neuronal size, number or length of CA3 pyramidal cell dendrites or synaptic connections. Though Δ^9-THC could not be construed to be neurotoxic to CA3 neurons in these monkeys, further studies in the CA1, dentate granule cells, and cerebellar granule cells were being conducted to rule out other potential neurotoxic effects which were suggested elsewhere (Eldridge and Landfield 1992; Scallet et al. 1987). However, these largely negative results were obtained following a 1 year period of inhalation exposure of male rhesus monkeys (Slikker et al. 1992). It is quite possible that this period of treatment was too short to produce effects. There have also been attempts to monitor neurological changes in rats but the conditions (duration of exposure, marihuana vs THC, dose, etc.) have differed from those described above for monkeys. Administration of Δ^9-THC for a minimum period of 3 months was required before histochemical alterations were observed in the rat (Ali et al. 1991; Scallet 1991; Scallet et al. 1987). Comparatively, a 3 month period is a large portion (8%–10%) of the rat life span, and to obtain a similar exposure period in monkeys would require a 3 year exposure period and in humans would correspond to a 7 or 10 year period. A review of data in rats following lengths of Δ^9-THC administration of 3 months or greater indicated the formation in the CA3 region of the hippocampus of short, broken, axodendritic connections; a significant degree of extracellular space; and subcellular organelles (vesicles, mitochondria) were not separated from extracellular space by intact membranes (Scallet 1991). Other observations included a smaller neuronal size and fewer synaptic densities in the CA3 region. Reduced neuronal density was observed in the CA1 stratum pyramidal cells as was an increase in the proportion of opaque material within the cytoplasm

of astroglia. It is important to point out that the degree of histological change was greater in peripubertal animals than in young adults. Though it is entirely possible that these neurotoxic effects involved the cannabinoid receptor, it is important to realize that other possibilities exist. Additionally, it is possible that these structural changes resulted from indirect effects. The observed alterations could also have been produced by large increases in plasma corticosterone, which might have produced neurotoxic effects in the hippocampus via specific glucocorticoid receptors (Scallet 1991).

IV. Tolerance

1. Animals

Specific cannabis-mediated effects to which tolerance develops in a variety of species have been reviewed elsewhere (Harris et al. 1977; Jones 1983). Generally, tolerance develops to some degree to all cannabis-induced effects. However, there are exceptions in which the degree of tolerance development is so slight as to be considered not of physiological importance. Tolerance development has been shown to occur in all species studied. Parameters to which tolerance develops include simple physiological indices and complex behaviors mediated via the central nervous system. Some of these parameters in laboratory animals include Δ^9-THC-induced anticonvulsant activity, catalepsy, depression of locomotor activity, hypothermia, hypotension, immunosuppression, static ataxia in dogs, and alteration of response rates and accuracy on schedule-controlled behaviors.

The degree of tolerance that can be developed to Δ^9-THC is quite high. A 100-fold development of tolerance has clearly been observed in pigeons, dogs, and rodents. However, some reports actually indicate lack of activity with doses following chronic treatment which were 300- or 6000-fold higher than those initially effective in producing an effect. Tolerance has also been shown to the lethal effect of Δ^9-THC in pigeons. Similarly, tolerance to the toxic effects of oral doses of Δ^9-THC as high as 250 mg/kg per day in rats has been reported.

The onset of tolerance can be very rapid or may require a prolonged treatment period. However, generally, only 1 week of daily administration is required to observe tolerance to most simple parameters measured in rodents, dogs, or monkeys. Examples of rapid onset of tolerance include rodent hypothermia and decreased locomotor activity. In these cases, a decrement in response can be observed 24–48 h later upon administration of a second dose of Δ^9-THC, with nearly complete tolerance observed after a third treatment. In monkeys, tolerance develops to the sedative properties of Δ^9-THC after 2 weeks of oral treatment, while tolerance to some excitatory components of behavior required 2 months of treatment prior to the

development of tolerance. Thus, tolerance develops differentially in all species as a function of the parameter measured. It is also not necessary to treat animals on a frequent basis in order to develop tolerance. The administration interval of 7–9 days in the pigeon and 8 days in the dog has proven sufficient to maintain tolerance. Similarly, one Δ^9-THC treatment per week, for a period of 7 weeks, is sufficient to produce tolerance in pigeons to the suppressive effect of Δ^9-THC on response rate in positive reinforcement paradigms. Additionally, the tolerance developed using these kinds of treatment protocols is long-lasting. The tolerance development observed in the dog clearly was still present for at least 23 days. Though tolerance may be observed for a period of months in some parameters, the tolerance developed to other effects of Δ^9-THC have been shown to last for only up to 24 h.

2. Humans

Most investigators suggest that pronounced tolerance must occur prior to the development of physical dependence to a drug. There is convincing evidence of tolerance development to Δ^9-THC in humans (Jones et al. 1976). Tolerance developed to cannabis-induced increases in cardiovascular and autonomic functions, to decreased intraocular pressure, to sleep disturbances and sleep EEG, as well as mood and behavioral changes in those subjects receiving oral Δ^9-THC. It is not too surprising that there is less agreement with regard to the development of behavioral tolerance to cannabis. Psychological effects are highly complex and dependent upon many factors, not the least of which is the interaction between the subject and the environmental situation. In the studies in which high doses of Δ^9-THC have been employed, behavioral tolerance has been found. For example, studies with oral administration of Δ^9-THC revealed tolerance development to the subjective effects following a few days of 10 mg Δ^9-THC treatment administered several times each day (Jones 1983). Ten days of treatment with repetitive 30 mg doses of Δ^9-THC produced even greater tolerance to the behavioral effects. Tolerance to Δ^9-THC can best be summarized as relatively little tolerance development when the doses are small or infrequent and the drug exposure is of limited duration (Hollister 1986). Tolerance clearly develops when individuals are exposed to high doses for a sustained period of time.

V. Dependence

1. Animals

The most robust demonstration of physical dependence in laboratory animals has been made with chronic administration of drugs possessing a relatively short half-life. The long half-life and resultant long duration of action of Δ^9-

THC precludes the rapid induction of a drug-free system necessary for producing readily observable withdrawal signs and symptoms. Generally, the chronic administration of a drug with a half-life of greater than 35 h tends not to be followed by a withdrawal syndrome upon abrupt cessation of abuse. With these considerations in mind, studies were conducted in monkeys by intravenously administering Δ^9-THC every 6 h, with increasingly larger doses for 14 days. Administration at the highest dose attained for 12 more days (36 day regimen) produced significant physiological effects during drug abstinence (Kaymakcalan 1973). Symptoms appeared 12 h after the last drug treatment, and continued for 5 days. Symptoms included anorexia, hyperirritability, aggressiveness, tremors and twitching, penile erection, and masturbation with ejaculation, as well as behaviors interpreted as hallucinations. However, it is not clear that the observed behaviors were in fact withdrawal, since Δ^9-THC was not clearly shown to reverse the effects. Similarly, after continuous intravenous infusion of Δ^9-THC (daily dose of 1.2 mg/kg) for 10 days, three of four monkeys suffered a disruption of schedule-controlled behavior (Beardsley et al. 1986). Observers also noted that animals were aggressive and hyperactive during abstinence. Additionally, this withdrawal syndrome could be reversed by administration of Δ^9-THC. These studies may indeed suggest that cannabis is capable of producing dependence in animals, though in either experiment the symptoms were not severe.

Δ^9-THC produces a unique behavioral change in dogs first described as static ataxia (Walton et al. 1937). The administration of effective doses of Δ^9-THC on a daily basis produced tolerance to this effect. Increasing the dose to very high levels did not overcome this tolerance. However, the administration of increasingly large doses of Δ^9-THC to dogs over 11 days did not produce withdrawal symptoms during an 8 day period of drug abstinence (McMillan and Dewey 1972). Likewise, pigeons given daily intramuscular injections of very high doses of Δ^9-THC did not show withdrawal signs when the drug was removed (McMillan et al. 1970). Soon after the end of this treatment regimen there was a decrement in the operant behavior of the pigeons. However, this interruption of behavior was not felt to be an indication of withdrawal, since normal behavior was not reestablished when the drug was readministered.

It is, of course, impossible to measure psychological dependence in laboratory animals. Self-administration of drugs may be an indication of psychological dependence and/or abuse potential or craving. Yet, there are few reports which claim to have established experimental models in which animals self-administer Δ^9-THC or any of the majority of its analogs. The inability to maintain self-administration of Δ^9-THC was best shown when only a portion of the animals treated would self-administer Δ^9-THC after having had the drug administered to them for a long period of time prior to allowing the animal control of its drug supply (Kaymakcalan 1973). Despite

the opportunity to self-administer Δ^9-THC to prevent possible symptoms of withdrawal, only a small portion of the monkeys self-administered during the abstinence period. Instead, when given a choice, some monkeys self-administered cocaine rather than Δ^9-THC. This choice suggests that, even when experiments are designed to enhance Δ^9-THC self-administration, the abuse potential and possible development of psychological dependence to Δ^9-THC is tremendously less than to cocaine. Δ^9-THC (as well as related analogs) did not substitute for drugs with strong reinforcing properties (Carney et al. 1977). This failure also suggests limited potential for development of physical cross-dependence as well as limited psychological dependence due to weak reinforcing properties.

2. Humans

It is well established that chronic heavy use of either cannabis or hashish does not result in a withdrawal syndrome with severe symptomatology. The number of well controlled studies on the development of psychological or physical dependence to cannabis in humans is much less than those in various animal species. However, cannabis has been used for centuries, and there are a considerable number of reports in the literature, regarding the long term use of this material. The occurrence of a psychological dependence, abuse liability, or craving is more probable than physical dependence. It has been difficult to draw conclusions from epidemiological data on the psychological dependence of marihuana given the plethora of social and legal factors that impact on the drug abuser. However, there are numerous case reports of psychological dependence to cannabis (Jones 1983).

The early evidence for "dependence" upon cannabis arose from uncontrolled clinical observations following cessation of chronic drug intake. Most reports originated in countries such as India, Greece or Jamaica where cannabis or hashish had been used for long periods and was much higher in potency than the material smoked in the U.S. There were very early reports that smokers suddenly deprived of cannabis became hyperirritable, experienced auditory and visual hallucinations, and masturbated incessantly for 3–5 weeks (Fraser 1949). Abstinence symptoms in Egyptian hashish smokers were characterized as dysphoria, hyperirritability and insomnia (Soueif 1976). South African smokers reportedly experienced anxiety, restlessness, nausea, and cramps when cannabis was suddenly unavailable. Not too surprisingly, the conclusions reached by different investigators vary considerably. However, there are some commonalities among the descriptions of cannabis withdrawal which include hyperirritability, tremors, sweating, auditory and visual hallucinations, dysphoria, anxiety, negativism, insomnia or abnormal sleep patterns.

Some of the symptoms of cannabis "withdrawal" that have been described in uncontrolled clinical studies have also been reported in more

controlled experiments. In a very early study, subjects smoked an average of 17 marihuana cigarettes daily for 39 days and reported feeling "jittery" upon withdrawal, although observers were not able to detect any symptoms (Williams et al. 1946). Almost 30 years later, studies were conducted in which subjects were placed in a controlled environment and allowed them to smoke a self-determined number of marihuana cigarettes for a 21-day period (Greenberg et al. 1976; Mendelson et al. 1976). Upon termination of the smoking period, some subjects experienced rapid weight loss, decreased appetite, tremor, increased anxiety, hostility, decreased friendliness, etc. In another study, volunteers smoked marihuana for 64 days in a hospital. These subjects were allowed to self-medicate by smoking as many cigarettes as they wished, each containing 20 mg of Δ^9-THC (Cohen et al. 1976). Sleeplessness, anorexia, nausea and irritability developed after cessation of smoking. While similar conclusions can be drawn from all of the above studies, the results should be interpreted cautiously. These studies lacked placebo or double-blind controls, and attempts were not made to reverse the withdrawal symptoms by reinitiation of marihuana smoking. Additionally, there are always problems with confining individuals for long periods of time. It may well be that some of the subjects exhibited mood changes as a consequence of confinement. It should be kept in mind that the subjects were aware of the treatment schedule, and therefore could anticipate termination of drug administration. The issue of self-administration has both advantages and disadvantages. Self-administration may well provide the most realistic treatment regimen for marihuana users, and therefore cessation of such treatment would have relevance to the real world situation.

The development of tolerance and dependence to cannabis and Δ^9-THC has been examined under a more rigorous treatment paradigm (Jones 1983; Jones and Benowitz 1976; Jones et al. 1976, 1981). The premise was that if dependence did not result under these conditions, then it was highly unlikely to occur under less stringent conditions. Either Δ^9-THC or cannabis extract was administered orally to volunteers every 3 or 4 h, 24 h a day, for up to 21 days. The 30 mg dose of Δ^9-THC resulted in peak blood levels that were comparable to those obtained by smoking a marihuana cigarette. Cessation of treatment usually resulted in subjective effects which were first reported within 5–6 h after the last dose of Δ^9-THC. The most prominent and frequent symptoms were increased irritability and restlessness. Other prominent and somewhat variable symptoms were insomnia, anorexia, increased sweating and mild nausea. Objective changes included body weight loss, increased body temperature, and hand tremor. Both the subjective and objective changes could be diminished by smoking a marihuana cigarette or by readministration of oral Δ^9-THC, suggesting establishment of a withdrawal syndrome. The intensity of the effects observed was dependent upon the length of the treatment time and the dose of Δ^9-THC.

VI. Δ^9-THC During Pregnancy

1. Effect on Dams and Litters

Marihuana use in humans has been attributed to the low birth weight and small gestational size of infants (Hatch and Bracken 1986). In animals, Δ^9-THC during pregnancy increases the frequency of stillbirths (Gal and Sharpless 1984; Hutchings et al. 1989b) and decreases litter size (Wenger et al. 1992). Resorption rates increased in mice but not rats following in utero exposure. A decrease in maternal food and water consumption occurred and led to decreased maternal weight gain, which may be the cause of various effects associated with prenatal exposure (Abel 1985b). Prenatal exposure to cannabinoids led to a decrease in pup birth weight, which may be the only postnatal effect on offspring reliably demonstrated. In rodents, increased resorption of fetuses, perinatal death, and altered sex ratio (Morgan et al. 1988) also affects the final characteristics of the litter (Wenger et al. 1992). Some of these effects have been attributed to altered LH, FSH, progesterone, placental steroid excretion and/or inhibited prostaglandin synthesis (Dalterio et al. 1984; Wenger et al. 1992) In mice and rats low to moderate doses of Δ^9-THC did not affect the length of gestation, maternal viability, or maternal weight gain, though high doses of Δ^9-THC did prevent maternal weight gain (Bloch 1983). The decreased production of milk following birth of the litter has been linked to high neonatal mortality. Diminished lactation appears to be due to disruption of prolactin release and disruption of hypothalamic neuronal controls (Bloch 1983).

