BASIC SCIENCE


The potential activity of cannabidiol, a non-psychoactive constituent of marijuana, in preventing damage caused by cerebral ischemia was studied. Cannabidiol (1.25-20 mg/kg) was given 5 min after 10 min bilateral carotid occlusion in freely-moving awake gerbils. Seven days after ischemia, it antagonized the electroencephalographic flattening of total spectral power, with a dose-dependent bell-shaped curve; the neuroprotective effect was greatest with 5 mg/kg. One day after ischemia cannabidiol completely antagonized ischemia-induced hyperlocomotion, at all doses. Rectal temperature did not change during the first hour after occlusion. Histological examination showed complete survival of CA1 neurons in cannabidiol-treated gerbils. These findings suggest a potential therapeutic role of cannabidiol in cerebral ischemia, though the clear mechanism of action remains to be elucidated.


Many types of neurons release endocannabinoids from their dendrites in response to elevation of intracellular calcium levels. Endocannabinoids then activate presynaptic cannabinoid receptors, thereby inhibiting neurotransmitter release for tens of seconds. A crucial step in understanding the physiological role of this retrograde signaling is to determine its sensitivity to elevations of postsynaptic calcium. Here we determine and compare the calcium dependence of endocannabinoid-mediated retrograde inhibition at three types of synapses onto cerebellar Purkinje cells. Previous studies have shown that Purkinje cell depolarization results in endocannabinoid-mediated retrograde inhibition of synapses received from climbing fibers, granule cell parallel fibers, and inhibitory interneurons. Using several calcium indicators with a range of affinities, we performed a series of in situ and in vitro calibrations to quantify calcium levels in Purkinje cells. We found that postsynaptic calcium levels of approximately 15 microM are required for half-maximal retrograde inhibition at all of these synapses. In contrast, previous studies had suggested that endocannabinoid release could occur with slight elevations of calcium above resting levels, which implies that inhibition should be widespread and continuously modulated by subtle changes in intracellular calcium levels. However, our results indicate that such small changes in intracellular calcium are not sufficient to evoke endocannabinoid release. Instead, because of its high requirement for calcium, retrograde inhibition mediated by calcium-dependent endocannabinoid release from Purkinje cells will occur under more restricted conditions and with greater spatial localization than previously appreciated.


Glucocorticoid negative feedback in the brain controls stress, feeding, and neural-immune interactions by regulating the hypothalamic-pituitary-adrenal axis, but the mechanisms of inhibition of hypothalamic neurosecretory cells have never been elucidated. Using whole-cell patch-clamp recordings in an acute hypothalamic slice preparation, we demonstrate a rapid
suppression of excitatory glutamatergic synaptic inputs to parvocellular neurosecretory neurons of the hypothalamic paraventricular nucleus (PVN) by the glucocorticoids dexamethasone and corticosterone. The effect was maintained with dexamethasone conjugated to bovine serum albumin and was not seen with direct intracellular glucocorticoid perfusion via the patch pipette, suggesting actions at a membrane receptor. The presynaptic inhibition of glutamate release by glucocorticoids was blocked by postsynaptic inhibition of G-protein activity with intracellular GDP-beta-S application, implicating a postsynaptic G-protein-coupled receptor and the release of a retrograde messenger. The glucocorticoid effect was not blocked by the nitric oxide synthesis antagonist (N)-nitro-L-arginine methyl ester hydrochloride or by hemoglobin but was blocked completely by the CB1 cannabinoid receptor antagonists AM251 [N-(piperidin-1-yl)-5-(4-iodophenyl)-1-(2,4-dichlorophenyl)-4-methyl-1H-pyrazole-3-carboxamide] and AM281 [1-(2,4-dichlorophenyl)-5-(4-iodophenyl)-4-methyl-N-4-morpholinyl-1H-pyrazole-3-carboxamide] and mimicked and occluded by the cannabinoid receptor agonist WIN55,212-2 [(beta)-[2,3-dihydro-5-methyl-3-(4-morpholinoethyl)pyrrolo][1,2,3-de] -1,4-benzoxazin-6-yl]-1-naphthalenymethanone mesylate, indicating that it was mediated by retrograde endocannabinoid release. Several peptidergic subtypes of parvocellular neuron, identified by single-cell reverse transcription-PCR analysis, were subject to rapid inhibitory glucocorticoid regulation, including corticotropin-releasing hormone-, thyrotropin-releasing hormone-, vasopressin-, and oxytocin-expressing neurons. Therefore, our findings reveal a mechanism of rapid glucocorticoid feedback inhibition of hypothalamic hormone secretion via endocannabinoid release in the PVN and provide a link between the actions of glucocorticoids and cannabinoids in the hypothalamus that regulate stress and energy homeostasis.


Objective and Design: In the present study we examined the effects of the cannabinoid receptor agonist HU210 on histamine-evoked somatosensory and vascular responses in humans. SUBJECTS: Two sets of experiments were performed, in which twelve (Study 1, iontophoresis) and six participants (Study 2, microdialysis) were recruited. TREATMENT: HU210 was administered peripherally by skin patch (50 mM) or dermal microdialysis (5 mM), whereas histamine was applied by iontophoresis (50 &mgr;Amps) or dermal microdialysis (5 &mgr;M). METHODS: Skin blood flow was monitored by laser Doppler, widespread flare reaction was evaluated planimetrically, extravasation of plasma proteins was measured in the dialysate and perceived itch was recorded using a visual analogue scale. Data were evaluated by analysis of variance. RESULTS: Experimentally induced itch was significantly reduced by peripheral administration of HU210 (p < 0.05). Additionally, skin blood flow and neurogenic mediated flare responses were attenuated (p < 0.003 and p < 0.03, respectively), whereas protein extravasation due to histamine was enhanced by co-administration of HU210, as investigated by dermal microdialysis. CONCLUSIONS: In humans peripheral administration of a cannabinoid receptor agonist attenuates histamine-induced itch. The observation that protein extravasation was not decreased demonstrates that the alleviation of itch is not due to an anti-histaminergic property of HU210. The reduced neurogenic flare reaction indicates an attenuated antidromic nerve fibre activation and neuropeptide release.


The salivary glands and saliva from the lone star tick Amblyomma americanum (L.) were analyzed for the presence of the two endogenous agonists of cannabinoid receptors, N-arachidonoylethanolamine (anandamide) and 2-arachidonoylglycerol (2-AG), as well as of the anandamide congener, N-palmitoylethanolamine (PEA), an anti-inflammatory and analgesic mediator that is inactive at cannabinoid receptors. Two very sensitive mass-spectrometric techniques were used for this purpose. Both 2-AG and PEA, as well as other N-acyethanolamines (NAEs), were identified in salivary glands, but anandamide was below detection. The levels of 2-AG were considerably higher in the salivary glands of partially fed than replete females. Ex vivo gland stimulation with arachidonic acid increased the levels of 2-AG, but not of PEA or other NAEs, and caused the formation of anandamide and of the potent analgesic...
compound N-arachidonoylglycine. Instead, the amounts of anandamide, 2-AG and PEA were not influenced by treatment of salivary glands with dopamine, which stimulates saliva secretion. The possible biosynthetic precursors of anandamide, PEA and other NAEs were also detected in salivary glands, whereas only PEA was detected in tick saliva. These data demonstrate for the first time that the salivary glands of an obligate ectoparasite species can make endocannabinoids and/or related congeners with analgesic and anti-inflammatory activity, which possibly participate in the inhibition of the host defense reactions.


