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SHORT REPORT

Oral Consumption of Cannabidiol During Pregnancy Alters Behavior in Mouse Offspring

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Abstract

Background: Pregnant people use cannabidiol (CBD) to treat nausea, insomnia, anxiety, and pain. However, CBD crosses the placenta and enters the fetal brain, where it can affect several targets important for brain development. While consumption of high doses of CBD during pregnancy has been shown to disrupt offspring neurodevelopment, pain sensitivity, and cognitive behavior in mice, lower doses have not been assessed.

Methodology: We administered 10 mg/kg/day CBD by oral gavage to pregnant C57Bl6 mice from embryonic day 5 through birth. We used the puzzle box, the forced swim test, and Hargreaves thermal sensitivity behavior tests and electrophysiology to determine how prenatal CBD exposure affects postnatal behavior and prefrontal cortex physiology.

Results: We show that oral consumption of 10 mg/kg/day CBD during pregnancy increases thermal pain sensitivity in male mouse offspring. Furthermore, the same dose impairs cognition and reduces excitability of the prefrontal cortex in female mouse offspring.

Conclusion: These data show that lower doses of CBD consumption during pregnancy can impair fetal brain development and postnatal behavior.

Keywords: CBD; cannabidiol; prenatal exposure; fetal exposure; behavior; brain development

Introduction

Pregnancy is accompanied by nausea, pain, anxiety, and insomnia.[1,2] To counter these effects, many self-medicate with cannabis due to its antiemetic and anti-anxiety properties.[1,2] Cannabis is composed of two primary cannabinoids, tetrahydrocannabinol (THC) and cannabidiol (CBD). THC is psychoactive while CBD is nonpsychoactive and is legal in many parts of the world.[3] Self-reported cannabis use increased from 6.80% to 12.50% among pregnant women from 2009 to 2017[4] and rose to 17% in the United States by 2024.[2] In addition to those who consume cannabis, 20% of pregnant women report using CBD-only products.[5] However, little is known about the consequences of prenatal CBD exposure on fetal development.

CBD crosses the placenta, reaches the fetal brain, and acts upon targets relevant to fetal development.[6] Several neurodevelopmental disorders have been associated with prenatal cannabis consumption, suggesting that CBD exposure could be partly responsible for behavioral outcomes.[7,8] Many of these behaviors are mediated by the prefrontal cortex (PFC), a brain region that importantly expresses CBD targets implicated in neuronal development.[9,10] This suggests that prenatal CBD exposure could impact brain development and postnatal behaviors.

We previously found that administration of 50 mg/kg/day CBD to pregnant mice dams results in increased thermal sensitivity in male, but not female, offspring and cognitive deficits in female, but not male, offspring.[10] The same dose resulted in reduced excitability of pyramidal neurons in the PFC of the female, but not male, CBD-exposed offspring.[10] Daily 10 mg/kg/day CBD is prescribed for human patients.[11]

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However, whether lower doses of CBD disrupt fetal brain development remained an open question. Here, we assess the consequences of a lower dose (10 mg/kg/day) on behavior and neuronal activity. We found that gestational exposure to 10 mg/kg/day CBD increased thermal sensitivity in male offspring and caused PFC-associated cognitive deficits and changes in excitability in female offspring. This suggests that prenatal exposure to lower doses of CBD results in adverse effects on fetal development.

Materials and Methods

CBD

CBD powder was obtained from the National Institute of Drugs of Addiction with Drug Enforcement Administration Schedule 1 drug license (#RB0605026). CBD powder was diluted to 2.5 mg/ml in pure sunflower oil in an amber glass container. The iC42 lab at the University of Colorado Anschutz Medical Campus confirmed purity, and concentration of diluted CBD by liquid chromatography-tandem mass spectrometry.

Mice

Eight-week-old female and male C57Bl6 (Jackson Labs, Maine, Strain #000664) mice were crossed. Upon observation of a vaginal plug (embryonic day 0.5), mice were individually housed to monitor weight gain and embryonic timepoint. Dams were administered between 60 and 175 microliters of 2.5 mg/mL CBD to reach 10 mg/kg/day or sunflower oil by oral gavage depending on dam weight from embryonic day 5 through birth. Dams were weighed daily after E5, and those that did not gain weight after E11 were removed from the study. Experiments were approved by the University of Colorado Institutional Animal Care and Use Committee (protocols #139 and 721). Male offspring were used for the thermal sensitivity test, while the same female offspring were used for both the forced swim and puzzle box tests.

