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Effects of the synthetic cannabinoid nabilone on spatial learning and hippocampal neurotransmission

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Abstract

Cannabinoids, the active components of marijuana, affect memory and hippocampal neurotransmission. It has been claimed that nabilone, a synthetic cannabinoid endowed with antiemetic properties, has a peculiar profile of actions. We studied the effects of the drug on spatial learning and in vitro hippocampal CA1 electrophysiology in the rat. Nabilone (0.1, 0.5, and 1.0 mg/kg ip) does not impair place learning in a water maze task, whereas Δ^8-tetrahydrocannabinol (Δ^8-THC) disrupts this function. At concentrations ranging from 1 nM to 10 μM nabilone does not influence basal glutamatergic neurotransmission, which is decreased by Δ^8-THC. Although cannabinoids have been consistently reported to affect synaptic plasticity, nabilone 1 μM does not change paired-pulse facilitation, long-term potentiation and the magnitude of long-term depression. However, the time course of the latter phenomenon is significantly changed by the drug, the depression being lower than in control experiments from 7 to 35 min postinduction. Altogether, our data indicate that there might be differences in the effects of agonists for central cannabinoid receptors, which could help to understand the pharmacology of this class of molecules. The results also suggest that amnesia induced by cannabinoids be possibly related to their effects on hippocampal neurotransmission. The study supports the use of nabilone in conditions the course of which is complicated by cognitive impairment.

Keywords: Nabilone; Cannabinoids; Learning; Water maze; Long-term potentiation; Long-term depression; Rat

1. Introduction

Potential therapeutic properties of cannabinoids, the active components of marijuana, are outweighed by their side effects. It has been claimed that nabilone, a benzopyrane derivative synthetic cannabinoid, used to control nausea and vomiting in patients undergoing anticancer chemotherapy, could separate antiemetic activity from mental effects of cannabinoids (Ahmedzai et al., 1983; Herman et al., 1979; Johansson et al., 1982). The peculiar pharmacological profile of this molecule is possibly explained either by the dimethylepthylic side chain or by the ketonic group in position 9 of the dibenzopyranic group (Ward and Holmes, 1985). Nabilone shows several properties, including cardiovascular (Ward and Holmes, 1985), respiratory (Gong et al., 1983), opioid (Gilbert, 1981; Hamann and di Vadi, 1999; Johnson and Jasinski, 1983), and behavioral effects (Stark and Dews, 1980; Stark and Henderson, 1966). Understanding the mechanisms of action of a cannabinoid endowed with such a characteristic spectrum of actions might provide hints into the pharmacology of this class of compounds.

A common problem with cannabinoids is represented by amnesia. Although some data exist about the amnesic effects of nabilone (Glass et al., 1980, 1981), the literature on this topic is quite scanty, and, to our knowledge, no study has addressed this problem expressly. Recently, the issue has found a new reason for attention. The drug, which is currently marketed in some countries, has been tried for the treatment of Parkinson's disease (Sieradzan et al., 2001), the course of which is sometimes complicated by cognitive impairment.

To add information to the pharmacological profile of nabilone, we studied its effects on rat place learning in the Morris water maze (Morris, 1984). This test has been widely used in pharmacology research due to the reduced influence of nonspecific cues, such as olfactory traces, and its sensitivity in revealing the amnesic effect of drugs (Arolfo and Brioni, 1991; Da and Takahashi, 2002; Varvel et al., 2001). We also studied the effects of nabilone on in vitro hippo-

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campal electrophysiology. The crucial importance of hippocampus in learning is established (Pearce et al., 1998), and this brain structure is believed to play a role in cannabinoid-induced amnesia (Sullivan, 2000). In addition to basal hippocampal neurotransmission, we investigated some effects of the drug on two forms of long-term synaptic efficacy changes, i.e., long-term potentiation (LTP) and long-term depression (LTD). Both phenomena represent experimental models of putative mechanisms of learning and memory.