2. Developmental Toxicity

A great deal of effort has been expended on investigation of the effects of perinatal exposure to Δ^9-THC, largely based upon initial data suggesting the existence of various deficits in humans (O'Connell and Fried 1991; Tennes et al. 1985) and with the anticipation of finding a definable syndrome equivalent to the fetal alcohol syndrome. For example, in humans, prenatal marihuana exposure was reported to be related to tremors, increased startle, and poorer habituation to visual stimuli of offspring (Fried and Makin 1987). Investigations of minor physical abnormalities indicated that there was no correlation between the number of anomalies present in an individual and marihuana use, though two anomalies (true ocular hypertelorism and severe epicanthus) were found only among children of heavy users of cannabis (O'Connell and Fried 1984). However, a survery of the literature indicates that prenatal exposure to cannabinoids does not produce malformations in humans and only does so in mice following exposure to high doses administered by the intraperitoneal route (Abel 1985b). Long-term studies on postnatal effects have produced generally inconsistent results, which may be due to methodological flaws in experimental design (Abel

1985b). Experiments that do not consider the confounding influences of maternal toxicity (prenatal and postnatal) are likely to yield a high rate of false-positive results, which has been observed in studies of cannabis that preceded current concerns for pair-feeding and surrogate fostering (Abel 1985b; Hutchings and Dow-Edwards 1991). Nearly all such studies found neurobehavioral effects that included changes in activity as well as impairments in learning and memory (Hutchings and Dow-Edwards 1991). Thus, it is now generally concluded that there are no significant lasting effects that can be demonstrated on marihuana exposed offspring (Hutchings and Dow-Edwards 1991). Transient decrements on rodent body growth (Hutchings et al. 1989a) and brain potein synthesis (Morgan et al. 1988) have been observed in neonates following perinatal marihuana exposure, but these effects appeared to be due to maternal toxicity (Hutchings and Dow-Edwards 1991). When marihuana is administered via smoke inhalation a ventilation/perfusion imbalance is created and fetal oxygen availability limited (Clapp et al. 1987), but the effect appears to be related to the 30% reduction in maternal respiration, suggesting any fetal effects are an indirect toxicity, and this decrement has not been related to any developmental toxicity.

3. Neural Development

The effects of perinatal cannabinoid exposure on development, with special emphasis on disruption of dopaminergic neurons of the nigrostriatal, mesolimbic, and tuberoinfundibular systems, has been reviewed (Rodriguez de Fonseca et al. 1991a, 1992a,b). Alterations in these systems have been suggested to result in changes in locomotor activity. However, numerous other studies failed to detect changes in motor and endocrine function (Brake et al. 1987; Hutchings et al. 1989b). It is unclear whether any of these events on dopaminergic neurons are mediated by the cannabinoid CB1 receptor, especially considering the sexual dimorphism described. The dopaminergic effects of perinatal cannabinoids on males is more pronounced and prolonged than the effects observed in females (Rodriguez de Fonseca et al. 1992b). However, the presence of the cannabinoid receptor during the critical time of early development has been described (Rodriguez de Fonseca et al. 1993).

4. Teratogenicity

Though Δ^9-THC possesses some potential for production of teratogenic effects (Dalterio 1986), such alterations were only observed after very high doses of Δ^9-THC administered specifically before the end of organogenesis (Wenger et al. 1992). Many other studies find little evidence to support this contention (Rosenkrantz et al. 1986), and clinical studies have also failed to resolve this issue (Tuchmann-Duplessis 1993; Wenger et al. 1992). A

review of animal data has not provided convincing evidence of teratogenesis (Gal and Sharpless 1984; Rosenkrantz et al. 1986). Some data would suggest that marihuana abuse during pregnancy can induce fetal stress and hypoxia, and despite evidence suggesting enhanced startle or tremors in babies, a follow-up study at 1 year of age indicated no adverse mental or motor effects (Gal and Sharpless 1984). Fetotoxicity has been suggested (Nahas 1993a). However, reviews by others (Rosenkrantz 1985), indicate there is a lack of solid evidence supporting embryotoxicity in women despite findings of increased resorption of fetuses and perinatal death in rodents (Rosenkrantz 1985; Wenger et al. 1992).

5. Fetotoxicity – Interactions with Ethanol

A review of the neurobehavioral and developmental effects of fetal drug exposure indicates that the drugs most commonly associated with an adverse developmental outcome are alcohol, anticonvulsants, narcotics, etc. (Gal and Sharpless 1984). However, the potential for an interaction between marihuana and other substances suggests many potential dangers. One potential danger in pregnancy is suggested by the fact that combination treatment with marihuana and alcohol, at doses that were inactive alone, produced complete fetal mortality in mice and a 73% fetal mortality in rats (Abel 1985a). A superadditive effect was also suggested when alcohol (1 g/kg) and marihuana extract (50 or 100 mg/kg Δ^9-THC) were coadministered (Abel and Dintcheff 1986).

D. Behavioral Pharmacology

I. Unlearned Behaviors/Ethology

1. General Comments

Dewey (1986) has indicated that "little if any conclusive evidence has been presented which shows that the cannabinoids affect any peripheral system without working at least indirectly through the central nervous system (CNS)." Although cannabinoids have been shown to produce direct cellular actions on peripheral tissue, most effects of interest to researchers appear to involve a neural component of the CNS or autonomic system.

A variety of centrally mediated phenomena have been observed in mouse, rat, dog, rabbit and monkey and are reviewed elsewhere (Dewey 1986; Hollister 1986; Razdan 1986), but include measures of spontaneous and forced (rotorod) locomotion, hypothermia, immobility, antinociception, drug discrimination, static ataxia, anticonvulsant actions and operant behavioral measures. Also, hypersensitivity to auditory or tactile stimulation has been observed (Ferri et al. 1981). By defining the spectrum of activity

(efficacy, potency, etc.) of naturally occurring cannabinoids in a series of procedures (Martin et al. 1987), it has been possible to determine whether new synthetic and structurally diverse chemical structures are cannabimimetic (Compton et al. 1992a,b; Martin et al. 1987).

2. Consummatory Behavior

It is well known that marihuana users consistently report an increased hunger during acute intoxication of the drug (Halikas et al. 1985). The numerous anecdotal accounts indicating that marihuana increases feeding behavior and body weight have suggested its potential thereapeutic use as an appetite stimulant for cancer or AIDS patients (Plasse et al. 1991). In actuality, there is a scarcity of experimental evidence supporting this cannabinoid action. In a previous review (Meyer 1978), the weight gain associated with marihuana smoking has been suggested to result from an increased appetite for sweets and carbohydrate-containing fruit drinks. Whether the self-reported appetite-enhancing effects of marihuana in humans is a direct drug effect or results from a complex interaction between the drug and social influences is an unresolved issue. Some research supports the latter explanation (Foltin et al. 1986). In an attempt to simulate a natural setting, human subjects were housed in a residential laboratory and allowed to smoke marihuana cigarettes prior to a private work period and during a social access period. A single active marihuana cigarette prior to the private work period had no effect on food intake. The administration of two or three active marihuana cigarettes during the social access period did not increase meal size but did increase daily caloric intake as between-meal snack items. Further research is needed to ascertain the mechanism by which cannabinoids enhance appetite.

In contrast, there is little experimental support in the animal literature for appetite enhancing effects of the cannabinoids. These compounds are typically reported to decrease food consumption and weight gain relative to the vehicle-treated subjects (Abel and Schiff 1969; Sofia and Barry 1974). The initial anorectic effect of Δ^9-THC by either i.p. or i.v. route of administration in rats, was found to diminish after 5 days of frequent administration; however, daily weight gain remained suppressed compared to the controls (Miczek and Dihit 1980; Verbene et al. 1980).

3. Motor Behavior

Comparison of changes in motor activity between animals and humans has not been easy (Rosenkrantz 1983). Human motor activity is highly variable, and greatly affected by prior drug exposure, psychosocial setting, cultural customs, etc. However, when high doses of cannabinoids are administered intravenously to humans a definite lethargy and sedation has been demon-

strated which would seem to resemble animal results (Rosenkrantz 1983). In laboratory animals cannabinoids have been shown to elicit locomotor effects in a variety of tasks including the static ataxia test in dogs, alterations in spontaneous activity in mice, catalepsy in mice and rats, and impairment in the rotorod test (Consroe and Mechoulam 1987; Little et al. 1988). The static ataxia test was one of the earlier behavioral tests employed to evaluate the psychoactivity of cannabinoids (Walton et al. 1937). In this paradigm, a dog is administered an intravenous dose of cannabinoid and the degree to which the animal exhibits motor dysfunction is rated by observers. Although noncannabinoid substances also produce ataxia, this paradigm was useful in identifying psychoactivity in both naturally occurring and synthetic cannabinoids (Martin et al. 1976, 1984; Wilson and May 1974, 1979). Recently, the dog static ataxia test for the assessment of cannabinoids has been largely replaced by rodent models of motor behavior that also reliably predict psychoactivity.

One measure that is used to identify cannabinoid activity is the assessment of spontaneous locomotor activity. Cannabinoids generally lead to decreases in spontaneous locomotion (Little et al. 1988). A variety of cannabinoids including the naturally occurring compounds as well as the synthetic agents from either the aminoalkylindole class (Compton et al. 1992a) or the bicyclic class (Compton et al. 1992b) have all been shown to produce hypomotility. Similarly, cannabinoids produce decreases in response rates under different schedules of reinforcement in a variety of species (Carney et al. 1979; Zuardi and Karniol 1983). Several studies have demonstrated biphasic effects on spontaneous locomotion. Cannabinoids are known to produce hypomotility at medium to high dose and increases in activity after treatment with low doses. A similar phenomenon has been observed using the low rate differential reinforcement schedule of operant behavior in rats (Hiltunen et al. 1989). The basis for the dissociation between the inhibitory and stimulatory effects of marihuana on locomotor activity may be related to its effects in different brain structures. However, the phenomenon is also found with other central depressants, such as the barbiturates and minor tranquilizers (Harris et al. 1966).

Another well established motor effect of marihuana is its propensity to cause animals to maintain a rigid posture or catalepsy. Cataleptic effects of cannabinoids have been assessed both the bar immobility test (Ferri et al. 1981) and the ring immobility test (Pertwee 1972). Clearly, the extrapyramidal system seems to play an important role in these effects. Intracerebral administration of either Δ^9-THC or 11-OH-Δ^9-THC into the caudate putamen had a moderate cataleptic effect (Gough and Olley 1978). Although neither of these drugs produced catalepsy in the globus pallidus, intrapallidal injections of the potent analog 11-OH-Δ^8-THC-dimethylheptyl produced catalepsy (Pertwee and Wickens 1991).

4. Social Behavior

a) Motivation in Humans

The belief that "frequent use of marihuana by young adolescents can impede normal maturation and cause or contribute to an amotivational syndrome" has sometimes been expressed (Schwartz et al. 1987; Tunving 1987; Watanabe et al. 1984), but a controversy exists concerning the existence of an "amotivational syndrome" as associated with long-term marihuana abuse (Hollister 1986; Maykut 1984; Page 1983; Solomons and Neppe 1989). An amotivational syndrome could generally be described as a condition of apathy, lethargy, a flattening of affect, and a lack of goal-oriented behavior. Attempts to verify the existence of such an effect in controlled humans studies or epidemiological studies in localities of great abuse have either failed to provide evidence of such a syndrome or observed other factors which could potentially produce the phenomenon observed or only found residual effects of acute Δ^9-THC administration (Dewey 1986; Foltin et al. 1989, 1990; Hollister 1986; Maykut 1984). Additionally, some changes that could be observed in an individual's character during long-term abuse of marihuana did not appear different from that produced by the abuse of any other licit or illicit drug (Taschner 1983). It seems likely that the lack of "motivation" in humans is more a function of drug abuse and psychosocial issues than of marihuana abuse per se.

b) Sensory and Other Effects in Animals

A 1 year period of repeated marihuana inhalation exposure in male rhesus monkeys appeared to reduce the "motivational" aspects of food reinforced responding in a progressive ratio protocol of an operant behavioral task (Paule et al. 1992; Slikker et al. 1992). The general health of the animals was not compromised, though both short- and long-term treatment stressed animals significantly as evidenced by urinary cortisol output. Similarly, cessation produced a physiological stress response that could have been indicative of a "withdrawal" phenomenon. There were no residual behavioral effects of chronic marihuana treatment 7 months after the termination of treatments. Similar studies in rodents (Scallet 1991) indicated altered performances in mazes, avoidance of footshock by motor activity, performance in memory tasks (in an eight-arm radial maze), deficits on differential reinforcement of a low lever-pressing response rate operant schedules, and decrements in rotorod performance.

One of the most notable cannabinoid effects is their ability to inhibit the perception of noxious sensory stimulation. Treatment with Δ^9-THC has been reported to decrease pain in patients suffering from neoplastic disease with a potency similar to that of codeine; however, the sedative and other intoxicating effects of the drug limited its clinical usefulness (Noyes et al. 1975).

In the nonhuman animal literature, the antinociceptive properties of cannabinoids have been demonstrated in a variety of pain assays including the tail-flick test (Martin 1985b), the hot plate test (Fride and Mechoulam 1993; Welch and Stevens 1992), and the *p*-phenylquinone writhing test (Formukong et al. 1988; Haubrich et al. 1990). Much of the research examining neuroanatomical and neurochemical mechanisms of cannabinoid-induced antinociception have employed the tail-flick test. Cannabinoids appear to produce antinociception through both spinal and supraspinal components of action because spinal transection attenuated but did not completely block the antinociceptive effects of intravenously administered cannabinoids (Lichtman and Martin 1991; Smith and Martin 1992). In addition, spinal administration and i.c.v. administration of cannabinoids were found to produce antinociception in a variety of species (Martin et al. 1993; Welch et al. 1994). Although those results indicate brain involvement, additional studies are required to elucidate the neural substrates of cannabinoid-induced antinociception. The occurrence of a higher concentration of cannabinoid receptors in the periaqueductal gray, a structure that plays an important role in antinociception, than other brainstem structures suggested its involvement in cannabinoid-induced antinociception. Consequently, intracerebral administration of CP-55,940 into the ventrolateral periaqueductal gray in the region of the dorsal raphe was found to elicit a potent antinociceptive effect, thus indicating its involvement in the antinociceptive effects of cannabinoids. Whether other brain areas also contribute to the antinociceptive effects of cannabinoids is an issue for additional research.

II. Conditioned Effects

1. Drug Discrimination

Despite the lack of methodological means to measure euphoria in animals (see self-administration below), the drug discrimination paradigm (Balster and Prescott 1992) has been a very useful model to assess the intoxicating effects of cannabinoids. In this paradigm, nonhuman primates, rats, or pigeons are trained to make two different responses for reinforcement contingent upon whether the training drug or vehicle were administered (Gold et al. 1992; Järbe and Hiltunen 1987; Weissman 1978). Once the subjects are able to discriminate successfully they can be administered other drugs to determine if these substances produce similar or different stimulus characteristics from the training drug. Drugs found to generalize to Δ^9-THC in the drug discrimination paradigm have also been reported to be marihuana-like in humans or bind to the cannabinoid receptor. Cannabinoids that are known to possess distinct structures but nonetheless bind to the cannabinoid receptor, including the aminoalkylindole (Compton et al. 1992a) and bicyclic

(Gold et al. 1992) compounds, have been shown to substitute for Δ^9-THC. This paradigm elicited a high degree of specificity because substances belonging to other drug classes, including dopaminergic, benzodiazepine, opioid, cholinergic, and noradrenergic, do not reliably substitute for THC (Balster and Prescott 1992). The relative potencies of drugs that generalize to THC also exhibit similar binding affinity to the cannabinoid receptor.

