INTRODUCTION

Here is the latest summary of research abstracts. The IACM conference scheduled for September 2004 in Oxford UK has been cancelled.

BASIC SCIENCE


Cannabinoid CB1 receptors in the brain are expressed on axon terminals presynaptic to neurons that express fatty acid amide hydrolase (FAAH). Postsynaptic FAAH catabolizes endocannabinoids which act as short-range transmitters. It has been previously shown that FAAH is also expressed in the epithelial cells of the choroid plexus. Using immunohistochemistry, we found that CB1 receptor protein is also expressed in choroid plexus epithelia. This is consistent with the hypothesis that FAAH in choroid plexus epithelial cells catabolizes endocannabinoids close to their site of action. Cannabinoids may then act directly on choroid plexus cells, and thereby contribute to the regulation of the composition of the CSF.


Previous studies have indicated that cannabinoids inhibit presynaptic neurotransmitter release in brain through CB(1) receptors. To examine this issue in a primary neuronal culture system, rat cerebellar granule cells (CGCs) were prepared. [(35)S]GTPgammaS binding assays in saponin-permeabilized CGCs showed that G-protein activation by the CB(1) agonist, WIN55212-2, and adenosine A1 agonist, phenylisopropyladenosine, was maximal during the second week in culture. ?(9)-tetrahydrocannabinol stimulated [(35)S]GTPgammaS binding to a lesser degree than WIN55212-2, and the antagonists SR141716A and AM281 acted as inverse agonists in intact CGCs, but not in CGC membrane preparations. Ten micromolar WIN55212-2 and ?(9)-tetrahydrocannabinol decreased depolarization-evoked efflux of [(3)H]-d-aspartate from CGCs by 32% and 13%, respectively. SR141716A and AM281 increased [(3)H]-d-aspartate release by 28%. The fatty acid amide hydrolase (FAAH) inhibitor phenylmethylsulfonyl fluoride (PMSF) and the anandamide uptake inhibitor AM404 inhibited transmitter release, implying that the antagonist effects were mediated by blockade of endocannabinoid activity. Levels of endocannabinoids (both anandamide and 2-arachidonyl glycerol [2-AG]) in extracts of the cells and cell incubation buffer were increased by PMSF pre-treatment. Depolarization with KCl significantly decreased the amount of anandamide and 2-AG in PMSF-treated CGCs. These results suggest that endogenous cannabinoids inhibit neurotransmitter release in CGCs, which may also release endocannabinoids upon neural stimulation.

At many central synapses, endocannabinoids released by postsynaptic cells inhibit neurotransmitter release by activating presynaptic cannabinoid receptors. The mechanisms underlying this important means of synaptic regulation are not fully understood. It has been shown at several synapses that endocannabinoids inhibit neurotransmitter release by reducing calcium influx into presynaptic terminals. One hypothesis maintains that endocannabinoids indirectly reduce calcium influx by modulating potassium channels and narrowing the presynaptic action potential. An alternative hypothesis is that endocannabinoids directly and selectively inhibit N-type calcium channels in presynaptic terminals. Here we test these hypotheses at the granule cell to Purkinje cell synapse in cerebellar brain slices. By monitoring optically the presynaptic calcium influx (Ca(influx)) and measuring the EPSC amplitudes, we found that cannabinoid-mediated inhibition arises solely from reduced presynaptic Ca(influx). Next we found that cannabinoid receptor activation does not affect the time course of presynaptic calcium entry, indicating that the reduced Ca(influx) reflects inhibition of presynaptic calcium channels. Finally, we assessed the classes of presynaptic calcium channels inhibited by cannabinoid receptor activation via peptide calcium channel antagonists. Previous studies established that N-type, P/Q-type, and R-type calcium channels are all present in granule cell presynaptic boutons. We found that cannabinoid activation reduced Ca(influx) through N-type, P/Q-type, and R-type calcium channels to 29, 60, and 55% of control, respectively. Thus, rather than narrowing the presynaptic action potential or exclusively modulating N-type calcium channels, CB1 receptor activation inhibits synaptic transmission by modulating all classes of calcium channels present in the presynaptic terminal of the granule cell to Purkinje cell synapse.


The cannabinoid CB(1) receptor antagonist, SR 141716 (Rimonabant), has been reported to stimulate, when acutely administered, intestinal motility in mice. The present study was aimed at determining whether tolerance develops to its repeated administration. Mice were treated twice a day for up to 8 consecutive days with 0, 3 and 5.6 mg/kg SR 141716 (i.p.). On days 1, 3, 5 and 8, separate groups of mice were treated intragastrically with a non-absorbable colored marker (carmine). The distance traveled by the head of the marker in the small intestine was recorded. On day 1, SR 141716 markedly activated intestinal peristalsis, but complete tolerance to this effect developed within the third day of treatment. The results may have some relevance to the proposed future clinical use of SR 141716.


Anandamide (N-arachidonoyl ethanolamine, AEA), an endogenous cannabinoid receptor agonist, causes potent vasodilation in the cerebral circulation through an endothelial-dependent or -independent mechanism. We have investigated the processing of [3H]AEA in cultured mouse cerebral microvascular endothelial cells (MEC) in order to better understand its mechanism of action in the cerebral vasculature. These cells took up anandamide very quickly, reaching a maximum value in 5 min and remaining at that level for at least 8 h. Analysis of the cell lipids demonstrated that, in addition to free anandamide, radioactivity was incorporated into phosphatidylcholine (PC), phosphatidylinositol (PI), and phosphatidylethanolamine (PE) in a time-dependent manner. Analysis of the hydrolyzed cell lipids indicated that anandamide was converted to arachidonic acid, a process that was inhibited by the selective fatty acid amide hydrolase inhibitor oleyl trifluoromethyl ketone (OTMK). Phospholipase A2 (PLA2) hydrolysis of the PC, PI, and PE fractions indicated that the arachidonic acid formed from anandamide was esterified predominately into sn-2 position of the endothelial phospholipids. Furthermore, anandamide and arachidonic acid were released when the cells were incubated with A23187. These results suggest that the biological activity of anandamide might be regulated by its rapid uptake and calcium-dependent release in endothelial cells, and conversion of anandamide to arachidonic acid might serve as an inactivation process in the cerebral microcirculation.

Abstract DREAM (downstream regulatory element antagonistic modulator) is a novel transcriptional repressor for the prodynorphin gene, and genetic deletion of DREAM in mice results in a phenotype of ongoing analgesia by virtue of its effect on opioid gene expression. In the present study, we evaluated the motivational effects of opioids (morphine), cannabinoids [Delta(9)-tetrahydrocannabinol (THC)] and cocaine in mice lacking the dream gene (dream(-/-)). The aversive effects of THC were potentiated in dream(-/-) mice in a kappa-opioid receptor-dependent fashion, whereas morphine reward and the aversive effects of morphine withdrawal remained intact. The rewarding and aversive effects of cocaine were likewise unperturbed in dream(-/-) mice. Moreover, the aversive properties of lithium chloride and naloxone were unaffected by the absence of DREAM, indicating that the effect of DREAM on THC-induced dysphoria is not due to a general involvement in the behavioral response to aversive stimuli. Additionally, physical dependence to morphine and the locomotor-sensitizing effects of cocaine were unaltered in these animals. Finally, whereas the absence of DREAM reduced the analgesic efficacy of THC, morphine analgesia was unaffected in dream(-/-) mice.


Administration of morphine and cannabinoids stimulates alcohol intake in rats. The present study investigated whether the promoting effect of morphine and of the cannabinoid receptor agonist, WIN 55,212-2 [(R)-(+)-(2,3-dihydro-5-methyl-3-(4-morpholinylmethyl)pyrrolo[1,2,3-de]-1, 4-benzoxazin-6-yl]-1-naphthalenylmethanone], on alcohol intake was prevented by the gamma-aminobutyric (GABA)(B) receptor agonist, baclofen. Sardinian alcohol-preferring (sP) rats were given alcohol (10%, v/v) and water under the standard homecage two-bottle-free choice regimen with unlimited access for 24 h/day. Baclofen (0, 0.5 and 1 mg/kg; i.p.) was administered acutely 30 min before lights off. Morphine (0 and 1 mg/kg, s.c.) or WIN 55,212-2 (0 and 2 mg/kg, i.p.) was administered acutely 10 min after baclofen. Alcohol intake was recorded 60 min after lights off. As predicted, both morphine and WIN 55,212-2 produced a specific and marked increase in alcohol intake. Pretreatment with baclofen, which failed to alter alcohol intake when given alone, dose-dependently suppressed morphine- and WIN 55,212-2-induced promotion of alcohol drinking. These results suggest the involvement of the GABA(B) receptor in the neural circuitry mediating the stimulating effect of morphine and cannabinoids on alcohol consumption in sP rats.