In the rat, antinociception of supraspinal origin is observed in response to administration of cocaine or an antagonist of the NMDA receptor for glutamate. The current study was conducted to determine if endocannabinoids are involved in the antinociceptive effect of cocaine, or antagonism of NMDA receptor binding. Intraperitoneal (i.p.) administration to male rats of cocaine, or the NMDA receptor antagonist, MK-801, resulted in a significant antinociceptive response of supraspinal origin, as indicated by a significant increase in reaction time in the hot plate test of analgesia (increase in the amount of time before the animal reacted to the hot plate by licking its paws or jumping). Treatment with SR141716A, a specific antagonist of the cannabinoid (CB1) receptor, resulted in a complete reversal of cocaine-induced antinociception when administered at a dose of 5.0mg/kg. Although the 2.5 and 5.0mg/kg doses of SR141716A produced a significant reduction in the antinociceptive effect of MK-801, the effect was incomplete since the reaction time in the hot plate test remained greater than that observed in vehicle-treated controls. These findings suggest that activation of the CB1 receptor participates significantly in antinociception resulting from treatment with cocaine and with the NMDA receptor antagonist, MK-801. The partial reversal of the antinociceptive effect of MK-801 by CB1 receptor antagonism indicates other mediators of nociception, in addition to the endocannabinoids, appear to be active in the antinociceptive response to NMDA receptor antagonism.


Research of cannabinoid actions was boosted in the 1990s by remarkable discoveries including identification of endogenous compounds with cannabimimetic activity (endocannabinoids) and the cloning of their molecular targets, the CB1 and CB2 receptors. Although the existence of an endogenous cannabinoid signaling system has been established for a decade, its physiological roles have just begun to unfold. In addition, the behavioral effects of exogenous cannabinoids such as delta-9-tetrahydrocannabinol, the major active compound of hashish and marijuana, await explanation at the cellular and network levels. Recent physiological, pharmacological, and high-resolution anatomical studies provided evidence that the major physiological effect of cannabinoids is the regulation of neurotransmitter release via activation of presynaptic CB1 receptors located on distinct types of axon terminals throughout the brain. Subsequent discoveries shed light on the functional consequences of this localization by demonstrating the involvement of endocannabinoids in retrograde signaling at GABAergic and glutamatergic synapses. In this review, we aim to synthesize recent progress in our understanding of the physiological roles of endocannabinoids in the brain. First, the synthetic pathways of endocannabinoids are discussed, along with the putative mechanisms of their release, uptake, and degradation. The fine-grain anatomical distribution of the neuronal cannabinoid receptor CB1 is described in most brain areas, emphasizing its general presynaptic localization and role in controlling neurotransmitter release. Finally, the possible functions of endocannabinoids as retrograde synaptic signal molecules are discussed in relation to synaptic plasticity and network activity patterns.


It has been suggested recently that the endocannabinoid system might be a component of the brain reward circuitry and thus play a role not only in cannabinoid tolerance/dependence,
but also in dependence/withdrawal to other drugs of abuse. Here we have examined the changes in endocannabinoid ligands and their receptors in different brain regions, with particular attention to those areas related to reinforcement processes, during dependence on the powerful addictive drug, morphine. Thus, we analysed the brain contents of N-arachidonylethanolamine (anandamide, AEA), the first discovered endocannabinoid, in rats subjected to daily injections of increasing doses of morphine, according to a schedule designed to render the animals opiate-dependent. Although evidence of physical dependence was assured by the appearance of somatic and neurovegetative responses in these animals after an acute challenge with naloxone, there were no changes in the contents of this endocannabinoid in any of the brain regions analysed. By contrast, we observed a significant decrease in the specific binding for CB(1) receptors in the midbrain and the cerebral cortex of morphine-dependent rats, with no changes in the other regions. The decrease in the cerebral cortex was, however, accompanied by a rise in the activation of signalling mechanisms by CB(1) receptor agonists, as revealed by WIN-55,212-2-stimulated [(35)S]GTPgammaS binding, whereas a reduction in this parameter was measured in the brainstem of morphine-dependent rats. In summary, the present data are indicative of the existence of an alteration of the endocannabinoid transmission during morphine dependence in rats, although the changes observed were region-dependent and affected exclusively CB(1) receptors with no changes in endocannabinoid levels. Because the changes occurred in regions of the midbrain, the cerebral cortex and the brainstem, which have been implicated in drug dependence, our data suggest that pharmacological manipulation of the endocannabinoid system might be a novel tool to reduce morphine addiction.


The rewarding properties of the psychoactive constituents of marijuana, termed "cannabinoids," may reflect actions on synaptic transmission in the nucleus accumbens (NAc). Furthermore, long-term changes in these synapses may support the addictive process. Excitatory and inhibitory synapses are acutely inhibited by cannabinoids in the NAc, and endogenous cannabinoids (endocannabinoids) play a critical role in the expression of long-term depression (LTD) of excitatory cortical afferents in this structure. Because humans often use marijuana for prolonged periods, we examined the impact of long-term cannabinoid exposure on synaptic processes in an animal model. Electrophysiological recordings in rat brain slices containing the NAc were performed after chronic exposure to vehicle solution, Delta9-tetrahydrocannabinol (THC), or the cannabinoid agonist R(+)-[2,3-dihydro-5-methyl-3-[(morpholino)methyl]pyrrolo[1,2,3-de]-1,4-benzoxazin-6-yl]-1-(1-naphthalenyl)methanone mesylate (WIN55,212-2). Extracellular glutamatergic postsynaptic potentials and whole-cell GABAergic IPSCs were concentration-dependently inhibited by WIN55,212-2 in slices from naive or vehicle-treated animals. However, the sensitivity to WIN55,212-2 was diminished in chronic agonist-treated animals. In addition, cross-tolerance to the inhibitory effect of the mu-opioid agonist Tyr-D-Ala2, N-CH3-Phe4,Gly-ol-enkephalin was observed. Endocannabinoid-mediated LTD was initiated via electrical stimulation (5 min, 10 Hz) of glutamatergic afferents to the NAc and was completely blocked by the cannabinoid receptor antagonist SR141716A [N-piperidino-5-(4-chlorophenyl)-1-(2,4-dichlorophenyl)-4-methylpyrazole-3-carboxamide] in vehicle-treated animals. LTD was not observed in brain slices from rats chronically treated with Delta9-THC or WIN55,212-2. These data demonstrate that long-term exposure to the active ingredient of marijuana blocks synaptic plasticity in the NAc and reduces the sensitivity of GABAergic and glutamatergic synapses to both cannabinoids and opioids.


RATIONALE. Interest in therapeutic activities of cannabinoids has been restrained by the fact that they are most often mediated through activation of cannabinoid CB(1) receptors, the same receptors that mediate the effects of Delta(9)-tetrahydrocannabinol (THC) and are responsible for the abuse liability of marijuana. Persistent intravenous self-administration of THC by animals was first demonstrated in squirrel monkeys and shown to be mediated by CB(1)
receptors, but monkeys in the study had a history of cocaine self-administration, raising the possibility that persistent neurobiological adaptations might subsequently predispose animals to self-administer THC. OBJECTIVES. To demonstrate persistent intravenous self-administration of THC in drug-naive squirrel monkeys. METHODS. Monkeys with no history of exposure to other drugs learned to press a lever for intravenous injections (0.2 ml in 0.2 s) of THC under a 10-response, fixed-ratio schedule with a 60-s time-out after each injection. Acquisition of THC self-administration was rapid and the final schedule was reached in 11-34 sessions. Dose of THC was then varied from 1 to 16 micro g/kg per injection with vehicle extinction following each dose of THC. RESULTS. THC maintained significantly higher numbers of self-administered injections per session and higher rates of responding than vehicle at doses of 2, 4 and 8 micro g/kg per injection, with maximal rates of responding at 4 micro g/kg per injection. Response rates, injections per session and total THC intake per session were two- to three-fold greater in monkeys with no history of exposure to other drugs compared to previous findings in monkeys with a history of cocaine self-administration. CONCLUSIONS. THC can act as an effective reinforcer of drug-taking behavior in monkeys with no history of exposure to other drugs, suggesting that self-administration of THC by monkeys provides a reliable animal model of human marijuana abuse.