2. Self-Administration

The facts that 60% of all young adults in the United States have used marijuana in their lifetime and more than 10% of this age group use it on a regular basis (Johnston et al. 1993) strongly suggest that this drug is a positive reinforcer. The self-administration paradigm in animals has been a valuable tool in predicting the abuse liabilities of drugs. However, there has been a general inability to obtain cannabinoid self-administration in non-human animals. The few published studies which employed this paradigm reported that Δ^9-THC was an ineffective reinforcer in rhesus monkeys (Harris et al. 1974). Because of the relatively delayed onset and the long duration of effect of cannabinoids and their rate decreasing effects, attempts were made to adapt the self-administration procedure by using a fixed interval schedule to circumvent response rate suppression (Mansbach et al., in press). Similar to studies employing the fixed ratio schedule, this attempt also failed to establish self-administration in laboratory animals. The apparent inability to establish cannabinoids as a reinforcer in the self-administration paradigm suggests a limitation in this model to predict the abuse liability of drugs in humans or that they have a low abuse liability.

Alternatively, cannabinoids have also been documented to elicit various aversive effects. In laboratory rodents, Δ^9-THC has been shown to act as an unconditioned stimulus in the taste aversion paradigm. Moreover, Δ^9-THC has been found to act as an anxiogenic agent in the elevated plus maze (Onaivi et al. 1990). Therefore, it may be that these apparent negative hedonic properties may mask the appetitive properties of cannabinoids and thus account for their failure to serve as positive reinforcers in the self-administration paradigm. Other research has demonstrated that cannabinoids act upon brain regions involved with reinforcement. Relatively high concentrations of cannabinoid receptors have been found in the nucleus accumbens (Herkenham et al. 1991b; Jansen et al. 1992; Thomas et al. 1992). Gardner and his colleagues have provided convincing evidence that cannabinoids produce effects upon the mesotelencephalic dopamine reward pathway similar to other rewarding drugs (Gardner and Lowinson 1991). Δ^9-THC was found to reduce the amount of electric current required for self-stimulation in the medial forebrain bundle (Gardner et al. 1988). In addition, systemic administration of Δ^9-THC was found to increase DA efflux in the nucleus accumbens (Chen et al. 1990). These effects are similar to those of other drugs which are reported to have positive hedonic effects in humans.

3. Performance, Memory and Learning

a) Intoxication and Performance Impairment

It seems safe to assume that the goal of most marihuana abusers is to attain a state of intoxication (Chait and Zacny 1992; Jones 1971). The possible role of cannabinoids regarding the brain reward system has been summarized by others (Gardner 1992; Gardner and Lowinson 1991). The euphoria coincides with adverse effects of behavioral toxicity, including alteration of motor control, sensory functions, and the cognitive process (Nahas 1993a; Nahas and Latour 1992). Impairment of both motor control and cognitive processes could easily lead to accidents and traffic fatalities (Aussedat and Niziolek-Reinhardt 1993). Nonvehicular accidents (Soderstrom et al. 1993) have been linked to abuse of marihuana. However, the question asked should be: What is the relationship between marihuana consumption, blood or urine levels of drug, and the degree of incoordination or loss of function that is produced? (Hollister et al. 1981; Soderstrom et al. 1993). The resolution of this issue would more clearly substantiate the detrimental effects of marihuana abuse by establishing the causal relationship between the period of psychomotor disruption and in vivo levels of Δ^9-THC or metabolites, which has obvious medico-legal implications.

The impact of marihuana on task performance in humans has been reviewed extensively (Chait and Pierri 1992; Leirer et al. 1991, 1993). Although there are innumerable problems interpreting a large number of studies when a diversity of methods and approaches have been taken, the authors were able to draw several general conclusions. In summary, at moderate levels of intoxication, there is a weak correlation between the incidences of heart rate increases and levels of euphoria. Marihuana and Δ^9-THC adversely affect gross and simple motor ability (body sway as measured on a "wobble board" and hand tremor), as well as some psychomotor behaviors (rotary pursuit, digit symbol substitution test, reaction time in choice reaction time tasks, accuracy in divided attention tasks, sustained attention) while not adversely affecting other tasks (simple reaction time, hand-eye coordination). Interestingly, in some studies in which chronic abusers were evaluated, the results suggested, in comparison to similar studies not using chronic abusers, that a large degree of tolerance may develop in humans to some of these acute effects. In conclusion, similar to the situation with alcohol consumption, cannabis intoxication of an experienced abuser may be difficult to detect except in performance tasks for which they have had no previous training or in tasks requiring a great deal of skill and/or manual dexterity. However, cannabis intoxication in an inexperienced individual would be readily detectable, but not necessarily on all performance measures.

Cannabinoid-induced impairment of flying (Leirer et al. 1991) and driving (Hollister 1986; Moskowitz 1985) have been documented. Each of

these tasks presumably require a great deal of manual dexterity and undisrupted cognition and therefore might be expected to readily detect the intoxicating effects of any drugs. A review of the impaired flying studies (Leirer et al. 1993) suggests that individuals trained on computerized flight simulations perform less well than controls on five of the eight variables measured for up to 24 h after treatment. However, in a second more sophisticated experiment, the researchers failed to replicate those results. Yet, in a third study where the level of flight difficulty increased, and subjects were allowed less training on the simulation than in the first study, the global score (aggregate of six variables) for simulated flight was significantly altered at times up to 24 h. It is interesting that these latter studies did not attempt to replicate the detrimental effects of age and Δ^9-THC consumption on simulated flight, during which older "pilots" faired worse than their younger counterparts. The data suggested that either the level of impairment (though statistically significant) was not of functional relevance in terms of performance (at least in younger pilots) or the testing procedure was not appropriate for measuring impairment in humans. Also, it should be noted that impairment was not observed in all individuals (Chait and Pierri 1992).

There is little doubt that automobile accidents have been linked to intoxication of the driver by marihuana and a variety of other drugs, sometimes used in combination (Mason and McBay 1984; Moskowitz 1985). Coabuse of marihuana with either alcohol (Wechsler et al. 1984) or with phencyclidine (Poklis et al. 1987) is common. However, it is also true that abuse of marihuana alone can disrupt driving performance if the task is of sufficient difficulty or the dose high enough. A summary of these results (Hollister 1986) suggests that intoxicating levels of alcohol impairs performance more than does marihuana. Unlike alcohol intoxication, not all driving measures were affected by marihuana and not all subjects were affected. The combination of alcohol with marihuana was more detrimental than either drug alone. Interestingly, when allowed to smoke marihuana until intoxicated, 94% of the individuals failed a roadside sobriety test 90 min after smoking, and 60% failed 150 min after smoking.

b) Memory and Time Perception

Δ^9-THC impairs memory and learning (Chait and Pierri 1992; Schwartz 1993), but results on specific evaluations are often inconsistent and test specific (Chait and Pierri 1992). The paradigms in which Δ^9-THC produces its greatest effects (10%–50% decrement) are in free recall tasks or short-term memory function (Chait and Pierri 1992). Some reviewers believe that data indicate long-term (possibly permanent) impairment of short-term memory in adolescent age chronic marihuana abusers (Schwartz 1993). It also appears that some individuals suffer no memory impairment at all and that as a group those with any type of learning disability are more affected than the exceptionally gifted student group (Schwartz 1993). Thus, the

question could be asked: Are marihuana abusers unsuccessful students because they smoke cannabis, or do they smoke cannabis because they are underachievers? Preliminary data support the latter contention and also suggest that continued abuse of marihuana and other substances also involves other factors (Johnson 1988; Johnson and Pandina 1991; Labouvie 1990).

A thorough review of the literature indicated that Δ^9-THC reliably alters the perception of time (Chait and Pierri 1992). Subjects overestimated time elapsed relative to real clock time or experienced an increase in the subjective rate of time. Attempts to demonstrate other behavioral effects on mental function have not met with such certainty (Chait and Pierri 1992). Mixed or inconsistent results have been obtained on the Stroop (color and word) test, mental arithmetic capability, and various "creativity" tasks, although significant effects of marihuana administration were observed on an embedded figures task (finding geometric figures within a more complex design) and on verbal output tests. Thus, psychomotor performance would be expected to the impaired if short-term memory and/or time perception were required for that task. Perhaps this is true and is reflected in driving or piloting studies, but evaluation of work productivity in groups of heavy marihuana abusers has indicated no decrement in performance (Hollister 1986).

c) Memory in Animals

Cannabinoids have long been known to impair learning and memory in a variety of tasks in rodents (Carlini et al. 1970), nonhuman primates (Ferraro and Grilly 1973), and humans (Abel 1971). In rats, Δ^9-THC has been found to disrupt memory as assessed in the delayed match-to-sample (DMTS) task (Heyser et al. 1993), Lashley III maze (Carlini et al. 1970), and the eight-arm radial-maze (Lichtman et al., in press; Nakamura et al. 1991). In nonhuman primates Δ^9-THC has been found to disrupt chaining behavior and the DMTS (Rupniak et al. 1991). In addition to Δ^9-THC, the structurally distinct synthetic compounds CP-55,940 and WIN-55,212-2 were also found to impair working memory in rats. Interestingly, anandamide failed to impair working memory in either the eight-arm radial maze or the delayed nonmatch to sample tasks (Crawley et al. 1993). This difference between Δ^9-THC and the endgoenous ligand of cannabinoid in memory function underscores the other differences found between these compounds (Smith et al. 1994).

The high concentration of cannabinoid receptors found in the hippocampus (Herkenham et al. 1991b; Jansen et al. 1992; Thomas et al. 1992) may mediate the disruptive effects of these drugs on cognition. Research from a convergence of in vitro and in vivo studies further implicates the involvement of the hippocampus in cannabinoid-induced memory impairment. Δ^9-THC applied to hippocampal tissue biphasically affected long-term potentiation (Nowicky et al. 1987), a neural mechanism believed to play a

prominent role in information storage in the brain. Long-term administration of Δ^9-THC decreased the concentration of synapses in the CA3 region of the hippocampus (Scallet et al. 1987). Δ^9-THC-induced impairment in the DMTS task was associated with a specific decrease in hippocampal cell discharge (Heyser et al. 1993). Direct evidence implicating hippocampal involvement was that intrahippocampal administration of CP-55,940 led to a dose-related increase in the number of errors committed in the eight-arm radial maze task. The effects of intrahippocampal CP-55,940 were apparently specific to cognition because no other cannabinoid pharmacological effects (e.g., antinociception, hypothermia, and catalepsy) were elicited. Δ^9-THC has been shown to alter cerebral metabolism in several brain regions including the hippocampus (Margulies and Hammer 1991).

D. Conclusions

Marihuana remains one of the most widely abused substances throughout the world. Despite a wide range of pharmacological effects on most organ systems, the health consequences of chronic marihuana abuse are relatively mild when compared to those of most other abused substances. There is no question that marihuana is capable of producing impairment of performance in individuals while intoxicated. Memory and learning decrements are well documented under specific circumstances. Attempts to establish neurochemical and histological damage produced by cannabinoids have not resulted in definitive conclusions. However, the current data suggest that a rigorous treatment during a long exposure period will be required if permanent neurological deficits are produced.

Considerable progress has been made regarding the characterization of cannabinoid receptor subtypes in brain and peripheral tissues, and there has been some insight into second messenger systems. The discovery of endogenous cannabinoids and the characterization of their synthetic and metabolic enzymes provides the basic foundation for establishing an endogenous cannabinoid system. The recent development of a cannabinoid antagonist will greatly facilitate this undertaking. The physiological role of cannabinoids should emerge in the not too distant future.

Acknowledgements. Portions of this research were supported by NIDA grants DA-00490 and DA-03672.

References

Abel EL (1971) Retrieval of information after use of marihuana. Nature 231:58

Abel EL (1985a) Alcohol enhancement of marihuana-induced fetotoxicity. Teratology 31:35–40

Abel EL (1985b) Effects of prenatal exposure to cannabinoids. In: Pinkert TM (ed) Current research on the consequences of maternal drug abuse. US Government Printing Office, Washington DC, pp 20–35

Abel EL, Dintcheff BA (1986) Increased marihuana-induced fetotoxicity by a low dose of concomitant alcohol administration. J Stud Alcohol 47:440–443
Abel EL, Schiff BB (1969) Effects of the marihuana homologue, parahexyl, on food and water intake and curiousity in the rat. Psychon Sci 16:38
Abood ME, Sauss C, Fan F, Tilton CL, Martin BR (1993) Development of behavioral tolerance of Δ^9-THC without alteration of cannabinoid receptor binding or mRNA levels in the whole brain. Pharmacol Biochem Behav 46:575–579
Adams IB, Thomas BF, Compton DR, Razdan RK, Martin BR (in press) Evaluation of cannabinoid receptor binding and in vivo activities for anandamide analogs. J Pharmacol Exp Ther
Adams MD, Earnhardt JT, Dewey WL, Harris LS (1976) Vasoconstrictor actions of Δ^8- and Δ^9-tetrahydrocannabinol in the rat. J Pharmacol Exp Ther 196:649–656
Adams MD, Earnhardt JT, Martin BR, Harris LS, Dewey WL, Razdan RK (1977) A cannabinoid with cardiovascular activity but no overt bahavioral effects. Experientia 33:1204–1205
Adams R (1942) Marihuana. Harvey Lect 37:168–197
Ali SF, Newport GD, Scallet AC, Paule MG, Bailey JR, Slikker W (1991) Chronic marijuana smoke exposure in the rhesus monkey. IV. Neurochemical effects and comparison to acute and chronic exposure to Δ^9-tetrahydrocannabinol (THC) in rats. Pharmacol Biochem Behav 40:677–682
Allebeck P (1993) Schizophrenia and cannabis: cause-effect relationship? In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 113–117
Arata S, Newton C, Klein T, Friedman H (1992) Enhanced growth of Legionella pneumophila in tetrahydrocannabinol-treated macrophages. Proc Soc Exp Biol Med 199:65–67
Aronow WS, Cassidy J (1975) Effect of smoking marihuana and of a high-nicotine cigarette on angina pectoris. Clin Pharmacol Ther 17(S):549–554
Askew WE, Ho BT (1974) The effects of tetrahydrocannabinols on cyclic AMP levels in rat brain areas. Experientia 30:879–880
Audette CA, Burstein SH, Doyle SA, Hunter SA (1991) G-protein mediation of cannabinoid-induced phospholipase activation. Pharmacol Biochem Behav 40: 559–563
Aussedat M, Niziolek-Reinhardt S (1993) Detection of cannabis and other drugs in 120 victims of road accidents. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 73–77
Balster RL, Prescott WR (1992) Δ^9-tetrahydrocannabinol discrimination in rats as a model for cannabis intoxication. Neurosci Biobehav Rev 16:55–62
Barnett C, Chiang C, Perez-Reyes M, Owens S (1982) Kinetic study of smoking marijuana. J Pharmacokinet Biopharm 10:495–506
Barnett G, Licko V, Thompson T (1985) Behavioral pharmacokinetics of marijuana. Psychopharmacology 85:51–56
Bartolucci G, Fryer L, Perris C, Shagass C (1969) Marijuana psychosis: a case report. Can Psychiatr Assoc J 14:77–79
Beardsley PM, Balster RL, Harris LS (1986) Dependence on tetrahydrocannabinol in rhesus monkeys. J Pharmacol Exp Ther 239:311–319
Benowitz NL, Rosenberg J, Rogers W, Bachman J, Jones RT (1979) Cardiovascular effects of intavenous Δ^9-tetrahydrocannabinol: autonomic nervous mechanisms. Clin Pharmacol Ther 25:440–446
Bhargava HN (1976) Effect of some cannabinoids on naloxone-precipitated abstinence in morphine-dependent mice. Psychopharmacology 49:267–270
Bhargava HN (1978) Time course of the effects of naturally occurring cannabinoids on morphine abstinence syndrome. Pharmacol Biochem Behav 8:7–11
Bhattacharya SK (1986) Δ^9-Tetrahydrocannabinol increases brain prostaglandins in the rat. Psychopharmacology 90:499
Bhattacharya SK, Ghosh P, Sanyal AK (1980) Effects of prostaglandins on some central pharmacological actions of cannabis. Indian J Med Res 71:955–960

