The T cell co-receptor CD5 alters mouse behavior and gut microbiome composition

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Abstract

Behavior is affected by psychological and biological factors. The central nervous system, immune system, and gut microbiota work together to affect our mental health and cognitive behaviors. Immune cell development is influenced by the gut microbiota. T cells help regulate immune responses to foreign microbes. T cell activation is enhanced or inhibited by co-receptor proteins on the cell membrane, which can dramatically affect T cell function and immune responses. CD5, a T cell co-receptor, modulates T cell activation by inhibiting T cells from being able to be activated by self-proteins in order to prevent potential autoimmune effects. Our project studies the CD5 protein co-receptor and its effects on the microbiota, immune response, and central nervous system by testing mice that have the protein (CD5WT), mice that don't have the protein (CD5KO) and their progeny. *We hypothesize that CD5 alters the gut microbiota which, in turn, alters the cognitive behavior of these mice.* To investigate this hypothesis, we will conduct a series of behavioral tests based on both genotype and microbiome composition. This project will bring greater understanding to the intersection of the immune system, gut microbiota, and the central nervous system.

Introduction

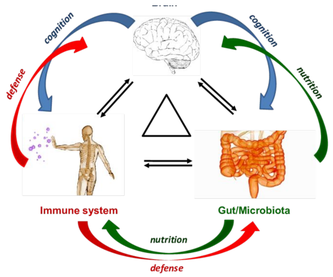

Figure 1. Diagram showing the relationship between the Central Nervous System, Gut Microbiota, and the Immune System

The central nervous system, the immune system, and the gut microbiota all work together to keep us functioning properly. Early in development, neonates are exposed to maternal microbes through the birth canal and begin gut microbiota colonization. Bacterial diversity in the gut microbiota is regulated through the activation state of the immune system and T cells specifically. CD5, a T cell co-receptor, is responsible for modulating T cell responses by inhibiting activation by self-peptides and regulating mature immune responses to the environment. Mice lacking the CD5 co-receptor (CD5KO) have shown increased activation profiles. We hypothesize that the co-receptor CD5 has a direct relationship with the gut microbiota and, in turn, affects cognitive behavior in mice. We will continue our investigation of CD5 by performing a variety of behavioral tests on co-housed CD5KO and CD5WT mice to determine the influence of the microbiome on their behavior.

Methodology

The marble-burying test measures OCD and anxiety-like behaviors. Each mouse is individually placed inside a cage that is filled with 4-cm of bedding throughout. On top of the bedding layer, we will place 20 evenly spaced marbles and allow each mouse to be in the cage for 10 minutes. After testing, a photograph will be taken of the cage and the amount of marbles buried 2/3rds of the marble size or more will be counted. 8-10 lab assistants will also count based on the picture taken. Mice with more anxiety and OCD-like behaviors bury more marbles throughout the test.

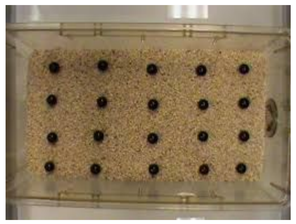

Figure 2. Image showing the the mouse cages prior to the marble burying test.

The open field test measures anxiety-like behaviors because mice generally distrust open spaces. Each mouse will have 10 minutes inside a square white PVC arena. A camera above the arena/box will track where the mice spend a majority of their time, duration in each spot, speed of travel, and duration of travel. Mice with more anxiety spend less time in the center of the arena and move less throughout the arena.

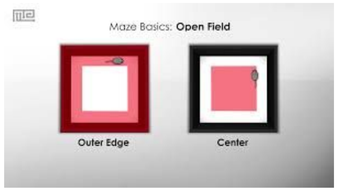

Figure 3. Representation of the gates to analyxe movement and position of mice within the open field arena, center and edge spaces.

An elevated plus maze experiment was also performed in order to measure fear based behaviors. A plus shaped maze is suspended off the ground. Two of the arms of the plus have are enclosed with walls while two arms are not enclosed at all. Each mouse is tested for a period of 10 minutes in the maze. The data is also recorded and analyzed in the same manner as the open field test. The results of our data show that there is no significant change in behavior due to fear.

Results

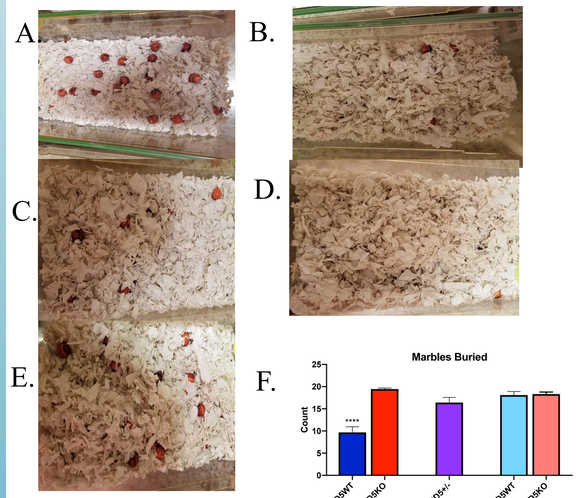

Figure 4. CD5KO Mice demonstrate OCD like behaviors during marble burying test. A) CD5WT mice bury very few marbles demonstrating less anxiety-like behaviors. B) CD5KO. C) Heterozygous mice. D) F2 positive mice. E) F2 Negative mice. F) CD5KO mice bury a statistically significant less amount of marbles than the CD5WT, demonstrating a more anxiety-like behavior. Heterozygous and F2 generation show no significant differences from the CD5KO.

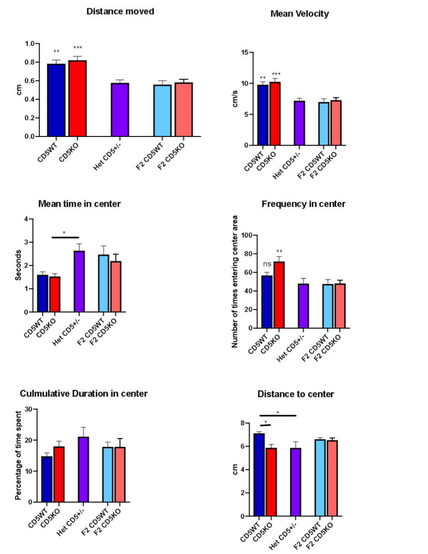

Figure 5. Open Field Test results. The distance moved, mean velocity, and the mean time in center show htat there is no significant difference between the WT and KO but that both WT and KO have significant differences from the other genotypes. It seems as though the specific genotype does not regulate the behavior in these figures but it could potentially be regulated by the gut-microbiome.

Conclusion and Future Directions

- When mice do not have the gene for CD5 (CD5KO) they exhibit behaviors parallel with increased OCD.
- It is still unknown if CD5 directly affects the Central Nervous System or if CD5 affects the gut-microbiome, which then affects the central nervous system. Figure 4F suggests that the gut-microbiome may play the biggest role in cognitive behaviors, this is because the co-housed F2 mice (having the same microbiome) gave similar results in their tests.
- We will be running more rounds of more tests to confirm our findings from past experiments.
- Later, we will also be treating the mice with antibiotics that will block the communication between the gut-microbiome and the central nervous system. We hypothesize that further behavioral tests after antibiotic treatment will result in a normalization of behaviors due to a decreased microbiota influence on the central nervous system.
- Introduction of a new 3 chamber socialization test to measure aggressive behaviors when mice are introduced to new objects both animate and inanimate.

Figure 6. Future directions. A) image of the 3 chamber socialization test. B) Antibiotics.

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