

Chronic Adolescent Δ^9 -Tetrahydrocannabinol Treatment of Male Mice Leads to Long-Term Cognitive and Behavioral Dysfunction, Which Are Prevented by Concurrent Cannabidiol Treatment

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Abstract

Introduction: The high prevalence of adolescent cannabis use, the association between this use and later psychiatric disease, and increased access to high-potency cannabis highlight the need for a better understanding of the long-term effects of adolescent cannabis use on cognitive and behavioral outcomes. Furthermore, increasing Δ^9 -tetrahydrocannabinol (THC) in high-potency cannabis is accompanied by a decrease in cannabidiol (CBD), thus an understanding of the interactions between CBD and THC in the neurodevelopmental effects of THC is also important. The current study examined the immediate and long-term behavioral consequences of THC, CBD, and their combination in a mouse model of adolescent cannabis use.

Materials and Methods: Male CD1 mice received daily injections of THC (3 mg/kg), CBD (3 mg/kg), CBD + THC (3 mg/kg each), vehicle, or remained undisturbed in their home cage (no handling/injections), either during adolescence (postnatal day [PND] 28–48) or during early adulthood (PND 69–89). Animals were then evaluated with a battery of behavioral tests 1 day after drug treatment, and again after 42 drug-free days. The tests included the following: open field (day 1), novel object recognition (NOR; day 2), marble burying (day 3), elevated plus maze (EPM; day 4), and Nestlet shredding (day 5).

Results: Chronic administration of THC during adolescence led to immediate and long-term impairments in object recognition/working memory, as measured by the NOR task. In contrast, adult administration of THC caused immediate, but not long term, impairment of object/working memory. Adolescent chronic exposure to THC increased repetitive and compulsive-like behaviors, as measured by the Nestlet shredding task. Chronic administration of THC, either during adolescence or during adulthood, led to a delayed increase in anxiety as measured by the EPM. All THC-induced behavioral abnormalities were prevented by the coadministration of CBD + THC, whereas CBD alone did not influence behavioral outcomes.

Conclusion: These data suggest that chronic exposure to THC during adolescence leads to some of the behavioral abnormalities common in schizophrenia. Interestingly, CBD appeared to antagonize all THC-induced behavioral abnormalities. These findings support the hypothesis that adolescent THC use can impart long-term behavioral deficits; however, cotreatment with CBD prevents these deficits.

Keywords: cannabis; neurodevelopment; object recognition; schizophrenia; working memory

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Introduction

Cannabis contains more than 60 cannabinoids.^{1–3} Of these, tetrahydrocannabinol (THC) is the major psychoactive and cannabidiol (CBD) is the main nonpsychoactive component of cannabis. Through genetic interactions, breeding for high-THC content (to create high-potency cannabis) results in very low-CBD levels. The market demand for high-potency cannabis has seen the average THC content of U.S. cannabis increasing from ~4% to 12% in the past 20 years. This has been accompanied by a corresponding decrease in CBD content, increasing the average THC/CBD ratio ~20-fold over the same period.⁴

Cannabis is the most widely used illicit drug among adolescents in the United States.⁵ In 2016, 12.8% of 8th graders have used cannabis in their lifetime, and by 12th grade 44.5% have tried cannabis.⁵ Furthermore, daily use increases from 0.7% among 8th graders to 6% among 12th graders.⁵ The high prevalence of frequent adolescent cannabis use, and the increased presence of high-THC/low-CBD cannabis, emphasizes a need for research examining the long-lasting effects of chronic adolescent exposure to major cannabis constituents on neurobehavioral outcomes.

During human adolescence (age ~10–25), neuroanatomical and functional changes lead to increased neural and cognitive efficiency.^{6–8} As the prefrontal cortex (PFC) matures during adolescence, it undergoes significant synaptic remodeling and changes in myelination.^{9–12} The PFC is responsible for higher order cognitive function such as executive function,^{13–15} and disruption of maturation PFC can be expected to have long-lasting adverse consequences.

Because of the prominent role of endocannabinoids in neurodevelopment,¹⁶ cannabis exposure during adolescence might perturb PFC neurodevelopment and cause long-term cognitive, emotional, and behavioral impairments. Supporting this hypothesis, many studies indicate a positive correlation between adolescent cannabis use and aspects of schizophrenia (age of first psychotic episode, severity).^{17–22} Factors such as onset of use, frequency of use, and the THC/CBD ratio in cannabis consumed have been shown to modulate this risk.^{17,23,24} For instance, after controlling for confounding variables (e.g., IQ, psychiatric illness in family members), a large population-based study found a dose-dependent relationship of adolescent cannabis use and the development of schizophrenia.¹⁹ In addition, the risk of developing a psychotic disorder was shown to increase threefold among users of high-potency cannabis compared with subjects who never used canna-

bis.²⁵ In this study, daily use of high-potency cannabis conferred an even higher (>fivefold) risk for developing schizophrenia.²⁵

A causal relationship between adolescent cannabis use and an increased risk for schizophrenia is unlikely to be proven given ethical constraints. Thus, research utilizing animal models, rigorously controlled experimental design, and examination of specific cannabis components can identify potential mechanisms linking frequent adolescent cannabis use and an increased risk for schizophrenia. Furthermore, examining the variables involved in producing schizophrenia-associated phenotypes—such as impairment of working and object recognition memory, anxiety, and repetitive and compulsive behaviors—might provide valuable insight into schizophrenia's etiology. Working^{26–29} and object recognition memory^{30–32} deficits are frequent in schizophrenia. Furthermore, repetitive and compulsive behaviors^{33,34} as well as anxiety^{35,36} are common in schizophrenia.

