INTRODUCTION

Here is the second installment of the research update. There were 56 references this month which is a reflection of the current pace of cannabinoid research. This means this is 14 pages long! I would like to hear from interested recipients of this summary if all these are relevant; I may have to exercise further editorial control so let me know what you prefer to receive.

BASIC SCIENCE


The striatum functions critically in movement control and habit formation. The development and function of cortical input to the striatum are thought to be regulated by activity-dependent plasticity of corticostriatal glutamatergic synapses. Here we show that the induction of a form of striatal synaptic plasticity, long-term depression (LTD), is dependent on activation of the CB1 cannabinoid receptor. LTD was facilitated by blocking cellular endocannabinoid uptake, and postsynaptic loading of anandamide (AEA) produced presynaptic depression. The endocannabinoid necessary for striatal LTD is thus likely to be released postsynaptically as a retrograde messenger. These findings demonstrate a new role for endocannabinoids in the induction of long-term synaptic plasticity in a circuit necessary for habit formation and motor control.


This article represents the proceedings of a symposium at the 2001 annual meeting of the Research Society on Alcoholism in Montreal, Canada. The chairpersons were Appa Hungund and George Koob. The presentations were (1) Role of endocannabinoids in ethanol tolerance, by Appa Hungund; (2) Modulation of cannabinoid receptor and its signal transduction in chronic alcoholism, by B. S. Basavarajappa; (3) Endocannabinoid involvement in the control of appetitive behavior, by George Kunos; (4) Regulation of voluntary ethanol intake by cannabinoid receptor agonists and antagonists in alcohol-preferring sP rats, by Giancarlo Colombo; (5) Role of endogenous cannabinoid system in alcoholism, by Fernando Rodriguez de Fonseca; and (6) Endocannabinoids and dopamine interactions in vivo, by Loren Parsons and George Koob.


The CB1 cannabinoid receptor has been shown to couple with pertussis toxin (PTX)-sensitive Gi/o proteins and inhibit adenyl cyclase. However, in certain conditions, CB1 mediates adenyl cyclase activation, possibly through Gs-type G proteins. In rat B103 neuroblastoma cells in which CBI gene was endogenously expressed, anandamide inhibited forskolin-induced cAMP accumulation via PTX-sensitive pathways. When CB1 was heterologously over-expressed using a retroviral transfer, high concentrations of anandamide increased forskolin-induced cAMP accumulation, and this effect was more prominent when cells were pretreated with PTX. In CB1-over-expressing B103 cells, anandamide induced cell rounding via a PTX-insensitive/Rho kinase inhibitor-sensitive pathway. These results suggest that the CB1 receptor could couple with G proteins that activate Rho (possibly G12/13) as well as Gi/o and Gs.

The study was undertaken to explore the effect of CP55,940 ((-)-cis-3-[2-Hydroxy4-(1,1-dimethylheptyl) phenyl]-trans-4-(3-hydroxypropyl)cyclohexanol), a drug commonly used as a CB1/CB2 cannabinoid receptor agonist, on intracellular free Ca2+ levels ([Ca2+]i) in several cell types [Ca2+]i was measured in suspended cells by using the fluorescent dye fura-2 as an indicator. At concentrations between 1-50 microM, CP55,940 increased [Ca2+]i in a concentration-dependent manner with an EC50 of 8 microM. The [Ca2+]i signal comprised an initial rise, a slow decay, and a sustained phase. CP55940 (10 microM)-induced [Ca2+]i signal was not altered by 5 microM of two cannabinoid receptor antagonists (AM-251, N-(Piperidin-1-yl)-5-(4-iodophenyl)-1-(2,4-dichlorophenyl)-4-methyl-1H-pyrazole-3-carboxamide; AM-281, 1-(2,4-Dichlorophenyl)-5-(4-iodophenyl)-4-m3thyl-N-4-morpholinyl-1H-pyrazole-3-carboxamide).

Extracellular Ca2+ removal decreased the maximum value of the Ca2+ signals by 50%. CPS5,940 (10 microM)-induced [Ca2+]i increase in Ca2+-free medium was inhibited by 80% by pretreatment with 1 microM thapsigargin, an endoplasmic reticulum Ca2+ pump inhibitor. Conversely, pretreatment with 10 microM CP55,940 in Ca2+-free medium for 6 min abolished thapsigargin-induced [Ca2+]i increase. Nifedipine (10 microM) and verapamil (10 microM) did not alter CP55,940 (10 microM)-induced [Ca2+]i increase. CP55, 940 (10 microM)-induced Ca2+ release was not affected when phospholipase C was inhibited by 2 microM U73122 (1-(6-((17beta-3-methoxyestra-1,3,5(10)-trien-17-yl)amino)hexyl)-1H-pyrrol e-2,5-dione). CP55,940 (5 microM) also increased [Ca22+]i in Madin-Darby canine kidney cells, MG63 human osteosarcoma cells, and IMR-32 neuroblastoma cells. Collectively, CP,55940 induced significant [Ca2+]i increases in several cell types by releasing store Ca2+ from thapsigargin-sensitive pools and by causing Ca2+ entry. The CP55,940's action appears to be dissociated from stimulation of cannabinoid receptors.