Behavior

Thermal sensitivity. Given our previous findings that prenatal exposure to 50 mg/kg/day CBD affected thermal sensitivity in male but not female offspring,[10] we tested male mice exposed to 10 mg/kg/day CBD or vehicle for thermal sensitivity at 20 weeks of age using the Hargreaves test. The Hargreaves test measures latency to response in seconds to a calibrated heat source that is directed at the center of each mouse hindpaw.[12–14] Removal of the paw or flinching is recorded as a response. Three measurements per mouse were averaged.

Forced swim. At 14 weeks of age, female mice exposed to CBD or vehicle prenatally underwent the forced swim test.[15] The forced swim test is the most widely accepted model of depression-like behavior in mice. It is performed only once per animal. The characteristic behavior of the test, termed immobility, develops when a rodent is placed in a tank of water for an extended time and "makes only those movements necessary to keep its head above water."[16] Forced swim sessions are conducted by placing mice individually in large glass cylinders (45 cm height × 20 cm diameter) containing 24–25C° water approximately 20 cm deep. The mouse cannot escape or touch the bottom of the container. The mouse is placed in the cylinder for 6 min. Latency to float and amount of time spent struggling are measured. Trials are video recorded and later scored by a blind observer. During the entire duration of the task, the experimenter is present and watching the mice. If there is any indication that the animal is in danger of drowning, it is immediately removed from the cylinder and excluded from the study. At the end of the swim session, the mice are towel-dried and returned to their home cage. The water in the test arena is changed between each subject.[16] The Forced Swim test was performed 2 weeks after the Puzzle Box test, with the same female offspring.

Puzzle box. Female mice exposed to CBD or vehicle prenatally underwent the puzzle box test at 12 weeks of age. The puzzle box is performed in a Plexiglas white box divided by a removable barrier into two compartments: a brightly lit start zone (58 cm long, 28 cm wide) and a smaller covered goal zone (15 cm long, 28 cm wide). Mice are introduced into the start zone and are trained to move into the goal zone through a narrow underpass (∼4 cm wide) located under the barrier. Mice are motivated by their aversion to the bright light to reach the goal zone. Mice undergo nine trials (T1–T9) over 3 consecutive days in which they are challenged with increasingly difficult obstructions. This sequence assesses problem-solving abilities (T2, T5, and T8), learning/short-term memory (T3, T6, and T9), and repetition on the next day measures long-term memory (T4 and T7).[10,17]

Preparation of acute prefrontal cortex slices

Acute coronal PFC slices were obtained from postnatal day (P) 15 female mice prenatally exposed to either CBD (10 mg/kg/day) or vehicle. Mice were

anesthetized with isoflurane and euthanized by decapitation. Immediately after decapitation, the brain was extracted and placed in icy cutting solution containing (in mM) 215 sucrose, 20 glucose, 26 $NaHCO_3$, 4 $MgCl_2$, 4 $MgSO_4$, 1.6 NaH_2PO_4, 1 $CaCl_2$, and 2.5 KCl. Using a Leica VT1000S vibratome, the PFC was sectioned into 300 μm thick slices. PFC slices were incubated at 32°C for 30 min in 50% cutting solution and 50% artificial cerebrospinal fluid (ACSF) composed (in mM) of 127 NaCl, 25 $NaHCO_3$, 25 D-glucose, 2.5 KCl, 1.25 NaH_2PO_4, 2 $CaCl_2$, and 1 $MgCl_2$. After 30 min, this solution was replaced with ACSF at room temperature. For all electrophysiology experiments, the slices were placed in a recording chamber and bathed in recirculating ACSF aerated with 95% O_2/5% CO_2 at 30°C and allowed to equilibrate for at least 30 min prior to the start of experiments.

Electrophysiology

PFC layer 2/3 pyramidal neurons were identified by morphology and distance from layer 1 and patched in the whole-cell (8–10 MΩ electrode resistance; 20–40 MΩ series resistance) current clamp configuration (MultiClamp 700B, Molecular Devices). Using a potassium-based internal solution (in mM: 136 K-gluconate, 10 HEPES, 17.5 KCl, 9 NaCl, 1 $MgCl_2$, 4 Na_2-ATP, 0.4 Na-GTP; $\sim$300 mOsm, and $\sim$pH 7.26), spiking properties were examined at 30°C in recirculating ACSF with 2 mM $CaCl_2$ and 1 mM $MgCl_2$. Excitability was measured by injection of depolarizing current steps (25–250 pA, 300 ms). Resting membrane potential was recorded prior to the first depolarizing current step. The minimum current required to elicit an action potential was defined as the smallest current step triggering at least one spike.[10] For each neuron, two trains of current injections were delivered per current step as technical replicates and averaged before further analysis.