Our study addressed the hypothesis that spinal release of endogenous opioids underlies Delta(9)-tetrahydrocannabinol (Delta(9)-THC)-induced antinociception in Freund's adjuvant-induced arthritic and nonarthritic rats. The paw-pressure test was used to assess the antinociceptive effects of Delta(9)-THC versus those of morphine, and opioid and cannabinoid receptor-selective antagonists were used to characterize the involved receptors. Cerebrospinal fluid was collected after Delta(9)-THC injection (i.p.) for the measurement of endogenous opioid peptides. Our results indicate that morphine or Delta(9)-THC is equally potent and efficacious in both nonarthritic and arthritic rats. Delta(9)-THC-induced antinociception is attenuated by the kappa opioid receptor antagonist, nor-binaltorphimine, in arthritic rats only. Delta(9)-THC induces increased immunoreactive dynorphin A (idyn A) levels in nonarthritic rats while decreasing idyn A in arthritic rats. We hypothesize that the elevated idyn A level in arthritic rats contributes to hyperalgesia by interaction with N-methyl-d-aspartate receptors, and that Delta(9)-THC induces antinociception by decreasing idyn A release.

The present study investigated the effects of different classes of cannabinoid receptor ligands on sensory neurotransmission in the rat isolated mesenteric arterial bed. Electrical field stimulation of the mesenteric bed evoked frequency-dependent vasorelaxation due to the activation of capsaicin-sensitive sensory nerves and release of calcitonin gene-related peptide (CGRP). The CB1/CB2 cannabinoid agonists WIN55,212 and CP55,940 (0.01-1 micro M) attenuated sensory neurogenic relaxation in a concentration-dependent manner. At 0.1 micro M, WIN55,212 and CP55,940 were largely ineffective in the presence of the CB1 antagonists SR141716A and LY320135 (1 micro M) but their inhibitory actions remained in the presence of the CB2 selective antagonist SR144528 (1 micro M). The CB1/CB2 agonist THC (1 micro M) attenuated sensory neurogenic relaxations, as did the CB2 agonist JWH-015. The inhibitory actions of both THC and JWH-015 were still evident in the presence of SR141716A (1 micro M) and SR144528 (1 micro M). None of the cannabinoid agonists investigated had an effect on vasorelaxation elicited by exogenous CGRP indicating a prejunctional mechanism. These data demonstrate that different classes of cannabinoid agonists attenuate sensory neurotransmission via a prejunctional site and provide evidence for mediation by a CB1 and/or a non CB1/CB2 receptor.


Brain cannabinoid CB(1) receptors are expressed in neural areas that contribute to movement such as basal ganglia, where they co-localize with dopamine D(1) and D(2) receptors. The objective of the present study was to further study the functional role of CB(1) receptors along with D(1) and D(2) dopamine receptors of basal ganglia by local injections of SR141716A (CB(1) receptor antagonist), SKF-38393 (D(1) agonist), and quinpirole (D(2) agonist), in a rat Parkinson's model. Turning response after amphetamine was considered as the parkinsonian variable for quantifying motor effects of drugs. The findings indicated that, after intrastratal infusions, both D(1) or D(2) dopamine receptor agonists alone reduced turning in parkinsonian rats. At the pallidal and subthalamic levels, D(1) (not D(2)) receptor stimulation also reduced rotation. Regarding SR141716A-induced effects, CB(1) antagonism reduced motor asymmetry in parkinsonian rats after injections into striatum, globus pallidus, and to a lesser extent, subthalamic nucleus. At the level of dorsal striatum, effects of SR141716A were mediated through an opposite modulation of D(1) and D(2) dopamine receptor function. At the pallidal and subthalamic nucleus levels, motor effects after SR14716A are not associated to modulation of D(1) and D(2) receptor function.


Depolarization-induced suppression of inhibition (DSI) is a form of retrograde signaling at GABAergic synapses that is initiated by the calcium- and depolarization-dependent release of endocannabinoids from postsynaptic neurons. In the neocortex, pyramidal neurons (PNs) appear to use DSI as a mechanism for regulating somatic inhibition from a subpopulation of GABAergic inputs that express the type 1 cannabinoid receptor. Although postsynaptic control of afferent inhibition may directly influence the integrative properties of neocortical PNs, little is known regarding the patterns of activity that evoke endocannabinoid release and the impact such disinhibition may have on the excitability of PNs. Here we provide the first systematic survey of AP-induced DSI in the neocortex. The magnitude and time course of DSI was directly related to the number and frequency of postsynaptic APs and was enhanced in the presence of the cholinergic receptor agonist carbachol. This AP-induced DSI was mediated by endocannabinoids, as it was prevented by the cannabinoid receptor antagonist AM251 and potentiated by the endocannabinoid transport inhibitor AM404. We also explored the consequences of neocortical DSI on PN excitability by examining the responsiveness of PNs to evoked synaptic stimulation. We found that endocannabinoid-mediated DSI markedly increased PN responsiveness to excitatory synaptic inputs and promoted AP discharge with a time course that paralleled DSI expression. Taken together, our data suggest a role for endocannabinoids in regulating the output of cortical PNs.

RATIONALE. Animals trained to lick for a sucrose solution of a given incentive value that subsequently encounter an incentive downshift (i.e., 32-4% sucrose) display an exaggerated decrease in the amount consumed, relative to unshifted controls. This change has been classified as a successive negative contrast (SNC) effect. The emotional component to this robust behavioural change is dynamic and changes from post-shift day (PSD) 1 to 2. Anxiolytics block SNC, but the possible link between anxiety and SNC needs further exploration. Both nicotine and a cannabinoid receptor agonist have been reported to change anxiety and both have actions on the reward process, but their effects on SNC have not been investigated. OBJECTIVES. To determine: (1) whether exposure to SNC evokes an anxiogenic response; (2) whether an anxiolytic dose of nicotine has the same effects on SNC as those of chlordiazepoxide; (3) the effects of a low (anxiolytic) and a high (anxiogenic) dose of the cannabinoid receptor agonist CP 55,940 on SNC. METHODS. Two groups of animals were given access to high (32%) or low (4%) sucrose solutions for 5 min per day for 10 days. On PSD 1 and 2, the shifted group had access to a devalued incentive (from 32 to 4% sucrose) and the unshifted group remained at 4% sucrose. The volumes (ml) of sucrose solution consumed were measured pre-shift and on PSD 1 and 2. In experiment 1, immediately after SNC testing on PSD 1 and 2, the rats were tested in the social interaction and elevated plus-maze tests of anxiety. In experiment 2, the effects of chlordiazepoxide (5 and 7.5 mg/kg) and nicotine (0.1 mg/kg) were examined on PSD 1 and 2. In experiment 3, the effects of CP 55,940 (5 and 40 microg/kg) were examined on PSD 1 and 2. RESULTS. There were no anxiogenic effects of shift in either test of anxiety on either test day. However, on PSD 1, the shifted group had significantly higher locomotor activity and spent a higher percentage of time on the open arms, perhaps reflecting search strategies. Nicotine was without significant effect on SNC on either test day. On PSD 1, chlordiazepoxide (5 mg/kg) and CP 55,940 (5 and 40 microg/kg, IP) blocked SNC. On PSD 2, both doses of chlordiazepoxide and the low, anxiolytic dose of CP 55,940 (5 microg/kg) blocked SNC, the high dose of CP 55,940 was without effect. CONCLUSIONS. The pattern of results allows for the separation between effects on anxiety and SNC. The block of contrast on PSD 1 was independent of changes in anxiety, since both anxiolytic and anxiogenic drug doses were effective. It is suggested that this may provide an animal model of disappointment in which the cannabinoid system plays an important role. An anxiolytic action would seem to be a necessary, but not a sufficient, action to block SNC on PSD 2.