Anandamide (N-arachidonoylethanolamine, AEA) and 2-arachidonoylglycerol (2-AG) are the most active endocannabinoids at brain (CB1) cannabinoid receptors. CD1 mice lacking the CB1 receptors ('knockout' [KO] mutants) were compared with wildtype (WT) littermates for their ability to degrade AEA through an AEA membrane transporter (AMT) and an AEA hydrolase (fatty acid amide hydrolase, FAAH). The age dependence of AMT and FAAH activity were investigated in 1- or 4-month-old WT and KO animals, and found to increase with age in KO, but not WT, mice and to be higher in the hippocampus than in the cortex of all animals. AEA and 2-AG were detected in nmol/mg protein (&mgr;m) concentrations in both regions, though the hippocampus showed approximately twice the amount found in the cortex. In the same regions, 2-AG failed to change across groups, while AEA was significantly decreased (approximately 30%) in hippocampus, but not in cortex, of old KO mice, when compared with young KO or age-matched WT animals. In the open-field test under bright light and in the lit-dark exploration model of anxiety, young KO mice, compared with old KO, exhibited a mild anxiety-related behaviour. In contrast, neither the increase in memory performance assessed by the object recognition test, nor the reduction of morphine withdrawal symptoms, showed age dependence in CB1 KO mice. These results suggest that invalidation of the CB1 receptor gene is associated with age-dependent adaptive changes of endocannabinoid metabolism which appear to correlate with the waning of the anxiety-like behaviour exhibited by young CB1 KO mice.


Scientific progress in the biological sciences increasingly relies on an integration of behavioral, pharmacological, cellular, and molecular approaches, particularly in translating basic
research observations into therapeutic potential. The strength of in vivo model systems lies in the direct assessment of physiological function. However, they only allow indirect evidence for mechanism of action. Frequently, in vitro models provide just the opposite. A combination of both in vitro and in vivo approaches are often essential for establishing the underlying mechanisms of a specific pharmacological effect. In recent times, an endogenous cannabinoid system has been characterized due to the combined efforts of chemists, pharmacologists, molecular and cellular biologists, and biochemists. This endogenous cannabinoid system is providing a basis for systematically addressing the pharmacological controversies surrounding marijuana. The description of this endogenous cannabinoid system and the strategies for establishing the physiological function of this system are the subjects of this article.


Traumatic brain injury (TBI) releases harmful mediators that lead to secondary damage. On the other hand, neuroprotective mediators are also released, and the balance between these classes of mediators determines the final outcome after injury. Recently, it was shown that the endogenous brain cannabinoids anandamide and 2-Arachidonoyl glycerol (2-AG) are also formed after TBI in rat and mouse respectively, and when administered after TBI, they reduce brain damage. In the case of 2-AG, better results are seen when it is administered together with related fatty acid glycerol esters. Significant reduction of brain edema, better clinical recovery, and reduced infarct volume and hippocampal cell death are noted. This new neuroprotective mechanism may involve inhibition of transmitter release and of inflammatory response. 2-AG is also a potent modulator of vascular tone, and counteracts the endothelin (ET-1)-induced vasoconstriction that aggravates brain damage; it may thus help to restore blood supply to the injured brain.


The endogenous cannabinoid anandamide (arachidonylethanolamide) produces vasorelaxation in different vascular beds. In the present study, we found that anandamide and a metabolically stable analog, methanandamide, produced dose-dependent (10 nM-10 &mgr;M) vasorelaxation of ~80% in a rabbit aortic ring preparation in an endothelium-dependent manner. Non-endothelium-dependent vasorelaxation was observed to be a maximum of 20-22% at >10 &mgr;M methanandamide. The efficacious CB(1) receptor analogs desacetylevonantradol (10 &mgr;M) and WIN55212-2 (10 &mgr;M) failed to produce vasorelaxation; however, the endothelium-dependent vasorelaxation evoked by methanandamide was partially (75%) blocked by the CB(1) receptor antagonist SR141716A. The VR(1) vanilloid receptor antagonist capsaicin or the calcitonin gene-related peptide (CGRP) antagonist CGRP-(8-37) partially attenuated (25%) the vasorelaxation in endothelium-intact preparations and greatly reduced the response in endothelium-denuded preparations. Pretreatment of aortic rings with N(G)-nitro-L-arginine methyl ester completely blocked the methanandamide-, capsaicin-, and CGRP-induced vasorelaxation. Pretreatment of aortic rings with pertussis toxin attenuated the methanandamide-induced vasorelaxation in endothelium-intact aortic rings, indicating the involvement of G(i/o) proteins in the vasorelaxation; however, pertussis toxin treatment failed to block the endothelium-independent response. Thus, in the rabbit aorta, methanandamide-induced vasorelaxation exhibits two components: 1) in endothelium-intact rings, an SR141716A-sensitive, non-CB(1) receptor component that requires pertussis toxin-sensitive G proteins and nitric oxide (NO) production; and 2) in endothelium-denuded rings, a component that is mediated by VR(1) vanilloid receptors and possibly by the subsequent release of CGRP that requires NO production but is independent of pertussis toxin-sensitive G proteins.


Recent studies have clarified that endogenous cannabinoids (endocannabinoids) are
released from depolarized postsynaptic neurons in a Ca(2+)-dependent manner and act retrogradely on presynaptic cannabinoid receptors to suppress inhibitory or excitatory neurotransmitter release. This type of modulation has been found in the hippocampus and cerebellum and was called depolarization-induced suppression of inhibition (DSI) or excitation (DSE). In this study, we quantitatively examined the effects of postsynaptic depolarization and a cannabinoid agonist on excitatory and inhibitory synapses in rat hippocampal slices and cultures. We found that both DSE and DSI can be induced, but DSE was much less prominent than DSI. For the induction of DSE, the necessary duration of depolarization was longer than for DSI. The magnitude of DSE was much smaller than that of DSI. To explore the reasons for these differences, we tested the sensitivity of EPSCs and IPSCs to a cannabinoid agonist, WIN55,212-2, in hippocampal cultures. IPSCs were dichotomized into two distinct populations, one with a high sensitivity to WIN55,212-2 (50% block at 2 nm) and the other with no sensitivity. In contrast, EPSCs were homogeneous and exhibited a low sensitivity to WIN55,212-2 (50% block at 60 nm). We estimated that the 5 sec depolarization elevated the local endocannabinoid concentration to a level equivalent to several nanomoles of WIN55,212-2. Using CB1 knock-out mice, we verified that both DSI and DSE were mediated by the cannabinoid CB1 receptor. These results indicate that presynaptic cannabinoid sensitivity is a major factor that determines the extent of DSI and DSE.