Statistics and reproducibility

The alpha value was set at 0.05. Datasets were compared using an unpaired two-tailed *t* test, Mann–Whitney test, or two-way ANOVA depending on normality and equal variance. For neuronal excitability and resting membrane potential, statistical comparisons were performed using individual neurons as independent biological units. For the puzzle box behavioral test, we used multiple *t*-tests to test for significance of individual trials and for overall performance of each mouse. Each trial tests a different cognitive ability (problem-solving at increasing difficulty, short-term memory, and long-term memory). Thus, trials were analyzed separately.

Results

10 mg/kg/day CBD exposure does not affect gestational length, litter size, or survival

To model human oral consumption, we administered 10 mg/kg of CBD dissolved in sunflower oil or sunflower oil alone (control) daily to C57BL6J pregnant dams from E5 to birth by oral gavage (Fig. 1A).[5] We quantified the gestational length, gestational weight gain, and size of litters (Fig. 1B). CBD consumption during pregnancy did not significantly change these litter factors.

10 mg/kg/day CBD prenatal exposure increases thermal pain sensitivity in male offspring

50 mg/kg/day of CBD during prenatal development led to heightened responses of male offspring to the Hargreaves test.[10] We tested whether prenatal 10 mg/kg CBD daily exposure was sufficient to elicit the same effect. We found that latency to respond to the thermal stimuli was significantly shorter in males exposed to CBD during pregnancy (Fig. 1C).

10 mg/kg/day CBD prenatal exposure does not result in depression-like behaviors in female offspring

We assessed whether female offspring exposed to 10 mg/kg/day CBD during pregnancy had increased depression-like behaviors in the forced swim depression test.[18] CBD-exposed females did not spend significantly more time inactive compared to controls (Fig. 1D).

10 mg/kg/day CBD prenatal exposure decreases problem-solving abilities in female offspring

Another possible long-term consequence of prenatal CBD exposure is its effects on cognitive abilities (problem-solving)[7,10] To evaluate whether the 10 mg/kg/day CBD exposure impacts problem-solving abilities, CBD-exposed and control female offspring underwent the puzzle box test. Female offspring exposed to 10 mg/kg/day CBD took significantly more time to reach the goal area in trial eight (Fig. 1E, left). CBD-exposed female offspring took more time to reach the goal area across trials (Fig. 1E, right).