2. Materials and methods

2.1. Animals and drugs

Experiments were performed on male, 3-month old Sprague–Dawley rats (Charles River, Calco-Como). The animals were housed in cages in groups of 4–5 at constant temperature (22 ± 1 °C) and humidity (60 ± 10%), with access to food and water ad libitum and in a 12-h light–dark cycle. Animal care and use followed the directives of The Council of the European Communities (1986). All efforts were made to minimize animal suffering and to reduce the number of animals used. Nabilone is a water insoluble white powder. Δ^8-Tetrahydrocannabinol (Δ^8-THC) was prepared from an ethyl alcohol solution. At the beginning of the study, the structure of the drugs was checked with ^{1}H-NMR spectroscopy.

2.2. Behavior

2.2.1. Apparatus

The behavioral test was performed in a silent room at the temperature of 24 ± 1 °C. The experimenter and the devices for data acquisition and analysis were located in an adjacent room.

Water maze is a circular pool of 140-cm diameter, 50-cm height, arbitrarily divided in four quadrants named according to the cardinal points (NE, NW, SE, SW) and filled with water at room temperature up to the height of 32 cm.

In one quadrant, at the center of the line from the pool wall to the pool center, an 11-cm diameter, 30-cm high, water-filled Plexiglas cylinder was placed. The cylinder's upper surface, which had been made rough to facilitate climbing, was 2 cm under water and provided a platform on which the rat could climb to escape from water during the experiments. In these conditions the platform was invisible to the animals.

During the experiments the operator was in the adjacent room and measured the escape latencies by a stopwatch. A camera mounted perpendicularly over the pool's center acquired the rat behavior. The camera was connected with a videorecorder, a monitor, and a contrast sensitive video-tracker (Videomex-V, Columbus Inst.—Ohio), which tracked the rat swimming paths and measured the swim distances. All experiments were recorded on tape for subsequent analyses. The videotracker data were saved on a personal computer with an ad hoc program.

2.2.2. Behavioral procedure

The platform was held in a fixed position during the whole place learning. The rats were trained to learn the position in daily blocks of four consecutive trials. Altogether, the rats underwent 20 learning trials over five consecutive days. The drugs, dissolved in polyethylenglycol (PEG) 300, 50% final volume, were injected intraperitoneally 30 min before each block of time. The following groups ($n = 5$) were studied: vehicle, nabilone 0.1, 0.5 and 1.0 mg/kg, Δ^8-THC 5 mg/kg. The effects of doses of nabilone higher than 1.0 mg/kg were not investigated, owing to the emergence of ataxia, which appeared at 2.0 mg/kg ip and precluded the proper execution of the test. Ataxia displayed a dose–effect relationship up to the dose of 8.0 mg/kg, which was the highest we tried. The ataxic effects of doses between 1.0 and 2.0 mg/kg were not tested.

At the beginning of each trial, the animals were placed in water with the head facing the pool wall, in the middle of one of the four wall segments. The starting point varied across trials according to a pseudo random sequence that was identical for all the rats. The rats were left in water until they reached the invisible platform and climbed on it; then they were left on the platform for a 10-s reinforcement period. If the platform had not been found within 70 s (cutoff time), the experimenter placed them on it.

Three days after the place learning, the platform was removed and the rats were allowed to swim (spatial probe). The time spent in each pool quadrant during the first 60 s of swimming was measured for subsequent analyses.

On the following day, 30 min after a drug treatment equal to the one used before each block of the place learning, the rats were trained to find a platform that had been made visible with a sharply contrasted cylinder (5-cm diameter, 12-cm height) placed on it. The training consisted of four trials (30-min intertrial interval). During this ''cued learning'', the platform position and the starting point were changed every trial. This experiment was performed to test the animal's visual acuity, motor ability and motivation to locate the platform.