Recent studies implicate dendritic endocannabinoid release from subsynaptic dendrites and subsequent inhibition of neurotransmitter release from nerve terminals as a means of retrograde signaling in multiple brain regions. Here we show that type 1 cannabinoid receptor-mediated endocannabinoid signaling is not involved in the retrograde control of synaptic efficacy at inhibitory synapses between fast-spiking interneurons and pyramidal cells in layer 2/3 of the neocortex. Vesicular neurotransmitter transporters, such as vesicular glutamate transporters (VGLUTs) 1 and 2, are localized to presynaptic terminals and accumulate neurotransmitters into synaptic vesicles. A third subtype of VGLUTs (VGLUT3) was recently identified and found localized to dendrites of various cell types. We demonstrate, using multiple immunofluorescence labeling and confocal laser-scanning microscopy, that VGLUT3-like immunoreactivity is present in dendrites of layer 2/3 pyramidal neurons in the rat neocortex. Electron microscopy analysis confirmed that VGLUT3-like labeling is localized to vesicular structures, which show a tendency to accumulate in close proximity to postsynaptic specializations in dendritic shafts of pyramidal cells. Dual whole-cell recordings revealed that retrograde signaling between fast-spiking interneurons and pyramidal cells was enhanced under conditions of maximal efficacy of VGLUT3-mediated glutamate uptake, whereas it was reduced when glutamate uptake was inhibited by incrementing concentrations of the nonselective VGLUT inhibitor Evans blue (0.5-5.0 microm) or intracellular Cl- concentrations (4-145 mm). Our results present further evidence that dendritic
vesicular glutamate release, controlled by novel VGLUT isoforms, provides fast negative feedback at inhibitory neocortical synapses, and demonstrate that glutamate can act as a retrograde messenger in the CNS.


A (1)H-NMR method has been developed for the quantitative analysis of pure cannabinoids and for cannabinoids present in Cannabis sativa plant material without any chromatographic purification. The experiment was performed by the analysis of singlets in the range of delta 4.0-7.0 in the (1)H-NMR spectrum, in which distinguishable signals of each cannabinoid are shown. Quantitation was performed by calculating the relative ratio of the peak area of selected proton signals of the target compounds to the known amount of the internal standard, anthracene. For this method no reference compounds are needed. It allows rapid and simple quantitation of cannabinoids with a final analysis time of only 5 min without the need for a pre-purification step.


The rDNA intergenic spacer (IGS) structure of Cannabis sativa was established and can be used for classification and identification of this species. In this study, DNA fragments of rDNA IGS were amplified by PCR from Cannabis sativa plant extracts and a 1387 bp fragment was obtained. DNA sequence analysis revealed six different repeat motifs. In the middle of the IGS sequence, there were three sequence motifs, and the same three sections of DNA were then repeated with minor variation in sequence. The terminal region of the IGS was composed of another three different repeat units; multiple copies of these terminal repeat motifs were present in no discernible order. Within six repeat motifs, point variations were observed in five. The DNA sequence of the locus was compared with all the plant sequences registered in GenBank by the Fasta program of GCG software with the result that this DNA fragment was significantly different from any other DNA sequence recorded to date. The most similar sequence was that of Hops (Humulus lupulus), but with a similarity of only 88.9% over 579 bp. These specific and complex variations of IGS may be related to the species and geographic distributions.


OBJECTIVE. To examine the discriminative stimulus effects of (i) the cannabinoid CB(1) receptor antagonist SR-141716 (SR, 5.6 mg/kg) and vehicle, and (ii) the cannabinoid receptor agonist Delta(9)-THC (THC, 1.8 mg/kg) and vehicle using a discriminated taste aversion (DTA) procedure. METHODS. Two groups of rats (n=6) were trained to discriminate between these drugs and vehicle in DTA (t=20 min). The 30-min drinking bout of tap water following drug (SR or THC) treatment was followed by an injection of lithium chloride (LiCl, 120 mg/kg) in the experimental animals. When offered water after vehicle pretreatment, experimental animals subsequently were given i.p. saline (NaCl, 10 ml/kg). Post-drinking treatment for controls (n=6) was NaCl, irrespective of the pretreatment condition (SR, THC or vehicle). Additional water was provided during the afternoon (30 min) with no other manipulations. Food was available ad lib at all times. When the discriminations were established other doses and drugs were examined (t=20 min). In testing there were no post-drinking treatments. RESULTS. The SR-related analog AM-251 (dose range: 1-5.6 mg/kg) substituted for SR, whereas other drugs such as the cannabinoid CB(2) receptor antagonist SR-144528 (3 and 10 mg/kg), THC (1-10 mg/kg), flumazenil (1-10 mg/kg), naloxone (1-10 mg/kg), morphine (10 and 18 mg/kg) and d-amphetamine (1 and 3 mg/kg) did not. There was a dose-related attenuation of SR-induced suppression of drinking when THC (1.8-10 mg/kg) was given together with SR (5.6 mg/kg). In the THC trained rats, SR (1-10 mg/kg), morphine (10 and 18 mg/kg) and d-amphetamine (1 and 3 mg/kg) did not substitute for THC. SR (1 mg/kg) attenuated the THC (1.8 mg/kg) induced suppression of drinking. Together with 3 mg/kg SR and 1.8 mg/kg THC, drinking was roughly equally suppressed in both the experimental group and the controls. CONCLUSION. SR-141716
induces a discriminative stimulus complex in DTA that shows potential for further examination of cannabinoid receptor antagonism.


In hippocampal pyramidal cells, a rise in Ca(2+) releases endocannabinoids that activate the presynaptic cannabinoid receptor (CB1R) and transiently reduce GABAergic transmission-a process called depolarization-induced suppression of inhibition (DSI). The mechanism that limits the duration of endocannabinoid action in intact cells is unknown. Here we show that inhibition of cyclooxygenase-2 (COX-2), not fatty acid amide hydrolase (FAAH), prolongs DSI, suggesting that COX-2 limits endocannabinoid action.


Cannabinoids have been shown to affect various immune functions. To date, almost no data exist on PMN, which provide the first line antimicrobial defense. The objective of the present study was to investigate the effects of the synthetic dibenzopyrane ligand CP55 940, the endogenous cannabinoid anandamide and methanandamide on the "respiratory burst" of isolated human PMN in vitro. After preincubation with high micromolar concentrations of CP55 940, fMLP-stimulated PMN showed a reduction in superoxide production, whereas the spontaneous burst activity of resting PMN remained unaffected. This inhibitory effect of CP55 940 was not CB-receptor-mediated. In contrast, anandamide and methanandamide did not alter the oxidative microbicidal PMN function.


The CB1 cannabinoid receptor (CB1R) displays a significant level of ligand-independent (i.e. constitutive) activity, either when heterologously expressed in non-neuronal cells or in neurons where CB1Rs are endogenous. The present study investigates the consequences of constitutive activity on the intracellular trafficking of CB1R. When transfected in HEK-293 cells, CB1R is present at the plasma membrane, but a substantial proportion (approximately 85%) of receptors is localized in intracellular vesicles. Detailed analysis of CB1-EGFP expressed in HEK-293 cells shows that the intracellular CB1R population is mostly of endocytic origin and that treatment with inverse agonist AM281 traps CB1R at the plasma membrane through a monensin-sensitive recycling pathway. Co-transfection with dominant positive or dominant negative mutants of the small GTPases Rab5 and Rab4, but not Rab11, profoundly modifies the steady-state and ligand-induced intracellular distribution of CB1R, indicating that constitutive endocytosis is Rab5 dependent while constitutive recycling is mediated by Rab4. In conclusion, our results indicate that, due to its natural constitutive activity, CB1R permanently and constitutively cycles between plasma membrane and endosomes, leading to a predominantly intracellular localization at steady-state.