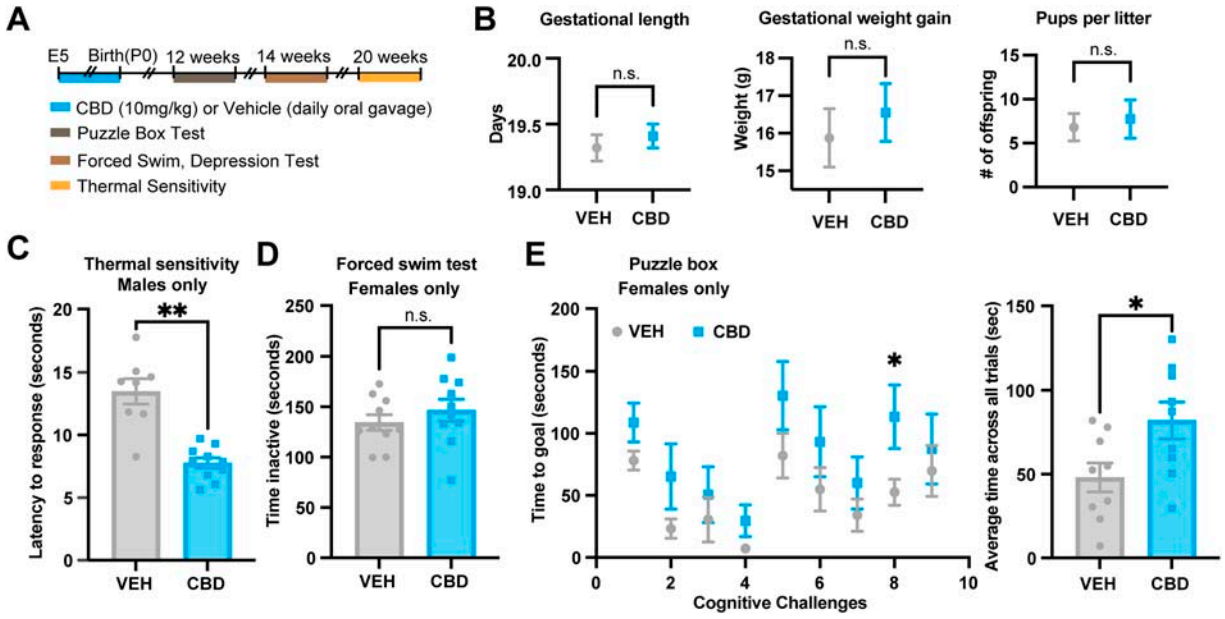

FIG. 1. Fetal CBD exposure alters postnatal behavior. **(A)** Experimental timeline for fetal CBD and vehicle treatment (daily oral gavage) and postnatal behavior paradigms. **(B)** Dams' gestational length ($p = 0.54$, *t*-test), gestational weight gain ($p = 0.57$, *t*-test), and litter size ($p = 0.296$, *t*-test) were unaffected by CBD (blue squares) compared to vehicle (gray circles) treatment (gestational length and weight, $n = 18$ vehicle and 11 CBD; litter size, $n = 10$ vehicle and 8 CBD). **(C)** The Hargreaves test displayed increased thermal sensitivity in fetal CBD exposed male offspring (vehicle: $n = 8$ male mice; CBD: $n = 10$ male mice; $p < 0.0001$, *t*-test). **(D)** The forced swim depression test showed no difference in vehicle and CBD treated female offspring (vehicle: n = 10 female mice; CBD: $n = 10$ female mice; $p = 0.38$, *t*-test). **(E)** Performance in puzzle box test was impacted in CBD exposed female offspring (vehicle: $n = 16$ female mice; CBD: $n = 16$ female mice; $p < 0.05$ by *t*-test for individual significant trials and overall performance). $^{*}p < 0.05$, $^{**}p < 0.01$; error bars represent SEM. n.s. not significant.

10 mg/kg/day CBD prenatal exposure decreases excitability of layer 2/3 pyramidal neurons in the PFC of female offspring

The PFC is a brain region critical for higher-order cognitive behaviors.[19,20] Given the effects of CBD on problem-solving behavior, we investigated the neuronal mechanisms underlying this impairment. We previously showed 50 mg/kg/day CBD decreased excitability of PFC layer 2/3 pyramidal neurons in female offspring.[10] To determine whether this effect remained following 10 mg/kg/day CBD exposure, we measured the intrinsic electrophysiological membrane properties of PFC layer 2/3 pyramidal neurons. CBD significantly decreased excitability and increased the minimum current required to trigger an action potential compared to controls without affecting the resting membrane potentials (Fig. 2C–E). 10 mg/kg/day CBD daily exposure during pregnancy is sufficient to reduce PFC neuronal excitability in female mouse offspring.

Discussion

We found that maternal consumption of 10 mg/kg/day CBD during pregnancy in mice increases thermal sensitivity in male offspring, while in female offspring it decreases problem-solving behaviors and reduces excitability of PFC layer 2/3 pyramidal neurons. These findings indicate that even low-dose prenatal CBD exposure produces neurobehavioral effects similar to those previously observed with high-dose exposure (50 mg/kg/day).[10]

Our previous work showed that prenatal exposure to 50 mg/kg/day CBD caused increased sensitivity to thermal pain dependent upon the TRPV1 receptor.[10] CBD desensitizes TRPV1 at lower doses but activates TRPV1 at high doses, presenting a question of whether