This study examined the immediate and long-term effects of chronic cannabinoid exposure on object recognition/working memory, repetitive and compulsive behaviors, and anxiety in adolescent and adult mice. We found that adolescent THC impaired selected behavioral domains and that this impairment was prevented by concurrent CBD treatment.

Materials and Methods

Subjects

Male CD1 mice ($n = 221$) were bred and housed three to four per cage under a standard reversed light cycle housing facility (light–dark cycle of 12 h: lights on at 7 p.m.). Food and water were available *ad libitum*. All experimental procedures were approved by Indiana University's Bloomington Institutional Animal Care and Use Committee.

Drugs

Injection solutions were made by dissolving THC, CBD, or CBD+THC in 100% ethanol to a concentration of 6 mg/mL for each drug. In a second tube, Kolliphor[®] EL (synonym: Cremophor[®] EL; Sigma-Aldrich) was mixed with sterile 0.9% saline. The dissolved cannabinoid was then mixed with the Cremophor/NaCl solution to a final ratio of 1:1:18 (cannabinoid/ethanol:Cremophor:saline). The final concentration of each component in the injection solutions was 0.3 mg/mL cannabinoid, 5% ethanol, 5% Cremophor, and 0.81% NaCl. Mice were injected intraperitoneally (i.p.) with 3 mg/kg of the



indicated drug (in a volume of 10 mL/kg). Controls received a vehicle consisting of 5% ethanol, 5% Cremophor, and 0.81% NaCl (also at a volume of 10 mL/kg).

Injection and behavioral testing schedule

Mice were randomly assigned to one of five groups: no treatment control (remained in home cages during 3 weeks of treatment, subjected only to routine cage changes); vehicle control; THC (3 mg/kg); CBD (3 mg/kg); or CBD + THC (3 mg/kg each). Animals underwent daily i.p. injections either during adolescence (postnatal day [PND] 28–48) or during adulthood (PND 69–89; Fig. 1). Animals were behaviorally tested starting 24 h after the last injection, and again 6 weeks later. The behavioral battery included open field (day 1), novel object recognition (NOR; day 2),

marble burying (day 3), elevated plus maze (EPM; day 4), and Nestlet shredding (day 5).

Procedures

Animals were habituated to the testing room for 30 min before testing. Testing was conducted within a 100 lux illuminated room, during the dark phase of the light–dark cycle. Conditions were counterbalanced across the testing sessions. All apparatuses were wiped down with a diluted 2% chlorhexidine solution (AgriLabs, St. Joseph, MO) between trials. An observer blind to group allocations manually scored video-recorded trials, except for the open field assay (OFA), which utilized an automated scoring system (The *Fusion* software program; Omnitech Electronics, Inc.; Columbus, OH).