2.3. Electrophysiology

Animals that had not been used for behavioral experiments were killed by decapitation under urethane anesthesia (1.5 g/kg ip). The head was rapidly put on ice and the brain removed and dissected. Transverse hippocampal slices, 400 μm thick, were prepared with a McIlwain tissue chopper (The Mickle Laboratory Engineering, Gomshall, Surrey, England). The slices were held for at least 2 h in a glass holding chamber containing an artificial cerebrospinal fluid solution (ACSF) saturated with a gas mixture (95% O_2, 5%

CO_2). ACSF is an aqueous solution, containing 126 mM NaCl, 3.5 mM KCl, 1.2 mM NaH_2PO_4, 25 mM $NaHCO_3$, 2 mM $CaCl_2$, 1.3 mM $MgCl_2$, 11 mM Glucose (pH 7.3). The slices were then placed in a submerged-type recording chamber, perfused with oxygenated ACSF (24 ± 1 °C) by a peristaltic pump (Gilson Minipuls 3) at a constant flow rate (2.5–3 ml/min).

Nabilone and Δ^8-THC were dissolved in dimethylsulfoxide (DMSO, Sigma Aldrich, Steinheim, Germany), 0.1% of final volume. As the 10-μM solution of nabilone showed an opalescent appearance, the concentration of 1 μM was chosen for LTP and LTD experiments. In control experiments, an ACSF solution containing 0.1% DMSO was used.

2.3.1. Field EPSP recordings

Field potentials were recorded in hippocampal CA1 area. A stimulating electrode (stainless steel, Teflon insulated, 2 MΩ) was placed into the stratum radiatum. The recording electrode, which was a glass pipette filled with Na acetate 0.5 M (OD 1.0 mm, ID 0.7 mm, 1.5–2 MΩ), was placed into the dendritic layer of CA1.

Excitatory postsynaptic potentials (EPSPs) were evoked by regular stimulation (0.033 Hz; squared waves, 60 μs; constant current, 20–200 μA). The depth of both electrodes was adjusted to maximize the height of EPSPs. After this procedure, slices that were not showing a steady response during a preliminary stabilization period were not used for the study.

The responses were amplified 1000 times and filtered at 5 kHz (LC low pass filter, -40 dB/decade). The signals were then sampled at 20 kHz, digitized (A/D board NB MIO 16 by National Instruments on personal computer Apple Macintosh IIfx) and stored on disk for subsequent off-line analyses. The slope of EPSP ascending phase was measured and used for statistical analysis.

In preliminary experiments, the effects of different concentrations of nabilone on field potentials were studied in 4 hippocampal slices. After a 30-min stabilization period, which was repeated every time the solution was changed, a stimulus–response (S–R) curve was produced by 11 consecutive stimuli. The stimuli intensity ranged from 0 to 200 μA and it was increased according to a linear progression (0, 20, 40, 60, 80, 100, 120, 140, 160, 180 and 200 μA). This sequence was repeated for each solution. The solutions were changed following this sequence: ACSF, DMSO 0.1% in ACSF (vehicle), nabilone at increasing concentrations (1 nM, 10 nM, 100 nM, 1 μM, and 10 μM), and Δ^8-THC 10 μM. Altogether, each solution was perfused for 35 min and 30 s.

LTP was studied in seven slices perfused with vehicle (DMSO 0.1%) and seven in slices perfused with nabilone 1 μM. Following a stabilization period of 30 min, at a stimulus intensity such that EPSP initial slopes ranged between 40% and 60% of the maximum, LTP was induced by three trains of tetanic stimuli (100 pulses, 100 Hz, 30-s intertrain interval, basal intensity). The recordings were prolonged for at least 1 h after this tetanus. Responses recorded within 1 min after last tetanus were assumed to be posttetanic potentiation (PTP).

LTD was studied in six slices perfused with vehicle and in five slices perfused with nabilone 1 μM. The depression was induced by a low-frequency, repeated paired-pulse, stimulation (200 ms paired-pulse interval, 900 pairs, 1 Hz, stimulation intensity doubled). This protocol induces a long-lasting decrease of evoked field potentials in slices from adult animals. Moreover, this form of LTD is sensitive to NMDA antagonists (Kemp et al., 2000). The recording was prolonged for at least 1 h post induction.