This study was undertaken to investigate the effect of some cannabinoid agonists on the bovine ciliary muscle. Both anandamide and CP 55,940 (cis-3-(2-hydroxy-4-(1,1-dimethyl heptyl) phenyl)-trans-4-(3-hydroxypropyl) cyclohexanol) produced a concentration-dependent contractile response in ciliary muscle. These responses were inhibited by SR 141716A (N-[piperidin-1-yl]-5-(4-cholophenyl)-1-(2,4-dichlorophenyl)-4-methyl-1H-p yrazole-3-carboxamide) (0.1 and 1 microM) but not by SR 144528 (N-[1S)-endo-1,3,3-trimethyl bicyclo[2.2.1] heptan-2-yl] 5-(4-chloro-3-methylphenyl)-1-(4 methoxy benzyl)-pyrazole-3-carboxamide) (1 and 10 microM). A preincubation with G(i/o) protein inhibitor pertussis toxin (500 ng/ml) for 20 min inhibited the contractile action of anandamide and CP 55,940. In addition, the phospholipase C inhibitor U73122 (1(6)-[(17beta)-3-methoxyestra-1,3,5(10)-tri-en-17-yl] amino] hexyl]-1H-pyrrole-2,5-dione) blocked the anandamide-
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and CP 55,940-induced contractions, whereas the protein kinase C activator, phorbol 12,13 dibutyrate (PDBu) significantly potentiated the contractions evoked by cannabinoid receptor agonists. We evaluated the binding of [(3)H]CP 55,940, which specifically labelled a single class of cannabinoid sites with affinity in low subnanomolar range (K(d)=0.6 nM) and the maximal number of binding sites of 1243 fmol/mg protein. Binding of [(3)H]CP 55,940 was inhibited by ligands having a major selectivity for cannabinoid (CB(1)) receptors. These findings provide strong evidence of the involvement of cannabinoid CB(1) receptors promoting contraction in the bovine ciliary muscle. Furthermore, the action of cannabinoid receptor agonists appears to be mediated via phospholipase C. These data also contribute to elucidate the cannabinoid CB(1) receptor pivotal role in the modulation of intraocular pressure and to show that cannabinoid receptor agonists may be regarded as potential antiglaucoma agents.


The influence of the endocannabinoid system on the sensitisation to the rewarding effects of morphine in the place conditioning paradigm was evaluated. In mice pretreated with morphine this drug induces place preference with lower doses. Pretreatment with non-rewarding doses of the cannabinoid agonist WIN 55,212-2 (0.5 and 1 mg/kg) induces sensitisation to the rewarding effects of morphine. However, the pretreatment with the cannabinoid antagonist SR 144716A plus morphine or WIN 55,212-2 blocks it. Our results suggest the existence of cross-talk between cannabinoids and opiates on the sensitisation to morphine and the implication of the endocannabinoid system in the process of sensitisation to opiates.


A rapid and sensitive method for the simultaneous confirmatory analysis of three forensic most relevant cannabinoids, Delta(9)-tetrahydrocannabinol (THC), 11-hydroxy-Delta(9)-tetrahydrocannabinol (11-OH-THC) and 11-nor-9-carboxy-Delta(9)-tetrahydrocannabinol (THC-COOH), by means of high-performance liquid chromatography/tandem mass spectrometry (LC/MS/MS) in human plasma was developed and fully validated. Sample clean-up was performed by automated silica-based solid-phase extraction and the separation was carried out using a PhenylHexyl column (50 x 2 mm i.d., 3 micro m) and acetonitrile-5 mM ammonium acetate gradient elution. Data were acquired with an API 3000 LC/MS/MS system equipped with a turboionspray interface and triple quadrupole mass analyzer using positive electrospray ionization and multiple reaction monitoring. Two MS/MS transitions for each substance were monitored and deuterated analogues of analytes were used as internal standards for quantitation. The limit of quantitation was 0.8 ng ml(-1) for THC, 0.8 ng ml(-1) for 11-OH-THC and 4.3 ng ml(-1) for THC-COOH and linearity with a correlation coefficient r(2) = 0.999 was achieved up to 100 ng ml(-1) for THC and 11-OH-THC and 500 ng ml(-1) for THC-COOH. The limits of detection were 0.2 ng ml(-1) for THC, 0.2 ng ml(-1) for 11-OH-THC and 1.6 ng ml(-1) for THC-COOH. The developed LC/MS/MS method was also successfully used for the determination of THC-COOH-glucuronide, the phase II metabolite of THC-COOH.


Considerable plasticity exists in the endogenous cannabinoid system, as evidenced by the high degree of tolerance that develops following repetitive exposure to exogenously administered cannabinoid receptor agonists. This tolerance development is accompanied by cannabinoid CB(1) receptor downregulation and attenuation of G-protein activation. The biological processes responsible for CB(1) receptor downregulation remain to be fully understood. However, recent evidence suggests that several protein kinases participate in the development of cannabinoid tolerance. These observations implicate a role for protein kinases in cannabinoid signaling pathways. It remains to be established whether these protein kinases are directly
involved in CB(1) receptor regulation or whether they contribute to tolerance by modulating additional signaling pathways.


Oleamide is a lipid with diverse properties, including cannabinoid-like activity. For example, it induces the classic triad of effects attributable to these molecules: decrease in core temperature, hypolocomotion, and reduction in pain perception. However, as it binds to the cannabinoid receptors (CB1) only at high concentrations, it is not considered an actual endocannabinoid. In this study, we tested the effect of oleamide on food intake and sexual behavior and compared it to the effect induced by anandamide. Results indicate that oleamide and anandamide increased food intake during the 3h post-injection. In addition, anandamide but not oleamide induced changes in sexual performance. This study further supports the role of endocannabinoids in food ingestion and male sexual behavior and gives additional support to the notion that, although oleamide might not be an endocannabinoid, it shares some effects with them.


The cellular inactivation of the endogenous cannabinoid (endocannabinoid) anandamide (AEA) represents a controversial and intensely investigated subject. This process has been proposed to involve two proteins, a transporter that promotes the cellular uptake of AEA and fatty acid amide hydrolase (FAAH), which hydrolyzes AEA to arachidonic acid. However, whereas the role of FAAH in AEA metabolism is well-characterized, the identity of the putative AEA transporter remains enigmatic. Indeed, the indirect pharmacological evidence used to support the existence of an AEA transporter has been suggested also to be compatible with a model in which AEA uptake is driven by simple diffusion coupled to FAAH metabolism. Here, we have directly addressed the contribution of FAAH to AEA uptake by examining this process in neuronal preparations from FAAH(-/-) mice and in the presence of the uptake inhibitor UCM707. The results of these studies reveal that (i) care should be taken to avoid the presence of artifacts when studying the cellular uptake of lipophilic molecules like AEA, (ii) FAAH significantly contributes to AEA uptake, especially with longer incubation times, and (iii) a UCM707-sensitive protein(s) distinct from FAAH also participates in AEA uptake. Interestingly, the FAAH-independent component of AEA transport was significantly reduced by pretreatment of neurons with the cannabinoid receptor 1 (CB1) antagonist SR141716A. Collectively, these results indicate that the protein-dependent uptake of AEA is largely mediated by known constituents of the endocannabinoid system (FAAH and the CB1 receptor), although a partial contribution of an additional UCM707-sensitive protein is also suggested.


AIM: To specify the functional activity of cannabinoid CB(1) receptor in alcohol-preferring Fawn Hooded and alcohol nonpreferring Wistar rats under naive conditions. METHOD: Cannabinoid CB(1) (WIN-55,212)-stimulated [(35)S]-GTPgammast binding autoradiography, and cannabinoid CB(1) receptor gene expression were measured in rats of both strains that received only water. RESULTS: Cannabinoid CB(1) receptor stimulated [(35)S]-GTPgammast binding was significantly lower in cingulate cortex (Cg), caudate-putamen (CPu), nucleus accumbens (Acc), ventromedial hypothalamic nucleus (VMN), amygdaloid area (AMG), fields (CA1, CA3) of the hippocampus and dentate gyrus (DG) in Fawn Hooded than in Wistar rats, whereas no differences were found either in substantia nigra pars reticulata (SNr) nor CA2 field of the hippocampus. In addition, cannabinoid CB(1) receptor gene expression was lower in Cg, CPu, VMN and CA3 field of the hippocampus in Fawn Hooded than in Wistar rats. CONCLUSIONS: We speculate that lower cannabinoid function appears to be related to greater vulnerability to
alcohol consumption. Cannabinoid CB(1) receptor may represent a key target in the treatment of alcohol dependence.