This review presents the remarkable advances that have been achieved in marijuana (cannabinoid) research, with the discovery of specific receptors and the existence of naturally occurring cannabis-like substances in the human body and brain. The last decade has seen more rapid progress in marijuana research than any time in the thousands of years that marijuana has been used by humans, particularly in cannabinoid genomics. The cDNA and genomic sequences encoding G protein-coupled cannabinoid receptors (Cnrs) from several species have now been cloned. Endogenous cannabinoids (endocannabinoids), synthetic and hydrolyzing enzymes and transporters that define neurochemically-specific cannabinoid brain pathways have been identified. Endocannabinoid lipid signaling molecules alter activity at G protein-coupled receptors (GPCR) and possibly at anandamide-gated ion channels, such as vanilloid receptors. Availability of increasingly-specific CB1 and CB2 Cnr antagonists and of CB1 and CB2 Cnr knockout mice have increased our understanding of these cannabinoid systems and provides tantalizing evidence for even more G protein-coupled Cnrs. Initial studies of the Cnr gene structure, regulation and polymorphisms whet our appetite for more information about these interesting genes, their variants and roles in vulnerabilities to addictions and other neuropsychiatric disorders. Behavioral studies of cannabinoids document the complex interactions between rewarding and aversive effects of these drugs. Pursuing cannabinoid-related molecular, pharmacological and behavioral leads will add greatly to our understanding of endogenous brain neuromodulator systems, abused substances and potential therapeutics. This review of CB1 and CB2 Cnr genes in human and animal brain and their neurobiological effects provide a basis for many of these studies. Therefore, understanding the physiological cannabinoid control system in the human body and brain will contribute to elucidating this natural regulatory mechanism in health and disease.


Rats display conditioned rejection reactions during an oral infusion of a flavor previously paired with an emetic drug; considerable evidence indicates that these rejection reactions reflect nausea. Here we report that cannabidiol, a major non-psychoactive cannabinoid found in marijuana and its synthetic dimethylheptyl homolog interfere with nausea elicited by lithium chloride and with conditioned nausea elicited by a flavor paired with lithium chloride. These results suggest that cannabinoids without psychoactive side-effects may have therapeutic value in the treatment of chemotherapy-induced nausea.

The first endocannabinoid, anandamide, was discovered in 1992. Since then, two other endocannabinoid agonists have been identified, 2-arachidonyl glycerol and, more recently, noladin ether. Here, we report the identification and pharmacological characterization of a novel endocannabinoid, virodhamine, with antagonist properties at the CB1 cannabinoid receptor. Virodhamine is arachidonic acid and ethanolamine joined by an ester linkage. Concentrations of virodhamine measured by liquid chromatography atmospheric pressure chemical ionization-tandem mass spectrometry in rat brain and human hippocampus were similar to anandamide. In peripheral tissues that express the CB2 cannabinoid receptor, virodhamine concentrations were 2- to 9-fold higher than anandamide. In contrast to previously described endocannabinoids, virodhamine was a partial agonist with in vivo antagonist activity at the CB1 receptor. However, at the CB2 receptor, virodhamine acted as a full agonist. Transport of [(14)C]anandamide by RBL-2H3 cells was inhibited by virodhamine. Virodhamine produced hypothermia in the mouse and acted as an antagonist in the presence of anandamide both in vivo and in vitro. As a potential endogenous antagonist at the CB1 receptor, virodhamine adds a new form of regulation to the endocannabinoid system.


The present study investigated the effect of the selective cannabinoid agonist, WIN 55212-2 [4,5-dihydro-2-methyl-4(4-morpholinylmethyl)-1-(1-naphthalenyl-carbonyl)-6H-pyrrolo[3,2,1-ij]quinolin-6-one], on body temperature. WIN 55212-2 (1, 2.5, 5, and 10 mg/kg, i.m.) induced hypothermia in a dose-dependent manner. The peak hypothermia occurred 60 to 180 min postinjection. Body temperature was still suppressed 5 h after the injection of the highest dose of WIN 55212-2. The selective CB(1) antagonist, SR141716A [N-(piperidin-1-yl)-5-(4-chlorophenyl)-1-(2,4-dichlorophenyl)-4-methyl-1H-pyrazole-3-carboxamide hydrochloride] (5 and 10 mg/kg, i.m.), blocked the WIN 55212-2-induced hypothermia, suggesting that CB(1) receptor activation mediated the hypothermia. In contrast, the selective CB(2) antagonist, SR144528 [N-((1S)-endo-1,3,3-trimethyl bicyclo heptan-2-yl)-5-(4-chloro-3-methylphenyl)-1-(4-methylbenzyl)-pyrazole-3-carboxamide] (5 mg/kg, i.m.), did not alter the WIN 55212-2-induced hypothermia. Neither SR141716A nor SR144528 alone altered body temperature. WIN 55212-2 (1-30 &mgr;g/&mgr;l) injected directly into the preoptic anterior hypothalamic nucleus (POAH) induced hypothermia in an immediate and dose-dependent fashion. The hypothermia produced by intra-POAH injection of WIN 55212-2 was brief, with body temperature returning to baseline 60 min postinjection. SR141716A (5 mg/kg, i.m.) abolished the hypothermia induced by intra-POAH injection of WIN 55212-2 (30 &mgr;g/&mgr;l), indicating that CB(1) receptors in the POAH mediated the hypothermia. The present results confirm the idea that CB(1) receptors mediate the hypothemic response to cannabinoid agonists. Moreover, the present data suggest that 1) the POAH is the central locus for thermoregulation, and 2) CB(1) receptors within the POAH are the primary mediators of cannabinoid-induced hypothermia.