Bidaut-Russell M, Howlett A (1988) Opioid and cannabinoid analgetics both inhibit cyclic AMP production in the rat striatum. In: Hamon M, Cros I, Meunier J (eds) Advances in the biosciences: proceedings of the international narcotics research conference. Pergamon, Oxford

Bidaut-Russell M, Howlett AC (1991) Cannabinoid receptor-regulated cyclic AMP accumulation in the rat striatum. J Neurochem 57:1769–1773

Bloch E (1983) Effects of marihuana and cannabinoids on reproduction, endocrine function, development, and chromosomes. In: Fehr KO, Kalant H (eds) Cannabis and health hazards: proceedings of an ARF/WHO scientific meeting on adverse health and behavioral consequences of cannabis use. Addiction Research Foundation, Toronto, pp 355–432

Bloom AS, Dewey WL (1978) A comparison of some pharmacological actions of morphine and Δ^9-tetrahydrocannabinol in the mouse. Psychopharmacology 57: 243–248

Brake SC, Hutchings DE, Morgan B, Lasalle E, Shi T (1987) Delta-9-tetrahydrocannabinol during pregnancy in the rat: II. Effects on ontogeny of locomotor activity and nipple attachment in the offspring. Neurotoxicol Teratol 9:45–49

Brostrom MA, Brostrom CO, Breckenridge BM, Wolff DJ (1978) Calcium-dependent regulation of brain adenylate cyclase. Adv Cyclic Nucleotide Res 9:85–99

Buchanan JF, Davis LJ (1984) Drug-induced infertility. Drug Intell Clin Pharm 18:122–132

Burstein S (1992) Eicosanoids as mediators of cannabinoid action. In: Murphy L, Bartke A (eds) Marijuana/cannabinoids: neurobiology and Neurophysiology. CRC Press, Boca Raton, pp 73–91

Burstein S, Hunter SA (1981) Prostaglandins and cannabis – VIII. Elevation of phospholipase A_2 activity by cannabinoids in whole cells and subcellular preparations. J Clin Pharmacol 21:240S–248S

Burstein S, Raz A (1972) Inhibition of prostaglandin E2 biosynthesis by Δ^1-THC. Prostaglandins 2:369–374

Burstein S, Levin E, Varanelli C (1973) Prostaglandins and cannabis-II: inhibition of biosynthesis by the naturally occuring cannabinoids. Biochem Pharmacol 22:2905–2910

Burstein S, Varanelli C, Slade LT (1974) Prostaglandins and cannabis-III. Inhibition of biosynthesis by essential oil components of marihuana. Biochem Pharmacol 24:1053–1054

Burstein SH, Hunter SA, Shoupe TS, Taylor P (1978) Cannabinoid inhibition of testosterone synthesis by mouse Leydig cells. Res Commun Chem Pathol Pharmacol 19:557–560

Burstein S, Hunter SA, Shoupe TS (1979) Site of inhibition of leydig cell testosterone synthesis by Δ^9-tetrahydrocannabinol. Mol Pharmacol 15:633–640

Burstein S, Hunter SA, Sedor C (1980) Further studies on the inhibition of Leydig cell testosterone production by cannabinoids. Biochem Pharmacol 29:2153–2154

Burstein S, Ozman K, Burstein E, Palermo N, Smith E (1982) Prostaglandins and cannabis – XI: inhibition of Δ^1-tetrahydrocannabinol-induced hypotension by aspirin. Biochem Pharmacol 31:591–592

Burstein S, Hunter SA, Renzulli L (1985) Prostaglandins and cannabis XIV. Tolerance to the stimulatory actions of cannabinoids on arachidonate metabolism. J Pharmacol Exp Ther 235:87–91

Burstein S, Hunter SA, Latham V, Renzulli L (1987) A major metabolite of Δ^1-tetrahydrocannabinol reduces its cataleptic effect in mice. Experientia 43: 402–403

Burstein SH, Hull K, Hunter SA, Shilstone J (1989) Immunization against prostaglandins reduces Δ^1-THC-induced catalepsy in mice. Mol Pharmacol 35: 6–9

Burstein SH, Audette CA, Charalambous A, Doyle SA, Guo Y, Hunter SA, Makriyannis A (1991) Detection of cannabinoid receptors by photoaffinity labelling. Biochem Biophys Res Commun 176:492–497

Cabral GA, Vasquez R (1993) Δ^9-tetrahydrocannabinol suppresses macrophage extrinsic anti-herpesvirus activity. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 137–153

Carlini EA, Hamaoui A, Bieniek D, Korte F (1970) Effects of $(-)\Delta^9$-trans-tetrahydrocannabinol and a synthetic derivative on maze performance of rats. Pharmacology 4:359–368

Carney JM, Uwayday IM, Balster RL (1977) Evaluation of a suspension system for intravenous self-administration studies with water-insoluble compounds in the Rhesus monkey. Pharmacol Biochem Behav 7:357–364

Carney JM, Balster RL, Martin BR, Harris LS (1979) Effects of systemic and intraventricular administration of cannabinoids on schedule-controlled responding in the squirrel monkey. J Pharmacol Exp Ther 210:399–404

Caulfield MP, Brown DA (1992) Cannabinoid receptor agonists inhibit Ca current in NG108-15 neuroblastoma cells via a pertussis toxin-sensitive mechanism. Br J Pharmacol 106:231–232

Cavero I, Buckley JP, Bhagavan SJ (1973) Hemodynamic and myocardial effects of $(-)$-Δ^9-trans-tetrahydrocannabinol in anesthetized dogs. Eur J Pharmacol 24:243–251

Chait LD, Pierri J (1992) Effects of smoked marijuana on human performance: a critical review. In: Murphy L, Bartke A (eds) Marijuana/cannabinoids: neurobiology and neurophysiology. CRC Press, Boca Raton, pp 387–423

Chait LD, Zacny JP (1992) Reinforcing and subjective effects of oral Δ^9-THC and smoked marijuana in humans. Psychopharmacology 107:255–262

Charalambous A, Yan G, Houston DB, Howlett AC, Compton DR, Martin BR, Makriyannis A (1992) 5′-Azido-Δ^8-THC: A novel photoaffinity label for cannabinoid receptor. J Med Chem 35:3076–3079

Chaudry A, Thompson RH, Rubin RP, Laychock SG (1988) Relationship between Δ^9-tetrahydrocannabinol-induced arachidonic acid release and secretagogue-evoked phosphoinositide breakdown and Ca^{2+} mobilization of exocrine pancreas. Mol Pharmacol 34:543–548

Chen J, Paredes W, Lowinson JH, Gardner EL (1990) Δ^9-Tetrahydrocannabinol enhances presynaptic dopamine efflux in medial prefrontal cortex. Eur J Pharmacol 190:259–262

Chen J, Marmur R, Pulles A, Paredes W, Gardner E (1993) Ventral tegmental microinjection of Δ^9-tetrahydrocannabinol enhances ventral tegmental somatodendritic dopamine levels but not forebrain dopamine levels: evidence for local neural action by marijuana's psychoactive ingredient. Brain Res 621:65–70

Chesher GB, Jackson DM (1985) The quasimorphine withdrawal syndrome: effect of cannabinol, cannabidiol and tetrahydrocannabinol. Pharmacol Biochem Behav 23:13–15

Chesher GB, Dahl CJ, Everingham M, Jackson DM, Merchant-Williams H, Starmer GA (1973) The effect of cannabinoids on intestinal motility and their antinociceptive effect in mice. Br J Pharmacol 49:588–594

Childers SR, Fleming L, Konkoy C, Marckel D, Pacheo M, Sexton T, Ward S (1992) Opioid and cannabinoid receptor inhibition of adenylyl cyclase in brain. Ann NY Acad Sci 654:33–51

Childers SR, Sexton T, Roy MB (1994) Effects of anandamide on cannabinoid receptors in rat brain membranes. Biochem Pharmacol 47:711–715

Chkili T, Ktiouet JE (1993) Prospective study of 104 psychiatric cases associated with cannabis use in a Moroccan medical center. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 101–104

Clapp JFI, Wesley M, Cooke R, Pekala R, Holstein C (1987) The effects of marijuana smoke on gas exchange in ovine pregnancy. Alcohol Drug Res 7:85–92

Cocchetto D, Owens S, Perez-Reyes M, Di Guiseppi S, Miller L (1981) Relationship between plasma delta-9-tetrahydrocannabinol concentration and pharmacologic effects in man. Psychopharmacology 75:158–164

Cohen S, Lessin P, Hahn PM, Tyrell ED (1976) A 94-day cannabis study. In: Braude MC, Szara S (eds) Pharmacology of marihuana. Raven, New York, pp 621–626

Compton DR, Dewey WL, Martin BR (1990a) Cannabis dependence and tolerance production. In: Erickson CK, Javors MA, Morgan WW (eds) Addiction potential of abused drugs and drug classes. Hayworth, Binghampton, pp 129–147

Compton DR, Little PJ, Martin BR, Gilman JW, Saha JK, Jorapur VS, Sard HP, Razdan RK (1990b) Synthesis and pharmacological evaluation of amino, azido, and nitrogen mustard analogues of 10-substituted cannabidiol and 11- or 12-substituted Δ^8-tetrahydrocannabinol. J Med Chem 33:1437–1443

Compton DR, Gold LH, Ward SJ, Balster RL, Martin BR (1992a) Aminoalkylindole analogs: cannabimimetic activity of a class of compounds structurally distinct from Δ^9-tetrahydrocannabinol. J Pharmacol Exp Ther 263:1118–1126

Compton DR, Johnson MR, Melvin LS, Martin BR (1992b) Pharmacological profile of a series of bicyclic cannabinoid analogs: classification as cannabimimetic agents. J Pharmacol Exp Ther 260:201–209

Compton DR, Rice KC, De Costa BR, Razdan RK, Melvin LS, Johnson MR, Martin BR (1993) Cannabinoid structure-activity relationships: correlation of receptor binding and in vivo activities. J Pharmacol Exp Ther 265:218–226

Cone E, Huestis M (1993) Relating blood concentrations of tetrahydrocannabinol and metabolites to pharmacologic effects and time of marihuana usage. Ther Drug Mon 15:527–532

Cone EJ, Johnson RE, Paul BD, Mell LD, Mitchell J (1988) Marijuana-laced brownies: behavioral effects, physiologic effects, and urinalysis in humans following ingestion. J Anal Toxicol 12:169–175

Consroe P, Mechoulam R (1987) Anticonvulsant and neurotoxic effects of tetrahydrocannabinol stereoisomers. In: Rapaka RS, Markiyannis A (eds) Structure-activity relationships of the cannabinoids. US Government Printing Office, Washington DC, pp 59–66

Crawley J, Corwin R, Robinson J, Felder C, Devane W, Axelrod J (1993) Anandamide, an endogenous ligand of the cannabinoid receptor, induces hypomotility and hypothermia in vivo in rodents. Pharmacol Biochem Behav 46:967–972

Dalterio SL (1986) Cannabinoid exposure: effects on development. Neurobehav Toxicol Teratol 8:345–352

Dalterio S, Bartke A, Harper MJK, Huffman R, Sweeney C (1981) Effects of cannabinoids and female exposure on the pituitary-testicular axis in mice: possible involvement of prostaglandins. Biol Reproduct 24:315–322

Dalterio S, Steger R, Mayfield D, Bartke A (1984) Early cannabinoid exposure influences neuroendocrine and reproductive functions in male mice: I. Prenatal exposure. Pharmacol Biochem Behav 20:107–113

Defer B (1993) Cannabis and schizophrenia: how causal a relationship? In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 119–121

Deutsch DG, Chin SA (1993) Enzymatic synthesis and degradation of anadamide, a cannabinoid receptor agonist. Biochem Pharmacol 46:791–796

Devane W, Axelrod J (1994) Enzymatic synthesis of anandamide, an endogenous ligand for the cannabinoid receptor by brain membranes. Proc Natl Acad Sci USA 91:6698–6701

Devane WA, Spain JW, Coscia CJ, Howlett AC (1986) An assessment of the role of opioid receptors in the response to cannabimimetic drugs. J Neurochem 46:1929–1935

Devane WA, Dysarz FA, Johnson MR, Melvin LS, Howlett AC (1988) Determination and characterization of a cannabinoid receptor in rat brain. Mol Pharmacol 34:605–613

Devane WA, Breuer A, Sheskin T, Järbe TUC, Eisen MS, Mechoulam R (1992a) A novel probe for the cannabinoid receptor. J Med Chem 35:2065–2069

Devane WA, Hanus L, Breuer A, Pertwee RG, Stevenson LA, Griffin G, Gibson D, Mandelbaum A, Etinger A, Mechoulam R (1992b) Isolation and structure of a brain constituent that binds to the cannabinoid receptor. Science 258: 1946–1949

Dewey WL (1986) Cannabinoid pharmacology. Pharmacol Rev 38:151–178

Dewey WL, Jenkins J, Rourke T, Harris LS (1972) The effects of chronic administration of *trans*-Δ^9-tetrahydrocannabinol on behavior and the cardiovascular system. Arch Int Pharmacol Ther 198:118–131

Dill JA, Howlett AC (1988) Regulation of adenylate cyclase by chronic exposure to cannabimimetic drugs. J Pharmacol Exp Ther 244:1157–1163

Dolby TW, Kleinsmith LJ (1974) Effects of Δ^9-tetrahydrocannabinol on the levels of cyclic adenosine 3′,5′-monophosphate in mouse brain. Biochem Pharmacol 23:1817–1825

Dolby TW, Kleinsmith LJ (1977) Cannabinoid effects on adynalate cyclase and phosphodiesterase activities of mouse brain. Can J Physiol Pharmacol 55:934–942

Drew WG, Miller LL (1974) Cannabis: neural mechanisms and behavior – a theoretical review. Pharmacology 11:12–32

Eldridge JC, Landfield PW (1990) Cannabinoid interactions with glucocorticoid receptors in rat hippocampus. Brain Res 534:135–141

Eldridge JC, Landfield PW (1992) Cannabinoid-glucocorticoid interactions in the hippocampal region of the brain. In: Murphy L, Bartke A (eds) Marijuana/ cannabinoids: neurobiology and neurophysiology. CRC Press, Boca Raton, pp 93–117

ElSohly MA, Ross SA (1994) Quaterly report, NIDA potency monitoring project. Report no 50

Evans D, Lake JT, Johnson MR, Howlett A (1994) Endogenous cannabinoid receptor binding activity released from rat brain slices by depolarization. J Pharmacol Exp Ther 268:1271–1277

Evans DM, Johnson MR, Howlett AC (1992) Ca^{2+}-dependent release from rat brain of cannabinoid receptor binding activity. J Neurochem 58:780–782

Fairbairn JW, Pickens JT (1979) The oral activity of Δ^9-tetrahydrocannabinol and its dependence on prostaglandin E2. Br J Pharmacol 67:379–385