Peroni, R. N., M. L. Orliac, et al. (2004). "Sex-linked differences in the vasorelaxant effects of anandamide in vascular mesenteric beds: role of oestrogens." Eur J Pharmacol 493(1-3): 151-60. Anandamide (0.01 to 10 microM) caused greater concentration-dependent reductions of the contractile-induced responses to noradrenaline in female than in male mesenteric vascular beds isolated from adult Sprague-Dawley rats. Greater relaxant responses in females were also induced by the vanilloid TRPV1 receptor agonist capsaicin (0.01 to 10 microM), whereas no sex differences were observed for the relaxations caused by either acetylcholine or sodium nitroprusside. The effect of anandamide in either sex was reduced by the vanilloid TRPV1 receptor antagonist capsazepine but not by the cannabinoid CB(1) receptor antagonist N-piperidino-5-(4-chlorophenyl)-1-(2,4-dichlorophenyl)-4-methyl-3-pyrazole carboxamide (SR141716A). In males, the anandamide-induced relaxations were potentiated by in vitro exposure during 5 min to 0.5 microM 17beta-oestradiol and unmodified by the protein synthesis inhibitor cycloheximide. The vasorelaxant effects of anandamide in female rats were decreased by ovariectomy. This decrease was prevented by in vivo treatment with 17beta-oestradiol-3-benzoate (450 microg/kg i.m., once a week during 3 weeks) and counteracted by in vitro exposure to oestrogen. In vivo treatment with 17beta-oestradiol also potentiated anandamide-induced responses in males. In conclusion, this study shows an oestrogen-dependent sensitivity to the vanilloid TRPV1 receptor-mediated vasorelaxant effects of anandamide in the mesenteric vasculature of Sprague-Dawley rats, that could be mediated by both genomic and non-genomic mechanisms.

Raharjo, T. J., I. Widjaja, et al. (2004). "Comparative Proteomics of Cannabis sativa Plant Tissues." J Biomol Tech 15(2): 97-106. Comparative proteomics of leaves, flowers, and glands of Cannabis sativa have been used to identify specific tissue-expressed proteins. These tissues have significantly different levels of cannabinoids. Cannabinoids accumulate primarily in the glands but can also be found in flowers and leaves. Proteins extracted from glands, flowers, and leaves were separated using two-dimensional gel electrophoresis. Over 800 protein spots were reproducibly resolved in the two-dimensional gels from leaves and flowers. The patterns of the gels were different and little correlation among the proteins could be observed. Some proteins that were only expressed in flowers were chosen for identification by matrix-assisted laser desorption/ionization time-of-flight mass spectrometry and peptide mass fingerprint database searching. Flower and gland proteomes were also compared, with the finding that less than half of the proteins expressed in flowers were also expressed in glands. Some selected gland protein spots were identified: F1D9.26-unknown prot. (Arabidopsis thaliana), phospholipase D beta 1 isoform 1a (Gossypium hirsutum), and PG1 (Hordeum vulgare). Western blotting was employed to identify a polyketide synthase, an enzyme believed to be involved in cannabinoid biosynthesis, resulting in detection of a single protein.

Raman, C., S. D. McAllister, et al. (2004). "Amyotrophic lateral sclerosis: delayed disease progression in mice by treatment with a cannabinoid." Amyotroph Lateral Scler Other Motor Neuron Disord 5(1): 33-9. Effective treatment for amyotrophic lateral sclerosis (ALS) remains elusive. Two of the primary hypotheses underlying motor neuron vulnerability are susceptibility to excitotoxicity and oxidative damage. There is rapidly emerging evidence that the cannabinoid receptor system has the potential to reduce both excitotoxic and oxidative cell damage. Here we report that treatment with Delta(9)-tetrahydrocannabinol (Delta(9)-THC) was effective if administered either before or after onset of signs in the ALS mouse model (hSOD(G93A) transgenic mice). Administration at the onset of tremors delayed motor impairment and prolonged survival in Delta(9)-THC treated mice when compared to vehicle controls. In addition, we present an improved method for the analysis of disease progression in the ALS mouse model. This logistic model provides an estimate of the age at which muscle endurance has declined by 50% with much greater accuracy than could be attained for any other measure of decline. In vitro, Delta(9)-THC was extremely
effective at reducing oxidative damage in spinal cord cultures. Additionally, Delta(9)-THC is anti-excitotoxic in vitro. These cellular mechanisms may underlie the presumed neuroprotective effect in ALS. As Delta(9)-THC is well tolerated, it and other cannabinoids may prove to be novel therapeutic targets for the treatment of ALS.


A novel comparison model of the human cannabinoid CB1 receptor has been constructed using the bovine rhodopsin X-ray structure as a template. The model was subjected to a 500-ps molecular dynamics simulation, and thereafter new conformers of the receptor model were produced in a simulated annealing procedure. Using an automated docking procedure, well-known cannabinoids were docked into six different model conformers, of which one was chosen for a detailed study of receptor-ligand interactions. The docking results confirm, for example, the importance of lysine K3.28(192) in the binding of these ligands. Also, other experimental data are fairly consistent with the present model, though there are some differences when compared to other recent CB1 comparison models. The present model will serve as a tool to investigate the receptor-ligand interactions and facilitate the design of novel cannabinimimetic drugs.


1. Several G-protein-coupled receptors (GPCRs) have been localized to various layers of the vertebrate retina, using autoradiographic and immunohistochemical techniques, but the functional data concerning G protein activation are limited. Here, we establish optimized assay conditions to detect receptor-dependent G protein activity in membranes and tissue sections of the rat retina. 2. Agonist-stimulated [35S]GTPgammaS-binding responses were characterized for the Gi/o-linked adenosine A1, cannabinoid CB1, m2/m4 muscarinic acetylcholine, and GABA(B) receptors. Initial assumption was that G protein activity under "basal conditions" is high due to enrichment and activity of rhodopsin and transducin in this tissue. 3. We found that pretreatment of retina membranes with hydroxylamine (10 mM), a rhodopsin-inactivating drug, substantially (up to 60%) reduced basal G protein activity, thereby improving signal-to-noise ratio to detect agonist-stimulated G protein activation for all studied receptors. [35S]GTPgammaS autoradiography revealed that hydroxylamine specifically reduced basal binding in the transducin-enriched photoreceptor layer. In contrast, hydroxylamine did not affect GPCR signaling in brain membranes, indicating specific action on retinal transducin. 4. For all studied receptors, [35S]GTPgammaS autoradiography allowed localization of G protein activity to different retinal layers, with the bulk of signal detected in the ganglion cell layer. Strongest responses were observed for adenosine and muscarinic receptor agonists. Additional G protein activity was detected in the inner plexiform layer. 5. Responses to all tested agonists were reversed in the presence of appropriate receptor-selective antagonists, indicating receptor-mediated G protein activation.


Delta(1)-Tetrahydrocannabinolic-acid (THCA) synthase is the enzyme that catalyzes oxidative cyclization of cannabigerolic-acid into THCA, the precursor of Delta(1)-tetrahydrocannabinol. We cloned a novel cDNA (Genbank(TM) accession number AB057805) encoding THCA synthase by reverse transcription and polymerase chain reactions from rapidly expanding leaves of Cannabis sativa. This gene consists of a 1635-nucleotide open reading frame, encoding a 545-amino acid polypeptide of which the first 28 amino acid residues constitute the signal peptide. The predicted molecular weight of the 517-amino acid mature polypeptide is 58,597 Da. Interestingly, the deduced amino acid sequence exhibited high homology to berberine bridge enzyme from Eschscholtzia californica, which is involved in alkaloid biosynthesis. The
liquid culture of transgenic tobacco hairy roots harboring the cDNA produced THCA upon feeding of cannabigerolic-acid, demonstrating unequivocally that this gene encodes an active THCA synthase. Overexpression of the recombinant THCA synthase was achieved using a baculovirus-insect expression system. The purified recombinant enzyme contained covalently attached FAD cofactor at a molar ratio of FAD to protein of 1:1. The mutant enzyme constructed by changing His-114 of the wild-type enzyme to Ala-114 exhibited neither absorption characteristics of flavoproteins nor THCA synthase activity. Thus we concluded that the FAD binding residue is His-114, and that the THCA synthase reaction is FAD-dependent. This is the first report on molecular characterization of an enzyme specific to cannabinoid biosynthesis.


