The Effects of Delta-8-tetrahydrocannibinol on *Danio rerio*

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Abstract

Background: Delta-8-tetrahydrocannabinol (THC), although less potent than THC, can reduce memory cognition and anxiety. However, Delta-8 can have adverse effects at high doses, like inducing anxiety, confusion, and hallucinations. The brain of *Danio rerio* (zebrafish) resembles human neuroanatomical and neurochemical pathways and demonstrates robust behavioral phenotypes, making them an excellent model organism for memory and anxiety.

Methods: Subjects will be fed Delta 8-THC before entering the experimental apparatus. Fish for both memory and anxiety tests will be recorded through Noldus Ethovision Software. For testing memory, ten male and female zebrafish will be individually placed in a three-compartment memory tank. One side of the memory tank will have a star shape, while the other will have a circle. Commercial fish flakes will be administered to the fish on the circle side which will be on the left in the first trial, then on the right in the second trial. Correct responses will be when the fish are present on the side of the tank used to present the fish flakes, and the fish will be rewarded with shrimp brine. We hypothesize that there will be fewer correct responses for fish fed with Delta-8-THC than the control. For testing anxiety, zebrafish will be fed Delta 8-THC before entering the diving tank. Zebrafish experiencing anxiety will stay at the bottom of the diving tank, but if they are relaxed, they will be at the top. We hypothesize that fish fed with Delta-8-THC will spend more time at the top of the diving tank than the control.

Results: We expect to see fewer correct responses in the memory tasks for the Delta 8 zebrafish in comparison to the control group. We also expect to see the experimental zebrafish stay at the top of the diving tank for a longer time than those in the control group. The treated zebrafish will have less mobility and a lower velocity than those in the control group.

Conclusion: We hypothesize that there will be fewer correct responses for fish fed with Delta-8-THC than the control. We hypothesize that fish fed with Delta-8-THC will spend a higher percentage of time at the top of the diving tank than the control. The treated zebrafish should have more mobility and spend more time in the upper zone than those in the control group, indicating lower anxiety.

Introduction

Cannabis sativa has been used medicinally and recreationally by humans and remains the most commonly used drug in the United States. Over 40% of the cannabis plants grown in the United States are cultivated in the Appalachia region, with Kentucky being a significant producer. The Food and Drug Association (FDA) has approved at least three synthetic formulations for medicinal use in treating nausea and vomiting associated with chemotherapy and anorexia associated with AIDS. However, most recreational users indulge in cannabis for relaxation, including victims of posttraumatic stress disorder (PTSD). Delta9-tetrahydrocannabiniol (THC) is a major psychoactive cannabinoid with anti-anxiolytic and memory reducing properties. It has been found that in low doses, THC can enhance reduce anxiety responses. Despite its therapeutic properties, delta9-THC can cause impairments in short term memory. The negative effects on memory of delta9-THC are mediated by cannabinoid type-1 (CB1) receptors. The CB1 selective antagonist is used to block the memory-impairing effects of delta9-THC.

The zebrafish is becoming a promising tool for modeling human brain disorders in the disciplines of biology and pharmacology, including neuroscience and genetics. The first study that used zebrafish was published in 1954 and their use has been steadily increasing since. Over 80% of genes associated with human disease have a zebrafish counterpart. Testing with zebrafish is also substantially simpler and cheaper exercise than with laboratory mammals. The brain of the zebrafish is similar to the basic neuroanatomical layout of other vertebrates. Its neurotransmitter systems are also comparable to mammals. Clinical drugs created for humans are also effective in zebrafish acting through similar and/or identical biochemical pathways.

Delta8-THC is a less potent version of Delta9, and the lack of studies on it drove our interest in testing the substance in zebrafish. In addition, delta-8-THC is more widely available in the United States since it is not prohibited by laws that ban the use of delta-9-THC and/or marijuana.

Methods

3 Compartment Tank

Diving Tank with Zebrafish

Subjects will be fed .3 mg/kg of Delta 8-THC before entering the experimental apparatus. Fish for both memory and anxiety tests will be observed and recorded through Noldus Ethovision Software. For testing memory, five female and five male zebrafish will be individually placed in a three-compartment memory tank. One side of the memory tank will have a star shape, while the other will have a circle shape. About .5mg of commercial fish flakes will be administered to the fish on the circle side iwhich will be on the left n the first trial, then on the right side in the second trial, and continue on either side for 20 trials. Correct responses will be scored when the fish are present on the side of the tank used to present the fish flakes. These correct responses will be compared to those of the control group, who will complete the same tasks but without Delta 8. For testing anxiety, ten male and female zebrafish will be fed .3 mg/kg of Delta 8-THC before entering the diving tank. Typically, zebrafish experiencing anxiety will stay at the bottom of the diving tank, but if they are relaxed, they will be at the top. The experimental zebrafish will be compared to the control regarding which zone of the tank they spend most of their time in.