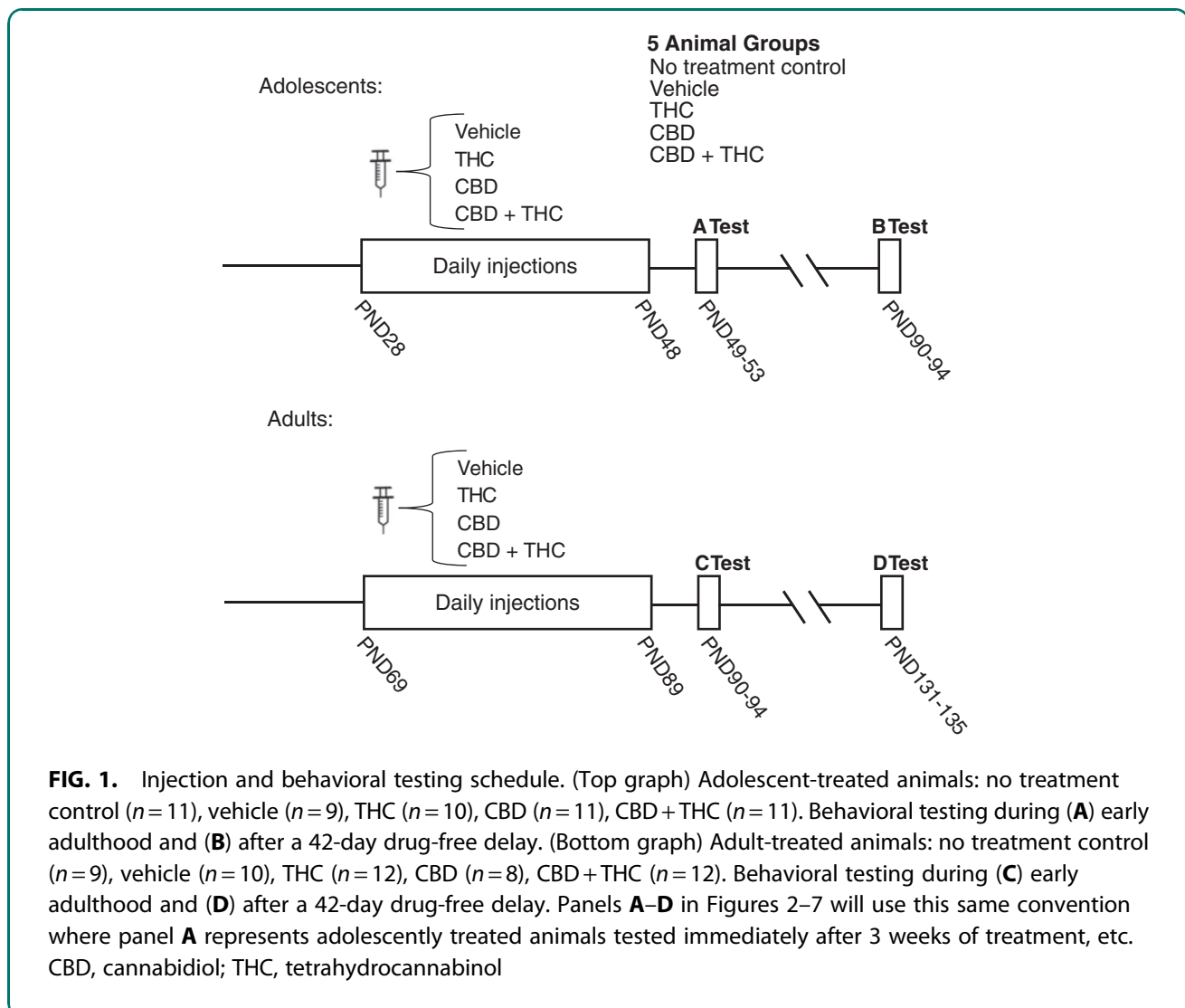


FIG. 1. Injection and behavioral testing schedule. (Top graph) Adolescent-treated animals: no treatment control ($n = 11$), vehicle ($n = 9$), THC ($n = 10$), CBD ($n = 11$), CBD + THC ($n = 11$). Behavioral testing during (A) early adulthood and (B) after a 42-day drug-free delay. (Bottom graph) Adult-treated animals: no treatment control ($n = 9$), vehicle ($n = 10$), THC ($n = 12$), CBD ($n = 8$), CBD + THC ($n = 12$). Behavioral testing during (C) early adulthood and (D) after a 42-day drug-free delay. Panels A–D in Figures 2–7 will use this same convention where panel A represents adolescently treated animals tested immediately after 3 weeks of treatment, etc. CBD, cannabidiol; THC, tetrahydrocannabinol



Novel object recognition

The NOR task evaluates working and object recognition memory and takes advantage of a mouse's inherent tendency to explore novel more than familiar objects.³⁷ During the NOR's familiarization phase, mice were exposed to two equivalent objects within a Plexiglass chamber (42L×42W×32H cm; AccuScan Instruments, Columbus, OH) for 5 min. (Untreated mice spent an equal amount of time exploring the two objects used in the NOR study; Supplementary Fig. S1.) The same pairs of objects were used for all testing. After a 105-min intersession interval, mice were returned to the chamber; however, one of the previously shown "familiar" objects was removed and replaced with a novel object. Familiar and novel object positions were counterbalanced across testing sessions. Testing lasted 3 min and was recorded using a webcam (Logitech Pro 9000 PC Internet Camera Webcam, 2.0-Megapixel Video Resolution and Carl Zeiss Lens Optics; Romanel-sur-Morges, Switzerland).

Exploration was defined by the mouse's nose pointed toward and within a 2-cm radius of an object. To assess performance in this task, a discrimination index (DI) was calculated. The DI quantified the difference between time spent exploring the novel and familiar object during testing as a fraction of total exploratory time, $DI = (T_N - T_F) / (T_N + T_F)$, where novel object exploration time is T_N and familiar object exploration time is T_F . The DI can range between +1 and -1, where a positive score indicates more time exploring the novel object, a negative score indicates more time exploring the familiar object, and zero indicates null preference.³⁷ Important to the interpretation of these studies, chronic THC administration to mice leads to impaired object recognition, which takes several days to resolve.³⁸

Nestlet shredding

The Nestlet shredding task is a measure of compulsive and repetitive-like behavior in mice, which takes

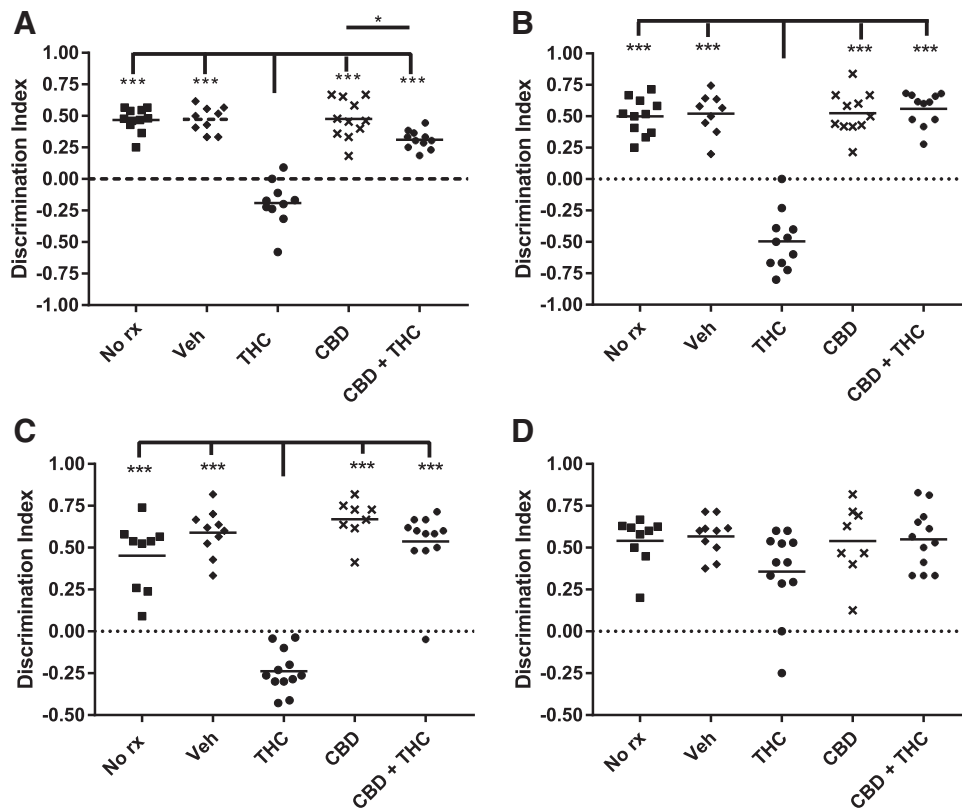


FIG. 2. Adolescent THC impairs NOR performance. Object recognition was scored using a discrimination index (see Materials and Methods section). Adolescent treated and (A) immediately tested, (B) delay tested. Adult treated and (C) immediately tested, (D) delay tested. *** $p < 0.001$ and * $p < 0.05$. NOR, novel object recognition.