2.4. Statistical analysis

ANOVA was performed in the behavioral data gathered. Swim distances, which were used to evaluate learning performances, were transformed into the ratio between the distance from the animal's starting point and the platform location, and the distance that the animal actually swam to reach it. In this way, the optimal performance resulted in a value of 1; if the animals had not been able to reach the escape platform within the cutoff time, the value was assumed to be 0. This transform is aimed to solve the problem of the statistical treatment of cutoff values and, at the same time, it corrects for differences among the distances the rat has to swim to reach the platform. The transformed place learning data and the spatial probe results underwent angular transform. Repeated measurement designs were implemented to analyze the transformed learning data (one way on between-group comparison; two ways on repetition for place learning, one way on repetition for cued learning). Bonferroni's test was used for post hoc individual comparisons.

ANOVA was performed on S–R curves for repeated measurements, whereas ANCOVA was performed on LTP and LTD experiments, using the average of EPSP slope values from the 10 preinduction field potentials as a covariate and assigning two ways on repetition (the first way with 4 levels, to put together values from two consecutive min of recording, the second with 30 levels to estimate the effect of time). Bonferroni's test was used for post hoc individual comparisons. All calculations were performed with Statistica 5.1 for Windows.

3. Results

3.1. Behavior

Average swim speeds (mean ± S.E.M.) resulted to be 25.48 ± 0.45 and 30.31 ± 1.13 for rats treated with vehicle and Δ^8-THC, respectively. The swim speeds of nabilone treated rats were 26.42 ± 0.65, 26.20 ± 0.74 and 25.35 ± 0.71 cm/s at the dose levels of 0.1, 0.5, and 1.0 mg/kg, respectively. The differences were significant by ANOVA [$F(4,20) = 7.062$, $P = .001$]. Individual comparisons revealed that the speeds of Δ^8-THC-treated rats were significantly

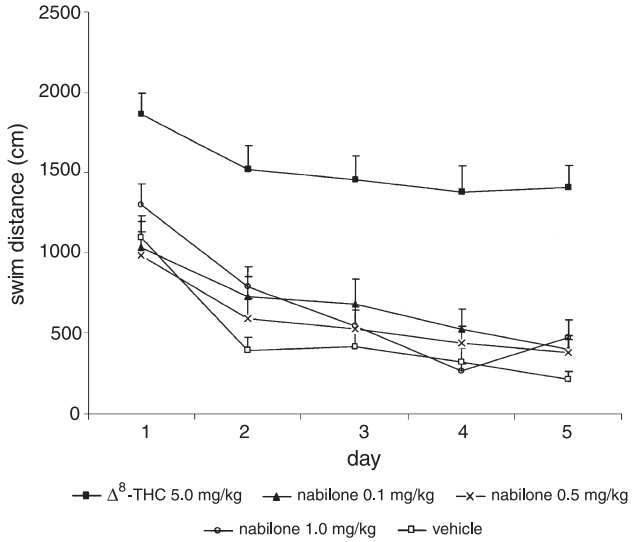

Fig. 1. Effects of nabilone on place learning. Swim distances to reach the hidden platform are displayed as a function of the day of learning. Each plot is the mean of data obtained from four consecutive learning trial. Data are obtained from five rats/group. Error bars: +1 S.E.M.

higher that those of any other group. On the contrary, no significant differences were found in either the duration or the number of periods of immobility of rats. Thigmotaxis, i.e., the tendency to swim close to the pool wall, was also studied. This phenomenon was expressed as percent of swim time along the pool wall. The results did not demonstrate any significant difference among groups (data not shown).

