

Jimmy M. Pham

L2 Medical Student

LSU Health Science Center, New Orleans, LA

Mentor: Tiffany Wills, Ph. D

Louisiana State University Health Sciences Center, Department of Cell Biology and Anatomy

“Protein Kinase C Delta Expression in the Bed Nucleus of the Stria Terminalis in Adolescent Mice Exposed to Intermittent Ethanol”

Background: The adolescence phase is a critical period of neural development. Alcohol use during this period can damage and produce long-lasting changes in the brain, including increased risk for the later development of an alcohol use disorder. Recent research suggests a bidirectional interaction of alcohol use and pain. Previously published data from our lab reports a long-lasting mechanical and thermal hypersensitivity after Adolescent Intermittent Ethanol (AIE) exposure. To investigate this persistent hypersensitivity after AIE, the aim of this study will investigate the neuronal population of Protein Kinase C delta (PKCd) positive cells in the Bed Nucleus of the Stria Terminalis (BNST), a brain region known to mediate the perception of pain.

Method: Postnatal day (PND) 28 – 39 C57BL/6J male and female mice were treated with volatilized ethanol or water vapor. This gold standard paradigm for AIE exposure consists of a two 4-day cycle of ethanol vapor (16 hours in vapor chamber and 8 hours outside of chamber per day). These two cycle were separated by a 3-day no alcohol outside of chamber. Adolescence mice's blood alcohol level was maintained around 200 mg/dL range. Mechanical hypersensitivity was assessed by von Frey at five different time point (24 hours, 7, 14, 21, and 28 days) post-vapor exposure. Hargraves, a test for thermal hypersensitivity was conducted. On day 28, thirty minutes after the final von Frey assessment, mice's brain tissue was collected after decapitation. The obtained brains were flash-frozen and sliced at 14uM for RNAscope assay. RNAscope was performed using probes for PKCd and mouse c-fos genes as molecular markers.

Result: AIE induced thermal hypersensitivity in male mice and mechanical hypersensitivity in both males and female mice. Mice exposed to AIE showed a strong trend towards increased activation of the dorsolateral BNST cells, as well as increased in activation of PKCd cells. No significant differences were found for the oval BNST region.

Conclusion: These findings suggest that a history of AIE may alter the activation of the PKCd cells in the dorsolateral BNST of males, potentially driving thermal hypersensitivity.

Future direction: Increase the number of animals/group and use chemogenetic approaches to selectively manipulate PKCd-expressing cells in the dorsolateral BNST and evaluate changes in thermal hypersensitivity.