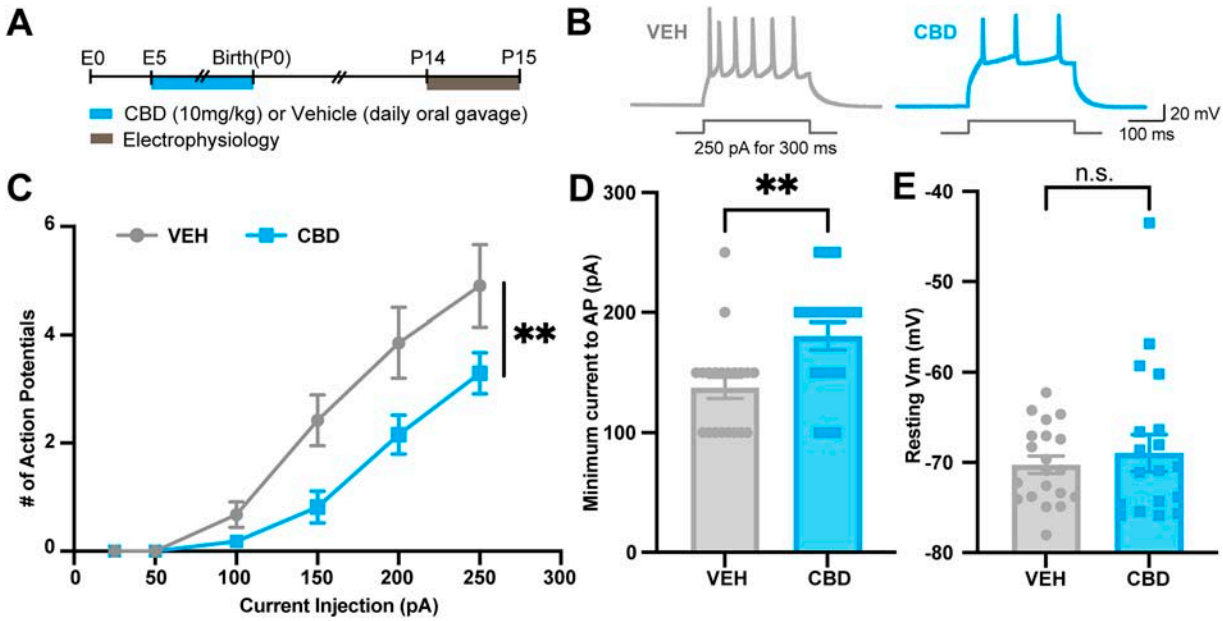

FIG. 2. Fetal CBD exposure decreases excitability of layer 2/3 pyramidal neurons in female offspring. **(A)** Experimental timeline of fetal CBD exposure (daily oral gavage) and postnatal electrophysiology. **(B)** Representative traces of an action potential train elicited by 250 pA current injection for 300 ms in vehicle (gray) and CBD (blue) exposed female offspring. **(C)** Fetal CBD exposure significantly reduced intrinsic excitability of PFC layer 2/3 pyramidal neurons compared to vehicle exposed controls (vehicle: $n = 20$ cells, 3 mice; CBD: $n = 19$ cells, 3 mice; treatment effect: $p < 0.0001$, 2-way ANOVA). **(D)** The minimum current required to elicit action potential spikes was increased in CBD exposed female offspring (vehicle: $n = 20$ cells, 3 mice; CBD: $n = 18$ cells, 3 mice; $p = 0.0049$, Mann–Whitney test). **(E)** Resting membrane potential was unaffected by CBD (vehicle: $n = 20$ cells, 3 mice; CBD, $n = 19$ cells, 3 mice; $p = 0.7919$, Mann–Whitney test). $^{*}p < 0.05$, $^{**}p < 0.01$; error bars represent SEM. n.s. not significant.

a lower dose of CBD would similarly affect thermal pain sensitivity in mouse offspring. Here we show that male mouse offspring exposed to 10 mg/kg/day CBD throughout gestation are more sensitive to thermal pain than control offspring, similar to when they were exposed to higher doses of CBD.

Our work adds to the body of evidence that prenatal CBD exposure impacts fetal neurodevelopment and postnatal behavior. Previous work has shown that maternal injections of 30 mg/kg/day CBD impair social and object recognition and increase anxiety-like behaviors in female rat offspring.[21] Furthermore, this same treatment decreased the firing frequency of pyramidal neurons in the PFC of rat offspring of both sexes.[21] These results are similar to our own findings, showing that a lower dose of CBD decreases excitability of pyramidal neurons of the PFC, but in female offspring only. Prenatal CBD exposure has been shown to affect communication, motor skills, and memory in early life for mouse offspring.[22,23] A recent study showed that fetal exposure to even 3 mg/kg/day CBD is sufficient to increase anxiety-like and reward-seeking behaviors in mice.[24] Our data adds to this work by showing that cognitive function and electrophysiological properties are altered in offspring exposed to 10 mg/kg/day CBD. Anxiety is typically comorbid with depression, but we found that our paradigm for fetal CBD exposure was not sufficient to induce anxiety-like behaviors in mice. Our results corroborate studies that show subcutaneous daily injection of 3 mg/kg CBD into pregnant mice does not significantly alter performance in the forced swim test in mouse offspring.[25]

The PFC is important for higher-order cognitive behaviors, and its function is crucial for brain

development.[19,20] CBD exposure may affect PFC development, potentially contributing to reduced problem-solving behavior in female offspring. One potential mechanism through which CBD could elicit this effect is through binding to serotonin 1A receptors (5-HT1ARs), which are highly expressed in the fetal PFC.[26–30] CBD activates 5-HT1ARs, and their activation decreases neuronal activity.[27,29,31–33] We observed that 10 mg/kg/day of CBD during fetal development decreases excitability of PFC layer 2/3 pyramidal neurons. Average resting membrane potential did not differ between groups, indicating it does not account for the reduced excitability. Thus, prenatal CBD exposure could activate 5-HT1ARs, resulting in a higher threshold for action potential firing in the PFC and decreased problem-solving abilities.[32,33] This part of our study focused on female offspring because previous work has shown that the effects of fetal CBD exposure on the central nervous system are more pronounced in female rodents.[10,22–24] One limitation of this study is that we did not monitor the estrus cycle of female offspring during behavioral tests, given that the estrus cycle can affect behavior. However, electrophysiological recordings were performed before the estrus cycle commences, suggesting that prenatal CBD exposure reduces excitability of the layer 2/3 pyramidal neurons of the PFC independent of estrus cycle contributions.