Agonist-induced regulation of cannabinoid CB1 receptors was examined in HEK-293 cells transfected with CB1 receptors and in neuroblastoma N18TG2 cells that naturally express CB1 receptors. In HEK-293 cells, CB1 receptors internalization proceeded, in parallel, via clathrin-coated pits and caveolae. Simultaneous disruption of both pathways induced compensatory endocytic mechanism(s). In N18TG2 cells, endocytosis was not mediated by caveolae-like membrane domains. Heterologous, opioid-induced, downregulation of CB1 receptors was evident in HEK-293 but not N18TG2 cells. The data demonstrate the existence of multiple pathways of CB1 receptors regulation.


The disc cell wall facing the secretory cavity in lipophilic glands of Cannabis was studied for origin and distribution of hyaline areas, secretory vesicles, fibrillar matrix and particulate material. Secretions evident as light areas in the disc cell cytoplasm pass through modified regions in the plasma membrane and appear as hyaline areas in the cell wall. Hyaline areas, surrounded with a filamentous outline, accumulate near the wall surface facing the secretory cavity where they fuse to form enlarged hyaline areas. Fibrillar matrix is related to and may originate from the dense outer layer of the plasma membrane. This matrix becomes distributed throughout the wall material and contributes in part to the composition of the surface feature of secretory vesicles. Thickening of the cell wall is associated with secretions from the disc cells that facilitates movement of hyaline areas, fibrillar matrix and other possible secretions through the wall to form secretory vesicles and intervesicular materials in the secretory cavity. The outer wall of disc cells in aggregate forms the basilar wall surface of the secretory cavity which facilitates the organization of secretory vesicles that fill the secretory cavity.


The endocannabinoid arachidonylethanolamide (AEA, anandamide) is an endogenous ligand for the cannabinoid receptors and has been shown to be oxygenated by cyclooxygenase-2 (COX-2). We examined the structural requirements for COX-mediated, AEA oxygenation using a number of substrate analogues and site-directed mutants of COX-2. Fourteen AEA analogues were synthesized and tested as COX substrates. These studies identified the hydroxyl moiety of AEA as a critical determinant in the ability of COX enzymes to effect robust endocannabinoid oxygenation. In addition, these studies suggest that subtle structural modifications of AEA analogues near the ethanolamide moiety can result in pronounced changes in their ability to serve as COX-2 substrates. Site-directed mutagenesis studies have permitted the development
of a model of AEA binding within the COX-2 active site. As with arachidonic acid, the omega-terminus of AEA binds in a hydrophobic alcove near the top of the COX-2 active site. The polar ethanolamide moiety of AEA, like the carboxylate of arachidonate, interacts with Arg-120 at the bottom of the COX-2 active site. Mutation of Tyr-385 prevents AEA oxygenation, suggesting that, as in the case of other COX substrates, AEA metabolism is initiated by Tyr-385-mediated hydrogen abstraction. Thus, AEA binds within the COX-2 active site in a conformation roughly similar to that of arachidonic acid. However, important differences have been identified that account for the isoform selectivity of AEA oxygenation. Importantly, the COX-2 side pocket and Arg-513 in particular are critical determinants of the ability of COX-2 to efficiently generate prostaglandin H(2) ethanolamide. The reduced efficiency of COX-1-mediated, AEA oxygenation can thus be explained by the absence of an arginine residue at position 513 in this isoform. Mutational analysis of Leu-531, an amino acid located directly across from the COX-2 side pocket, suggests that AEA is shifted away from this hydrophobic residue and toward Arg-513 relative to arachidonic acid. Coupled with earlier observations with the endocannabinoid 2-arachidonylglycerol, these results indicate that one possible function of the highly conserved COX-2 active site side pocket is to promote endocannabinoid oxygenation.


Cannabinoids could provide neuroprotection in neurodegenerative disorders. In this study, we examined whether a treatment with Delta9-tetrahydrocannabinol, a non-selective cannabinoid receptor agonist, or with SR141716, a selective antagonist for the cannabinoid CB(1) receptor subtype, could affect the toxicity of the complex II reversible inhibitor malonate injected into the striatum, which replicates the mitochondrial complex II deficiency seen in Huntington's disease patients. As expected, malonate injection produced a significant reduction in cytochrome oxidase activity in the striatum consistent with the expected neurodegeneration caused by this toxin. The administration of Delta9-tetrahydrocannabinol increased malonate-induced striatal lesions compared to vehicle and, surprisingly, SR141716, far from producing effects opposite to those of Delta9-tetrahydrocannabinol, also enhanced malonate effects, and to an even greater extent. In summary, our results are compatible with the idea that manipulating the endocannabinoid system can modify neurodegeneration in Huntington's disease, and suggest that highly selective CB(1) receptor agonists might be necessary to produce neuroprotective effects against indirect excitotoxicity.


BACKGROUND AND PURPOSE: Cannabinoids confer neuroprotection in several experimental paradigms, but the responsible mechanisms remain unknown. Therefore, we sought to examine whether the synthetic CB1 agonist HU-210 is capable of reducing ischemic damage and to determine the mechanisms responsible for such protection. METHODS: Sprague-Dawley rats underwent permanent middle cerebral artery occlusion (PMCAO). After dose-response and therapeutic time window-finding experiments, the rats were injected with HU-210 (45 micro g/kg IV) or vehicle 1 hour after PMCAO. Physiological parameters and cerebral blood flow in the peri-infarct zone were monitored. The animals were examined with a motor disability scale, and the infarct volumes were measured 72 hours later. We also examined the effects of the selective CB1 antagonist SR-141716 and of controlled warming on the neuroprotection conferred by HU-210. RESULTS: HU-210 reduced blood pressure and heart rate but did not alter the cerebral blood flow in the infarct border zone. Motor disability and infarct volumes were significantly reduced (by up to 77%; P<0.05) in animals treated with HU-210. A single injection of HU-210 significantly lowered the body temperature compared with vehicle as measured both at 1 hour (32.3+/−1.3 degrees C versus 35+/−1.6 degrees C; P=0.0024) and at 24 hours (31.5+/−2.5 degrees C versus
37.25+/-.3 degrees C; P=0.0031) after PMCAO. The protective effects of HU-210 were partially reversed by pretreatment with SR-141716 but were completely abolished by warming of the animals to the levels observed in controls. CONCLUSIONS: HU-210 confers robust protection against ischemic damage. This protection is mediated at least in part by binding to CB1 receptors and is also associated with the indirect protective effects of hypothermia.


Cannabinoids have been shown to disrupt memory processes and these effects occur primarily through cannabinoid CB1 receptors in the brain. The present study investigates, for the first time, the regional variations and age-related changes in CB1 protein expression in the hippocampus and its neighbouring entorhinal, perirhinal, postrhinal and temporal cortices using Western blotting. In young adult rats, CB1 protein was highly expressed in the hippocampus and within the hippocampus, the greatest density of CB1 protein was located in CA1. When a comparison was made between young (4-month-old) and aged (24-month-old) rats, CB1 protein expression was significantly increased in the aged entorhinal and temporal cortices and was significantly decreased in the aged postrhinal cortex. The present study demonstrates region-specific changes in CB1 protein expression during ageing and further suggests that cannabinoid CB1 receptors may contribute to the aging process.


This study was designed to evaluate the neuroprotective effect of the cannabinoid agonist WIN-55212 after inducing acute severe asphyxia in newborn rats. The left common carotid artery was ligated in anaesthetised 7-day-old Wistar rats, which were then asphyxiated by inhaling 100% nitrogen for 10 min. Pups recovering from asphyxia were s.c. administered vehicle (n=23), WIN-55212 (0.1 mg/kg, n=18), or WIN-55212 plus the CB(1) receptor antagonist SR141716 (3 mg/kg, n=10). Pups undergoing a sham operation served as controls (n=12). Coronal sections of the brain were obtained on the 14th day after surgery and observed under light microscope after Nissl or Fluoro-Jade B (FJB) staining, to respectively quantify surviving or degenerating neurones in the CA1 area of the hippocampus and parietal cortex. Acute asphyxia led to early neurone loss amounting to 19% in the hippocampus and 29% in the cortex (both ANOVA P<0.05 vs. control). Delayed neurone loss occurred in the proportions 13% in the hippocampus and 20% in the cortex (both ANOVA P<0.05 vs. control). Neuronal loss was fully prevented by WIN-55212 administration. Co-administration of SR141716 failed to modify the protective effect of WIN-55212 on early neuronal death, but abolished the WIN-55212-induced prevention of delayed neuronal death. We conclude that when administered after acute severe asphyxia in newborn rats, WIN-55212 shows a neuroprotective effect, reducing both early and delayed neurone loss. This effect is achieved through two parallel CB(1)-dependent and -independent mechanisms.