advantage of a mouse's inherent tendency to shred material for nest building.³⁹ In this task, mice were individually placed in a clean mouse cage with ~2 cm (height) of woodchip bedding material and one 2×2 in. packed cotton Nestlet (Ancare Corp; Bellmore, NY) laid on top of the bedding. Testing lasted 75 min. The remaining Nestlet was collected, dried overnight, and weighed, and the percentage of the Nestlet shredded was calculated.

Marble burying

The marble burying task is a measure of repetitive-like behavior in rodents, and takes advantage of a mouse's inherent tendency to bury objects,^{39,40} although marble burying can also be affected by changes in aversion to novelty, novelty-induced anxiety, excessive locomotion, etc. Mice were individually placed into a clean mouse cage containing 5 cm (height) of corncob bedding, with 20 evenly placed blue marbles (5×4) laid on top. Testing was for 8 min. Marbles that were at least three-fourth covered by bedding were considered to be buried.

Elevated plus maze

The EPM measures anxiety-like behavior in rodents.⁴¹ Mice were individually placed in the central-open area within the EPM. Testing was for 5 min. Scoring was based on (1) time spent in the open versus closed arms and (2) entries into the open versus closed arms. As an index of locomotor activity, the total number of entries into all four EPM arms was also counted. Behavior was recorded using the same webcam as above.

Open field

The OFA was used to assess locomotion and thigmotaxis, the latter as an alternative measure of anxiety.⁴² Mice were placed into a Plexiglass chamber (42L×42W×32H cm) and allowed to acclimate to the chamber for ~3 min. Activity was then measured for three, consecutive 10-min bins. The *Fusion* software program collected data from sensors located along the bottom periphery of the chamber. *Fusion* distinguishes the *interior* (21×21 cm within the center of the chamber) versus *peripheral zones* (10.5 cm from edge to center) of the chamber,

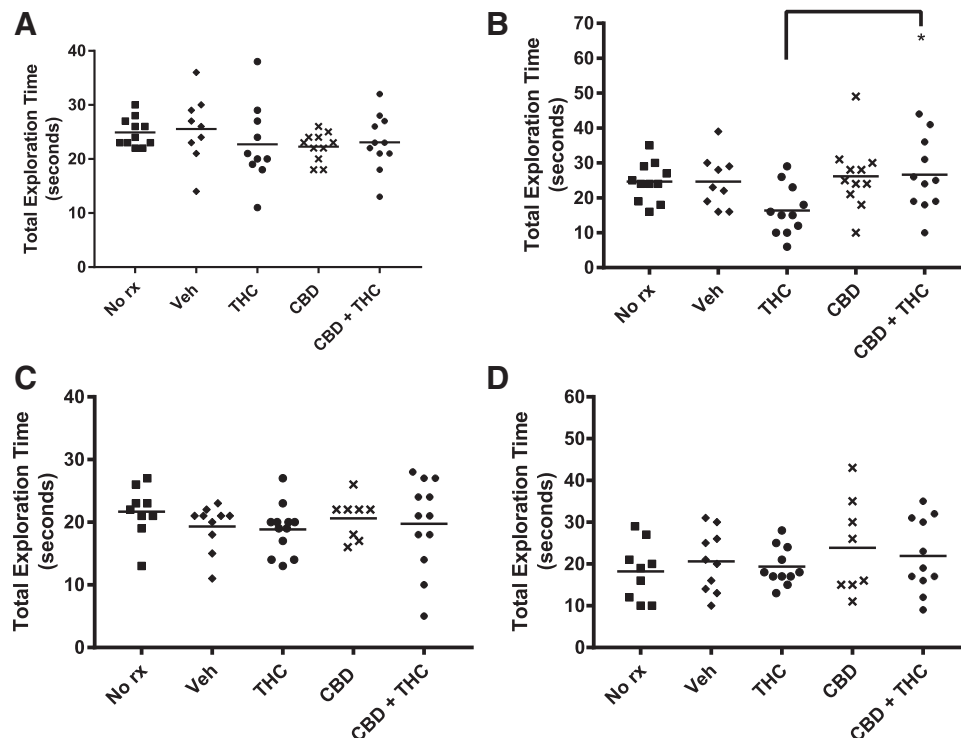


FIG. 3. Similar exploration times in the NOR test among all treatment groups. Exploration time was the time the mouse spent engaged with either object (see Materials and Methods section for details). Adolescent treated and (A) immediately tested, (B) delay tested. Adult treated and (C) immediately tested, (D) delay tested. * $p < 0.05$.



which is used for scoring. Two variables were analyzed in this study: duration (whole body) and distance traveled (whole body) within each zone.