Recent discoveries have opened new fields for research on the biochemistry and pharmacology of cannabinoids. Among them, and most importantly, are the characterization and molecular cloning of central and peripheral cannabinoid receptors as well as the isolation of the first putative endogenous ligands that bind to them, anandamide and 2-arachidonylglycerol. The enzyme that degrades these so-called 'endocannabinoids' is an integral membrane protein, fatty acid amide hydrolase. Its distribution and biochemistry in rat brain suggest that it plays a critical role in the regulation of the endocannabinoid system. However, few data exist regarding its distribution and mechanism of action in human tissues. To that end, we have studied its cellular
distribution in the human central nervous system by immunohistochemistry. Using an affinity-purified antibody, we report that fatty acid amide hydrolase is localized to specific and well-delimited cell populations, including cortical pyramidal neurons, subcortical white matter astrocytes, striatal and striatoefferent projecting neurons, hypothalamic and midbrain nuclei, granular and molecular layers of the cerebellum, Purkinje neurons, dentate cerebellar nucleus, inferior olivary nuclei and others. This distribution resembles that of the central cannabinoid receptors as well as that of the enzyme distribution in the rat brain. In summary, the cellular localization of the degradative enzyme of the endogenous cannabinoid ligands in human central nervous system reveals its presence on both neuronal and glial elements and shows a significant overlapping with that of central cannabinoid receptors, mainly in areas related with motor control, confirming the notion that the endocannabinoid system plays a critical role in the control of movement.


The present study demonstrates a novel stimulatory effect of a cannabinoid agonist on calcium channels. DALN (1 nM) potentiated 45Ca(2+)-uptake by N18TG2 neuroblastoma cells, an effect that was abolished by the specific CB1 receptor antagonist SR141716A. The stimulation of 45Ca(2+)-uptake by DALN was resistant to pertussis toxin (PTX), suggesting that Gi/Go GTP-binding proteins did not mediate this effect. Furthermore, PTX unmasked a stimulatory effect of a high concentration of DALN (1 &mgr;M), which by itself failed to stimulate calcium uptake in naive cells. The stimulatory effect of DALN on calcium entry to the cells was blocked by nicardipine but not by omega-conotoxin GVIA, indicating the entry of calcium through L-type voltage-dependent calcium channels. Blocking cAMP-dependent protein kinase (PKA) by H-89 completely eliminated the elevation in calcium uptake, while blocking protein kinase C (PKC) by chelerythrine and calphostine-C only partially attenuated the stimulation. Blocking calmodulin by W-7 revealed a similar partial inhibition of the stimulatory effect of DALN. Hence, we suggest a cannabinoid-specific, PTX-insensitive, stimulatory effect on L-type voltage-dependent calcium channels, which is mediated by PKA and modulated by PKC and calmodulin.


Cannabinoids can disrupt short-term memory in humans and animals and induce learning deficits and other cognitive impairments. In the present study we examined the role of a full cannabinoid agonist in short-term memory, sensorimotor gating, and the acquisition and expression of an operant learning paradigm in rats. We tested the effects of the synthetic cannabinoid WIN 55,212-2 (0.6 and 1.2 mg/kg) on short-term memory in social and object recognition tests, on prepulse inhibition (PPI) of startle, as well as on lever pressing for palatable food. Injections of 0.6 and 1.2 mg/kg WIN 55,212-2 impaired recognition memory and PPI in a dose-dependent manner, but had no effect on lever-pressing acquisition or expression, or on food preference. The PPI deficit was reversed by the administration of 0.1 mg/kg haloperidol. These data suggest that the synthetic cannabinoid WIN 55,212-2 does not lead to a general impairment of learning in an appetitive instrumental task, but significantly affects short-term memory and sensorimotor integration. The impairment in recognition and PPI might be due to deficits in attention-based short-term information processing.


It has been suggested that cannabinoid agonists increase dopamine (DA) transmission in the mesolimbic dopamine system. However, evidence for such an effect is inconsistent. Prepulse inhibition (PPI) of the acoustic startle reflex is a behavioural paradigm that is modulated by an increase of mesolimbic dopamine. This study sought to ascertain whether or not a cannabinoid agonist, CP 55,940, mimicked the effects of amphetamine (a drug which increases dopamine release) on PPI. The first experiment measured the PPI of 16 male Wistar rats injected (i.p.) with
different doses of CP 55,940 in a Latin-square design. A second experiment replicated the effects of the first experiment in a between-subjects design, and also examined the effects of using a 5% alcohol solution as a solvent for cannabinoid agonists, in comparison to the more inert detergent, Tween 80. In both experiments, CP 55,940 in Tween 80 significantly reduced basal activity, increased startle onset latencies and increased PPI, effects opposite to those of amphetamine. These results suggest that the net behavioural effects of cannabinoids are opposite to those of amphetamine. In addition, it was found that 1 ml/kg of a 5% alcohol solution has significant behavioural effects on its own, and reverses the effects of CP 55,940 on PPI.