Objectives: Endocannabinoids have been shown to play a role in the regulation of vascular tone. The effects of 2-arachidonoyl glycerol (2-AG) on induced-tone were examined in rat aortic rings in vitro. Methods: Aortic rings from Wistar Kyoto (WKY) rats were suspended in organ chambers for recording isometric tension development in response to 2-AG. The production of TXA(2) in response to 2-AG was also assessed by enzyme immunoassay. Results: In endothelium-intact rings pre-contracted to PGF(2alpha), 2-AG (10 nM-30 microM) induced a biphasic effect: a weak relaxation from 10 nM to 0.1 microM, which turned into a concentration-dependent contraction from 3 to 30 microM. Endothelium-denudation did not change 2-AG-mediated vascular effects. 2-AG-induced contraction was unaffected by both the cannabinoid CB1 receptor antagonist SR141716A (3 microM) and the CB2 receptor antagonist SR144528 (1 microM). In contrast, the anandamide transport inhibitor (AM404, 100 microM) and the amino hydrodase inhibitor (PMSF, 30 microM) attenuated (P<0.05) the contractile response evoked by 2-AG in endothelium-intact and rubbed aortic rings. In addition, the cyclooxygenase inhibitor (indomethacin, 10 microM) and the thromboxane A(2) (TXA(2)) receptor (TP receptor) antagonist GR32191 (0.3 microM) totally abolished the contraction elicited by 2-AG in endothelium-intact and rubbed aortic rings. Conclusion: These data suggested that the contraction elicited by 2-AG resulted from the vascular smooth muscle cell uptake and conversion of 2-AG to constrictor prostanoid TXA(2), which in turn caused vasoconstriction through the stimulation of TP receptor.


There is evidence that cannabinoids modulate the reuptake of some neurotransmitters in the central nervous system. In this study, we investigated the effects of the synthetic cannabinoid receptor agonist WIN55212-2, the endocannabinoid anandamide and the chemically related arachidonic acid on serotonin (5-HT) and dopamine (DA) uptake into rat neocortical synaptosomes. At micromolar concentrations, anandamide and arachidonic acid produced steep inhibition curves with Hill coefficients above unity. WIN55212-2 inhibited both DA and 5-HT uptake with Hill coefficients near unity, also within the micromolar range. The effect of WIN55212-2 was not mediated by cannabinoid receptors, since the CB1 receptor antagonist AM251 failed to diminish uptake inhibition by WIN55212-2 and since the Ki estimates of WIN55212-2 were outside the range of the dissociation constants of WIN55212-2 at both CB1 and CB2 receptors. A 100-fold higher concentration of DA, respectively 5-HT, did not induce a shift to the right of the WIN55212-2 concentration-inhibition curves, suggesting a carrier-independent mechanism. The Na(+)/K(+)-ATPase inhibitor ouabain concentration dependently inhibited 5-HT uptake. Possible drug effects on commercial Na(+)/K(+)-ATPase and synaptosomal ATP consumption were investigated using an ATP bioluminescence assay. Ouabain inhibited both commercial and synaptosomal Na(+)/K(+)-ATPase. WIN55212-2 had no effect on commercial Na(+)/K(+)-ATPase, but inhibited synaptosomal ATP consumption. Anandamide produced a sharp decrease in the activity of commercial Na(+)/K(+)-ATPase and on synaptosomal ATP consumption. Presence of ouabain significantly reduced the inhibitory effect of anandamide on synaptosomal ATP consumption, whereas the effect of WIN55212-2 remained unchanged. Our results show
that cannabinoids and arachidonic acid inhibit DA and 5-HT uptake into rat neocortical synaptosomes. This effect is neither cannabinoid receptor-mediated nor due to competitive inhibition of membrane transporters, but is partly effected by a decreased Na(+)/K(+)-ATPase activity.


The antigen-induced release of histamine from sensitised guinea-pig mast cells was dose-dependently reduced by endogenous (2-arachidonylglycerol, 2AG) and exogenous (CP55,940) cannabinoids. The inhibitory action afforded by 2AG and CP55,940 was reversed by SR144528, a selective CB2 receptor antagonist, and left unchanged by the selective CB1 antagonist AM251. The inhibitory action of 2AG and CP55,940 was reduced by a nitric oxide synthase (NOS) inhibitor, N-monomethyl-L-arginine methylester (L-NAME), and reinstated by L-arginine, the physiological substrate. The inhibitory action of 2AG and CP55,940 was also reduced by the unselective cyclooxygenase (COX) inhibitor, indomethacin and by the selective COX-2 blocker rofecoxib. Both 2AG and CP55,940 significantly increased the production of nitrite from mast cells, which was abrogated by L-NAME and by 1400W, a selective iNOS inhibitor. Consistently, nitrite production was parallel to a CP55,940-induced increase in the expression of iNOS protein in mast cells. Both 2AG and CP55,940 increased the generation of PGE2 from mast cells, which was abrogated by indomethacin and by rofecoxib and was parallel to the CP55,940-induced expression of COX-2 protein. Mast cell challenge with antigen was accompanied by a net increase in intracellular calcium levels. Both cannabinoid receptor ligands decreased the intracellular calcium levels which were reversed by SR144528 and L-NAME. In unstimulated mast cells both ligands increased cGMP levels. The increase was abrogated by SR144528, L-NAME, indomethacin and rofecoxib. Our results suggest that 2AG and CP55,940 decreased mast cell activation in a manner that is susceptible to a CB2 receptor antagonist and to inhibition of nitric oxide and prostanoid pathways.

CLINICAL SCIENCE


To estimate the patterns and prevalence of cannabis use among patients with multiple sclerosis (MS), 220 patients were surveyed in Halifax, Nova Scotia. Seventy-two subjects (36%) reported ever having used cannabis for any purpose; 29 respondents (14%) reported continuing use of cannabis for symptom treatment. Medical cannabis use was associated with male gender, tobacco use, and recreational cannabis use. The symptoms reported by medical cannabis users to be most effectively relieved were stress, sleep, mood, stiffness/spasm, and pain.


Recent advances in the understanding of brain cannabinoid receptor function have renewed interest in the association between cannabinoid compounds and psychosis. In a 3-day, double-blind, randomized, and counterbalanced study, the behavioral, cognitive, and endocrine effects of 0, 2.5, and 5 mg intravenous delta-9-tetrahydrocannabinol (Delta-9-THC) were characterized in 22 healthy individuals, who had been exposed to cannabis but had never been diagnosed with a cannabis abuse disorder. Prospective safety data at 1, 3, and 6 months poststudy was also collected. Delta-9-THC (1) produced schizophrenia-like positive and negative
symptoms; (2) altered perception; (3) increased anxiety; (4) produced euphoria; (5) disrupted immediate and delayed word recall, sparing recognition recall; (6) impaired performance on tests of distractibility, verbal fluency, and working memory (7) did not impair orientation; (8) increased plasma cortisol. These data indicate that Delta-9-THC produces a broad range of transient symptoms, behaviors, and cognitive deficits in healthy individuals that resemble some aspects of endogenous psychoses. These data warrant further study of whether brain cannabinoid receptor function contributes to the pathophysiology of psychotic disorders. Neuropsychopharmacology advance online publication, 2 June 2004; doi:10.1038/sj.npp.1300496


The authors sought to determine the prevalence of marijuana use in patients with epilepsy by performing a telephone survey in a tertiary care epilepsy center. Twenty-one percent of subjects had used marijuana in the past year with the majority of active users reporting beneficial effects on seizures. Twenty-four percent of all subjects believed marijuana was an effective therapy for epilepsy. Despite limited evidence of efficacy, many patients with epilepsy believe marijuana is an effective therapy for epilepsy and are actively using it.