Interleukin-1 receptor antagonist (IL-1ra) is an important anti-inflammatory cytokine that blocks all known actions of IL-1 and markedly protects against experimentally induced ischemic, excitotoxic, and traumatic brain insults. Cannabinoids (CBs) also exert potent anti-inflammatory and neuroprotective effects, but the mechanisms of their actions are unknown. Here we tested the hypothesis that the actions of CBs are mediated by endogenous IL-1ra. We report for the first time that both CB1 and CB2 receptors modulate release of endogenous IL-1ra from primary cultured glial cells. Activation of CB1 or CB2 receptors increased lipopolysaccharide-induced IL-1ra release, and specific CB1 or CB2 antagonists blocked lipopolysaccharide-induced production of IL-1ra from glial cells. Comparison of neuronal cultures from wild-type mice and mice lacking IL-1ra (knock-out) indicates that endogenous IL-1ra is essential for the neuro-protective effects of CBs against excessive activation of glutamate receptors (excitotoxicity) in response to S-AMPA.
or NMDA. Similarly, analysis of mixed glial cultures from IL-1ra knock-out mice indicates that endogenous IL-1ra is required for the CB-induced inhibition of nitric oxide production in response to bacterial lipopolysaccharide. These data suggest a novel neuroprotective mechanism of action for CBs in response to inflammatory or excitotoxic insults that is mediated by both CB1 and CB2 receptor-dependent pathways.


Anandamide is a prominent member of the endocannabinoids, a group of diffusible lipid molecules which influences neuronal excitability. In this context, endocannabinoids are known to modulate certain presynaptic Ca(2+) and K(+) channels, either through cannabinoid (CB1) receptor stimulation and second messenger pathway activation or by direct action. We investigated the susceptibility of voltage-sensitive sodium channels to anandamide and other cannabimimetics using both biochemical and electrophysiological approaches. Here we report that anandamide, AM 404 and WIN 55,212-2 inhibit veratridine-dependent depolarization of synaptoneurosomes (IC(50)s, respectively 21.8, 9.3 and 21.1 &mgr;M) and veratridine-dependent release of L-glutamic acid and GABA from purified synaptosomes [IC(50)s: 5.1 &mgr;M (L-glu) and 16.5 &mgr;M (GABA) for anandamide; 1.6 &mgr;M (L-glu) and 3.3 &mgr;M (GABA) for AM 404, and 12.2 (L-glu) and 14.4 &mgr;M (GABA) for WIN 55,212-2]. The binding of [3H]batrachotoxinin A 20-alpha-benzoate to voltage-sensitive sodium channels was also inhibited by low to mid micromolar concentrations of anandamide, AM 404 and WIN 55,212-2. In addition, anandamide (10 &mgr;M), AM 404 (10 &mgr;M) and WIN 55,212-2 (1 &mgr;M) were found to markedly block TTX-sensitive sustained repetitive firing in cortical neurones without altering primary spikes, consistent with a state-dependent mechanism. None of the inhibitory effects we demonstrate on voltage-sensitive sodium channels are attenuated by the potent CB1 antagonist AM 251 (1-2 &mgr;M). Anandamide's action is reversible and its effects are enhanced by fatty acid amidohydrolase inhibition. We propose that voltage-sensitive sodium channels may participate in a novel signaling pathway involving anandamide. This mechanism has potential to depress synaptic transmission in brain by damping neuronal capacity to support action potentials and reducing evoked release of both excitatory and inhibitory transmitters.


The cholinergic system is crucial for higher brain functions including learning and memory. These functions are mediated primarily by muscarinic acetylcholine receptors (mAChRs) that consist of five subtypes (M1-M5). A recent study suggested a novel role of acetylcholine as a potent enhancer of endocannabinoid signalling that acts retrogradely from postsynaptic to presynaptic neurons. In the present study, we further investigated the mechanisms of this cholinergic effect on endocannabinoid signalling. We made paired whole-cell recordings from cultured hippocampal neurons, and monitored inhibitory postsynaptic currents (IPSCs). The postsynaptic depolarization induced a transient suppression of IPSCs (DSI), a phenomenon known to involve retrograde signalling by endocannabinoids. The cholinergic agonist carbachol (CCh) markedly enhanced DSI at 0.01-0.3 micro m without changing the presynaptic cannabinoid sensitivity. The facilitating effect of CCh on DSI was mimicked by the muscarinic agonist oxotremorine-M, whereas it was eliminated by the muscarinic antagonist atropine. It was also blocked by a non-hydrodizable analogue of GDP (GDP-beta-S) that was applied intracellularly to postsynaptic neurons. The muscarinic enhancement of DSI persisted to a substantial degree in the neurons prepared from M1-knockout and M3-knockout mice, but was virtually eliminated in the neurons from M1/M3-compound-knockout mice. CCh still enhanced DSI significantly under the blockade of postsynaptic K+ conductance, and did not significantly influence the depolarization-induced Ca2+ transients. These results indicate that the activation of postsynaptic M1 and M3 receptors facilitates the depolarization-induced release of endocannabinoids.

Earlier work from our laboratories has provided evidence for the existence of a subsite within the CB1 and CB2 cannabinoid receptor binding domain corresponding to substituents at the benzylic side chain position of classical cannabinoids. The existence and stereochemical features of this subsite have now been probed through the synthesis of a novel series of (-)-Delta(8)-tetrahydrocannabinol analogues bearing C1'-ring substituents. Of the compounds described here, those with C1'-dithiolane (1c), C1'-dioxolane (2d), and cyclopentyl (2a) substituents exhibited the highest affinities for CB1 and CB2. We used molecular modeling approaches to better define the stereochemical limits of the putative subsite. In vitro pharmacological testing found 1c to be a potent CB1 agonist.


In dioecious plants of hemp ( Cannabis sativa L.), males are regarded as heterogametic XY and females as homogametic XX, although it is difficult to discriminate the X cytologically from the Y. The Y chromosome is somewhat larger than the X. Our aim was to analyse AFLP markers on X and Y, and to use them to gain some insight into the structure of the sex chromosomes. Markers located on the sex chromosomes can be grouped into different classes, depending on the presence or absence of a fragment on the X and/or the Y. They are detected by separately analysing male and female progenies of a single cross. Five markers were found to be located on both chromosomes. A few recombinants were observed for marker pairs of this class in the male progenies. Two completely linked markers located on the Y chromosome in the male parent show a recombination rate of r = 0.25 with sex. Recombination must have occurred between the sex chromosomes in the male parent. The recombination analysis led to the conclusion that there is a pseudoautosomal region (PAR) on the sex chromosomes, allowing recombination between the X and the Y chromosome. The other regions of the sex chromosomes show only a few recombination events, for the Y as well as for the X. These results are discussed in comparison to other dioecious plants.