Determination of THC plasma levels

Lipid extractions of plasma obtained by exsanguination 24 h after the last cannabinoid injection were performed in a separate group of adolescent male CD1 mice receiving 21 days of THC or THC + CBD, as previously described.⁴³ Deuterium-labeled anandamide (d_8 AEA; Cayman Chemical, Ann Arbor, MI) was used as an internal standard to determine extraction efficiency. Samples were analyzed using an Applied Biosystems API 3000 triple quadrupole mass spectrometer with electrospray ionization (Foster City, CA). THC levels were determined by running each sample using a multiple reactions monitoring method to detect the 315.2 parent ion mass and the 123.2 fragment. Analysis of the HPLC/MS/MS data was per-

formed using Analyst software (Applied Biosystems) as previously described.⁴³

Statistical analysis

Analyses were conducted using Microsoft Excel 2016 (Redmond, WA) and GraphPad Prism 7 (La Jolla, CA), with the level of significance set at 0.05. Significance was assessed by one-way analysis of variance (ANOVA), or by Student's *t*-test if two samples were compared. If a significant difference in ANOVA was detected, Tukey's *post hoc* analyses were conducted.

Results

Adolescent THC leads to long-term impairment in NOR

Three weeks of THC treatment significantly decreased the DI in both adolescent and adult mice relative to all other treatment groups (Fig. 2). However, although the DI remained decreased in adolescent-treated mice 6

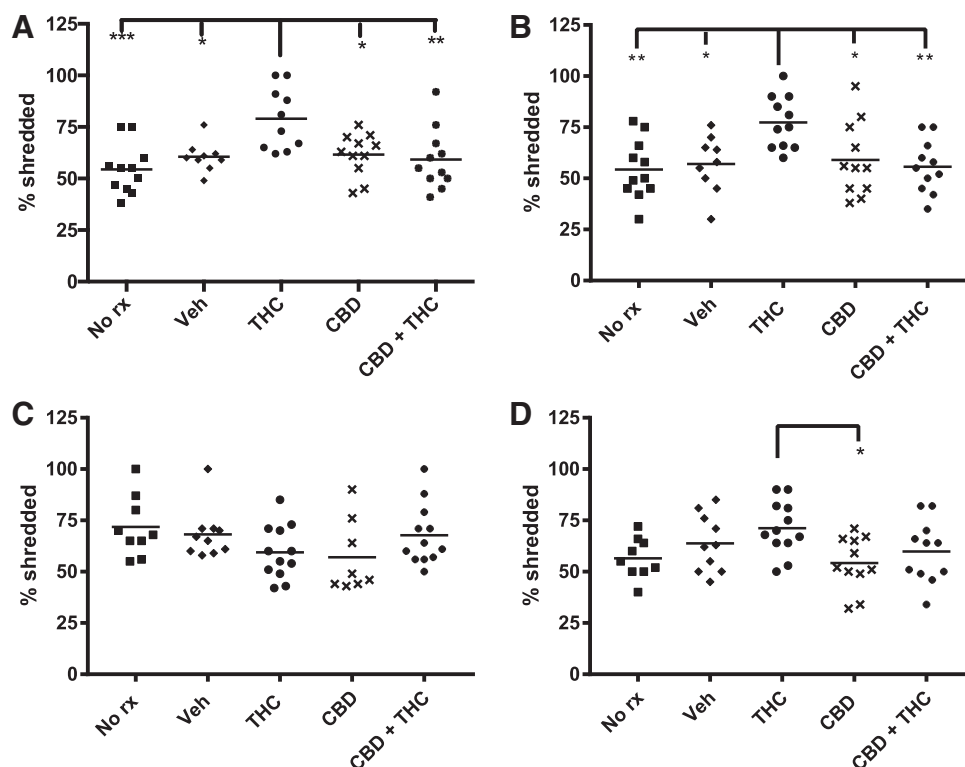


FIG. 4. Adolescent THC increases Nestlet shredding. The amount of Nestlet shred was determined by measuring unshred Nestlet 75 min after placing the Nestlet in with the mouse. Adolescent treated and (A) immediately tested, (B) delay tested. Adult treated and (C) immediately tested, (D) delay tested. *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$.



weeks after the final THC dose, the DI had recovered to control levels in the adult-treated mice (Fig. 2). Thus, 3 weeks of THC treatment similarly impaired NOR in adolescent and adult mice, but although the impairment was transient in the adult-treated mice, it was persistent in mice treated during adolescence. If a treatment markedly decreased exploratory time, then the DI may also be decreased. However, Figure 3 shows no major differences in total exploration times among treatment groups, or testing times.

CBD has been observed to attenuate various undesirable effects of THC,⁴⁴⁻⁴⁷ and the risk of developing schizophrenia among heavy cannabis users appears to increase as CBD content decreases.^{23-25,48} Therefore, we examined whether concurrently administered CBD would prevent either the immediate or long-term consequences of THC administration on NOR. Interestingly, chronic administration of CBD alone did not affect NOR performance (Fig. 2), whereas co-administration of CBD+THC completely abolished

the deleterious consequences of THC on DI among adolescent- and adult-treated mice (Fig. 2).

Nestlet shredding

Figure 4 shows that adolescent THC treatment increases Nestlet shredding at both time periods investigated. In contrast, adult THC treatment did not increase Nestlet shredding when compared with vehicle controls. As with impaired NOR, concurrent treatment with CBD prevented both the immediate and long-lasting THC-induced increases in Nestlet shredding (Fig. 4).