Fairbairn JW, Pickens JT (1980) The effect of conditions influencing endogenous prostaglandins on the activity of Δ^9-tetrahydrocannabinol in mice. Br J Pharmacol 69:491–493

Falkenstein BA, Holley DC (1992) Effect of acute intravenous administration of Δ^9-tetrahydrocannabinol on the episodic secretion of immunoassayable growth hormone in the rat. Life Sci 50:1109–1116

Feigenbaum JJ, Bergmann R, Richmond SA, Mechoulam R, Nadler V, Kloog Y, Sokolovsky M (1989) Nonpsychotropic cannabinoid acts as a functional N-methyl-D-aspartate receptor blocker. Proc Natl Acad Sci USA 86:9584–9587

Felder CC, Veluz JS, Williams HL, Briley EM, Matsuda LA (1992) Cannabinoid agonists stimulate both receptor- and non-receptor-mediated signal transduction pathways in cells transfected with and expressing cannabinoid receptor clones. Mol Pharmacol 42:838–845

Felder CC, Briley EM, Axelrod J, Simpson JT, Mackie K, Devane WA (1993) Anandamide, an endogenous cannabimimetic eicosanoid, binds to the cloned human cannabinoid receptor and stimulates receptor-mediated signal transduction. Proc Natl Acad Sci USA 90:7656–7660

Fernández-Ruiz JJ, de Fonseca FR, Navarro M, Ramos JA (1992) Maternal cannabinoid exposure and brain development: changes in the ontogeny of dopaminergic

neurons. In: Murphy L, Bartke A (eds) Marijuana/cannabinoids: neurobiology and neurophysiology. CRC Press, Boca Raton, pp 119–164
Ferraro DP, Grilly DM (1973) Lack of tolerance to Δ^9-tetrahydrocannabinol in chimpanzees. Science 179:490–492
Ferri S, Costa G, Murari G, Panico AM, Rapisarda E, Speroni E, Arrigo RR (1981) Investigations on behavioral effects of an extract of Cannabis sativa L. in the rat. Psychopharmacology 75:144–147
Fischer-Stenger K, Updegrove AW, Cabral GA (1992) Δ^9-tetrahydrocannabinol decreases cytotoxic T lymphocyte activity to herpes simplex virus type 1-infected cells. Proc Soc Exp Biol Med 200:422–430
Foltin RW, Brady JV, Fischman MW (1986) Behavioral analysis of marijuana effects on food intake in humans. Pharmacol Biochem Behav 25:577–582
Foltin RW, Fischman MW, Brady JV, Kelly TH, Bernstein DJ, Nellis MJ (1989) Motivational effects of smoked marihuana: behavioral contingencies and high-probability recreational activities. Pharmacol Biochem Behav 34:871–877
Foltin RW, Fischman MW, Brady JV, Bernstein DJ, Nellis MJ, Kelly TH (1990) Marijuana and behavioral contingencies. Drug Dev Res 20:67–80
Formukong EA, Evans AT, Evans FJ (1988) Analgesic and antiinflammatory activity of constituents of Cannabis sativa L. Inflammation 12:361–371
Forney RB, Kiplinger GF (1971) Toxicology and pharmacology of marijuana. Ann NY Acad Sci 191:74–82
Fraser JD (1949) Withdrawal symptoms in cannabis-indica addicts. Lancet 257: 747–748
Fride E, Mechoulam R (1993) Pharmacological activity of the cannabinoid receptor agonist, anandamide, a brain constituent. Eur J Pharmacol 231:313–314
Fried PA, Makin JE (1987) Neonatal behavioral correlates of prenatal exposure to marihuana, cigarettes and alcohol in a low risk population. Neurotoxicol Teratol 9:1–7
Gahlinger PM (1984) Gastrointestinal illness and cannabis use in a rural Canadian community. J Psychoactive Drugs 16:263–265
Gal P, Sharpless MK (1984) Fetal drug exposure-behavioral teratogenesis. Drug Intell Clin Pharm 18:186–201
Gamse R, Holzer P, Lembeck F (1979) Indirect evidence for presynaptic location of opiate receptors in chemosensitive primary sensory neurones. Arch Pharmacol 308:281–285
Gardner EL (1992) Cannabinoid interaction with brain reward systems – the neurobiological basis of cannabinoid abuse. In: Murphy L, Bartke A (eds) Marijuana/cannabinoids: neurobiology and neurophysiology. CRC Press, Boca Raton, pp 275–335
Gardner EL, Lowinson JH (1991) Marijuana's interaction with brain reward systems – update 1991. Pharmacol Biochem Behav 40:571–580
Gardner EL, Paredes W, Smith D, Donner A, Milling C, Cohen A, Morrison D (1988) Facilitation of brain stimulation reward by Δ^9-tetrahydrocannabinol. Psychopharmacology 96:142–144
Gascon AL, Peres MT (1973) Effect of Δ^9- and Δ^8-tetrahydrocannabinol on the peripheral autonomic nervous system in vitro. Can J Physiol Pharmacol 51:12–21
Gennings C, Carter JWH, Martin BR (1994) Response-surface analysis of morphine sulfate and Δ^9-tetrahydrocannabinol interaction in mice. In: Lange N, Ryan L (eds) Case studies in biometry. Wiley, New York, pp 429–451
George HR (1970) Two psychotic epidodes associated with cannabis. Br J Addict 65:119–121
Gérard C, Mollereau C, Vassart G, Parmentier M (1990) Nucleotide sequence of a human cannabinoid receptor cDNA. Nucleic Acids Res 18:7142
Gilbert PE (1981) A comparison of THC, nantradol, nabilone, and morphine in the chronic spinal dog. J Clin Pharmacol 21:311S–319S

Gold L, Balster RL, Barrett RL, Britt DT, Martin BR (1992) A comparison of the discriminative stimulus properties of Δ^9-THC and CP-55 940 in rats and rhesus monkeys. J Pharmacol Exp Ther 262:479–486

Gough AL, Olley JE (1978) Catalepsy induced by intrastriatal injections of Δ^9-THC and 11-OH-Δ^9-THC in the rat. Neuropharmacology 17:137–144

Greenberg I, Mendelson JH, Kuehnle JC, Mello N, Babor TF (1976) Psychiatric and behavioral observations of casual and heavy marijuana users in a controlled research setting. Ann NY Acad Sci 282:72–84

Halikas JA, Weller RA, Morse CL, Hoffmann RG (1985) A longitudinal study of marijuana effects. Int J Addict 20:701–711

Hanus L, Gopher A, Almog S, Mechoulam R (1993) Two new unsaturated fatty acid ethanolamides in brain that bind to the cannabinoid receptor. J Med Chem 36:3032–3034

Harclerode J (1984) Endocrine effects of marijuana in the male: preclinical studies. In: Braude MC, Ludford JP (eds) Marijuana effects on the endocrine and reproductive systems. US Government Printing Office, Washington DC, pp 46–64

Harris RA, Stokes JA (1982) Cannabinoids inhibit calcium uptake by brain synaptosomes. J Neurosci 2:443–447

Harris LS, Pearl J, Aceto MD (1966) Similarities in the effects of barbiturates. Psychonom Sci 4:267–268

Harris R, Waters W, McLendon D (1974) Evaluation of reinforcing capability of Δ^9-tetrahydrocannabinol in rhesus monkeys. Psychopharmacology 37:23–29

Harris LS, Dewey WL, Razdan RK (1977) Cannabis: its chemistry, pharmacology, and toxicology. In: Martin WR (ed) Drug addiction. Springer, Berlin Heidelberg New York, pp 371–429 (Handbook of experimental pharmacology, vol 45/2)

Harris LS, Carchman RA, Martin BR (1978) Evidence for the existence of specific cannabinoid binding sites. Life Sci 22:1131–1138

Hatch EE, Bracken MB (1986) Effect of marijuana use in pregnancy on fetal growth. Am J Epidemiol 124:986–993

Haubrich DR, Ward SJ, Baizman E, Bell MR, Bradford J, Ferrari R, Miller M, Perrone M, Pierson AK, Saelens JK, Luttinger D (1990) Pharmacology of pravadoline: a new analgesic agent. J Pharmacol Exp Ther 255:511–522

Heindel JJ, Keith WB (1989) Specific inhibition of FSH-stimulated cAMP accumulation by Δ^9-tetrahydrocannabinol in cultures of rat Sertoli cells. Toxicol Appl Pharmacol 101:124–134

Herkenham M, Lynn AB, Little MD, Johnson MR, Melvin LS, DeCosta BR, Rice KC (1990) Cannabinoid receptor localization in the brain. Proc Natl Acad Sci USA 87:1932–1936

Herkenham M, Lynn AB, DeCosta BR, Richfield EK (1991a) Neuronal localization of cannabinoid receptors in the basal ganglia of the rat. Brain Res 547:267–274

Herkenham M, Lynn AB, Johnson MR, Melvin LS, de Costa BR, Rice KC (1991b) Characterization and localization of cannabinoid receptors in rat brain: a quantitative in vitro autoradiographic study. J Neurosci 11:563–583

Heyser CJ, Hampson RE, Deadwyler SA (1993) Effects of Δ^9-tetrahydrocannabinol on delayed match to sample performance in rats: alterations in short-term memory associated with changes in task specific firing of hippocampal cells. J Pharmacol Exp Ther 264:294–307

Hillard CJ, Bloom AS (1983) Possible role of prostaglandins in the effects of the cannabinoids on adenylate cyclase activity. Eur J Pharmacol 91:21–27

Hillard CJ, Pounds JJ, Boyer DR, Bloom AS (1990) Studies of the role of membrane lipid order in the effects of Δ^9-tetrahydrocannabinol on adenylate cyclase activation in heart. J Pharmacol Exp Ther 252:1075–1082

Hiltunen AJ, Jarbe TU, Kamkar MR, Archer T (1989) Behaviour in rats maintained by low differential reinforcement rate: effects of Δ^1-tetrahydrocannabinol, cannabinol and cannabidiol, alone and in combination. Neuropharmacology 28:183–189

Hine B (1985) Morphine and Δ^9-tetrahydrocannabinol: two-way cross tolerance for antinociceptive and heart rate responses in the rat. Psychopharmacology 87:34–38

Hine B, Friedman E, Torellio M, Gershon S (1975) Morphine-dependent rats: blockade of precipitated abstinence by tetrahydrocannabinol. Science 187: 443–445

Hollister LE (1986) Health aspects of cannabis. Pharmacol Rev 38:1–20

Hollister LE (1988) Marijuana and immunity. J Psychoactive Drugs 20:3–8

Hollister LE, Gillespie HK, Ohlsson A, Lindgren J-E, Wahlen A, Agurell S (1981) Do plasma concentrations of Δ^9-tetrahydrocannabinol reflect the degree of intoxication? J Clin Pharmacol 21:171S–177S

Howes JF, Osgood PF (1976) Cannabinoids and the inhibition of prostaglandin synthesis. In: Nahas GG (ed) Marihuana: chemistry, biochemistry and cellular effects. Springer, Berlin Heidelberg New York, pp 415–424

Howlett AC (1984) Inhibition of neuroblastoma adenylate cyclase by cannabinoid and nantradol compounds. Life Sci 35:1803–1810

Howlett AC (1985) Cannabinoid inhibition of adenylate cyclase. Biochemistry of the response in neuroblastoma cell membranes. Mol Pharmacol 27:429–436

Howlett AC (1987) Cannabinoid inhibition of adenylate cyclase: relative activity of constituents and metabolites of marihuana. Neuropharmacology 26:507–512

Howlett AC, Fleming RM (1984) Cannabinoid inhibition of adenylate cyclase. Pharmacology of the response in neuroblastoma cell membranes. Mol Pharmacol 26:532–538

Howlett AC, Qualy JM, Khachatrian LL (1986) Involvement of Gi in the inhibition of adenylate cyclase by cannabimimetic drugs. Mol Pharmacol 29:307–313

Howlett AC, Johnson MR, Melvin LS, Milne GM (1988) Nonclassical cannabinoid analgetics inhibit adenylate cyclase: development of a cannabinoid receptor model. Mol Pharmacol 33:297–302

Howlett AC, Bidaut-Russell M, Devane WA, Melvin LS, Johnson MR, Herkenham M (1990a) The cannabinoid receptor: biochemical, anatomical and behavioral characterization. TINS 13:420–423

Howlett AC, Champion TM, Wilken GH, Mechoulam R (1990b) Stereochemical effects of 11-OH-Δ^8-tetrahydrocannabinol-dimethylheptyl to inhibit adenylate cyclase and bind to the cannabinoid receptor. Neuropharmacology 29:161–165

Howlett AC, Evans DM, Houston DB (1992) The cannabinoid receptor. In: Murphy L, Bartke A (eds) Marijuana/cannabinoids: neurobiology and neurophysiology. CRC Press, Boca Raton, pp 35–72

Huestis MA, Henningfield JE, Cone EJ (1992a) Blood Cannabinoids: I. Absorption of THC and formation of 11-OH-THC and THCCOOH during and after smoking marijuana. J Anal Toxicol 16:276–282

Huestis MA, Sampson AH, Holicky BJ, Henningfield JE, Cone EJ (1992b) Characterization of the absorption phase of marijuana smoking. Clin Pharmacol Ther 52:31–41

Hunter SA, Audette CA, Burstein S (1991) Elevation of brain prostaglandin E_2 levels in rodents by Δ^1-THC. Prostaglandins Leukot Essent Fatty Acids 43:185

Hutchings DE, Brake SC, Morgan B (1989a) Animal studies of prenatal Δ^8-tetrahydrocannabinol: female embryolethality and effects on somatic and brain growth. Ann NY Acad Sci 562:133–144

Hutchings DE, Gamagaris Z, Miller N, Fico TA (1989b) The effects of prenatal exposure to Δ^9-tetrahydrocannabinol to the rest-activity cycle of the preweanling rat. Neurotoxicol Teratol 11:353–356

Hutchings DE, Dow-Edwards D (1991) Animal models of opiate, cocaine, and cannabis use. Clin Perinatol 18:1–22

Iverfeldt K, Ögren SO, Barfai T (1988) Substance P receptors in the rat spinal cord: the effect of GTP and of chronic antidepressant treatment. Acta Physiol Scand 132:175–179

Jandhyala BS, Buckley JP (1977) Autonomic and cardiovascular effects of chronic Δ^9-tetrahydrocannabinol administration in mongrel dogs. Res Commun Chem Pathol Pharmacol 16:593–607

Jansen EM, Haycock DA, Ward SJ, Seybold VS (1992) Distribution of cannabinoid receptors in rat brain determined with aminoalkylindoles. Brain Res 575:93–102

Järbe TU, Hiltunen AJ (1987) Cannabimimetic activity of cannabinol in rats and pigeons. Neuropharmacol 26:219–228

Johansson E, Agurell S, Hollister L, Halldin M (1988) Prolonged apparent half-life of delta-9-tetrahydrocannabinol in plasma of chronic marijuana users. J Pharm Pharmacol 40:374–375

Johson MR, Melvin LS (1986) The discovery of nonclassical cannabinoid analgetics. In: Mechoulam R (ed) Cannabinoids as therapeutic agents. CRC Press, Boca Raton, pp 121–144

Johnson V (1988) Adolescent alcohol and marijuana use: a longitudinal assessment of a social learning perspective. Am J Drug Alcohol Abuse 14:419–439

Johnson V, Pandina RJ (1991) Effects of the family environment on adolescent substance use, delinquency, and coping styles. Am J Drug Alcohol Abuse 17:71–88