The ANOVA for place learning displayed significant differences among the five groups [$F(4,20 = 10.412$, $P=.0001$]. Individual comparisons demonstrated that Δ^8-THC affects learning, whereas nabilone has no effect at any of the doses studied. Effects of drug treatment on interactions between the group of treatment and both the repetition factors that were assigned to block [$F(16,80) = 1.916$, $P=.0306$] and to within-block data [$F(12,60 = 2.013$, $P=.0385$) were significant. The individual comparisons showed that this significance is explained by the effects of Δ^8-THC 5.0 mg/kg. No significant differences from control were observed for the two interactions in nabilone-treated groups, suggesting that the drug has no effect on both reference and working memory. Fig. 1 summarizes the results of place learning.

The results of the spatial probe demonstrate that the rats tend to swim in the platform quadrant after its removal. The times spent in this quadrant (mean ± S.E.M.) were 26.2 ± 1.2, 26.8 ± 1.6, 25.2 ± 2.0, 27.2 ± 3.2, and 18.6 ± 1.1 s for vehicle, nabilone 0.1 mg/kg, nabilone 0.5 mg/kg, nabilone 1.0 kg/kg, and Δ^8-THC 5.0 mg/kg, respectively. The differences were significant by the ANOVA [$F(4,20) = 3.295$, $P=.0315$ after angular transform). Assuming that four comparisons were relevant to the analysis, individual comparisons by Bonferroni's test demonstrated a significant difference between vehicle and Δ^8-THC groups ($df = 8$, $t = 4.851$, $P=.0013$).

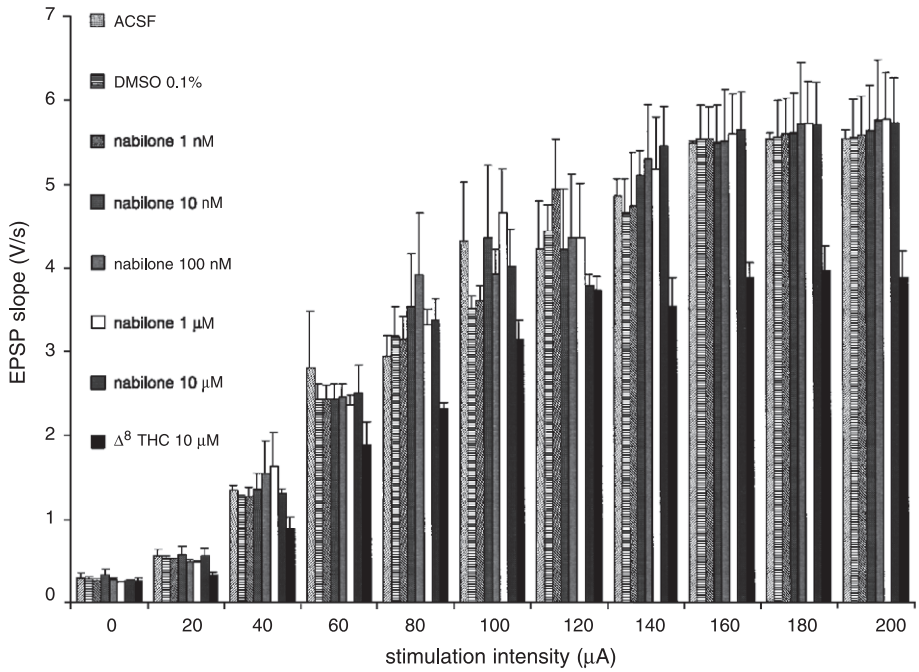

Fig. 2. Effects of nabilone on stimulus–response curves. The slopes of the field EPSPs are displayed as a function of stimulus intensity. Each bar represents the effects of one drug perfusion on the stimulus–response curve. Data are obtained from four slices. In each experiment a slice was perfused according to the following sequence: ACSF, ACSF + DMSO 0.1%, nabilone at increasing concentrations (From 1 nM to 10 μM), Δ^8-THC 10 μM. Nabilone and Δ^8-THC were dissolved in DMSO, 0.1% final volume. Error bars: +1 S.E.M.

During cued learning all rats performed optimally, proving that the drugs do not affect visual and motivational performances. ANOVA for repeated measurements failed to show any significant effect of treatments and their interactions.