Marble burying

Figure 5 shows marble burying was enhanced immediately after 3 weeks of THC treatment of adolescent mice. Although not reaching our threshold for statistical significance, there was a strong trend for a difference in the adolescent THC-treated mice versus vehicle ($p=0.06$). Six weeks later, there were no apparent differences between any of the groups receiving

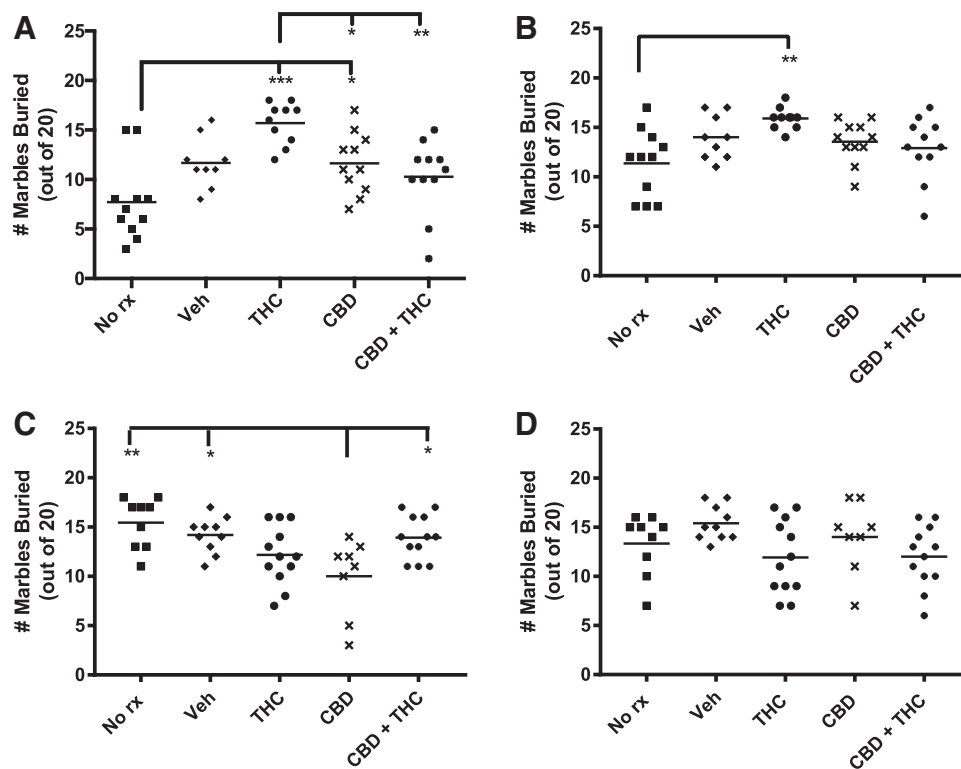


FIG. 5. Adolescent THC transiently enhances marble burying. Buried marbles were those that were at least three-fourth covered with bedding. Adolescent treated and (A) immediately tested, (B) delay tested. Adult treated and (C) immediately tested, (D) delay tested. *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$.



injections. The pattern was quite different in the adult-treated mice. Chronic CBD treatment resulted in fewer marbles buried relative to the two control groups and the combined CBD + THC group when tested immediately after the end of injections (Fig. 5). However, when adults were tested 6 weeks later, no significant differences were apparent between the various treatment groups.

Elevated plus maze

Both adolescent- and adult-treated mice exhibited decreased EPM open arm entries when tested 6 weeks after THC treatment, suggesting the late emergence of anxiety (Fig. 6). This was accompanied by a decrease in open arm time (Fig. 7). The THC-induced decrease in open arm entries and duration was prevented by cotreatment with CBD (Figs. 6 and 7). Adult THC-treated mice also showed a decrease in open arm entries immediately after chronic treatment. There were no consistent differences in total arm entries between the

various treatment groups (data not shown), suggesting that the various treatments did not have major effects on locomotor activity in the EPM.

Open field

There were no significant differences in total distance traveled or thigmotaxis within the OFA arena between any of the treatment groups (data not shown).

Plasma THC levels

One possible mechanism by which CBD could attenuate THC effect is if CBD increased THC metabolism and lowered THC plasma levels. This seems unlikely as most studies find that concurrent CBD+THC does not increase THC metabolism,⁴⁹ nonetheless, to test this possibility, THC plasma levels were measured after 3 weeks of THC or THC+CBD. As shown in Figure 8, concurrent CBD administration modestly, but significantly, increased plasma THC levels ($p=0.027$). Thus the attenuation of THC's effects across