This paper presents a GC-MS confirmation method, based on large-volume programmed-temperature vapourisation (PTV) injection, for the determination of cannabinoids in plasma samples (or whole blood) with deuterium-labelled internal standards using only 25 &mgr;l of biological fluid. The analytes, Delta(9)-tetrahydrocannabinol (THC), 11-hydroxy-Delta(9)-tetrahydrocannabinol (11-OH-THC) and 11-nor-9-carboxy-Delta9-tetrahydrocannabinol (THC-COOH), were enriched by means of solid-phase extraction cartridges containing octadecyl-bonded silica and were, subsequently, methylated. A 20 &mgr;l aliquot of an extract in hexane was injected into a PTV in solvent split mode. Method development and the results of the analyses of standard reference material and real samples are presented and discussed. This micro-method is precise and sensitive enough to assess relevant cannabinoid levels in human blood for forensic investigations as well as for clinical applications.


The effects of the principal psychoactive component of marijuana, Delta(9)-tetrahydrocannabinol (Delta(9)-THC), on endogenous extracellular glutamate levels in primary cultures of rat cerebral cortex neurons were investigated. Locally applied Delta(9)-THC (0.03, 3, 300, and 1,000 nM) concentration-dependently increased basal extracellular glutamate levels (+18% +/- 11%, +54% +/- 10%, +90% +/- 14%, +149% +/- 33% vs. basal). The facilitatory effects of Delta(9)-THC (3 and 300 nM) on cortical glutamate were fully counteracted in the presence of the selective CB(1) receptor antagonist SR141716A (10 nM) and by replacement of the normal Krebs-Ringer bicarbonate buffer with a low-Ca(2+) (0.2 mM) medium. Delta(9)-THC application also induced an enhancement in K(+)-evoked glutamate levels. These findings suggest that an increase in cortical glutamatergic transmission mediated by local CB(1) receptor activation may underlie some of the psychoactive and behavioral effects of acute marijuana consumption. Copyright 2002 Wiley-Liss, Inc.


The endocannabinoid system has been proposed to modulate a variety of physiological processes, including those that underlie cognition. The present study tested whether this system is tonically active in learning and memory by comparing CB(1) receptor knockout mice (CB(1)(-/-)) to wild-type mice (CB(1)(+/+)) in several Morris water maze tasks. Also, the effects of three cannabinoid agonists, Delta(9)-tetrahydrocannabinol (Delta(9)-THC), R-(+)-[2,3-dihydro-5-methyl-3[morpholinyl]methyl]pyrrolo[1,2,3-de]-1, 4-benzoxazinyl-(1-naphthalenyl)methanone mesylate (WIN 55,212-2), and methanandamide, were evaluated in a working memory procedure. Both genotypes exhibited identical acquisition rates in a fixed platform procedure; however, the CB(1)(+/-) mice demonstrated significant deficits in a reversal task in which the location of the hidden platform was moved to the opposite side of the tank. This phenotype difference was most likely due to an increased perseverance of the CB(1)(-/-) mice in that they continued to return to the
original platform location, despite being repeatedly shown the new platform location. In addition, Delta(9)-THC (ED(50) = 1.3 mg/kg), WIN 55,212-2 (ED(50) = 0.35 mg/kg), and methanandamide (ED(50) = 3.2 mg/kg) disrupted the performance of CB(1)(+/+) mice in the working memory task at doses that did not elicit motivational or sensorimotor impairment as assessed in a cued version of the task. Furthermore, doses of each drug that were maximally disruptive in CB(1)(+/+) mice were ineffective in either N-(piperidin-1-yl)-5-(4-chlorophenyl)-1-(2,4-dichlorophenyl)-4-methyl-1H-pyrazole-3-carboxamide HCl (SR 141716A)-treated CB(1)(+/+) or CB(1)(/-/-) mice. These results provide strong evidence that cannabinoids disrupt working memory through a CB(1) receptor mechanism of action, and suggest that the endocannabinoid system may have a role in facilitating extinction and/or forgetting processes.


The primary psychoactive ingredient in cannabis, Delta(9)-tetrahydrocannabinol (Delta(9)-THC), affects the brain mainly by activating a specific receptor (CB1). CB1 is expressed at high levels in many brain regions, and several endogenous brain lipids have been identified as CB1 ligands. In contrast to classical neurotransmitters, endogenous cannabinoids can function as retrograde synaptic messengers: They are released from postsynaptic neurons and travel backward across synapses, activating CB1 on presynaptic axons and suppressing neurotransmitter release. Cannabinoids may affect memory, cognition, and pain perception by means of this cellular mechanism.


Plasma levels of clozapine and olanzapine are lower in smokers than in nonsmokers, which is mainly due to induction of cytochrome P4501A2 (CYP1A2) by some smoke constituents. Smoking cessation in patients treated with antipsychotic drugs that are CYP1A2 substrates may result in increased plasma levels of the drug and, consequently, in adverse drug effects. Two cases of patients who smoked tobacco and cannabis are reported. The first patient, who was receiving clozapine treatment, developed confusion after tobacco and cannabis smoking cessation, which was related to increased clozapine plasma levels. The second patient, who was receiving olanzapine treatment, showed important extrapyramidal motor symptoms after reducing his tobacco consumption. The clinical implication of these observations is that smoking patients treated with CYP1A2 substrate antipsychotics should regularly be monitored with regard to their smoking consumption in order to adjust doses in cases of a reduction or increase in smoking.

CLINICAL SCIENCE


The use of cannabis in our society is a common problem and the subject of much medical and political debate. We present a case in which a 17-year-old male regular cannabis user developed a large swollen uvula (uvulitis) and partial upper airway obstruction after smoking cannabis. Symptoms resolved with the administration of corticosteroids and antihistamines.