RATIONALE. Many neuropsychological studies have documented deficits in working memory among recent heavy cannabis users. However, little is known about the effects of cannabis on brain activity. OBJECTIVE. We assessed brain function among recent heavy cannabis users while they performed a working memory task. METHODS. Functional magnetic resonance imaging was used to examine brain activity in 12 long-term heavy cannabis users, 6-36 h after last use, and in 10 control subjects while they performed a spatial working memory task. Regional brain activation was analyzed and compared using statistical parametric mapping techniques. RESULTS. Compared with controls, cannabis users exhibited increased activation of brain regions typically used for spatial working memory tasks (such as prefrontal cortex and anterior cingulate). Users also recruited additional regions not typically used for spatial working memory (such as regions in the basal ganglia). These findings remained essentially unchanged when re-analyzed using subjects' ages as a covariate. Brain activation showed little or no significant correlation with subjects' years of education, verbal IQ, lifetime episodes of cannabis use, or urinary cannabinoid levels at the time of scanning. CONCLUSIONS. Recent cannabis users displayed greater and more widespread brain activation than normal subjects when attempting to perform a spatial working memory task. This observation suggests that recent cannabis users may experience subtle neurophysiological deficits, and that they compensate for these deficits by "working harder"-calling upon additional brain regions to meet the demands of the task.


Introduction: The prevalence of recreational drug abuse among young adults, including women, has increased markedly over the last 2 decades. Nearly 90 % of these women are of childbearing age. Marijuana remains the drug most commonly used for recreational purposes in pregnancy. However, there appears to be an absence of uniform guidelines for obstetric and anaesthetic management of pregnant patients with a history of marijuana abuse. Materials and Methods: A Medline search for articles highlighting drug abuse in pregnancy, with particular emphasis on marijuana abuse in pregnancy, the drug 's impact on the fetus and implications for administration of obstetrical anaesthesia was performed. Results: Because the pharmacological actions of marijuana are complex and include a unique blend of effects, the clinical picture could
be very unpredictable, the diagnosis often difficult, and management at times controversial. Conclusion: In the absence of uniform anaesthetic guidelines for pregnant patients with a history of drug abuse, including abuse of marijuana, the decision regarding administration of peripartum analgesia or anaesthesia should be individualised and conducted on a case-by-case basis.


We report on a case of a 25-year-old woman with clusters of myoclonus induced by a single exposure to inhaled cannabis. Investigations excluded a structural abnormality of the spine. Multi-channel surface EMG with parallel frontal EEG recording confirmed the diagnosis of propriospinal myoclonus.


The neuropeptides neurokinin B, neurotensin, and anandamide, the endogenous ligands of NK3, NT1, and CB1 receptors respectively, are known to interact with brain dopaminergic transmission. This study evaluated the effects of these three antagonists of the NK3 (SR 142801), neurotensin (SR 48692), and cannabinoid (SR 141716) receptors on the severity of motor symptoms and levodopa-induced dyskinesias after administration of a single dose of levodopa in 24 patients with Parkinson disease. In this exploratory randomized, double-blind, placebo-controlled study, at the dose used, the drugs tested were well tolerated and could not improve parkinsonian motor disability.


The objective of this study was to evaluate the effect of the acute administration of marijuana (MJ) on cardiovascular (CV) function and CNS pharmacokinetics (PK) of [(15)O]water in occasional (O) versus chronic (C) MJ users. Each subject received four injections of [(15)O]water (one prior and three post-smoking) on two occasions in which they received active or placebo MJ. For each injection, measures of CV function and CNS PK [(15)O]water were made. Postsmoking, MJ influenced all measured CV and [(15)O]water PK parameters. C users reported significantly lower "highness" and smaller heart rate (HR) changes, which resulted in reduced rate pressure product (RPP) changes compared to O users, even though Delta(9)-tetrahydrocannabinol levels were higher, whereas changes in blood pressure (BP), arrival time, and [(15)O]water concentration were not significantly different between the groups. Significant CV changes resulted in changes in the whole-body distribution of cardiac output rather than changes in cerebral blood flow. Chronic MJ use produces tolerance to the HR increases induced by acute MJ smoking compared to changes observed in occasional users, without changing the effects on BP and [(15)O]water PK.


Previous laboratory investigations, case reports, and a hospital-based case-control study have suggested that marijuana use may be a risk factor for squamous cell head and neck cancer. We conducted a population-based case-control study to determine whether marijuana use is associated with the development of oral squamous cell carcinoma (OSCC). Case subjects (n = 407) were 18-65-year-old residents of three counties in western Washington State who were newly diagnosed with OSCC from 1985 through 1995. Control subjects (n = 615), who were similar to the cases with respect to age and sex, were selected from the general population using random-digit telephone dialing. Lifetime histories of marijuana use and exposure to known OSCC...
risk factors were ascertained using a structured questionnaire. Information on genetic polymorphisms in glutathione S-transferase enzymes was obtained from assays on participant DNA. Odds ratios for associations with features of marijuana use were adjusted for sex, education, birth year, alcohol consumption, and cigarette smoking. A similar proportion of case subjects (25.6%) and control subjects (24.4%) reported ever use of marijuana (adjusted odds ratio, 0.9; 95% confidence interval, 0.6-1.3). There were no trends in risk observed with increasing duration or average frequency of use or time since first or last use. No subgroup defined by known or suspected OSCC risk factors (age, cigarette smoking, alcohol consumption, and genetic polymorphisms) showed an increased risk. Marijuana use was not associated with OSCC risk in this large, population-based study.


Previous reports have documented an improvement in night vision among Jamaican fishermen after ingestion of a crude tincture of herbal cannabis, while two members of this group noted that Moroccan fishermen and mountain dwellers observe an analogous improvement after smoking kif, sifted Cannabis sativa mixed with tobacco (Nicotiana rustica). Field-testing of night vision has become possible with a portable device, the LKC Technologies Scotopic Sensitivity Tester-1 (SST-1). This study examines the results of double-blinded graduated THC administration 0-20mg (as Marinol((R))) versus placebo in one subject on measures of dark adaptometry and scotopic sensitivity. Analogous field studies were performed in Morocco with the SST-1 in three subjects before and after smoking kif. In both test situations, improvements in night vision measures were noted after THC or cannabis. It is believed that this effect is dose-dependent and cannabinoid-mediated at the retinal level. Further testing may assess possible clinical application of these results in retinitis pigmentosa or other conditions.


The neurophysiological effects of prenatal marijuana exposure on response inhibition were assessed in 18- to 22-year-olds. Thirty-one participants from the Ottawa Prenatal Prospective Study (OPPS) performed a blocked design Go/No-Go task while neural activity was imaged with functional magnetic resonance imaging (fMRI). The OPPS is a longitudinal study that provides a unique body of information collected from each participant over 20 years, including prenatal drug history, detailed cognitive/behavioral performance from infancy to young adulthood, and current and past drug usage. The fMRI results showed that with increased prenatal marijuana exposure, there was a significant increase in neural activity in bilateral prefrontal cortex and right premotor cortex during response inhibition. There was also an attenuation of activity in left cerebellum with increased prenatal exposure to marijuana when challenging the response inhibition neural circuitry. Prenatally exposed offspring had significantly more commission errors than nonexposed participants, but all participants were able to perform the task with more than 85% accuracy. These findings were observed when controlling for present marijuana use and prenatal exposure to nicotine, alcohol and caffeine, and suggest that prenatal marijuana exposure is related to changes in neural activity during response inhibition that last into young adulthood.


BEHAVIOURAL SCIENCE


Actually, guidelines for treatment of substance-related disorders were written under the overall control of the DG-Sucht e. V. and the DGPPN e. V. This appears within the framework of
the Arbeitsgemeinschaft der Wissenschaftlichen Medizinischen Fachgesellschaft (AWMF). The leading objective of these guidelines is the description of the current scientifically proven and evidence-based medicine in addiction to derive recommendations to therapy. In this context, the guideline for treatment of cannabis-related disorders is introduced.