CBD has been shown to inhibit voltage-gated sodium and calcium channels.[34–37] Thus, fetal CBD exposure could directly affect neuronal membrane potential and electrical activity. Electrical activity directly effects transcription through calcium-activated transcription factors.[38–41] Furthermore, it mediates bone morphogenetic protein (BMP) release and downstream signaling, which is crucial for cortical development.[42–47] In addition, CBD inhibits G-protein coupled receptors, potentially disrupting axon guidance and altering neuronal connectivity.[34,48] While these mechanisms suggest that CBD consumption during pregnancy could impact cortical development, they remain speculative, as none were directly examined in the present study. Future research will be necessary to experimentally test these hypotheses and determine the mechanisms underlying the neurodevelopmental effects of prenatal CBD exposure, including potential sex-dependent effects.

Conclusion

These results show that prenatal exposure to a lower dose of CBD induces adverse neurodevelopmental effects in mice.

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Authors' Contributions

E.A.B. and W.C.O. conceived ideas and designed experiments. E.A.B. conducted thermal sensitivity behavior tests and analyzed all behavior data. L.G.W. and V.N.C. performed and analyzed electrophysiology experiments. V.N.C. wrote the first draft of most of the article. E.A.B., W.C.O., and L.G.W. edited and provided text for portions of the article.

Author Disclosure Statement

No competing financial interests exist.

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References

1. Volkow ND, Han B, Compton WM, et al. Self-reported medical and nonmedical cannabis use among pregnant women in the United States. JAMA 2019;322(2):167–169; doi: 10.1001/jama.2019.7982
2. Zaugg C, Terplan M, Mailman K, et al. Reasons pregnant people use cannabis to self-treat health conditions during pregnancy: Results from a US population-based survey. Drug Alcohol Rev 2024;43(7):1742–1752; doi: 10.1111/dar.13934
3. Stella N. THC and CBD: Similarities and differences between siblings. Neuron 2023;111(3):302–327; doi: 10.1016/j.neuron.2022.12.022
4. Young-Wolff KC, Sarovar V, Tucker LY, et al. Self-reported daily, weekly, and monthly cannabis use among women before and during pregnancy. JAMA Netw Open 2019;2(7):e196471-e196471; doi: 10.1001/jamanetworkopen.2019.6471
5. Bhatia D, Battula S, Mikulich-Gilbertson S, et al. Cannabidiol-Only product use in pregnancy in the United States and Canada findings from the international cannabis policy study. Obstet Gynecol 2024;144(2):156–159; doi: 10.1097/AOG.0000000000005603