3.2. Electrophysiology

The results of S–R studies are summarized in Fig. 2. Stimulus intensity [$F(10,30) = 33.669$ $P < .0001$] and drug treatment [$F(7,21) = 7.353$, $P = .0002$] were associated with a significant effect. On the contrary, the effect of the interaction Drug Perfusion × Stimulus Intensity [$F(70,210) = 1.051$, $P = .3867$] was not significant. Individual comparison of groups of drug treatment by Bonferroni's test showed that the responses elicited during the perfusion of Δ^8-THC were significantly lower that those observed in any other group. No significant differences were observed in the comparisons of any other group.

Nabilone tends to reduce paired-pulse facilitation (PPF; 50-ms interstimulus interval; EPSP slopes +5.53%, $n = 18$, and +7.56%, $n = 14$, for nabilone and DMSO, respectively). The difference, however, was not significant by the ANCOVA.

In all slices used for the study of LTP a significant, long-lasting increase of synaptic efficacy after tetanic stimulation was observed. As displayed in Fig. 3, nabilone does not affect LTP [$F(1,11) = 0.099$, $P = .7586$ by repeated measurement ANCOVA) and its time course [Treatment × Time interaction: $F(29,348) = 0.172$, $P = 1.0000$], neither does the drug affect PTP [$F(1,11) = 0.411$, $P = .5346$ at 30-s posttetanus by ANCOVA].

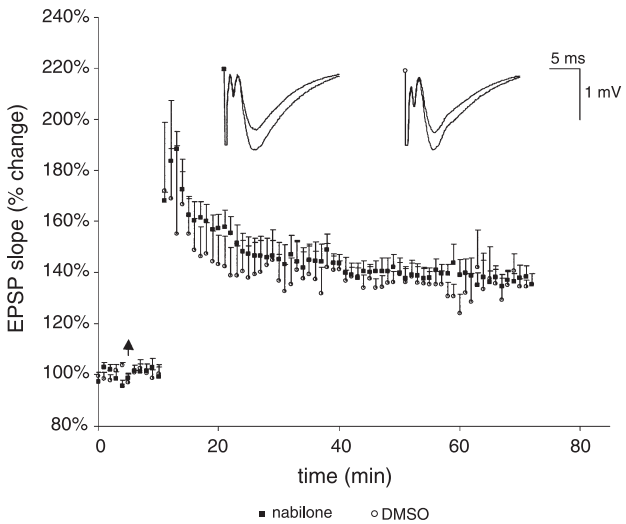

Fig. 3. Effects of nabilone on LTP. The normalized percentage changes in the slope of the field EPSP are displayed as a function of time. Each plot is normalized with respect to the mean of baseline period 10 min prior to the delivery of tetanus. The tetanus (three trains of 100 pulses at 100 Hz; 30 s intertrain interval) was applied at the time denoted by the arrow. Data are obtained from seven control slices perfused with DMSO 0.1% and seven slices perfused with nabilone 1 μM. Nabilone was dissolved in DMSO, 0.1% final volume. Error bars: +1 S.E.M. The insets display representative responses obtained 5 min pretetanus and 50 min posttetanus for nabilone (top left) and DMSO (top right).