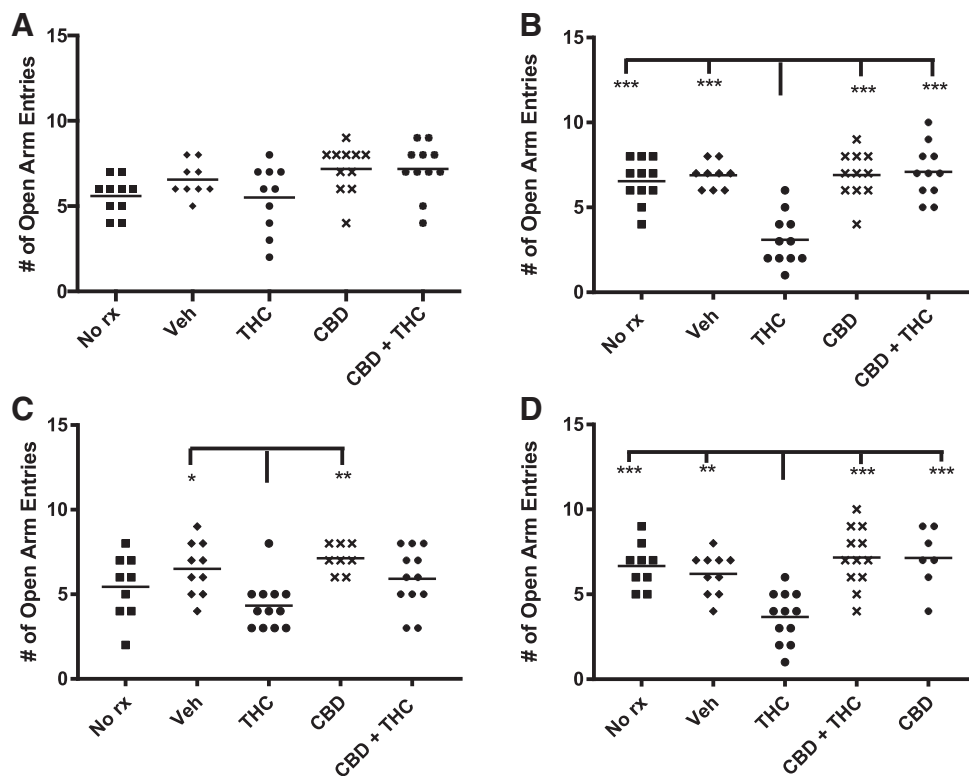


FIG. 6. Adult THC consistently decreases EPM open arm entries and adolescent THC decreases open arm entries after a delay. A movement was considered an entry into the maze arm if head and shoulders entered the arm. Adolescent treated and (A) immediately tested, (B) delay tested. Adult treated and (C) immediately tested, (D) delay tested. *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$. EPM, elevated plus maze.



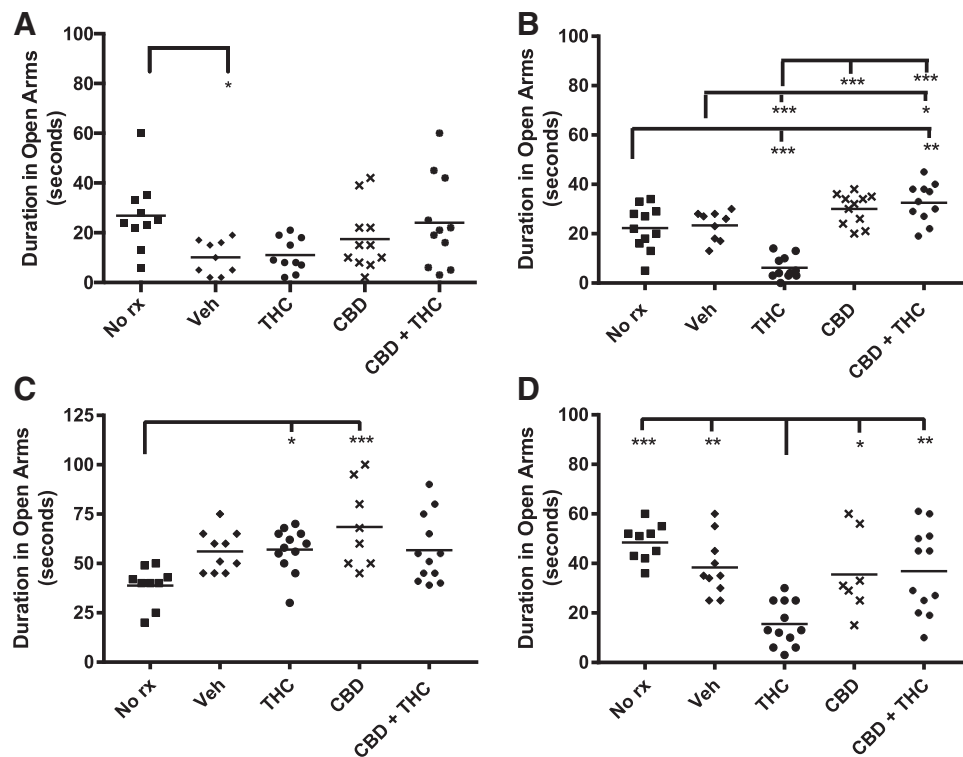


FIG. 7. Adolescent and adult THC decrease EPM open arm duration after a THC-free delay. Mice were considered to be within an arm until their entire body had exited the arm. Adolescent treated and (A) immediately tested, (B) delay tested. Adult treated and (C) immediately tested, (D) delay tested. *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$.

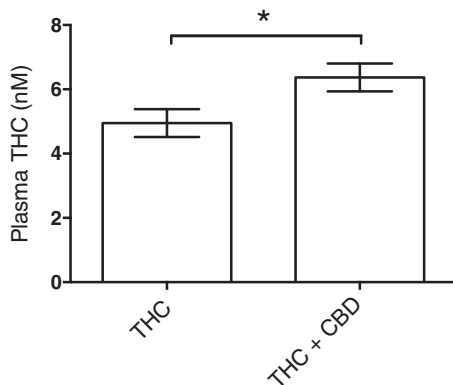


FIG. 8. Combining THC with CBD mildly increases THC plasma levels. THC levels were determined 24 h after 21 days of daily THC injections. Plasma was collected and THC levels were analyzed as described in Materials and Methods section. Concurrent CBD treatment increased plasma THC levels by ~30%. * $p < 0.027$.

multiple behavioral tests is not due to increased THC metabolism and is likely due to a pharmacodynamic interaction.

Discussion

This study found that THC administration to adolescent male mice for 3 weeks led to long-lasting deficits in NOR performance and repetitive behaviors, whereas a similar treatment of adults results in only transient abnormalities. Furthermore, chronic THC administration to both adolescent and adult mice induced anxiety behaviors after a delay. Very interestingly, co-administration of CBD prevented all of the THC-induced impairments observed in this study, whereas chronic CBD alone was inactive in the assays used here.