Cannabinoid compounds have been shown to produce antinociception and antihyperalgesia by acting upon cannabinoid receptors located in both the CNS and the periphery. A potential mechanism by which cannabinoids could inhibit nociception in the periphery is the activation of cannabinoid receptors located on one or more classes of primary nociceptive neurons. To address this hypothesis, we evaluated the neuronal distribution of cannabinoid receptor type 1 (CB1) in the trigeminal ganglion (TG) of the adult rat through combined in situ hybridization (ISH) and immunohistochemistry (IHC). CB1 receptor mRNA was localized mainly to medium and large diameter neurons of the maxillary and mandibular branches of the TG. Consistent with this distribution, in a de facto nociceptive sensory neuron population that exhibited vanilloid receptor type 1 immunoreactivity, colocalization with CB1 mRNA was also sparse (<5%). Furthermore, very few neurons (approximately 5%) in the peptidergic (defined as calcitonin gene-related peptide- or substance P-immunoreactive) or the isolectin B(4)-binding sensory neuron populations contained CB1 mRNA. In contrast, and consistent with the neuron-size distribution for CB1, nearly 75% of CB1-positive neurons exhibited N52-immunoreactivity, a marker of myelinated axons. These results indicate that in the rat TG, CB1 receptors are expressed predominantly in neurons that are not thought to subserve nociceptive neurotransmission in the noninjured animal. Taken together with the absence of an above background in situ signal for CB2 mRNA in TG neurons, these findings suggest that the peripherally mediated antinociceptive effects of cannabinoids may involve either as yet
unidentified receptors or interaction with afferent neuron populations that normally subserve non-nociceptive functions.


Multiple sclerosis is increasingly being recognized as a neurodegenerative disease that is triggered by inflammatory attack of the CNS. As yet there is no satisfactory treatment. Using experimental allergic encephalomyelitis (EAE), an animal model of multiple sclerosis, we demonstrate that the cannabinoid system is neuroprotective during EAE. Mice deficient in the cannabinoid receptor CB1 tolerate inflammatory and excitotoxic insults poorly and develop substantial neurodegeneration following immune attack in EAE. In addition, exogenous CB1 agonists can provide significant neuroprotection from the consequences of inflammatory CNS disease in an experimental allergic uveitis model. Therefore, in addition to symptom management, cannabis may also slow the neurodegenerative processes that ultimately lead to chronic disability in multiple sclerosis and probably other diseases.


Cannabinoids are cell membrane-derived signalling molecules that are released from nerves, blood cells and endothelial cells, and have diverse biological effects. They act at two distinct types of G-protein-coupled receptors, cannabinoid CB(1) and CB(2) receptors. Cannabinoid CB(1) receptors are highly localised in the central nervous system and are also found in some peripheral tissues, and cannabinoid CB(2) receptors are found outside the central nervous system, in particular in association with immune tissues. Novel actions of cannabinoids at non-CB(1) non-CB(2) cannabinoid-like receptors and vanilloid VR1 receptors have also recently been described. There is growing evidence that, among other roles, cannabinoids can act at prejunctional sites to modulate peripheral autonomic and sensory neurotransmission, and the present article is aimed at providing an overview of this. Inhibitory cannabinoid CB(1) receptors are expressed on the peripheral terminals of autonomic and sensory nerves. The role of cannabinoid receptor ligands in modulation of sensory neurotransmission is complex, as certain of these (anandamide, an "endocannabinoid", and N-arachidonoyl-dopamine, an "endovanilloid") also activate vanilloid VR1 receptors (coexpressed with cannabinoid CB(1) receptors), which excites sensory nerves and causes a release of sensory neurotransmitter. The fact that the activities of anandamide and N-arachidonoyl-dopamine span two distinct receptor families raises important questions about cannabinoid/vanilloid nomenclature, and as both compounds are structurally related to the archetypal vanilloid capsaicin, all three are arguably members of the same family of signalling molecules. Anandamide is released from nerves, but unlike classical neurotransmitters, it is not stored in and released from nerve vesicles, but is released on demand from the nerve cell membrane. In the central nervous system, cannabinoids function as retrograde signalling molecules, inhibiting via presynaptic cannabinoid CB(1) receptors the release of classical transmitter following release from the postsynaptic cell. At the neuroeffector junction, it is more likely that cannabinoids are released from prejunctional sites, as the neuroeffector junction is wide in some peripheral tissues and cannabinoids are rapidly taken up and inactivated. Understanding the actions of cannabinoids as modulators of peripheral neurotransmission is relevant to a variety of biological systems and possibly their disorders.


New data strengthen the idea of a prominent role for endocannabinoids in the modulation of a wide variety of neurobiological functions. Among these, two functions, control of movement and antinociception, have attracted the maximal interest because of the possibility that cannabinoids and related compounds might be used with a therapeutic purpose. However, the functions of endocannabinoids in the brain, and also in the periphery, are large and involve, not only the adulthood, but also the period of prenatal and postnatal development, when endocannabinoids have been reported to be significantly present and to play a role in processes of brain development as neuronal proliferation and migration, axonal elongation, synaptogenesis.
and/or myelogenesis. The present review article will summarize the different studies carried out on this topic and will suggest future lines of research to clarify the role of endocannabinoids and their receptors in the development.


The cannabinoid CB(1) and CB(2) receptors belong to the Class A, rhodopsin-like family of G protein-coupled receptors. Antagonists for each receptor sub-type, as well as four structural classes of agonists that bind to both receptors, have been identified. An extensive amount of SAR has been developed for agonists and antagonists that bind at CB1, while the SAR of CB2 ligands is only now emerging in the literature. Cannabinoid agonists have been suggested to have potential therapeutic uses as appetite stimulants, analgesics, anti-emetics, anti-diarrheals, anti-spasmodics, tumor anti-proliferative agents, anti-glaucoma agents and as agents for the treatment of diseases associated with inappropriate retention of aversive memories such as post-traumatic stress disorders and phobias. Cannabinoid CB1 antagonists have been suggested to have potential therapeutic uses as appetite suppressants and as agents that improve memory. This review focuses first on recent CB1 and CB2 SAR and on the pharmacophores that have been developed for ligand recognition at the CB1 receptor. Emerging ideas about how the cannabinoid receptors are activated by agonists or inactivated by inverse agonists are then presented. Challenges for future SAR and pharmacophore development are also identified.


Anandamide triggers various cellular activities by binding to cannabinoid (CB1/CB2) receptors or vanilloid receptor 1 (VR1). However, the role of these receptors in anandamide-induced apoptosis remains largely unknown. Here, we show that SR141716A, a specific inhibitor of cannabinoid receptor (CB1-R), did not block anandamide-induced cell death in endogenously CB1-R expressing cells. In addition, CB1-R-lacking Chinese hamster ovary (CHO) cells underwent cell death after anandamide treatment. SR144528, a specific inhibitor of CB2-R also failed to block anandamide-induced cell death in HL-60 cells. Capsazepine, a specific antagonist of VR1 could not prevent anandamide-induced cell death in constitutively and endogenously VR1 expressing PC12 cells. Moreover, anandamide noticeably triggered cell death in VR1-lacking human embryonic kidney (HEK) cells. In contrast, methyl-beta cyclodextrin (MCD), a membrane cholesterol depletor, completely blocked anandamide-induced cell death in a variety of cells, including PC12, C6, Neuro-2a, CHO, HEK, SMC, Jurkat and HL-60 cells. MCD also blocked anandamide-induced superoxide generation, phosphatidyl serine exposure and p38 MAPK/JNK activation. Thus, our data imply a novel role for of membrane lipid rafts in anandamide-induced cell death.


Anti-nociceptive effects of the endocannabinoid anandamide are well established. Anandamide has, however, also been shown to activate pro-nociceptive vanilloid 1 (VR1) receptors present on primary afferent nociceptors. The aim of the present study was to determine the effect of intraplantar injection of anandamide on dorsal spinal neuronal responses in control rats and rats with hindpaw carrageenan-induced inflammation. Effects of intraplantar administration of anandamide (50 microg in 50 microl) on peripheral mechanically-evoked responses of spinal neurones were studied in halothane-anaesthetised rats in vivo. Responses of spinal neurones to mechanical punctate stimulation (von Frey filaments, 8-80 g) of the peripheral receptive field were similar in non-inflamed rats and rats with hindpaw carrageenan-induced inflammation. Intraplantar injection of anandamide, but not vehicle, significantly (P<0.05) inhibited innocuous and noxious mechanically-evoked responses of spinal neurones in rats with hindpaw inflammation, but not in non-inflamed rats. Co-administration of the cannabinoid (2) (CB(2)) receptor antagonist, SR144528 (10 microg in 50 microl), but not the cannabinoid (1) (CB(1))
receptor antagonist, SR141716A (10 microg in 50 microl), significantly blocked inhibitory effects of anandamide on peripheral evoked neuronal responses in rats with hindpaw inflammation. This study demonstrates inhibitory effects of exogenous anandamide on mechanically-evoked responses under inflammatory conditions in vivo, which are mediated by peripheral CB(2) receptors.