Background: The purpose of this study was to investigate neuroendocrine function in ecstasy (3,4-methylenedioxyamphetamine = MDMA) users and controls.Methods: Prolactin response to d-fenfluramine was assessed in abstinent ecstasy users with concomitant use of cannabis only (n = 24, male/female 13/11) and in two control groups: healthy nonusers (n = 13, female) and exclusive cannabis users (n = 7, male).Results: Prolactin response to d-fenfluramine was slightly blunted in female ecstasy users. Both male user samples exhibited a weak prolactin response to d-fenfluramine, but this was weaker in the group of cannabis users. Baseline prolactin and prolactin response to d-fenfluramine were associated with the extent of previous cannabis use.Conclusions: Endocrinological abnormalities of ecstasy users may be closely
related to their coincident cannabis use. Cannabis use may be an important confound in endocrinological studies of ecstasy users and should be looked for more systematically in future studies.

The following letters appeared in JAMA following an earlier study published in that journal by Solowij et al:


OBJECTIVE: Evidence that mothers report higher levels of drinking retrospectively than during pregnancy has led some investigators to suggest that women systematically underreport alcohol antenatally and that alcohol-related deficits may actually reflect heavier prenatal exposure. This study is the first to compare the validity of antenatal and retrospective reports of pregnancy drinking, drug use, and smoking in relation to effects on infant neurobehavioral outcomes. METHODOLOGY: Three hundred fifty-four inner-city mothers were interviewed regarding their alcohol, drug use, and smoking during pregnancy and retrospectively at 13 months' postpartum. Their infants were assessed at 6.5, 12, and 13 months on a large battery of neurobehavioral assessments. RESULTS: Although higher levels of alcohol were reported retrospectively, the correlations of prenatal alcohol exposure with infant outcome were as strong or stronger for the antenatal measures and only the antenatal reports predicted poorer cognitive performance on the Bayley Scales and symbolic play, slower processing speed on the Fagan Test of Infant Intelligence and cross-modal transfer, and slower infant reaction time. Women also reported higher levels of cocaine and marijuana but not cigarette smoking retrospectively. Relations between cocaine use and smoking on birth size and gestational age were as strong for either report. No effects were detected in relation to either report of marijuana use during pregnancy. CONCLUSIONS: These findings suggest that antenatal alcohol interviews provide the most valid information and demonstrate the importance of assessing prenatal alcohol use during pregnancy to minimize the risk of failing to detect neurobehavioral deficits. Adverse effects were consistently seen at levels as low as 0.5 oz absolute alcohol/day (the equivalent of 7 drinks per week) based on maternal antenatal report. These data suggest that alcohol-related deficits do not reflect heavier prenatal exposure than that reported during pregnancy and that threshold values derived from antenatal reports are reasonably accurate.


The authors conducted a randomized, double-blind, placebo-controlled, twofold crossover study in 16 patients with MS who presented with severe spasticity to investigate safety, tolerability, and efficacy of oral Delta(9)-Tetrahydrocannabinol (THC) and Cannabis sativa plant extract. Both drugs were safe, but adverse events were more common with plant-extract treatment. Compared with placebo, neither THC nor plant-extract treatment reduced spasticity. Both THC and plant-extract treatment worsened the participant's global impression.

The use of cannabis sativa preparations as recreational drugs can be traced back to the earliest civilizations. However, animal models of cannabinoid addiction allowing the exploration of neural correlates of cannabinoid abuse have been developed only recently. We review these models and the role of the CB1 cannabinoid receptor, the main target of natural cannabinoids, and its interaction with opioid and dopamine transmission in reward circuits. Extensive reviews on the molecular basis of cannabinoid action are available elsewhere (Piomelli et al., 2000; Schlicker and Kathmann, 2001).


The effects of smoking marijuana on regional cerebral blood flow (rCBF) and cognitive performance were assessed in 12 recreational users in a double-blinded, placebo-controlled study. PET with [15Oxygen]-labeled water ([15O]H(2)O) was used to measure rCBF before and after smoking of marijuana and placebo cigarettes, as subjects repeatedly performed an auditory attention task. Smoking marijuana resulted in intoxication, as assessed by a behavioral rating scale, but did not significantly alter mean behavioral performance on the attention task. Heart rate and blood pressure increased dramatically following smoking of marijuana but not placebo cigarettes. However, mean global CBF did not change significantly. Increased rCBF was observed in orbital and mesial frontal lobes, insula, temporal poles, anterior cingulate, as well as in the cerebellum. The increases in rCBF in anterior brain regions were predominantly in "paralimbic" regions and may be related to marijuana's mood-related effects. Reduced rCBF was observed in temporal lobe auditory regions, in visual cortex, and in brain regions that may be part of an attentional network (parietal lobe, frontal lobe and thalamus). These rCBF decreases may be the neural basis of perceptual and cognitive alterations that occur with acute marijuana intoxication. There was no significant rCBF change in the nucleus accumbens or other reward-related brain regions, nor in basal ganglia or hippocampus, which have a high density of cannabinoid receptors.


This article provides an overview of the issues surrounding the use of cannabis for therapeutic purposes. Examples of some of the ethical issues related to professional practice are discussed. The authors do not advocate legalising cannabis for all, but the therapeutic advantages and disadvantages of using cannabis are highlighted.

**BEHAVIOURAL SCIENCE**


In view of implications of Kohlberg's theory of moral development, two hypotheses were considered in two independent studies: a) individuals who consider the use of potentially harmful substances to be morally wrong will be less likely to use such substances than peers who view such activities as a personal choice; and b) compared to those who are less mature, more mature moral reasoners display more consistency between their expressed beliefs about the morality of drug use and their reports of actual drug use. Two samples of college students, 29 men and 59 women in Study 1 and 46 men and 100 women in Study 2, served as participants. All completed questionnaires about their use of tobacco, alcohol, and illicit drugs and their beliefs about the
morality of using these substances. Participants in Study 2 also responded to the Defining Issues Test (DIT) to assess their level of moral thinking. Results from Study 1 supported hypothesis (a). Findings from Study 2 supported hypotheses (a) and (b).