6. Rokeby ACE, Natale BV, Natale DRC. Cannabinoids and the placenta: Receptors, signaling and outcomes. Placenta 2023;135:51–61; doi: 10.1016/j.placenta.2023.03.002
7. Paul SE, Hatoum AS, Fine JD, et al. Associations between prenatal cannabis exposure and childhood outcomes: Results from the ABCD study. JAMA Psychiatry 2021;78(1):64–76; doi: 10.1001/jamapsychiatry.2020.2902
8. Rompala G, Nomura Y, Hurd YL. Maternal cannabis use is associated with suppression of immune gene networks in placenta and increased anxiety phenotypes in offspring. Proc Natl Acad Sci U S A 2021;118(47):e2106115118; doi: 10.1073/pnas.2106115118
9. de Almeida DL, Devi LA. Diversity of molecular targets and signaling pathways for CBD. Pharmacol Res Perspect 2020;8(6):e00682; doi: 10.1002/prp2.682
10. Swenson KS, Gomez Wulschner LE, Hoelscher VM, et al. Fetal Cannabidiol (CBD) exposure alters thermal pain sensitivity, problem-solving, and prefrontal cortex excitability. Mol Psychiatry 2023;28(8):3397–3413; doi: 10.1038/s41380-023-02130-y
11. Devinsky O, Patel AD, Cross JH, et al.; GWPCARE3 Study Group. Effect of Cannabidiol on drop seizures in the lennox–gastaut syndrome. N Engl J Med 2018;378(20):1888–1897; doi: 10.1056/nejmoa1714631
12. Cheah M, Fawcett J, Andrews M. Assessment of thermal pain sensation in rats and mice using the hargreaves test. Bio Protoc 2017;7(16):e2506; doi: 10.21769/BioProtoc.2506
13. Bates EA, Nikai T, Brennan KC, et al. Sumatriptan alleviates nitroglycerin-induced mechanical and thermal allodynia in mice. Cephalalgia 2010;30(2):170–178; doi: 10.1111/j.1468-2982.2009.01864.x
14. Hargreaves K, Dubner R, Brown F, et al. A new and sensitive method for measuring thermal nociception in cutaneous hyperalgesia. Pain 1988;32(1):77–88; doi: 10.1016/0304-3959(88)90026-7
15. Can A, Dao DT, Arad M, et al. The mouse forced swim test. JoVE 2012(59):3638; doi: 10.3791/3638-v
16. Porsolt RD, Le Pichon M, Jalfre M. Depression: A new animal model sensitive to antidepressant treatments. Nature 1977;266(5604):730–732; doi: 10.1038/266730a0
17. O'Connor AM, Burton TJ, Leamey CA, et al. The use of the puzzle box as a means of assessing the efficacy of environmental enrichment. J Vis Exp 2014;94:e52225; doi: 10.3791/52225
18. Castagné V, Moser P, Roux S, et al. Rodent models of depression: Forced swim and tail suspension behavioral despair tests in rats and mice. Curr Protoc Pharmacol 2010;Chapter 5(1):Unit 5.8–Unit 5.8.14; doi: 10.1002/0471141755.ph0508s49
19. Yan Z, Rein B. Mechanisms of synaptic transmission dysregulation in the prefrontal cortex: Pathophysiological implications. Mol Psychiatry 2022;27(1):445–465; doi: 10.1038/s41380-021-01092-3
20. Menon V, D'Esposito M. The role of PFC networks in cognitive control and executive function. Neuropsychopharmacology 2022;47(1):90–103; doi: 10.1038/s41386-021-01152-w
21. DeVuono MV, Nashed MG, Sarikahya MH, et al. Prenatal tetrahydrocannabinol and cannabidiol exposure produce sex-specific pathophysiological phenotypes in the adolescent prefrontal cortex and hippocampus. Neurobiol Dis 2024;199:106588; doi: 10.1016/j.nbd.2024.106588
22. Iezzi D, Caceres-Rodriguez A, Chavis P, et al. In utero exposure to cannabidiol disrupts select early-life behaviors in a sex-specific manner. Transl Psychiatry 2022;12(1):501; doi: 10.1038/s41398-022-02271-8
23. Compagno MK, Silver CR, Cox-Holmes A, et al. Maternal ingestion of Cannabidiol (CBD) in mice leads to sex-dependent changes in memory, anxiety, and metabolism in the adult offspring, and causes a decrease in survival to weaning age. Pharmacol Biochem Behav 2025;247:173902; doi: 10.1016/j.pbb.2024.173902
24. Iezzi D, Cáceres-Rodríguez A, Chavis P, et al. Sex-specific disruptions in the developmental trajectory of anxiety-like behaviors due to prenatal cannabidiol exposure. Transl Psychiatry 2025;15(1):354; doi: 10.1038/s41398-025-03517-x
25. Maciel IdS, Abreu G D, Johnson CT, et al. Perinatal CBD or THC exposure results in lasting resistance to fluoxetine in the forced swim test: Reversal by fatty acid amide hydrolase inhibition. Cannabis Cannabinoid Res 2022;7(3):318–327; doi: 10.1089/can.2021.0015
26. Bar-Peled O, Gross-Isseroff R, Ben-Hur H, et al. Fetal human brain exhibits a prenatal peak in the density of serotonin 5-HT1A receptors. Neurosci Lett 1991;127(2):173–176; doi: 10.1016/0304-3940(91)90787-t
27. Bonnin A, Peng W, Hewlett W, et al. Expression mapping of 5-HT1 serotonin receptor subtypes during fetal and early postnatal mouse forebrain development. Neuroscience 2006;141(2):781–794; doi: 10.1016/j.neuroscience.2006.04.036