Johnston LD, O'Malley PM, Bachman JG (1994) National survey results on drug use from the monitoring the future study, 1975–1993, National survey results on drug use from the monitoring the future study, 1975–1993, vol 1 and 2. US Department of Health and Human Services, Washington DC

Jones RT (1971) Marihuana-induced "high": influence of expectation, setting and previous drug experience. Pharmacol Rev 23:359–369

Jones RT (1983) Cannabis tolerance and dependence. In: Fehr KO, Kalant H (eds) Cannabis and health hazards. Addiction Research Foundation, Toronto, pp 617–689

Jones RT, Benowitz N (1976) The 30-day trip – clinical studies of cannabis tolerance and dependence. In: Braude MC, Szara S (eds) Pharmacology of marihuana. Raven, New York, pp 627–642

Jones RT, Benowitz N, Bachman J (1976) Clinical studies of cannabis tolerance and dependence. Ann NY Acad Sci 282:221–239

Jones RT, Benowitz NL, Herning RI (1981) Clinical relevance of cannabis tolerance and dependence. J Clin Pharmacol 21:143S–152S

Jorapur VS, Khalil ZH, Duffley RP, Razdan RK, Martin BR, Harris LS, Dewey WL (1985) Hashish: Synthesis and central nervous system activity of some novel analogues of cannabidiol and oxepin derivatives of Δ^9-tetrahydrocannabinol. J Med Chem 28:783–787

Kaminski NE, Abood ME, Kessler FK, Martin BR, Schatz AR (1992) Identification of a functionally relevant cannabinoid receptor on mouse spleen cells that is involved in cannabinoid-mediated immune modulation. Mol Pharmacol 42:736–742

Kawakami Y, Klein TW, Newton C, Djeu JY, Dennert G, Spector S, Friedman H (1988) Suppression by cannabinoids of a cloned cell line with natural killer cell activity. Proc Soc Exp Biol Med 187:355–359

Kaymakcalan S (1973) Tolerance to and dependence on cannabis. Bull Narc 25:39–47

Kaymakcalan S, Deneau GA (1972) Some pharmacologic properties of synthetic Δ^9-tetrahydrocannabinol. Acta Med Turc [Suppl] 1:5

Kaymakcalan S, Ercan ZS, Turker RK (1975) The evidence of the release of prostaglandin-like material from rabbit kidney and guinea-pig lung by (−)-trans-Δ^9-THC. J Pharm Pharmacol 27:564–568

Kelly LA, Butcher RW (1973) The effects of Δ^1-tetrahydrocannabinol on cyclic AMP levels in WI-38 fibroblasts. Biochim Biophysica Acta 1973:540–544
Kelly LA, Butcher RW (1979) Effects of Δ^1-tetrahydrocannabinol (THC) on cyclic AMP metabolism in cultured human fibroblasts. In: Abood LG, Sharp CW (eds) Membrane mechanisms of drugs of abuse. Liss, New York, pp 227–236
Klein TW, Newton C, Friedman H (1987) Inhibition of natural killer cell function by marijuana components. J Toxicol Environ Health 20:321–332
Knudsen P, Vilmar T (1984) Cannabis and neuroleptic agents in schizophrenia. Acta Psychiatr Scand 69:162–174
Kochar MS, Hosko MJ (1973) Electrocardiographic effects of marihuana. J Am Med Assoc 225:25–27
Koe BK, Milne GM, Weissman A, Johnson MR, Melvin LS (1985) Enhancement of brain [^{3}H]flunitrazepam binding and analgesic activity of synthetic cannabimimetics. Eur J Pharmacol 109:201–212
Koutek B, Prestwich GD, Howlett AC, Chin SA, Salehani D, Akhavan N, Deutsch DG (1994) Inhibitors of arachidonyl ethanolamide hydrolysis. J Biol Chem 269:22937–22940
Kruszka K, Gross R (1994) The ATP- and CoA-independent synthesis of arachidonoylethanolamide. J Biol Chem 269:14345–14348
Kumbaraci NM, Nastuk WL (1980) Effects of Δ^9-tetrahydrocannabinol on excitable membranes and neuromuscular transmission. Mol Pharmacol 17:344–349
Kuster JE, Stevenson JI, Ward SJ, D'Ambra TE, Haycock DA (1993) Aminoalkylindole binding in rat cerebellum: selective displacement by natural and synthetic cannabinoids. J Pharmacol Exp Ther 264:1352–1363
Labouvie EW (1990) Personality and alcohol and marihuana use: patterns of convergence in young adulthood. Int J Addict 25:237–252
Landfield PW, Cadwallader LB, Vinsant S (1988) Quantitative changes in hippocampal structure following long-term exposure to Δ^9-tetrahydrocannabinol: possible mediation by glucocorticoid systems. Brain Res 443:47
Layman JM (1971) Some actions of Δ^1-tetrahydrocannabinol and cannabidiol at cholinergic junctions. Proc Br Pharmacol Soc 41:379p
Leader JP, Koe BK, Weisman A (1981) GABA-like actions of levonantradol. J Clin Pharmacol 21:2625–2705
Leirer VO, Yesavage JA, Morrow DG (1991) Marijuana carry-over effects on aircraft pilot performance. Aviat Space Environ Med 62:221–227
Leirer VO, Yesavage JA, Morrow DG (1993) Marijuana carry-over effects on psychomotor performance: a chronicle of research. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 47–60
Lemberger L, Crabtree RE, Rowe HM (1972a) 11-Hydroxy-Δ^9-tetrahydrocannabinol: pharmacology, disposition, and metabolism of a major metabolite of marihuana in man. Science 177:62–64
Lemberger L, Weiss JL, Watanabe AM, Galanter IM, Wyatt RJ, Cardon PV (1972b) Δ^9-Tetrahydrocannabinol: temporal correlation of the psychologic effects and blood levels after various routes of administration. N Eng J Med 286:685–688
Lemberger L, McMahon R, Archer R, Matsumoto K, Rowe H (1974) Pharmacologic effects and physiologic disposition of delta^{6a,10a} dimethylheptyl tetrahydrocannabinol (DMHP) in man. Clin Pharmacol Ther 15:380–386
Li DMF, Ng CKM (1984) Effects of Δ^1- and Δ^6-tetrahydrocannabinol on the adynalate cyclase activity in ventricular tissue of the rat heart. Clin Exp Pharmacol Physiol 11:81–85
Lichtman AH, Martin BR (1991) Spinal and supraspinal mechanisms of cannabinoid-induced antinociception. J Pharmacol Exp Ther 258:517–523
Lichtman AH, Dimen KR, Martin BR (1995) Systemic or intrahippocampal cannabinoid administration impairs spatial memory in rats. Psychopharmacology 119:282–290

Lindgren J, Ohlsson A, Agurell S, Hollister L, Gillespie H (1981) Clinical effects and plasma levels of delta-9-tetrahydrocannabinol in heavy and light users of cannabis. Psychopharmacology 74:208–212

Linton PH, Kuechenmeister CA, White HB, Travis RP (1975) Marijuana: heart rate and EEG response. Res Commun Chem Pathol Pharmacol 10:201–214

Little PJ, Martin BR (1991) The effects of Δ^9-tetrahydrocannabinol and other cannabinoids on cAMP accumulation in synaptosomes. Life Sci 48:1133–1141

Little PJ, Kaplan NC, Martin BR (1987) Pharmacological profile of Δ^9-THC carbamate. Alcohol Drug Res 7:517–523

Little PJ, Compton DR, Johnson MR, Melvin LS, Martin BR (1988) Pharmacology and stereoselectivity of structurally novel cannabinoids in mice. J Pharmacol Exp Ther 247:1046–1051

Little PJ, Compton DR, Mechoulam R, Martin BR (1989) Stereochemical effects of 11-OH-dimethylheptyl-Δ^8-tetrahydrocannabinol. Pharmacol Biochem Behav 32:661–666

Loewe S (1946) The rate of disappearance of marihuana-active substance from the circulating blood. J Pharmacol Exp Ther 294–296

Lynn A, Herkenham M (1994) Localization of cannabinoid receptors and nonsaturable high-density cannabinoid binding sites in peripheral tissues of the rat: implications for receptor-mediated immune modulation by cannabinoids. J Pharmacol Exp Ther 268:1612–1623

Mackie K, Hille B (1992) Cannabinoids inhibit N-type calcium channels in neurobalstoma-glioma cells. Proc Natl Acad Sci USA 89:3825–3829

Mackie K, Devane W, Hille B (1993) Anandamide, an endogenous cannabinoid, inhibits calcium currents as a partial agonist in N18 neuroblastoma cells. Mol Pharmacol 44:498–503

Mailleux P, Vanderhaeghen J (1993a) Dopaminergic regulation of cannabinoid receptor mRNA levels in the rat caudate-putamen: an in situ hybridization study. J Neurochem 61:1705–1712

Mailleux P, Vanderhaeghen JJ (1993b) Glucocorticoid regulation of cannabinoid receptor messenger RNA levels in the rat caudate-putamen. An in situ hydridization study. Neurosci Lett 156:51–53

Malit LA, Johnstone RE, Bourke DI, Kulp RA, Klein V, Smith TC (1975) Intravenous Δ^9-tetrahydrocannabinol: effects on ventilatory control and cardiovascular dynamics. Anesthesiology 42:666–673

Mansbach RS, Nicholson KL, Martin BR, Balster RL (in press) Failure of Δ^9-tetrahydrocannabinol and CP 55,940 to maintain intravenous self-administration under a fixed-interval schedule in rhesus monkeys. Behav Pharmacol

Margulies JE, Hammer RP (1991) Δ^9-tetrahydrocannabinol alters cerebral metabolism in a biphasic, dose-dependent manner in rat brain. Eur J Pharmacol 202:373–378

Martin BR (1985a) Characterization of the antinociceptive activity of intravenously administered Δ^9-tetrahydrocannabinol in mice. In: Harvey DJ (ed) Marihuana '84 proceedings of the oxford symposium on cannabis. IRL Press, Oxford, pp 685–692

Martin BR (1985b) Structural requirements for cannabinoid-induced antinociceptive activity in mice. Life Sci 36:1523–1530

Martin BR (1986) Cellular effects of cannabinoids. Pharmacol Rev 38:45–74

Martin BR, Dewey WL, Harris LS, Beckner JS (1976) ^{3}H-Δ^9-tetrahydrocannabinol tissue and subcellular distribution in the central nervous system and distribution in peripheral organs of tolerant and nontolerant dogs. J Pharmacol Exp Ther 196:128–144

Martin BR, Kallman MJ, Kaempf GF, Harris LS, Dewey WL, Razdan RK (1984) Pharmacological potency of R- and S-3′-hydroxy-Δ^9-tetrahydrocannabinol: additional structural requirement for cannabinoid activity. Pharmacol Biochem Behav 21:61–65

Martin BR, Compton DR, Little PJ, Martin TJ, Beardsley PM (1987) Pharmacological evaluation of agonistic and antagonistic activity of cannabinoids. In: Rapaka RS, Makriyannis A (eds) Structure-activity relationships of cannabinoids. US Government Printing Office, Washington DC, pp 108–122

Martin BR, Howlett AS, Welch SP (1989) Cannabinoid action in the central nervous system. In: Harris LS (ed) Problems of drug dependence 1988. Proceedings of the 50th annual scientific meeting. US Government Printing Office, Washington DC, 275–283

Martin BR, Compton DR, Thomas BF, Prescott WR, Little PJ, Razdan RK, Johnson MR, Melvin LS, Mechoulam R, Ward SJ (1991) Behavioral, biochemical, and molecular modeling evaluations of cannabinoid analogs. Pharmacol Biochem Behav 40:471–478

Martin BR, Welch SP, Abood M (1994) Progress toward understanding the cannabinoid receptor and its second messenger systems. In: August JT, Anders MW, Murad F (eds) Advances in pharmacology. Academic, San Diego, pp 341–397

Martin WJ, Lai NK, Patrick SL, Tsou K, Walker JM (1993) Antinociceptive actions of cannabinoids following intraventricular administration in rats. Brain Res 629:300–304

Mason AP, McBay AJ (1984) Ethanol, marijuana, and other drug use in 600 drivers killed in single-vehicle crashes in North Carolina, 1978–1981. J Forensic Sci 29:987–1026

Mason AP, McBay AJ (1985) Cannabis: pharmacology and interpretation of effects. J Forensic Sci 30:615–631

Mathew RJ, Wilson WH (1992) The effects of marijuana on cerebral blood flow and metabolism. In: Murphy L, Bartke A (eds) Maijuana/cannabinoids: neurobiology and neurophysiology. CRC Press, Boca Raton, pp 337–386

Mathew RJ, Wilson WH (1993) Acute changes in cerebral blood flow after smoking marijuana. Life Sci 52:757–767

Mathew RJ, Wilson WH, Humphreys DF, Lowe JV, Wiethe KE (1992) Regional cerebral blood flow after marijuana smoking. J Cereb Blood Flow Metab 12: 750–758

Matsuda LA, Lolait SJ, Brownstein MJ, Young AC, Bonner TI (1990) Structure of a cannabinoid receptor and functional expression of the cloned dDNA. Nature 346:561–564

Maykut MO (1984) Health consequences of acute and chronic marihuana use, health consequences of acute and chronic marihuana use. Pergamon, Oxford

McBay A (1986) Drug concentrations and traffic safety. Adv Alcohol Subst Abuse 2:51–59

McMillan DE, Dewey WL (1972) On the mechanism of tolerance to Δ^9-THC. In: Lewis MF (ed) Current research in marihuana. Academic, New York, pp 97–128

McMillan DE, Harris LS, Frankenheim JM, Kennedy JS (1970) L-Δ^9-trans-tetrahydrocannabinol in pigeons: tolerance to the behavioral effects. Science 169:501–503

McMillan DE, Dewey WL, Harris LS (1971) Characteristics of tetrahydrocannabinol tolerance. Ann NY Acad Sci 191:83–99

Mechoulam R, Lander N, Srebnik M, Zamir, I, Breuer A, Shalita B, Dikstein S, Carlini EA, Leite JR, Edery H, Porath G (1984) Recent advances in the use of cannabinoids as therapeutic agents. In: Agurell S, Dewey W, Willette (eds) The cannabinoids: chemical, pharmacologic and therapeutic aspects. Academic, New York, pp 777–793

Mechoulam R, Feigenbaum JJ, Lander N, Segal M, Jarbe TUC, Hiltunen AJ, Consroe P (1988) Enantiomeric cannabinoids: stereospecificity of psychotropic activity. Experientia 44:762–764

Mechoulam R, Hanus L, Ben-Shabat S, Fride E, Weidenfeld J (1994) The anandamides, a family of endogenous cannabinoid ligands – chemical and biological studies. Neuropsychopharmacology 10:145S–145S

Mellors A (1979) Cannabinoids and membrane-bound enzymes. In: Nahas GG, Paton WDM (eds) Marihuana biological effects-analysis, metabolsim, cellular responses, reproduction and brain. Pergamon, Oxford, pp 329–342

Melvin LS, Johnson MR, Harbert CA, Milne GM, Weissman A (1984) A cannabinoid derived prototypical analgesic. J Med Chem 27:67–71

Mendelson JH, Babor TF, Kuehenle JC, Rossi AM, Berstein JG, Mello NK, Greenberg I (1976) Behavioral biologic aspects of marihuana use. Ann NY Acad Sci 282:186–210