BACKGROUND AND RATIONALE. Starting with the discovery of an endogenous brain cannabinoid system with specific receptors and endogenous ligands, research in the cannabinoid field has accelerated dramatically over the last 15 years. Cannabis is the most used illicit psychotropic substance in the world but only recently have reliable preclinical models become available for investigating the rewarding and dependence-producing actions of its psychoactive constituent, Delta(9)-tetrahydrocannabinol (THC). OBJECTIVES. The goal of this review is to examine the various animal models currently available that are being used to facilitate our understanding of the rewarding and dependence-producing actions of cannabinoids, which are central to their abuse liability, and of the neurochemical mechanisms that may underlie these actions of cannabinoids. RESULTS AND CONCLUSIONS. Recent demonstrations that strong and persistent intravenous self-administration behavior can be obtained in squirrel monkeys using a range of THC doses that are in agreement with the total intake and the single doses of THC normally self-administered by humans smoking marijuana cigarettes provides a reliable and direct tool for assessing the reinforcing effects of THC that are central to its abuse liability. In addition, recent demonstrations of persistent intravenous self-administration of synthetic cannabinoid CB1 receptor agonists by rats and mice and the development of genetically modified mice lacking specific cannabinoid receptors provide convenient rodent models for exploring underlying neurochemical mechanisms. Repeated demonstrations in rats that THC and synthetic CB1 agonists can induce conditioned place preferences or aversions, depending on details of dose and spacing, can reduce the threshold for intracranial self-stimulation behavior under certain conditions, and can serve as effective discriminative stimuli for operant behavior provide less direct, but more rapidly established, measures for investigating the rewarding effects of cannabinoids. Finally, there have been numerous recent reports of major functional interactions between endogenous cannabinoid, opioid, and dopaminergic neurotransmitter systems in areas such as analgesia, physical dependence and tolerance development, and drug reinforcement or reward. This provides an opportunity to search for drugs with the beneficial therapeutic effects of currently available cannabinoids or opioids but without undesirable adverse effects such as abuse liability.


Endocannabinoids are thought to function as retrograde messengers, which modulate neurotransmitter release by activating presynaptic cannabinoid receptors. Anandamide and 2-arachidonoylglycerol (2-AG) are the two best studied endogenous lipids which can act as endocannabinoids. Together with the proteins responsible for their biosynthesis, inactivation and the cannabinoid receptors, these lipids constitute the endocannabinoid system. This system is proposed to be involved in various neurodegenerative diseases such as Parkinson's and Huntington's diseases as well as Multiple Sclerosis. It has been demonstrated that the endocannabinoid system can protect neurons against glutamate excitotoxicity and acute neuronal damage in both in vitro and in vivo models. In this paper we review the data concerning the involvement of the endocannabinoid system in neurodegenerative diseases in which neuronal cell death may be elicited by excitotoxicity. We focus on the biosynthesis of endocannabinoids and on their modes of action in animal models of these neurodegenerative diseases.

2-Arachidonoylglycerol (2-AG) is a putative endogenous ligand for cannabinoid receptors and was suggested to play an important role in both physiological and pathological events in the central nervous system (CNS) as well as in peripheral organs. The sequential hydrolysis of arachidonic acid (20:4n-6, AA)-containing phospholipids has been proposed as a major biosynthetic route of 2-AG. On the other hand, the manipulation of the dietary n-3 polyunsaturated fatty acid (PUFA) status changes the AA level in tissue phospholipids. We, therefore, conducted two separate experiments to confirm whether the dietary n-3 PUFA status influences the 2-AG level in the mouse brain. In the first experiment, we fed mice with n-3 PUFA-deficient diet, which resulted in a marked decrease in the docosahexaenoic acid (22:6n-3, DHA) levels without a change in the AA level in brain phospholipids as compared with the mice fed with an n-3 PUFA-sufficient diet. The brain 2-AG level in the n-3 PUFA-deficient group was significantly higher than in the n-3 PUFA sufficient group. In the second experiment, we found that short-term supplementation of DHA-rich fish oil reduced brain 2-AG level as compared with the supplementation with low n-3 PUFA. The decrease in the AA level and the increase in the DHA level in the major phospholipids occurred in the brains of the mice fed the fish oil diet compared with those fed the low n-3 PUFA diet. Our results indicate that the n-3 PUFA deficiency elevates and n-3 PUFA enrichment reduces the brain 2-AG level in mice, suggesting that physiological and pathological events mediated by 2-AG through cannabinoid receptor in the CNS could be modified by the manipulation of the dietary n-3 PUFA status.


The expression of central cannabinoid (CB(1)) receptors in tyrosine hydroxylase (TH) containing neurones was demonstrated. Co-localisation was present in different brain areas responsible for reward-related mechanisms. The immunohistochemical investigations have shown that co-localisation is present in parts of mesolimbic-mesocortical dopaminergic system like nucleus accumbens (Nacb), ventral tegmental area (VTA), in the striatum, pyriform cortex, respectively. The results suggest a functional role of CB(1) receptors in cannabis addiction by acting directly on reward-related structures.


A notable consequence of CB1 cannabinoid receptor activation in vertebrates is an impairment of cognitive function related to learning and short-term memory. The mechanisms of this impairment remain unclear, but one possibility is that cannabinoids influence encoding of stimuli at sensory and/or perceptual levels. Here, by treating zebra finches with the cannabinoid agonist WIN55212-2 and then measuring expression of the transcription factor zenk following presentation of novel zebra finch song, we show that cannabinoid receptor activation differentially influences zenk expression in sensory versus perceptual regions of the songbird auditory telencephalon. That is, WIN55212-2 dose-dependently inhibited zenk expression in a region for auditory perception (NCM, the caudomedial neostriatum), but had no effect on zenk expression in the primary auditory area, the Field L complex. The inhibitory effects of WIN55212-2 on zenk expression in NCM were reversed by coadministration of the CB1-selective antagonist SR141716A. Moreover, we found that the habituation of the NCM zenk response to repeated presentation of the same song, a well-established neural correlate of song recognition, was blocked when birds were treated with WIN55212-2 during habituation trials. Our data suggest that activation of CB1 cannabinoid receptors can selectively influence perceptual and mnemonic aspects of auditory experience.


Cannabinoids produce a characteristic profile of in vivo effects in mice, including suppression of spontaneous activity, antinociception, hypothermia, and catalepsy. Measurement of these four properties, commonly referred to as the tetrad test, has played a key role in establishing the structure-activity relationship of cannabinoids acting at cannabinoid CB(1)
receptors. The purpose of this study was to determine whether drugs acting at noncannabinoid CB(1) receptors produced a similar pharmacological profile. Mice were tested in this paradigm after being injected with Delta(9)-tetrahydrocannabinol and selected drugs from other drug classes. Delta(9)-Tetrahydrocannabinol dose-dependently produced all four effects with reversal by the cannabinoid CB(1) receptor antagonist N-(piperidin-1-yl)-5-(4-chlorophenyl)-1-(2,4-dichlorophenyl)-4-methyl-1H-pyrazole-3-carboxamide hydrochloride (SR 141716A). Amphetamine, scopolamine, morphine, desipramine, pimozide, pentobarbital, ethanol, and diazepam were not fully active in at least one of the tests. Antipsychotics showed the greatest similarity to those of cannabinoids in the tetrad tests, although there were also distinct differences. Clozapine, haloperidol, thioridazine, and chlorpromazine (but not pimozide) were fully active in all four tests; however, unlike with Delta(9)-tetrahydrocannabinol, their effects were not blocked by SR 141716A. Further, whereas antipsychotics produced nearly 100% catalepsy, maximal catalepsy produced by Delta(9)-tetrahydrocannabinol was 60%. The mechanism through which antipsychotics produce these effects in mice is uncertain, but it differs from cannabinoid CB(1) receptor activation that mediates the effects of cannabinoids. While results of previous research suggest that the tetrad tests are a useful tool in examination of structure-activity relationships of cannabinoid CB(1) receptor agonists, the present results suggest that they must be used cautiously in the search for novel cannabinoid receptors.