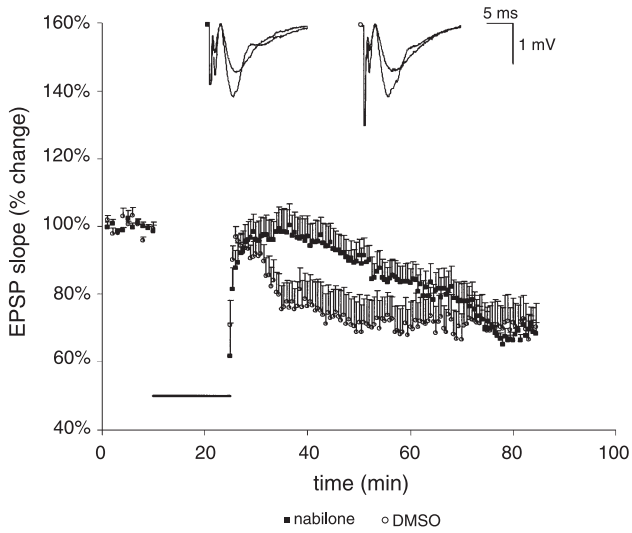

Fig. 4. Effects of nabilone on LTD. The normalized percentage changes in the slope of the field EPSP are displayed as a function of time. Each plot is normalized with respect to the mean of baseline period 10 min prior to LTD induction. LTD was induced at the time denoted by the solid line by 900 pairs of stimuli at 1 Hz (200 ms paired-pulse interval). Data are obtained from six control slices perfused with DMSO 0.1% and five slices perfused with nabilone 1 μM. Nabilone was dissolved in DMSO, 0.1% final volume. Error bars: +1 S.E.M. The insets display representative responses obtained 5 min preinduction and 50 min postinduction for nabilone (top left) and DMSO (top right).

Fig. 4 summarizes the results for LTD. The paired-pulse depression protocol induced an LTD in all slices used for the study. Although the depression was of a similar magnitude in the two treatment groups [$F(1,8) = 1.197$, $P = .3057$ by repeated measurement ANCOVA; at 60-min posttetanus −31.6% and −29.6% for nabilone and DMSO, respectively; $F(1,8) = 1.197$, $P = .3058$ by ANCOVA], the phenomenon showed a different time course, as it is demonstrated by the analysis of the Treatment × Time interaction [$F(29,261) = 2.849$, $P < .0001$]. In particular, nabilone 1 μM appears to affect the depression from 7 to 35 min by individual comparisons.

4. Discussion

The results of the behavioral experiments support the hypothesis that nabilone does not affect mnemonic functions. The drug did not reduce place learning at any of the three dose levels studied; the analysis of learning across trials and days suggested that the drug does not affect short- and long-term memory. However, the possibility that doses of nabilone higher than 1.0 mg/kg ip induce an amnesic effect cannot be rejected. These higher doses were not tried due to the effects of nabilone on motor coordination. Since humans are less prone than rodents to cannabinoid induced ataxia (Herkenham et al., 1991), we cannot exclude that the

drug impairs memory in humans. Yet, the doses we used in rats are considerably higher than those endowed with anxiolytic effects in mice (Onaivi et al., 1990) and with antiemetic effects in cats (London et al., 1979; McCarthy and Borison, 1981). It was estimated that nabilone is about seven times more potent than Δ^8-THC in causing psychic symptoms, such as euphoria, sedation and dysphoria in humans (Johnson and Jasinski, 1983). Also, Browne and Weissman (1981) found that nabilone was two- to threefold more potent than Δ^8-THC in rats evaluated in the drug discrimination paradigm. We observed that spatial learning is substantially disrupted by Δ^8-THC 5 mg/kg ip. Moreover, doses up to 8 mg/kg ip of the same molecule do not induce ataxia in rats (Diana, 1992). Therefore, sub-ataxing doses of Δ^8-THC produce an impairment of learning performances in rats. Altogether, this seems to confirm that nabilone shows a peculiar profile of behavioral effects as compared to ''typical'' cannabinoids.