28. Sargin D, Jeoung HS, Goodfellow NM, et al. Serotonin regulation of the prefrontal cortex: Cognitive relevance and the impact of developmental perturbation. ACS Chem Neurosci 2019;10(7):3078–3093; doi: 10.1021/acschemneuro.9b00073
29. Goodfellow NM, Benekareddy M, Vaidya VA, et al. Layer II/III of the prefrontal cortex: Inhibition by the serotonin 5-HT1A receptor in development and stress. J Neurosci 2009;29(32):10094–10103; doi: 10.1523/JNEUROSCI.1960-09.2009
30. Altieri SC, Garcia-Garcia AL, Leonardo ED, et al. Rethinking 5-HT1A receptors: Emerging modes of inhibitory feedback of relevance to emotion-related behavior. ACS Chem Neurosci 2013;4(1):72–83; doi: 10.1021/cn3002174
31. Béïque JC, Campbell B, Perring P, et al. Serotonergic regulation of membrane potential in developing rat prefrontal cortex: Coordinated expression of 5-hydroxytryptamine (5-HT)1A, 5-HT2A, and 5-HT7 receptors. J Neurosci 2004;24(20):4807–4817; doi: 10.1523/JNEUROSCI.5113-03.2004
32. Alexander C, Jeon J, Nickerson K, et al. CBD and the 5-HT1A receptor: A medicinal and pharmacological review. Biochem Pharmacol 2025;233:116742; doi: 10.1016/j.bcp.2025.116742
33. Russo EB, Burnett A, Hall B, et al. Agonistic properties of cannabidiol at 5-HT1a receptors. Neurochem Res 2005;30(8):1037–1043; doi: 10.1007/s11064-005-6978-1
34. Bates E, Oh WC. Molecular targets of cannabidiol warn against its consumption during pregnancy. MRAJ 2024;12(5); doi: 10.18103/mra.v12i5.5450
35. Harding EK, Souza IA, Gandini MA, et al. Differential regulation of Cav3.2 and Cav2.2 calcium channels by CB1 receptors and cannabidiol. Br J Pharmacol 2023;180(12):1616–1633.
36. Huang J, Fan X, Jin X, et al. Cannabidiol inhibits Nav channels through two distinct binding sites. Nat Commun 2023;14(1):3613.
37. Ghovanloo MR, Shuart NG, Mezeyova J, et al. Inhibitory effects of cannabidiol on voltage-dependent sodium currents. J Biol Chem 2018;293(43):16546–16558.
38. Iacobas DA, Iacobas S, Lee PR, et al. Coordinated activity of transcriptional networks responding to the pattern of action potential firing in neurons. Genes (Basel) 2019;10(10):754; doi: 10.3390/genes10100754
39. Tyssowski KM, DeStefino NR, Cho JH, et al. Different neuronal activity patterns induce different gene expression programs. Neuron 2018;98(3):530–546.e11; doi: 10.1016/j.neuron.2018.04.001
40. Lee PR, Cohen JE, Iacobas DA, et al. Gene networks activated by specific patterns of action potentials in dorsal root ganglia neurons. Sci Rep 2017;7(1):43765; doi: 10.1038/srep43765
41. Yap EL, Greenberg ME. Activity-Regulated transcription: Bridging the gap between neural activity and behavior. Neuron 2018;100(2):330–348; doi: 10.1016/j.neuron.2018.10.013
42. Dahal GR, Rawson J, Gassaway B, et al. An inwardly rectifying K+ channel is required for patterning. Development 2012;139(19):3653–3664; doi: 10.1242/dev.078592
43. Follmer ML, Isner TJ, Ozekin YH, et al. Depolarization induces calcium-dependent BMP4 release from mouse embryonic palate mesenchymal cells. Nat Commun 2024;15(1):9806; doi: 10.1038/s41467-024-53642-2
44. Dahal GR, Pradhan SJ, Bates EA. Inwardly rectifying potassium channels influence Drosophila wing morphogenesis by regulating Dpp release. Development 2017;144(15):2771–2783; doi: 10.1242/dev.146647
45. Berke B, Wittnam J, McNeill E, et al. Retrograde BMP signaling at the synapse: A permissive signal for synapse maturation and activity-dependent plasticity. J Neurosci 2013;33(45):17937–17950; doi: 10.1523/JNEUROSCI.6075-11.2013
46. Samanta J, Kessler JA. Interactions between ID and OLIG proteins mediate the inhibitory effects of BMP4 on oligodendroglial differentiation. Development 2004;131(17):4131–4142; doi: 10.1242/dev.01273

47. Mukhopadhyay A, McGuire T, Peng CY, et al. Differential effects of BMP signaling on parvalbumin and somatostatin interneuron differentiation. Development 2009;136(15):2633–2642; doi: 10.1242/dev.034439
48. Cherif H, Argaw A, Cécyre B, et al. Role of GPR55 during axon growth and target innervation. eNeuro 2015;2(5):ENEURO.0011–15.2015; doi: 10.1523/ENEURO.0011-15.2015

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Abbreviations Used

5-HT1ARs = serotonin 1A receptors
BMP = bone morphogenetic protein
CBD = Cannabidiol
cm = centimeter
DEA = Drug Enforcement Administration
Kg = kilogram
LC-MS/MS = liquid chromatography-tandem mass spectrometry
Mg = milligram
NIDA = National Institute of Drugs of Addiction
PFC = prefrontal cortex
T = trial
THC = THC
TRPV1 = transient receptor potential vanilloid 1