We analyzed nationally representative data from the 1998 National Alternative High School Youth Risk Behavior Survey, conducted by the Centers for Disease Control and Prevention, to determine the prevalence of substance use on school property among alternative high school students in the United States, to describe the characteristics of students who use substances on school property, and to examine the interrelationships of substance-use behaviors. During the 30 days preceding the survey, nearly 48 percent of students used at least one substance on school property and 17 percent used more than one substance on school property. Males were more likely than females and white students were more likely than black or Hispanic students to have used substances on school property. The results of this and other studies suggest that school administrators, public health practitioners, and policy makers should work to improve strategies for reducing substance use in this heterogeneous, hard-to-reach population.


This study examines marijuana use among children of male drug abusers. Subjects were 83 African-American and European-American male drug abusers, of whom the majority were injection drug users, and their children. Thirty-one of the fathers were HIV-positive and 52 were HIV-negative. Using logistic regression analyses, we explored cross-sectionally the relationship between four psychosocial domains (ie, paternal attributes, adolescent problem behaviors, father-adolescent relations, and environment) and adolescent marijuana use. The father's use of illegal drugs and his failure to cope adaptively predicted adolescent marijuana use, while a close father-child bond predicted less adolescent marijuana use. Adolescent problem behaviors predicted an increased likelihood of marijuana use. Furthermore, hierarchical regression analysis demonstrated that the adolescent's problem behavior mediated the associations between both the father-adolescent relationship and environmental factors with adolescent marijuana use. Reducing the risk factors and enhancing the protective factors within each of the domains could help reduce marijuana use among the adolescent children of drug-abusing fathers. Moreover, if a father is a drug abuser, it is important to help him establish a close bond with his child in order to help attenuate the influence of his drug use on the child's marijuana use.


Schizophrenia, intoxication with tetrahydrocannabinol (Delta-THC), and cannabis psychosis induce characteristic time and space distortions suggesting a common psychotic dysfunction. Since genetic research into schizophrenia has led into disappointing dead ends, the present study is focusing on this phenotype. It is shown that information theory can account for the dynamical basis of higher sensorimotor information processing and consciousness under physiologic as well as pathologic conditions. If Kolmogorov entropy (inherent in the processing of action and time) breaks down in acute psychosis, it is predicted that Shannon entropy (inherent in the processing of higher dimensional perception) will increase, provoking positive symptoms and altered states of consciousness. In the search for candidate genes and the protection of vulnerable individuals from cannabis abuse, non-linear EEG analysis of Kolmogorov information could thus present us with a novel diagnostic tool to directly assess the breakdown of information processing in schizophrenia. Copyright 2002 Elsevier Science Ltd. All rights reserved.


OBJECTIVE: To determine the relationships among socioeconomic status (SES), depression, and substance use among teenagers. We hypothesized that, among teenagers,
substance use was associated with SES in a graded fashion and that depression is a mechanism through which SES affects substance use behaviors. DESIGN: Linear regression analyses of cross-sectional data from Wave I of the National Longitudinal Study of Adolescent Health (1995). PARTICIPANTS: Fifteen thousand one hundred twelve adolescents whose parents answered questions assessing household income and parental education. MAIN OUTCOME MEASURES: Use of cigarettes, alcohol, marijuana, and cocaine. RESULTS: For all 4 substances, frequency of use varied by SES. In the total population, inverse SES gradients were present for cigarette use (education, mean change= -0.052; 95% confidence interval [CI], -0.081 to -0.023; income, mean change= -0.038; 95% CI, -0.069 to -0.007) and alcohol use (income, mean change= 0.044; 95% CI, 0.016-0.071). The relationship between marijuana use and education was also significant but inverse-U-shaped, not linear. This relationship was only present among nonwhite teenagers. Race/ethnicity also moderated the relationships between SES and cigarette use and SES and cocaine use. For cigarette use, stratification by race/ethnicity revealed an inverse graded relationship among white non-Hispanic teenagers and a direct, graded relationship among nonwhite teenagers (ie, mean change for education among white non-Hispanic teenagers, -0.012; 95% CI, -0.016 to -0.075; mean change for education among nonwhite teenagers, 0.040; 95% CI, 0.014-0.072). For cocaine use, a weak, inverse linear relationship existed only between education and cocaine use among white non-Hispanic teenagers (mean change for education, -0.013; 95% CI, -0.026 to -0.0004). The relationship between the SES indicator and substance use weakened when depressive symptoms were entered into the model for the SES-cigarette use relationship (23% decrease in mean change associated with a 1-unit change in both education and income) and for the association between education and cocaine use among white non-Hispanic teenagers (31% decrease). CONCLUSIONS: Socioeconomic status is associated with substance use among teenagers but the nature of the relationship is not consistent across SES indicators or across race/ethnicity groups. Depressive symptoms are a mechanism through which SES affects cigarette and cocaine use behaviors among teenagers. However, these data indicate that interventions targeted toward decreasing depressive symptoms will not have a strong impact on the effects of SES on teenage substance use.


More than half of US adolescents will experiment with marijuana. Of those who try marijuana more than once, approximately one third will subsequently use marijuana regularly, although most will have stopped by their late 20s. Although genetic predisposition plays the most important role in determining who will develop dependence, environmental factors influence who will initiate marijuana use. One of the challenges for prevention and treatment programs is that the immediate adverse effects of marijuana use are not extreme, and many adolescents have difficulty in making decisions based on future risks. Therefore, the consequences of leaving school early, having unprotected sex, and driving while intoxicated are often insufficient to deter adolescents from using marijuana. Thus, it is not surprising that current prevention and treatment programs have had limited success in decreasing the rates of initiation and regular use of marijuana among adolescents. However, the accumulation of data about marijuana use in adolescents has the potential to enable the development of more effective prevention and treatment programs.