**CLINICAL SCIENCE**


Research of the cannabinoid system has many similarities with that of the opioid system. In both instances, studies into drug-producing plants led to the discovery of an endogenous control system with a central role in neurobiology. Few compounds have had as much positive press from patients as those of the cannabinoid system. While these claims are investigated in disorders such as multiple sclerosis spasticity and pain, basic research is discovering interesting members of this family of compounds that have previously unknown qualities, the most notable of which is the capacity for neuroprotection. Large randomised clinical trials of the better known compounds are in progress. Even if the results of these studies are not as positive as many expect them to be, that we are only just beginning to appreciate the huge therapeutic potential of this family of compounds is clear.


The abuse of alcohol is associated with chronic cardiomyopathy, hypertension, and arrhythmia. Abstinence or using alcohol in moderation can reverse these cardiovascular problems. Alcohol is also distinguished among the substances of abuse by having possible protective effects against coronary artery disease and stroke when used in moderate amounts. Amphetamines (eg, speed, ice, ecstasy) have many of the cardiovascular toxicities seen with cocaine, including acute and chronic cardiovascular diseases. Heroin and other opiates can cause arrhythmias and noncardiac pulmonary edema, and may reduce cardiac output. Cardiovascular problems are less common with cannabis (marijuana) than with opiates, but major cognitive disorders may be seen with its chronic use. It is still controversial whether caffeine can cause hypertension and coronary artery disease, and questions have been raised about its safety in patients with heart failure and arrhythmia.


Cognitive, cerebrovascular, and psychiatric impairments have been documented with chronic marijuana users. To better understand the nature and duration of these neurocognitive changes in marijuana abusers, we recorded the resting EEG of 29 abstinent chronic marijuana abusers and 21 control subjects. The marijuana abusers were tested twice: the first evaluation
occurred within 72 hours of admission to the inpatient research unit; the second evaluation occurred after 28 to 30 days of monitored abstinence. A three-minute period of EEG was recorded during resting eyes-closed conditions from eight electrodes (F(3), C(3), P(3), O(1), F(4), C(4), P(4), and O(2)). The artifactual EEG was converted to six frequency bands (delta, theta, alpha(1), alpha(2), beta(1), and beta(2)) using a fast Fourier transform. During early abstinence, absolute power was significantly lower (p < 0.05) for the marijuana abusers than for the control subjects for the theta and alpha(1) bands. These reductions in theta and alpha(1) power persisted for 28 days of monitored abstinence. These EEG changes, together with cerebral blood flow deficits, might underlie the cognitive alterations observed in marijuana abusers. Additional research is needed to determine how long these deficits persist during abstinence and if treatment with neuroprotective agents may reverse them.


During the last few years, the debate over the use of marijuana for medical purposes has moved from the legislative arena into the public forum. Since 1996, eight states have passed ballot initiatives that allow people with certain medical conditions to use marijuana, and now most of those states are wrestling with ways to implement the laws. During the past 24 years, 35 states have enacted laws on medical marijuana; however, several states have either repealed their laws or allowed them to sunset.


Twenty-nine volunteers participated in a randomized, double-blind, placebo-controlled study. Cerebral blood velocity (CBV), pulse rate, blood pressure (BP), skin perfusion (SP) on forehead and plasma Delta(9) tetrahydrocannabinol (THC) levels were quantified during reclining and standing for 10 min before and after THC infusions and marijuana smoking. Both THC and marijuana induced postural dizziness, with 28% reporting severe symptoms. Intoxication and dizziness peaked immediately after drug. The severe dizziness group showed the most marked postural drop in CBV and BP and showed a drop in pulse rate after an initial increase during standing. Postural dizziness was unrelated to plasma levels of THC and other indices.


Until recently, relatively little research has focused on the treatment of marijuana abuse or dependence; however, marijuana use disorders are now receiving increased attention. This paper reviews the initial clinical trials evaluating the efficacy of outpatient treatments for adult marijuana dependence. Findings from five controlled trials of psychotherapeutic interventions suggest that this disorder appears responsive to the same types of treatment as other substance dependencies. Moreover, these initial studies suggest that many patients do not show a positive treatment response, indicating that marijuana dependence is not easily treated. Strengths and weaknesses of the data are presented. Preliminary data from less controlled studies relevant to the treatment of marijuana dependence are discussed to suggest future research areas. Although very few studies on treatment for marijuana abuse and dependence have been completed, the initial reports identify promising treatment approaches and demonstrate a need for more research on the development of effective interventions.


BEHAVIOURAL SCIENCE


The purpose of this study was to examine rates and patterns of illicit drug use among Canadian university undergraduates, to compare these rates with those for non-university samples, and to describe drug-use trends among university undergraduates in the province of Ontario between 1988 and 1998. A national mail survey was carried out based on stratified 2-stage sample design. The sample comprised 7,800 Canadian undergraduates from 16 universities (52% of eligible respondents). Approximately 47.5% reported use of illicit drug during their life, 29.6% in the previous 12 months, and 18.7% since the beginning of the academic year. Cannabis was by far the most widely used drug (47.0%, 28.7%, and 18.2%, respectively). Many of the gender and regional associations were similar to those found in general-populations surveys. Comparisons to non-university peers did not indicate elevated rates among university students. Among Ontario university undergraduates the use of cannabis, hallucinogens, methamphetamine, crack, and heroin remained stable between 1988 and 1998. The use of cocaine declined from 4.8% to 1.7%. Rates of illicit drug use were not appreciably higher than those among their non-university peers. Other public-health issues, such as heavy drinking and poor mental health, override those related to illicit drug use.


The main aim of this study was to identify adolescent/young adulthood factors that predicted persistent driving after drinking, persistent unsafe driving after drinking, and persistent cannabis use and driving among young adults. It was a longitudinal study of a birth cohort (n=933, 474 males and 459 females) and was based on data collected at ages 15, 18, 21 and 26 years. At each of these ages members of the cohort attended the research unit for a personal interview by a trained interviewer, using a standardised questionnaire. For this study, the data for the outcome measures (persistent driving after drinking, persistent unsafe driving after drinking, and persistent driving after using cannabis) were obtained at ages 21 and 26 years. The main explanatory measures were collected at ages 15, 18, 21 years and included demographic factors (academic qualifications, employment, parenting); personality measures; mental health measures (substance use, cannabis dependence, alcohol dependence, depression); anti-social behaviour (juvenile arrest, aggressive behaviour, court convictions); early driving behaviour and experiences (car and motorcycle licences, traffic crashes). The analyses were conducted by gender. The results showed that females who persisted in driving after drinking (13%, n=61) were more likely than the others to have a motorcycle licence at 18. The males who persisted in driving after
drinking (28%, n=135) were more likely than the other males to have some school academic qualifications and to be employed at age 26. Compared to the other males, those who persisted in unsafe driving after drinking (4%, n=17) were more likely to be aggressive at 18 and alcohol dependent at 21. Only six (1%) females persisted in unsafe driving after drinking so regression analyses were not conducted for this group. For persistent driving after using cannabis, the univariate analyses showed that females who persisted with this behaviour tended to have high substance use at 18, cannabis dependence at 21, police contact as a juvenile, and to be a parent at 21. For this group, because of the small numbers (3%, n=13) multivariate analyses were not appropriate. For the males who persisted in driving after using cannabis (14%, n=68) a wide range of variables were significant at the univariate stage. The multivariate analysis showed that the most important factors were dependence on cannabis at 21, at least one traffic conviction before 21, a non traffic conviction before 18, and low constraint at 18. CONCLUSION: These results show different characteristics were associated with persistence in each of these outcome behaviours. This indicates that different approaches would be required if intervention programmes were to be developed to target these behaviours.