The effects of prenatal marijuana and alcohol exposure on school achievement at 10 years of age were examined. Women were interviewed about their substance use at the end of each trimester of pregnancy, at 8 and 18 months, and at 3, 6, 10, 14, and 16 years. The women were of lower socioeconomic status, high-school-educated, and light-to-moderate users of marijuana and alcohol. The sample was equally divided between Caucasian and African-American women. At the 10-year follow-up, the effects of prenatal exposure to marijuana or alcohol on the academic performance of 606 children were assessed. Exposure to one or more marijuana joints per day during the first trimester predicted deficits in Wide Range Achievement Test-Revised (WRAT-R) reading and spelling scores and a lower rating on the teachers' evaluations of the children's performance. This relation was mediated by the effects of first-trimester marijuana exposure on the children's depression and anxiety symptoms. Second-trimester marijuana use was significantly associated with reading comprehension and underachievement. Exposure to alcohol during the first and second trimesters of pregnancy predicted poorer teachers' ratings of overall school performance. Second-trimester binge drinking predicted lower reading scores. There was no interaction between prenatal marijuana and alcohol exposure. Each was an independent predictor of academic performance.


In spite of having been formulated nearly two decades back, there is as yet no consensus on the validity of the clinically popular self-medication hypothesis (SMH) of substance use disorders in patients with dual diagnosis. SMH broadly proposes that patients use substances in a non-random fashion so that the psychopharmacologic characteristics of particular substances are used to alleviate a variety of psychiatric symptoms and emotional distress. In order to test the SMH empirically, it was broken down to five sub-hypotheses, which were tested in a group of dual-diagnosis schizophrenics (DDS) patients vis-à-vis a group of only-schizophrenia (S) patients (n = 22 each). The DDS group scored lower than the S group regarding general and some specific psychopathology. The DDS patients ascribed reasons for substance use more often for hedonistic pursuit but also for reduction in symptoms and distress. There was a trend for alcohol to be used more for self-medication purposes compared to opioids and cannabis. The perceived effects of these three substances were significantly different on several symptom/distress dimensions. Finally, there was some degree of "match" between symptom-oriented reasons for use of substances and the effect that was perceived. All of this evidence provides a consistent but modest support for the SMH for "some patients, some substances, and some symptoms." The implications are discussed. (Am J Addict 2004;13:139-150)


Alcohol-dependent women progress faster from onset of alcohol drinking to entry into treatment, experiencing an earlier onset (i.e., "telescoping") of alcohol-related complications. This phenomenon also appears to be evident in drug-dependent women, though the data available to support telescoping in drug dependence is less abundant. Objective: To evaluate gender effects on progression to treatment entry and on the frequency, severity and related complications of DSM-III-R drug and alcohol dependence among 271 substance-dependent patients (mean age: 32.6 years; 156 women). Method: Multivariate and univariate ANCOVA was used to compare age at onset of regular use of cocaine, opioids, cannabis and alcohol and time elapsed between initiation of regular use of each substance and entry into an index or current substance abuse treatment. Scores on the Addiction Severity Index (ASI) were also examined. Results: There was...
no gender difference among patients in the age at onset of regular use of any substance. Women experienced fewer years of regular use of opioids and cannabis, and fewer years of regular alcohol drinking before entering treatment. Although the severity of drug and alcohol dependence did not differ by gender, women reported more severe psychiatric, medical and employment complications. Conclusions: These findings support the notion of an accelerated progression to treatment entry among opioid-, cannabis- and alcohol-dependent women, and suggest that there exists a gender-based vulnerability to the adverse consequences of these disorders.


This paper reports findings from a qualitative study which explores the role of cannabis in young people's lives during their early teenage years. In particular, it focuses on the relationship between cannabis and tobacco-related beliefs and behaviour. Fifty-nine young people of both sexes, aged 13-15, from different socioeconomic backgrounds, and with a wide range of cigarette and cannabis use experience, took part in the study. All were recruited from youth club settings and most were interviewed in self-selected friendship pairs. The paper argues that, while many young people appear to hold predominantly negative views about cigarettes, particularly in relation to their potential to foster dependence, cannabis is often viewed as relatively benign. In spite of these beliefs, for some 'cannabis-oriented' young people, their cannabis use appears to support and reinforce their smoking habit. The paper concludes that a coordinated approach to the planning and delivery of services which addresses young people's health risk behaviours is required. Smoking cessation and drugs education practitioners need to break with tradition, and find ways of working more closely together.


This technical report provides historical perspectives and comparisons of various approaches to the legal status of marijuana to aid in forming public policy. Information on the impact that decriminalization and legalization of marijuana could have on adolescents, in addition to concerns surrounding medicinal use of marijuana, are also addressed in this report. Recommendations are included in the accompanying policy statement.


As experts in the health care of children and adolescents, pediatricians may be called on to advise legislators concerning the potential impact of changes in the legal status of marijuana on adolescents. Parents, too, may look to pediatricians for advice as they consider whether to support state-level initiatives that propose to legalize the use of marijuana for medical purposes or to decriminalize possession of small amounts of marijuana. This policy statement provides the position of the American Academy of Pediatrics on the issue of marijuana legalization, and the accompanying technical report (available online) reviews what is currently known about the relationship between adolescents' use of marijuana and its legal status to better understand how change might influence the degree of marijuana use by adolescents in the future.


Prior research has shown that those with alcohol problems have significantly elevated rates of traffic events (i.e. traffic violations and collisions) than licensed drivers from the general population and that treatment is associated with reductions in alcohol-related collisions. However, very little research exists on traffic events and the impact of treatment for cannabis or cocaine clients. The objectives of this research are: (1) to determine whether clients in treatment for a primary problem of alcohol, cannabis or cocaine have significantly elevated rates of traffic events than a matched control group of licensed drivers; and (2) to assess whether a significant reduction in traffic events occurs after treatment for each client group compared to a control group. Driver records of patients admitted to substance abuse treatment in 1994 for a primary problem of alcohol (Formula: see text), cannabis (Formula: see text) or cocaine (Formula: see text) were accessed from the Ministry of Transportation for Ontario, Canada. A comparison
group of 504 licensed drivers frequency matched by age, sex and place of residence, was also randomly selected. Data was collapsed into two 6-year time periods: 1988-1993 (i.e. before treatment) and 1995-2000 (i.e. after treatment). Six repeated measures analysis of variance tests were conducted where traffic violations and collisions of three treatment groups (i.e. alcohol, cannabis or cocaine) and a control group were compared before and after treatment. All three treatment groups had significantly more traffic violations than the control group and no significant interactions between time period and group membership were found. For collisions, there was a significant interaction between the alcohol and control groups and between the cocaine and control groups. The average number of collisions for the alcohol and cocaine groups decreased after completing treatment, whereas the number for the control group was stable over the same time periods. Neither the interaction term nor the between group effect was significant in the comparison of the cannabis and control groups. When rates of collisions were calculated based on the period that each driver had a valid license, the interaction term was still significant for the comparison of the alcohol and control groups but not for the cocaine and control groups. The results contribute to existing literature by demonstrating that cocaine and cannabis clients have a higher risk of traffic violations than matched controls and that reductions in collision risk was found after treatment for the alcohol and cocaine groups. More research is needed to better understand the reasons for the higher risk of traffic events and to determine reasons for declines.


Pharmacists, the most accessible of health care professionals, are well positioned to help prevent and treat substance use disorders and should prepare themselves to perform these functions. New research improves our knowledge about the pharmacological and behavioral risks of drug abuse, supports the clinical impression that drug dependence is associated with long-lasting neurochemical changes, and demonstrates effective pharmacological treatments for certain kinds of drug dependencies. The profession is evolving. Pharmacists are engaging in new practice behaviors such as helping patients manage their disease states. Collaborative practice agreements and new federal policies set the stage for pharmacists to assist in the clinical management of opioid and other drug dependencies. Pharmacists need to be well informed about issues related to addiction and prepared not only to screen, assess, and refer individual cases and to collaborate with physicians caring for chemically dependent patients, but also to be agents of change in their communities in the fight against drug abuse. At the end of this article the pharmacist will be better able to: 1. Explain the disease concept of chemical dependence. 2. Gather the information necessary to conduct a screen for chemical dependence. 3. Inform patients about the treatment options for chemical dependence. 4. Locate resources needed to answer questions about the effects of common drugs of abuse (alcohol, marijuana, narcotics, "ecstasy", and cocaine). 5. Develop a list of local resources for drug abuse treatment. 6. Counsel parents who are concerned about drug use by their children. 7. Counsel individuals who are concerned about drug use by a loved one. 8. Counsel individuals who are concerned about their own drug use.

This newsletter is supported in part by an unrestricted educational grant from ICN/Valeant Pharmaceuticals (Canada)