BACKGROUND: Three prior population-based twin studies, none of which was nationally representative, suggested that both genetic and familial-environmental factors contribute to family resemblance for lifetime cannabis use. We seek to replicate these results in a US national probability sample of twin and sibling pairs examining only last year cannabis use. METHODS: Cannabis use in the last year was assessed by self-report questionnaire. Biometrical twin analyses were performed. RESULTS: Twin and sibling resemblance for last-year cannabis use was substantial, and much higher in monozygotic pairs than in dizygotic and sibling pairs, where levels of resemblance were similar. Modeling suggested that sibling resemblance was due to genetic factors—with a heritability of at least 60% and probably family environmental factors. No
evidence was found that cannabis use was influenced by a special twin environment. CONCLUSIONS: Consistent with prior studies, use of cannabis is substantially influenced by genetic factors but family-environment is also possibly of importance.


The goal of this study is to examine in detail the relationship between pubertal timing and substance use onset using a sample of females from The National Longitudinal Study of Adolescent Health. The sample includes 966 females who were in 7th grade at Wave 1 and 8th grade at Wave 2. Participants in the sample are approximately 69% White, 20% African American, 4% Asian or Pacific Islander, 2% American Indian, 4% other, of Hispanic origin, and 1% other, not of Hispanic origin. Twenty percent of the females were identified as early maturers based on self-reports of body changes (increased breast size and body curviness) measured in 7th grade. These participants are hypothesized to be at increased risk for substance use onset. Important differences in substance use onset were found between early maturers and their on-time and late-maturing counterparts. During 7th grade, females in the early-maturing group are three times more likely to be in the most advanced stage of substance use (involving alcohol use, drunkenness, cigarette use, and marijuana use) than are those in the on-time/late group. Prevalence rates indicate that early maturers are more likely to have tried alcohol, tried cigarettes, been drunk, and tried marijuana. Prospective findings show that early developers are significantly more likely to transition out of the "No Substance Use" stage between 7th and 8th grade (47% for early developers vs. 22% for on-time and late developers). In addition, early developers are more likely to advance in substance use in general, regardless of their level of use at Grade 7.


OBJECTIVE: This article provides information on the extent of alcohol use and other drug use among American college students. METHOD: Five different sources of data are examined for estimating recent levels of alcohol (and other drug) use among college students: Harvard School of Public Health College Alcohol Study (CAS), the Core Institute (CORE), Monitoring the Future (MTF), National College Health Risk Behavior Survey (NCHRBS) and National Household Survey on Drug Abuse (NHSDA). RESULTS: Alcohol use rates are very high among college students. Approximately two of five American college students were heavy drinkers, defined as having had five or more drinks in a row in the past 2 weeks. Alcohol use is higher among male than female students. White students are highest in heavy drinking, black students are lowest and Hispanic students are intermediate. Use of alcohol--but not cigarettes, marijuana and cocaine--is higher among college students than among noncollege age-mates. Longitudinal data show that, while in high school, students who go on to attend college have lower rates of heavy drinking than do those who will not attend college. Both groups increase their heavy drinking after high school graduation, but the college students increase distinctly more and actually surpass their nonstudent age-mates. Trend data from 1980 to 1999 show some slight improvement in recent years. CONCLUSIONS: Despite improvements in the past 20 years, colleges need to do more to reduce heavy alcohol use among students.


Alcohol and other drug problems experienced by adolescents who use only alcohol compared to those who use both alcohol and marijuana (A + M) is studied. Using the national longitudinal survey of youth 1994 data, forward multiple regression analyses revealed that impulsivity, A + M use (compared to alcohol-only use), age, sex, religiosity, frequency of substance use were associated with a higher number of behavioral problems. Youth with more alcohol problems were found to be binge drinkers, impulsive, more frequent alcohol users, and
nonHispanic. Implications and future research needs are discussed.


Drawing upon an "exposure opportunity" concept described by Wade Hampton Frost, the authors studied two mechanisms to help account for prior observations about the "stepping-stone" or "gateway" sequences that link the use of alcohol, tobacco, marijuana, and cocaine. Data were obtained from four nationally representative and independent cross-sectional samples of US household residents (n = 44,624 persons aged 12-25 years). Data were gathered using standardized self-report methods and were analyzed via survival methods. Results indicated that users of tobacco and alcohol were more likely than nonusers to have an opportunity to try marijuana and were more likely to actually use marijuana once a marijuana opportunity had occurred. Opportunity to use cocaine was associated with prior marijuana smoking. Among young people with a cocaine opportunity, those who had used marijuana were more likely to use cocaine than were those with no history of marijuana use. The observed associations did not seem to arise solely as a result of young drug users' seeking out opportunities to use drugs. Applying Frost's epidemiologic concept of exposure opportunity, the authors offer new epidemiologic evidence on the sequences that link earlier use of alcohol and tobacco to later illegal drug involvement.


Drug legalization is a frequently-debated drug control policy alternative. It should come as little surprise, therefore, that the arguments in favor of both legalization and prohibition have resulted in a conceptual stalemate. While theoretical deliberations are unquestionably valuable, they seem to have propelled this particular issue to its limit. To date, no works have suggested any empirical studies that might test the framework and potential consequences of drug legalization. In the current study, the arguments surrounding the drug legalization debate are synthesized into a proposal for future research. Such a proposal illustrates that the core elements surrounding drug legalization are not only testable, but that the time may be right to consider such an empirical effort.

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