Results
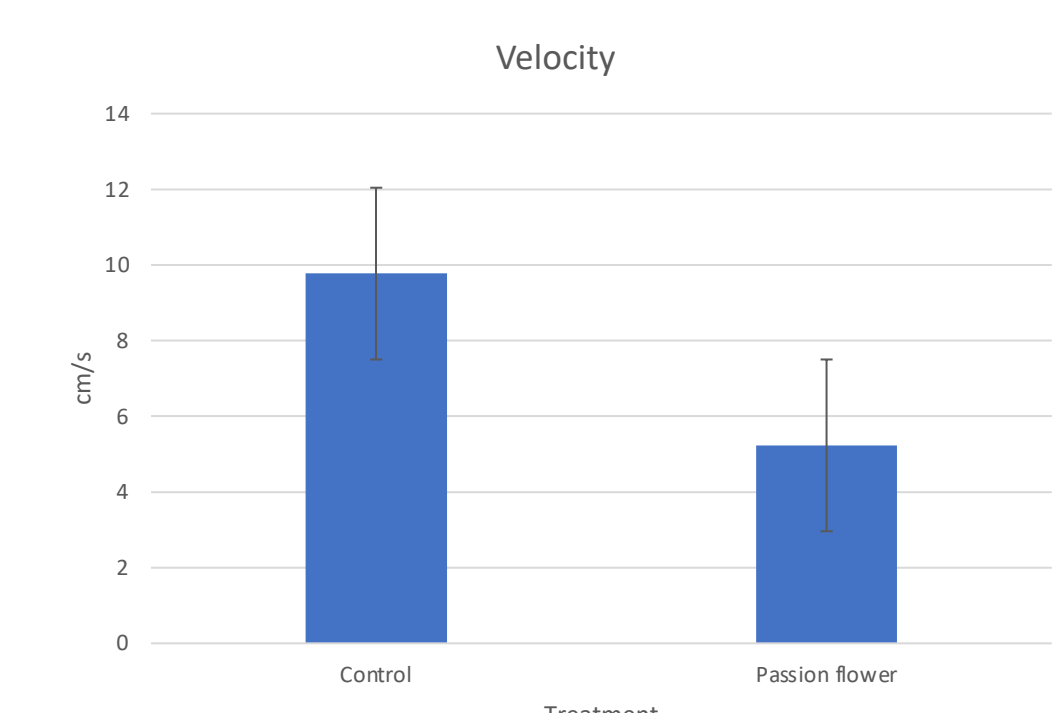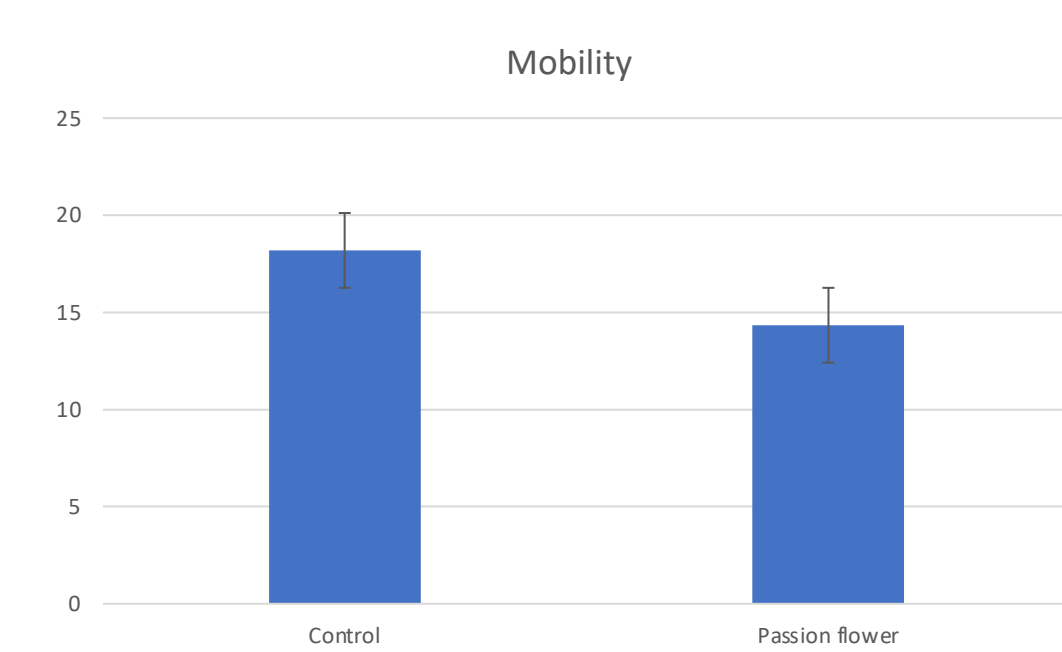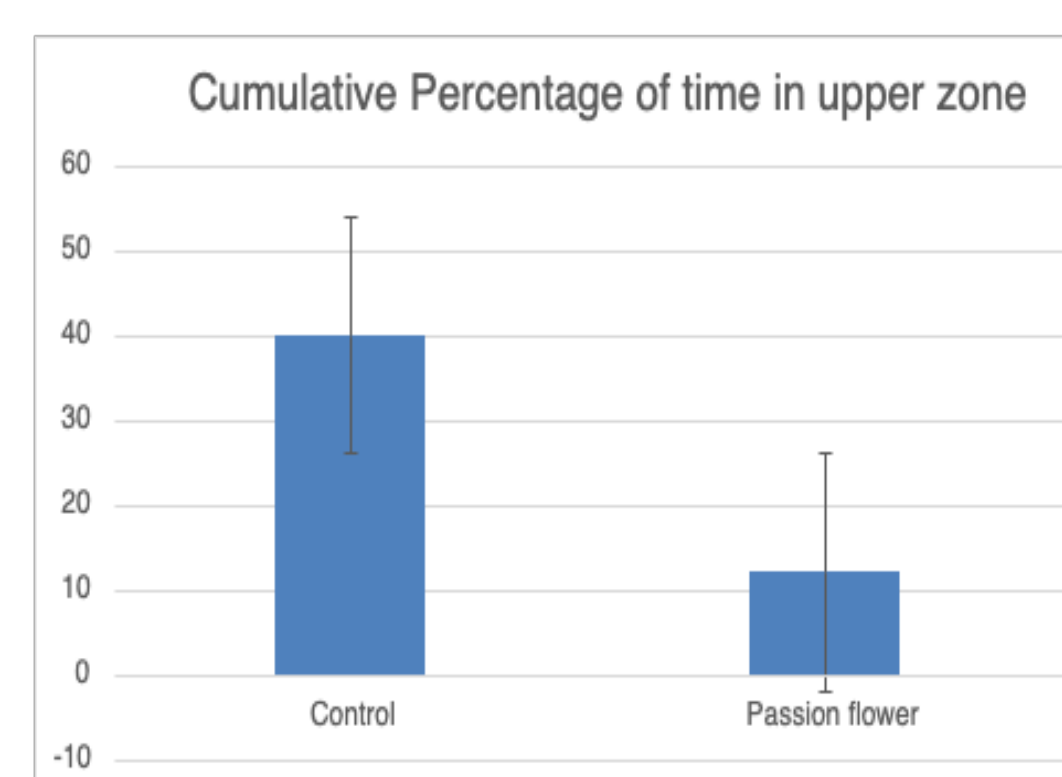
We expect to see fewer correct responses in the memory tasks for the Delta 8 zebrafish in comparison to the control group. We also expect to see the experimental zebrafish stay at the top of the diving tank for a longer time than those in the control group. Previous research from our lab has been done with passion flowers that support these predictions. On average, the control fish spent 9.8% of time in the upper zone with a standard deviation of 32.2. The fish treated with passion flower spent a lower percentage of time in the top zone in comparison to the control fish. The passion flower zebrafish had a lower velocity and less mobility compared to the control. The anxiety was overwhelming, causing the fish to freeze and become hesitant to move. Understanding the mechanism for Delta8-THC, we expect the opposite results.

Discussion

Discussion: We expect a statistically significant relationship between Delta8-THC and anxiety and memory decline. We expect the zebrafish treated with Delta8-THC to spend a longer time in the upper zone than the control group and to have more motility and higher velocity.

Limitations: We are testing in fish and not in humans. We expect similar effects in fish and humans, but we are unsure how pure the substance that we purchased will be. We plan to use HPTLC to investigate if the substance is contaminated or contains traces of Delta9-THC.

Future Studies: Delta9-THC in high doses has shown to have adverse effects, including inducing anxiety. Further research should investigate the effects of Delta8-THC in high doses on anxiety levels. Further research should also investigate the effects of Delta8-THC on psychosis-like behavior. Studies have shown that high doses of Delta9 can increase the risk of psychosis in humans.

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