The lack of cognitive effects of nabilone is associated with a lack of effects on hippocampal responses. The analysis of S–R curves demonstrates that field potentials are not affected by drug concentrations up to 10 μM, whereas the same concentration of Δ^8-THC reduces the responses. This is in accordance with the hypothesis that THC induced amnesia is caused by a decreased hippocampal neurotransmitter release (Sullivan, 2000). Previous studies indicated that CB1 agonists affect glutamate release in hippocampal synaptic buttons (Shen et al., 1996; Shen and Thayer, 1998). It was recently shown that a decrease of EPSP slope and PS amplitude of field potentials in CA1 is induced by both Win 55,212-2 and the endogenous CB1 ligand anandamide. These effects are blocked by the CB1 antagonist SR 141716 (Ameri et al., 1999). A study carried out on cultured glutamatergic neurons from hippocampal CA1 and CA3 provided some hints into the mechanisms of the inhibition. It was shown that cannabinoid receptor activation reduces excitatory postsynaptic currents without effects on spontaneous miniature excitatory postsynaptic currents. Moreover, it increases PPF at low Ca^{++} concentration, suggesting a reduction of glutamate release at the presynaptic side (Sullivan, 1999). In our study nabilone, at physiological Ca^{++} concentration, decreased PPF. However, this reduction does not reach the statistical significance.

Nabilone does not change the size of LTP and LTD in hippocampal CA1, suggesting that it does not affect long-lasting synaptic plasticity in a significant manner. This seems to be inconsistent with the effects of other cannabinoids, which appear to impair both LTP (Collins et al., 1994; Nowicky et al., 1987; Terranova et al., 1995) and LTD (Levenes et al., 1998; Misner and Sullivan, 1999). The figures do not display the effects of positive control drugs on these phenomena. Therefore, it cannot be directly assessed whether cannabinoids would have had an effect on these variables. However, in recent works from our laboratory, in which the experiments had been carried out under the same conditions as in the present experiments, we described the depressive effects of Win 55,212-2 10 μM on LTP (Diana et al., 2002). The effects of Win 55,212-2 were compared to those induced in an independent unpotentiated hippocampal pathway, the responses of which were recorded at the same site. Apparently, the decrease of the responses in the potentiated pathway paralleled the one observed in the control pathway, thus resulting in a fictitious reduction of LTP. We concluded that the effects of cannabinoids on synaptic plasticity be a mere consequence of reduced glutamatergic transmission. Recently, other reports questioned the effects of cannabinoids on synaptic plasticity (Barinaga, 2001; Misner and Sullivan, 1999). It has also been hypothesized that the effects of cannabinoids on LTP be explained by the reduction in glutamate release from synaptic buttons at levels that could not remove the block of Mg^{++} on NMDA receptors (Sullivan, 2000).

The results also show an effect of nabilone on the time course of LTD: The inhibition of this phenomenon is evident in early times, and it disappears after 35 min. The finding of a decreased LTD is difficult to explain, due to different induction schedules used elsewhere and the scanty literature about this topic. Since we induced LTD by repeated paired-pulse inhibition (Kemp et al., 2000), the depressive effect of cannabinoids on this phenomenon (Paton et al., 1998; Sullivan, 1999) might provide explanation for the effects of nabilone on the time course of LTD.

In conclusion, in validated experimental models, nabilone seems to be almost devoid of the cognitive and hippocampal electrophysiological effects of cannabinoids. This suggests an association between the reduction of hippocampal CA1 neurotransmission and the amnesia induced by natural cannabinoids.

Many data about the pharmacodynamics of cannabinoids come from studies on molecules, such as Δ^8-THC, which produce most of the effects of marijuana. However, there might exist differences in the activity of different compounds belonging to this class. Understanding these differences could provide information about the pharmacology of cannabinoids. Our findings do not explain the mechanisms involved in the selectivity of nabilone. It has been suggested that cannabinoid-induced amnesia is caused by the block of presynaptic Q- and N-type calcium channels (Sullivan, 1999). This mechanism, given the lack of amnesic effects of nabilone, might not be at the basis of the antiemetic and, possibly, other effects of cannabinoids. More recently, the existence of a new receptorial site for cannabinoids has been hypothesized (Breivogel et al., 2001; Hajos et al., 2001). Apparently, this site mediates the depression of glutamatergic neurotransmission in the hippocampus (Hajos et al., 2001), which is left unaffected by nabilone. Although this issue needs further studies, the specific effects of nabilone might be explained by a characteristic spectrum of binding to the different CNS receptorial sites for cannabinoids.

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