Meyer RE (1978) Behavioral pharmacology of marihuana. In: Lipton MA, DiMascio A, Killam KF (eds) Psychopharmacology: a generation of progress. Raven, New York, pp 1639–1652

Miczek KA, Dihit BN (1980) Behavioral and biochemical effects of chronic Δ^9-tetrahyrocannabinol in rats. Psychopharmacology 67:195–202

Molnar J, Petri I, Berek I, Shoyama Y, Nishioka I (1987) The effects of cannabinoids and cannabispiro compounds on Escherichia coli adhesion to tissue culture cells and on leukocyte functions in vitro. Acta Microbiol Hung 34:233–240

Morgan B, Brake SC, Hutchings DE, Miller N, Gamagaris Z (1988) Δ^9-tetrahydrocannabinol during pregnancy in the rat: effects on development of RNA, DNA, and protein in offspring brain. Pharmacol Biochem Behav 31:365–369

Moskowitz H (1985) Marihuana and driving. Accid Anal Prevent 17:323–345

Mountjoy KG, Robbins LS, Mortrud MT, Cone RD (1992) The cloning of a family of genes that encode the melanocortin receptors. Science 257:1248–1251

Munro S, Thomas KL, Abu-Shaar M (1993) Molecular characterization of a peripheral receptor for cannabinoids. Nature 365:61–64

Munson AE, Fehr KO (1983) Immunological effects of cannabis. In: Fehr KO, Kalant H (eds) Cannabis and health hazards: proceedings of an ARF/WHO scientific meeting on adverse health and behavioral consequences of cannabis use. Addiction Research Foundation, Toronto, pp 257–354

Murphy LL, Newton SC, Dhali J, Chavez D (1991) Evidence for a direct anterior pituitary site of Δ^9-tetrahydrocannabinol action. Pharmacol Biochem Behav 40:603–607

Nahas G (1993a) General toxicity of cannabis. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 5–17

Nahas G (1993b) Historical outlook of the psychopathology of cannabis. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 95–99

Nahas G, Latour C (1992) The human toxicity of marijuana. Med J Aust 156:495–497

Nakamura EM, Da Silva EA, Concilio GV, Wilkinson DA, Masur J (1991) Reversible effects of acute and lot-term administration of Δ^9-tetrahydrocannabinol (THC) on memory in the rat. Drug Alcohol Depend 28:167–175

Nakano Y, Pross SH, Friedman H (1992) Modulation of interleukin 2 activity by Δ^9-tetrahydrocannabinol after stimulation with concanavalin A, phytohemagglutinin, or anti-CD3 antibody. Proc Soc Exp Biol Med 201:165–168

Nakano Y, Pross S, Friedman H (1993) Contrasting effect of Δ^9-tetrahydrocannabinol on IL-2 activity in spleen and lymph node cells of mice of different ages. Life Sci 52:41–51

Naor Z (1990) Cyclic GMP stimulates inostitol phosphate production in cultured pituitary cells: Possible implications to signal transduction. Biochem Biophys Res Commun 167:982–992

Narimatsu S, Watanabe K, Yamamoto I, Yoshimura H (1988) Mechanism for inhibitory effect of cannabidiol on microsomal testosterone oxidation in male rat liver. Drug Metab Dispos 16:880–889

Navarro M, Fernandez-Ruiz JJ, De Miguel R, Hernandez ML, Cebeira M, Ramos JA (1993) Motor disturbances induced by an acute dose of Δ^9-tetrahydrocannabinol: possible involvement of nigrostriatal dopaminergic alterations. Pharmacol Biochem Behav 45:291–298

Negrete JC (1993) Effects of cannabis on schizophrenia. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 105–112

Newman LM, Lutz MP, Gould MH, Domino EF (1974) Δ^9-tetrahydrocannabinol and ethyl alcohol: evidence for cross-tolerance in the rat. Science 175:1022–1023

Niemi WD (1979) Effect of Δ^9-tetrahydrocannabinol on synaptic transmission in the electric eel electroplaque. Res Commun Chem Pathol Pharmacol 25:537–546

Nishizuka Y (1983) Calcium, phospholipid turnover and transmembrane signalling. Philos Trans R Soc Lond 302:101–112

Nishizuka Y (1984) The role of protein kinase C in cell surface signal transduction and tumour promotion: Nature 308:693–697

Nowicky AV, Teyloer TJ, Vardaris RM (1987) The modulation of long-term potentiation by Δ^9-tetrahydrocannabinol in the rat hippocampus, in vitro. Brain Res Bull 19:663–672

Noyes JR, Brunk SF, Avery DH, Canter A (1975) The analgesic properties of Δ^9-tetrahydrocannabinol and codeine. Clin Pharmacol Ther 18:84–89

Nye JS, Seltzman HH, Pitt CG, Snyder SH (1984) Labelling of a cannabinoid binding site in brain with a [^{3}H]quarternary ammonium analogue of Δ^8-THC. In: Harvey DJ (ed) Marijuana '84: proceedings of the oxford symposium on cannabis. IRL Press, Oxford, pp 253–262

Nye JS, Seltzman HH, Pitt CG, Snyder SS (1985) High-affinity cannabinoid binding sites in brain membranes labeled with [^{3}H]-5$'$-trimethylammonium-Δ^8-tetrahydrocannabinol. J Pharmacol Exp Ther 234:784–791

O'Connell CM, Fried PA (1984) An investigation of prenatal cannabis exposure and minor physical anomalies in a low risk population. Neurobehav Toxicol Teratol 6:345–350

O'Connell CM, Fried PA (1991) Prenatal exposure to cannabis: a preliminary report of postnatal consequences in school-age children. Neurotoxicol Teratol 13: 631–639

Ohlsson A, Lindgren J-E, Wahlen A, Agurell S, Hollister LE, Gillespie HK (1980) Plasma Δ^9-tetrahydrocannabinol concentrations and effect after oral and intravenous administration and smoking. Clin Pharmacol Ther 28:409–416

Ohlsson A, Lindgren J-E, Wahlen A, Agurell, S, Hollister L, Gillespie H (1982) Single dose kinetics of deuterium labelled Δ^1-tetrahydrocannabinol in heavy and light cannabis users. Biomed Mass Spectrom 9:6–11

Ohlsson A, Agurell S, Londgren J, Gillespie H, Hollister L (1985) Pharmacokinetics studies of delta-1-tetra hydrocannabinol in man. In: Barnett G, Chiang C (eds) Pharmacokinetics and pharmacodynamics of psychoactive drugs. Biomedical, Stony Brook, pp 75–92

Okada M, Mine K, Fujiwara M (1989) Relationship of calcium and adenylate cyclase messenger systems in rat brain synaptosomes. Brain Res 501:23–31

Okada M, Urae A, Mine K, Shoyama Y, Iwasaki K, Fujiwara M (1992) The facilitating and suppressing effects of Δ^9-tetrahydrocannabinol on the rise in intrasynaptosomal Ca^{2+} concentration in rats. Neurosci Lett 140: 55–58

Olson KG, Welch SP (1991) The effects of dynorphin A (1–13) and U50 488H on free intracellular calcium in guinea pig cerebellum. Life Sci 48:575–581

Onaivi ES, Green MR, Martin BR (1990) Pharmacological characterization of cannabinoids in the elevated plus maze. J Pharmacol Exp Ther 253:1002–1009

Oviedo A, Glowa J, Herkenham M (1993) Chronic cannabinoid administration alters cannabinoid receptor binding in rat brain: a quantitative autoradiographic study. Brain Res 616:293–302

Pacheco M, Childers SR, Arnold R, Casiano F, Ward SJ (1991) Aminoalkylindoles: actions on specific G-protein-linked receptors. J Pharmacol Exp Ther 257: 170–183

Pacheco M, Ward S, Childers S (1994) Differential requirements of sodium for coupling of cannabinoid receptors to adenylyl cyclase in rat brain membranes. J Neurochem 62:1773–1782

Page JB (1983) The amotivational syndrome hypothesis and the Costa Rica study: relationships between methods and results. J Psychoactive Drugs 15:261–267

Paria BC, Kapur S, Dey SK (1992) Effects of 9-ene-tetrahydrocannabinol on uterine estrogenicity in the mouse. J Steroid Biochem Mol Biol 42:713–719

Paule MG, Allen RR, Bailey JR, Scallet AC, Ali SF, Brown RM, Slikker W (1992) Chronic marijuana smoke exposure in the rhesus monkey II: effects on progressive ratio and conditioned position responding. J Pharmacol Exp Ther 260:210–222

Perez-Reyes M (1985) Pharmacodynamics of certain drugs of abuse. In: Barnett G, Chiang C (eds) Pharmacokinetics and pharmacodynamics of psychoactive drugs. Biomedical Publications, pp 287–310

Perez-Reyes M, Owens S, Di Guiseppi S (1981) The clinical pharmacology and dynamics of marijuana cigarette smoking. J Clin Pharmacol 21:201S–207S

Perez-Reyes M, Di Guiseppi S, Davis KH, Schindler VH, C.E. C (1982) Comparison of effects of marihuana cigarettes of three different potencies. Clin Pharmacol Ther 31:617–624

Perez-Reyes M, Burstein SH, White WR, McDonald SA, Hicks RE (1991) Antagonism of marihuana effects by indomethacin in humans. Life Sci 48:507–515

Pertwee RG (1972) The ring test: a quantitative method for assessing the "cataleptic" effect of cannabis in mice. Br J Pharmacol 46:753–763

Pertwee RG (1988) The central neuropharmacology of psychotropic cannabinoids. Pharmacol Ther 36:189–261

Pertwee (1990) The central neuropharmacology of psychotropic cannabinoids. In: Balfour DJK (ed) Psychotropic drugs of abuse. Pergamon, New York, p 355

Pertwee R (1992) In vivo interactions between psychotropic cannabinoids and other drugs involving central and peripheral neurochemical mediators. In: Murphy L, Bartke A (eds) Marihuana/cannabinoids: neurobiology and neurophysiology. CRC Press, Boca Raton, pp 165–218

Pertwee RG, Wickens AP (1991) Enhancement by chlordiazepoxide of catalepsy induced in rats by intravenous or intrapallidal injections of enantiomeric cannabinoids. Neuropharmacology 30:237–244

Plasse TF, Gorter RW, Krasnow SH, Lane M, Shepard KV, Wadleigh RG (1991) Recent clinical experience with dronabinol. Pharmacol Biochem Behav 40: 695–700

Poklis A, Maginn D, Barr JL (1987) Drug findings in "driving under the influence of drugs" cases: a problem of illicit drug use. Drug Alcohol Depend 20:57–62

Pradhan SN (1984) Pharmacology of some synthetic tetrahydrocannabinols. Neurosci Biobehav Rev 8:369–385

Pross S, Nakano Y, Smith J, Widen R, Rodriguez A, Newton C, Friedman H (1992a) Suppressive effect of tetrahydrocannabinol on specific T cell subpopulations in the thymus. Thymus 19:97–104

Pross SH, Nakano Y, McHugh S, Widen R, Klein TW, Friedman H (1992b) Contrasting effects of THC on adult murine lymph node and spleen cell populations stimulated with mitogen or anti-CD3 antibody. Immunopharmacol Immunotoxicol 14:675–687

Pross SH, Nakano Y, Widen R, McHugh S, Newton CA, Klein TW, Friedman H (1992c) Differing effects of Δ^9-tetrahydrocannabinol (THC) on murine spleen cell populations dependent upon stimulators. Int J Immunopharmacol 14: 1019–1027

Purohit V, Ahluwahlia BS, Vigersky RA (1980) Marihuana inhibits dihydrotestosterone binding to the androgen receptor. Endocrinology 107:848–850

Raffel G, Clarenbach P, Peskar BA, Hertting G (1976) Synthesis and release of prostaglandins by rat brain synaptosomal fractions. J Neurochem 26:493–498

Razdan RK (1986) Structure-activity relationships in cannabinoids. Pharmacol Rev 38:75–149

Reggio PH, Seltzman HH, Compton DR, Prescott WR, Martin BR (1990) An investigation of the role of the phenolic hydroxyl in cannabinoid activity. Mol Pharmacol 38:854–862

Reggio PH, McGaughey GB, Odear DF, Seltzman HH, Compton DR, Martin BR (1991) A rational search for the separation of psychoactivity and analgesia in cannabinoids. Pharmacol Biochem Behav 40:479–486

Reggio PH, Panu AM, Miles S (1993) Characterization of a region of steric interference at the cannabinoid receptor using the active analog approach. J Med Chem 36:1761–1771

Reichman M, Nen W, Hokin LE (1987) Effects of Δ^9-tetrahydrocannabinol on prostaglandin formation in brain. Mol Pharmacol 32:686–690

Reuter H (1983) Calcium channel modulation by neurotransmitters, enzymes and drugs. Nature 301:569–574

Rinaldi-Carmona M, Barth F, Héaulme M, Shire D, Calandra B, Congy C, Martinez S, Maruani J, Néliat G, Caput D, Ferrara P, Soubrié P, Brelière JC, Le Fur G (1994) SR141716A, a potent and selective antagonist of the brain cannabinoid receptor. FEBS Lett 350:240–244

Rodriguez de Fonseca F, Cebeira M, Fernandez RJJ, Navarro M, Ramos JA (1991a) Effects of pre- and perinatal exposure to hashish extracts on the ontogeny of brain dopaminergic neurons. Neuroscience 43:713–723

Rodriguez de Fonseca F, Fernandez RJJ, Murphy L, Eldridge JC, Steger RW, Bartke A (1991b) Effects of Δ^9-tetrahydrocannabinol exposure on adrenal medullary function: evidence of an acute effect and development of tolerance in chronic treatments. Pharmacol Biochem Behav 40:593–598

Rodriguez de Fonseca F, Fernández-Ruiz JJ, Murphy LL, Cebeira M, Steger RW, Bartke A, Ramos JA (1992a) Acute effects of Δ^9-tetrahydrocannabinol on dopaminergic activity in several rat brain areas. Pharmacol Biochem Behav 42:269–275

Rodriguez de Fonseca F, Hernandez ML, de MR, Fernandez RJJ, Ramos JA (1992b) Early changes in the development of dopaminergic neurotransmission after maternal exposure to cannabinoids. Pharmacol Biochem Behav 41:469–474

Rodriguez de Fonseca F, Ramos JA, Bonnin A, Fernandez RJJ (1993) Presence of cannabinoid binding sites in the brain from early postnatal ages. Neuroreport 4:135–138

Rodriguez de Fonseca F, Gorriti M, Fernandez RJJ, Palomo T, Ramos JA (1994) Downregulation of rat brain cannabinoid binding sites after chronic Δ^9-tetrahydrocannabinol treatment. Pharmacol Biochem Behav 47:33–40

Rosell S, Agurell S, Martin B (1976) Effects of cannabinoids on isolated smooth muscle preparations. In: Nahas GG (ed) Marihuana: chemistry, biochemistry, and cellular effects. Springer, Berlin Heidelberg New York, pp 397–406

Rosell S, Bjorkroth U, Agurell S, Leander K, Ohlsson A, Martin B, Mechoulam R (1979) Relation between effects of cannabinoid derivatives on the twitch response of the isolated guinea-pig ileum and their psychotropic properties. In: Nahas GG, Paton WDM (eds) Marihuana biological effects-analysis, metabolism, cellular responses, reproduction and brain. Pergamon, Oxford, pp 63–70