The incidence of alcohol and drugs in fatally injured drivers were determined in three Australian states, Victoria (VIC), New South Wales (NSW) and Western Australia (WA) for the period of 1990-1999. A total of 3398 driver fatalities were investigated which included 2609 car drivers, 650 motorcyclists and 139 truck drivers. Alcohol at or over 0.05g/100ml (%) was present in 29.1% of all drivers. The highest prevalence was in car drivers (30.3%) and the lowest in truckers (8.6%). WA had the highest rate of alcohol presence of the three states (35.8%). Almost 10% of the cases involved both alcohol and drugs. Drugs (other than alcohol) were present in 26.7% of cases and psychotropic drugs in 23.5%. These drugs comprised cannabis (13.5%), opioids (4.9%), stimulants (4.1%), benzodiazepines (4.1%) and other psychotropic drugs (2.7%). 8.5% of all drivers tested positive for Delta(9)-tetrahydrocannabinol (THC) and the balance of cannabis positive drivers were positive to only the 11-nor-Delta(9)-tetrahydrocannabinol-9-carboxylic acid (carboxy-THC) metabolite. The range of THC blood concentrations in drivers was 0.1-228ng/ml, with a median of 9ng/ml. Opioids consisted mainly of morphine (n=84), codeine (n=89) and methadone (n=33), while stimulants consisted mainly of methamphetamine (n=51), MDMA (n=6), cocaine (n=5), and the ephedrines (n=61). The prevalence of drugs increased over the decade, particularly cannabis and opioids, while alcohol decreased. Cannabis had a larger prevalence in motorcyclists (22.2%), whereas stimulants had a much larger presence in truckers (23%).


A simple method for the simultaneous determination of many drugs of abuse in saliva is referred [methadone, 2-ethyl-1,5-dimethyl-3,3-diphenylpyrrolinium perchlorate (EDDP), cocaine, cocaethylene, amphetamine, methamphetamine, 3,4-methylenedioxyamphetamine (MDMA), 3,4-methylenedioxyethylamphetamine (MDEA), N-methyl-1-(3,4-methylenedioxyphenyl)-2-butynamine (MBDB), cannabidiol (CBD), Delta(9)-tetrahydrocannabinol (THC), cannabinol (CBN)].Head space-solid phase microextraction (HS-SPME) and direct immersion-solid phase microextraction (DI-SPME) followed by gas chromatographic/mass spectrometric analyses (GC/MS) were employed, and results obtained with both techniques are discussed. The method was validated testing reproducibility, sensitivity, linearity.

There has been increased recognition of the clinical treatment needs of patients with co-occurring mental and substance use disorders and the heterogeneity of this group with regard to types of substances used and mental disorders. This article examines differences between men and women diagnosed with mood or psychotic disorders at admission to residential drug treatment, specifically regarding their addiction history, treatment history, perceived service needs, and psychosocial functioning. Males initiated drug use at a younger age and had higher levels of dependence on alcohol, cannabis, and opioids. There were no differences among groups in treatment history, motivation, or initiation. Males had higher rates of being under legal supervision and engaging in property crime, whereas females had higher rates of prostitution. Females had greater needs for family- and trauma-related services, and females with psychotic disorders had the highest needs for basic services. There were no differences among groups in barriers to treatment, quality of life, self-efficacy, or family support. Individuals with psychotic disorders had more symptoms of psychological distress; females had higher rates of posttraumatic stress disorder. Differences among dually-diagnosed individuals related to gender and diagnosis need to be considered in treatment planning and in matching services to patient needs.


In December 2002, the author conducted a comprehensive review of indicators of use of illicit substances in the San Francisco Bay Area. Cocaine use prevalence appears to be rising again, after a significant decline in the late 1990s. The shift away from smoking crack and toward snorting powder cocaine persists. The former predominance of Blacks among users continues to ebb. Heroin use indicators consistently show a peak in 1999, followed by a significant decline. The average age of users keeps increasing. Local street prices of heroin have risen considerably since 2001. Marijuana indicators suggest a continued increase in prevalence. Methamphetamine indicators are mixed. Usage is still widespread, and risky injection practices among gay/bisexual men remain a major factor for HIV incidence. Incidence of new HIV infection declined between 1997 and 2001 for heterosexual drug injectors, but increased for gay male and transsexual injectors.


The objective of this study was to investigate relationships between adolescent cannabis use and indices of parent-child attachment, family functioning and parent attitudes to drugs and delinquency. A total of 2848 year 9 and 2363 year 11 students participated in the Victorian Adolescent Health and Well-Being Survey (1999). The study was a school-based random sample of 535 metropolitan and rural, government and non-government secondary schools throughout Victoria, Australia. Cannabis use was defined as 'any' and 'weekly' use in the last 30 days. Multivariate logistic regression was used to identify independent associations between cannabis use and parent-child attachment, family functioning and parent attitudes to drugs and delinquency. Cannabis use in year 9 was associated with permissive parent attitudes to drugs and delinquency (any use: adjusted odds ratio (OR) = 8.1; weekly use: adjusted OR = 7.6), and was particularly sensitive to small changes in the quality of the parent-child relationship with risk increasing threefold for those describing their attachment as 'good' compared with 'very good' (any use: adjusted OR = 2.8, weekly use adjusted OR = 2.9). A similar, but more moderate pattern association was evident in year 11. After adjusting for other family and background factors, poor family functioning showed minimal association with level of cannabis use at both year levels. Results suggest that intervention efforts might sensibly target strengthening parent-child relationships and promoting less permissive parent attitudes to drug use.


A 10-item self-report measure of social self-control was examined for its association with substance use, controlling for its associations with 12 personality disorder indices and 4 demographic variables among a sample of 1050 high-risk youth. Social self-control was found to
be associated with 30-day cigarette smoking, alcohol use, marijuana use, and hard drug use, controlling for these other variables. The most consistent concurrent predictors of substance use were male gender, antisocial personality disorder, and social self-control. These results highlight the importance of social self-control as a unique concurrent predictor of substance use and suggest that social self-control skill training is relevant in substance abuse prevention programming.

This paper uses a unique dataset on the inhabitants of Amsterdam, to study the dynamics of the consumption of cannabis and cocaine. People are most likely to start using that drugs at ages 18-20 and 20-25. An analysis of the starting rates shows some evidence of cannabis being a "stepping-stone" for cocaine. However, the fact that some individuals use both cannabis and cocaine has to do mostly with correlation through (unobserved) personal characteristics and not with cannabis causing the use of cocaine.

The aim of this study was to investigate the prevalence of substance use and substance use disorders (SUDs) among incarcerated boys, and comorbidity patterns and the relationship between SUDs and violent offending and criminal recidivism. The presence of SUDs and other psychiatric disorders was assessed in a representative sample of 204 incarcerated boys aged 12 to 18 years using the Diagnostic Interview Schedule for Children (DISC). Ninety-two percent had used alcohol, 86% had used cannabis, and 33% had used other substances. The 6-month prevalence of SUDs was 55%, and 22% reported polysubstance abuse or dependence. SUDs were positively associated with comorbid externalizing and psychotic disorders. Substance dependence was negatively associated with violent offending but not with criminal recidivism. These high prevalence rates call for more attention to diagnosis and management of SUDs among incarcerated male adolescents. The negative association between substance dependence and the violent nature of the index offense needs further investigation.

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