THC-induced cognitive impairment

While the current study used mice, previous studies using rats found chronic cannabinoid (THC, CP 55940, or



WIN55,212-2) administration during adolescence, but not during adulthood, causes object recognition impairment after a drug-free delay.^{50–52} Similarly to our findings with mice and THC, chronic WIN55,212-2 administration during adolescence leads to immediate object recognition deficits in rats.⁵³ Therefore, this study provides additional support that THC exposure during adolescence can lead to enduring cognitive impairment that is absent if THC is given during adulthood.

THC-induced behavioral impairment

Although this study is the first to specifically examine the effects of chronic cannabinoid administration on Nestlet shredding in mice, other studies have found both acute⁵⁴ and chronic⁵⁵ THC disrupted nest building. Although shredding material is the first step in nest building, increases in shredding behavior detected by the Nestlet shredding task are considered a measure of repetitive and compulsive-like behavior⁵⁶ and as a model of obsessive-compulsive disorder.³⁹ This study is the first to find that chronic adolescent THC administration leads to a long-lasting increase in this repetitive behavior.

Our findings also suggest that THC administration during adolescence transiently increases repetitive behaviors, as assessed by the marble-burying task. This difference was clear when THC mice were compared with adolescent mice treated with CBD or CBD + THC. In addition, although just missing our significance threshold, chronic adolescent THC treatment likely leads to an immediate increase in marble burying compared with vehicle. No biologically significant differences in marble burying were found among adolescent-treated mice tested 6 weeks after the last injection. This is the first study to examine the effects of chronic THC on marble burying. Several earlier studies have found that a single dose of synthetic cannabinoid, CBD, and/or acutely increasing endocannabinoid levels, suppressed marble burying.^{57–59} The decrease in CBD-treated adult mice (Fig. 5C) is consistent with these earlier findings.

THC-induced anxiety

This study is the first to examine the persistent effects of chronic cannabinoid administration on EPM behaviors in adolescent or adult mice. We found that chronic THC administration to either adult or adolescent mice produced a delayed anxiogenic effect, as measured through EPM. An earlier study⁶⁰ using adult mice that examined the effects of THC or CBD on anxiety behaviors found that a chronic dose of THC (10 mg/kg), higher than that used in this study, increased anxiety when mice were tested im-

mediately after dosing (vs. 4 days later as in this study). A previous study examining the effects of chronic, incrementing (2.5 mg/kg PND 35–37; 5 mg/kg PND 38–41; 10 mg/kg PND 42–45) THC administration in adolescent rats of both sexes during the early light cycle found no differences in anxiety-like behavior in the EPM, when measured after a drug-free delay (PND 75).⁶¹ The difference between the cited studies and this study may be due to species, escalating dose, or delay in testing, etc.

In contrast to the EPM results, no evidence of anxiety was apparent in any of the open field measures, for any treatment, age, or post-test interval. Thus, these results are similar to the previously cited study in rats.⁶¹ Owing to the inclusion of a ~3 min acclimation period, it was not possible to determine whether the various treatments affected acclimation to a novel environment.

CBD protects against THC-induced deficits

Perhaps the most significant finding of our study is that coadministration of CBD + THC protected against all of the cognitive and behavioral impairments induced by THC. Interestingly, this was observed even though coadministration of CBD with THC actually increased plasma THC levels (Fig. 8). The mechanism of this protection is unclear, but does not appear to be pharmacokinetic. Numerous targets have been proposed for CBD.^{62,63} A particularly interesting potential mechanism that could explain the current results is CBD's negative allosteric modulation of CB1 cannabinoid receptors.⁶⁴ The finding that coadministration of CBD + THC blocks the deleterious effects of THC may help explain the higher psychosis/schizophrenia risk among users of high-potency THC (and low CBD) cannabis.^{23,25,48,65} We chose to administer equal amounts of CBD and THC for these experiments, as this approximates the ratio of the acid forms of THC and CBD present in *Cannabis sativa* strains expressing both CBD and THC syntheses.⁶⁶ However, it will be quite interesting to determine the minimal amount of CBD that is protective.

Conclusions

Chronic THC during adolescence in male mice produced a selective spectrum of persistent behavioral consequences. Based on the rat literature, it is likely that treatment of female mice will also lead to persistent behavioral abnormalities.^{67–70} Other than late-emerging anxiety, these abnormalities were absent in similarly treated adult male mice. These findings reinforce the notion that adolescence is a vulnerable



developmental period, and that sustained exposure to THC during this time may have long-lasting and detrimental consequences on cognition and behavior. The finding that coadministration of equal amounts of CBD with THC prevented the emergence of all behavioral deficits supports the epidemiological finding that higher CBD content in cannabis reduces risk for psychotic disorders.⁴⁸ Furthermore, encouraging the cultivation and consumption of “balanced” CBD cannabis (i.e., roughly equal proportions of CBD and THC), over lower CBD cannabis, may be a reasonable strategy for harm reduction.⁷¹

Acknowledgments

We thank Ben Cornett for his careful mouse husbandry for this study. This study was funded by the NIH: DA021696 and DA039463.

Author Disclosure Statement

The authors declare no competing financial interests exist.

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Cite this article as: Murphy M, Mills S, Winstone J, Leishman E, Wager-Miller J, Bradshaw H, Mackie K (2017) Chronic adolescent Δ^9 -tetrahydrocannabinol treatment of male mice leads to long-term cognitive and behavioral dysfunction, which are prevented by concurrent cannabidiol treatment, *Cannabis and Cannabinoid Research* 2:1, 235–246, DOI: 10.1089/can.2017.0034.

Abbreviations Used

CBD = cannabidiol
DI = discrimination index
EPM = elevated plus maze
NOR = novel object recognition
OFA = open field assay
PFC = prefrontal cortex
THC = tetrahydrocannabinol

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