Rosenkrantz H (1983) Cannabis, marihuana, and cannabinoid toxicological manifestations in man and animals. In: Fehr KO, Kalant H (eds) Cannabis and health hazards: proccedings of an ARF/WHO scientific meeting on adverse health and behavioral consequences of cannabis use. Addiction Research Foundation, Toronto, pp 91–176

Rosenkrantz H (1985) Cannabis components and responses of neuroendocrine-reproductive targets: An overview. In: Paton W, Nahas GG (eds) Marihuana

'84: proceedings of the Oxford symposium on cannabis. IRL Press, Oxford, pp 457–505
Rosenkrantz H, Grant RJ, Fleischman RW, Barker JR (1986) Marihuana-induced embryotoxicity in the rabbit. Fund Appl Toxicol 7:236–243
Roth SH, Williams PJ (1979) The non-specific membrane binding properties of Δ^9-tetrahydrocannabinol and the effects of various solubilizers. J Pharm Pharmacol 31:224–230
Rupniak NM, Samson NA, Steventon MJ, Iversen SD (1991) Induction of cognitive impairment by scopolamine and noncholinergic agents in rhesus monkeys. Life Sci 48:893–899
Sanders J, Jackson DM, Starmer GA (1979) Interactions among the cannabinoids in the antagonism of abdominal constriction response in the mouse. Psychopharmacology 61:281–285
Scallet AC (1991) Neurotoxicology of cannabis and THC: a review of chronic exposure studies in animals. Pharmacol Biochem Behav 40:671–676
Scallet AC, Uemura E, Andrews A, Ali SF, McMillan DE, Paule MG, Brown RM, Slikker W (1987) Morphometric studies of the rat hippocampus following chronic Δ^9-tetrahydrocannabinol (THC). Brain Res 436:193–198
Schwartz RH (1993) Chronic marihuana smoking and short-term memory impairment. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 61–71
Schwartz RH, Hoffmann NG, Jones R (1987) Behavioral, psychosocial, and academic correlates of marijuana usage in adolescence. A study of a cohort under treatment. Clin Pediatr (Phila) 26:264–270
Semus SF, Martin BR (1990) A computergraphic investigation into the pharmacological role of the THC-cannabinoid phenolic moiety. Life Sci 46:1781–1785
Shook JE, Burks TF (1989) Psychoactive cannabinoids reduce gastrointestinal propulsion and motility in rodents. J Pharmacol Exp Ther 249:444–449
Slikker W Jr, Paule MG, Ali SF, Scallet AC, Bailey JR (1992) Behavioral, neurochemical, and neurohistological effects of chronic marijuana smoke exposure in the nonhuman primate. In: Murphy L, Bartke A (eds) Marijuana/cannabinoids: neurobiology and neurophysiology. CRC Press, Boca Raton, pp 219–273
Smiley KA, Karler R, Turkanis SA (1976) Effects of cannabinoids on the perfused rat heart. Res Commun Chem Pathol Pharmacol 14:659–675
Smith PB, Martin BR (1992) Spinal mechanisms of Δ^9-tetrahydrocannabinol-induced analgesia. Brain Res 578:8–12
Smith PB, Welch SP, Martin BR (1993) nor-Binaltorphimine specifically inhibits Δ^9-tetrahydrocannabinol-induced antinociception in mice without altering other pharmacological effects. J Pharmacol Exp Ther
Smith PB, Compton DR, Welch SP, Razdan RK, Mechoulam R, Martin BR (1994) The pharmacological activity of anandamide, a putative endogenous cannabinoid, in mice. J Pharmacol Exp Ther 270:219–227
Soderstrom CA, Trifillis AL, Shankar BS, Clark WE, Cowley A (1993) Marijuana and alcohol use among 1023 trauma patients. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 79–92
Sofia RD, Barry H (1974) Acute and chronic effects of Δ^9-tetrahydrocannabinol on food intake by rats. Psychopharmacology 39:213–222
Sofia RD, Nalepa SD, Harakal JJ, Vassar HB (1973) Anti-edema and analgesic properties of Δ^9-tetrahydrocannabinol (THC). J Pharmacol Exp Ther 186: 646–655
Solomons K, Neppe VM (1989) Cannabis – its clinical effects. S Afr Med J 76:102–104
Soueif MI (1976) Cannabis-type dependence: the psychology of chronic heavy consumption. Ann NY Acad Sci 282:121–125

Specter SC, Klein TW, Newton C, Mondragon M, Widen R, Friedman H (1986) Marijuana effects on immunity: suppression of human natural killer cell activity of Δ^9-tetrahydrocannabinol. Int J Immunopharmacol 8:741–745

Spronck HJW, Luteijn JM, Salemink CA, Nugteren DH (1978) Inhibition of prostaglandin biosynthesis by derivatives of olivetol formed under pyrolysis of cannabidiol. Biochem Pharmacol 27:607–608

Substance Abuse and Mental Health Services Administration (1994) Preliminary estimates from the 1993 National Household Survey on Drug Abuse. US Department of Health and Human Services, Washington DC (Advance report no 7)

Talbott JA, Teague JW (1969) Marihuana Psychosis. JAMA 210:299–302

Tang JL, Lancz G, Specter S, Bullock H (1992) Marijuana and immunity: tetrahydrocannabinol-mediated inhibition of growth and phagocytic activity of the murine macrophage cell line, P388D1. Int J Immunopharmacol 14:253–262

Taschner KL (1983) Psychopathology and differential diagnosis of so-called Cannabis psychoses. Fortschr Neurol Psychiatr 51:235–248

Tennant FS (1983) Clinical toxicology of cannabis use. In: Fehr KO, Kalant H (eds) Cannabis and health hazards: proceedings of an ARF/WHO scientific meeting on adverse health and behavioral consequences of cannabis use. Addiction Research Foundation, Toronto, pp 69–90

Tennes K, Avitable N, Blackard C, Boyles C, Hassoun B, Holmes L, Kreye M (1985) Marijuana: prenatal and postnatal exposure in the human. In: Pinkert TM (ed) Current research on the consequences of maternal drug abuse. US Government Printing Office, Washington DC, pp 48–60

Thomas BF, Compton DR, Martin BF, Semus SF (1991) Modeling the cannabinoid receptor: a three-dimensional quantitative structure-activity analysis. Mol Pharmacol 40:656–665

Thomas BF, Wei X, Martin BR (1992) Characterization and autoradiographic localization of the cannabinoid binding site in rat brain using [^{3}H]11-OH-Δ^9-THC-DMH. J Pharmacol Exp ther 263:1383–1390

Thornicroft G (1990) Cannabis and psychosis: is there epidemiological evidence for an association? Br J Psychiatry 157:25–33

Tramposch A, Sangdee C, Franz DN, Karler R, Turkanis SA (1981) Cannabinoid-induced enhancement and depression of cat monosynaptic reflexes. Neuropharmacology 20:617–621

Tuchmann-Duplessis H (1993) Effects of cannabis on reproduction. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 187–192

Tulunay FC, Ayhan IH, Portoghese PS, Takemori AE (1981) Antagonism by chlornaltrexamine of some effects of Δ^9-tetrahydrocannabinol in rats. Eur J Pharmacol 70:219–224

Tulunay FC, Ayhan IH, Sparber SB (1982) The effects of morphine and Δ^9-tetrahydrocannabinol on motor activity in rats. Psychopharmacology 78:358–360

Tunving K (1987) Psychiatric aspects of cannabis use in adolescents and young adults. Pediatrician 14:83–91

Turkanis SA, Karler R (1986) Effects of Δ^9-tetrahydrocannabinol, 11-hydroxy-Δ^9-tetrahydrocannabinol and cannabidiol on neuromuscular transmission in the frog. Neuropharmacology 25:1273–1278

Turker RK, Kaymakcalan S, Ercan ZS (1975) Antihistaminic action of (−)-trans-Δ^9-tetrahydrocannabinol. Arch Int Pharmacodyn Ther 214:254–262

Tyrey L (1984) Endocrine aspects of cannabinoid action in female subprimates: search for sites of action. In: Braude MC, Ludfor JP (eds) Marijuana effects on the endocrine and reproductive systems. US Government Printing Office, Washington DC, pp 65–81

Tyrey L (1992) Δ^9-tetrahydrocannabinol attenuates luteinizing hormone release induced by electrochemical stimulation of the medial preoptic area. Biol Reprod 47:262–267

Uphouse LA, Welch SP, Ellis EF, Embrey JP (1993) Antinociceptive activity of intrathecal ketoralac is blocked by the kappa-opioid antagonist, nor-binaltorphimine. Eur J Pharmacol 242:53–58

Van der Kloot W (1994) Anandamide, a naturally-occurring agonist of the cannabinoid receptor, blocks adenylate cyclase at the frog neuromuscular junction. Brain Res 649:181–184

Vaysse PJ, Gardner EL, Zukin RS (1987) Modulation of rat brain opioid receptors by cannabinoids. J Pharmacol Exp Ther 241:534–539

Verbene AJM, Taylor DA, Fennessy MR (1980) Withdrawal-like behaviour induced by inhibitors of biogenic amine reuptake in rats treated chronically with Δ^9-tetrahydrocannabinol. Psychopharmacology 68:261–267

Vocci FJ, Petty SK, Dewey WL (1978) Antinociceptive action of butyryl derivatives of guanosine 3′:5′-cyclic monophosphate. J Pharmacol Exp Ther 207:892–898

Vogel Z, Barg J, Levy R, Saya D, Heldman E, Mechoulam R (1993) Anandamide, a brain endogenous compound, interacts specifically with cannabinoid receptors and inhibits adenylate cyclase. J Neurochem 61:352–355

Volkow ND, Fowler JS (1993) Use of positron emission tomography to study drugs of abuse. In: Nahas GG, Latour C (eds) Cannabis: physiopathology, epidemiology, detection. CRC Press, Boca Raton, pp 21–43

Wall ME, Sadler BM, Brine D, Taylor H, Perez-Reyes M (1983) Metabolism, disposition, and kinetics of Δ^9-tetrahydrocannabinol in men and women. Clin Pharmacol Ther 34:352–363

Walton RP, Martin LF, Keller JH (1937) The relative activity of various purified products obtained from American grown hashish. J Pharmacol Exp Ther 62: 239–251

Ward SJ, Baizman E, Bell M, Childers S, D'Ambra T, Eissenstat M, Estep K, Haycock D, Howlett A, Luttinger D, Miller M (1991) Aminoalkylindoles (AAIs): a new route to the cannabinoid receptor? In: Harris LS (ed) Problems of drug dependence 1990: proceedings of the 52nd annual scientific meeting. US Government Printing Office, Washington DC, pp 425–426

Watanabe N, Moroji T, Tada K, Aoki N (1984) A therapeutic trial of caerulein to a long-term heavy marihuana user with amotivational syndrome. Prog Neuropsychopharmacol Biol Psychiatry 8:419–421

Wechsler H, Rohman M, Kotch JB, Idelson RK (1984) Alcohol and other drug use and automobile safety: a survey of Boston-area teen-agers. J Sch Health 54: 201–203

Weidenfeld J, Feldman S, Mechoulam R (1994) The effect of the brain constituent anandamide, a cannabinoid receptor agonist, on the hypotholamo-pituitary-adrenal axis in the rat. Neuroendocrinology 59:110–112

Weiss JL, Watanabe AM, Lemberger L, Tamarkin NR, Cardon PV (1972) Cardiovascular effects of Δ^9-tetrahydrocannabinol in man. Clin Pharmacol Ther 13: 671–684

Weissman A (1978) Generalization of the discriminative stimulus properties of Δ^9-tetrahydrocannabinol to cannabinoids with therapeutic potential. In: Colpaert FC, Rosecrans JA (eds) Stimulus properties of drugs: ten years of progress. Elsevier/North-Holland Biomedical, Amsterdam, pp 99–122

Welch SP (1993) Blockade of cannabinoid-induced antinociception by nor-binaltorphimine, but not N,N-diallyl-tyrosine-aib-phenylalanine-leucine, ICI 174,864 or naloxone in mice. J Pharmacol Exp Ther 256:633–640

Welch SP, Stevens DL (1992) Antinociceptive activity of intrathecally administered cannabinoids alone, and in combination with morphine, in mice. J Pharmacol Exp Ther 262:10–18

Welch SP, Thomas C, Patrick GS (1994) Modulation of cannabinoid-induced antinociception following intracerebroventricular versus intrathecal administration to mice: possible mechanisms for interaction with morphine. J Pharmacol Exp Ther 272:310–331

Wenger T, Croix D, Tramu G, Leonardelli J (1992) Effects of Δ^9-tetrahydro cannabinol on pregnancy, puberty, and the neuroendocrine system. In: Murphy L, Bartke A (eds) Marijuana/cannabinoids: neurobiology and neurophysiology. CRC Press, Boca Raton, pp 539–560

Westlake TM, Howlett AC, Ali SF, Paule MG, Scallet AC, Slikker W Jr (1991) Chronic exposure to Δ^9-tetrahydrocannabinol fails to irreversibly alter brain cannabinoid receptors. Brain Res 544:145–149

White HL, Tansik RL (1980) Effects of Δ^9-tetrahydrocannabinol and cannabidiol on phospholipase and other enzymes regulating arachidonate metabolism. Prostagladins Med 4:409–411

Wickens AP, Pertwee RG (1993) Δ^9-Tetrahydrocannabinol and anandamide enhance the ability of muscimol to induce catalepsy in the globus pallidus of rats. Eur J Pharmacol 250:205–208

Williams EG, Himmelsbach CK, Wikler A, Ruble DC, Lloyd BJ Jr (1946) Studies on marihuana and pyrahexyl compound. Public Health Rep 61:1059–1083

Williams RB, Ng LKY, Lamprecht F, Roth K, Kopin IJ (1973) Δ^9-Tetrahydrocannabinol: A hypotensive effect in rats. Psychpharmacology 28:269–274

Wilson RS, May EL (1974) 9-Nor-Δ^8-tetrahydrocannabinol, a cannabinoid of metabolic interest. J Med Chem 17:475–476

Wilson RS, May EL (1975) Analgesic properties of the tetrahydrocannabinols, their metabolites, and analogs. J Med Chem 18:700–703

Wilson RS, May EL (1979) Some 9-hydroxycannabinoid-like compounds. Synthesis and evaluation of analgesic and behavioral properties. J Med Chem 22:886–888

Yahya MD, Watson RR (1987) Immunomodulation by morphine and marijuana. Life Sci 41:2503–2510

Yaksh TL (1981) The antinociceptive effects of intrathecally-administered levonantradol and desacetyllevonantradol in the rat. J Clin Pharmacol 21:3345–3405

Yaksh TL, Al-Rodhan NRF, Jensen TS (1988) In: Fields HL, Besson JM (eds) Sites of action of opiates in production of analgesia. Elsevier, Amsterdam, pp 371–394 (Progress in brain research, vol 77)

Yebra M, Klein TW, Friedman H (1992) Δ^9-tetrahydrocannabinol suppresses concanavalin A induced increase in cytoplasmic free calcium in mouse thymocytes. Life Sci 51:151–160

Zimmerman S, Zimmerman AM, Laurence H (1981) Effect of Δ^9-tetrahydrocannabinol on cyclic nucleotides in synchronously dividing Tetrahymena. Can J Biochem 59:489–493

Zuardi AW, Karniol IG (1983) Effects on variable-interval performance in rats of delta 9-tetrahydrocannabinol and cannabidiol, separately and in combination. Braz J Med Biol Res 16:141–146