# Examining The Role of Ventral Pallidal Inputs In Stress-induced Social Avoidance

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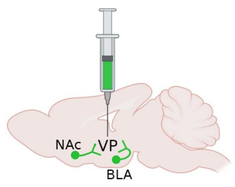
## Background

- The ventral pallidum (VP) is a structure within the reward pathway that processes rewarding and aversive signals. The VP receives inputs from regions such as the nucleus accumbens (NAc) and the basolateral amygdala (BLA) and projects to downstream regions to regulate motivated behaviors[1]. Behaviors related to depression, including social avoidance, have been linked to VP circuit activity.
- Chronic social defect stress (CSDS) has been shown to produce negative affective behavioral states in mice that are in line with depressive symptomatology in humans[2].
- It is not yet understood how VP inputs mediate stress-induced social avoidance. We aim to verify VP inputs and determine the impact of VP-projecting NAc & BLA neuronal activity during social interaction and assess whether CSDS impairs activity of VP inputs.

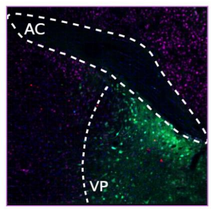
## Methods

- Viral retrograde labeling methods and immunohistochemistry was used to examine the changes in Fos expression within distinct VP circuits in male mice following a social interaction test. Fos was used as a measure of neuronal activity as its expression correlates with neuronal firing and has been shown to increase after SDS[3].
- After sample sectioning, we stained tissue from the VP, NAc, and BLA for DAPI, NeuN, c-FOS, and GFP (from Rg-GFP AAV)

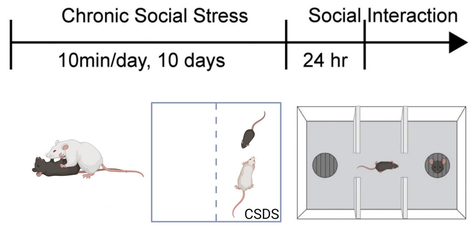
AAV Infusions



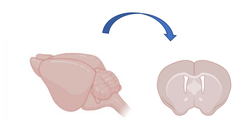

Expression at Injection Site




Behavioral Paradigm




Tissue Processing and Protein Detection



Perfusions, tissue fixation & sectioning

IHC & confocal microscopy

## Results

### Individual Differences in Social Interaction within Male Mice Following Chronic Social Defeat Stress

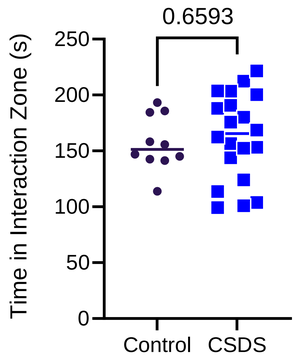



**Fig. 1A** There was no significant difference between control and CSDS mice in time spent interaction zone (un-paired t-test; t (28) = 0.4457, p = 0.6593).

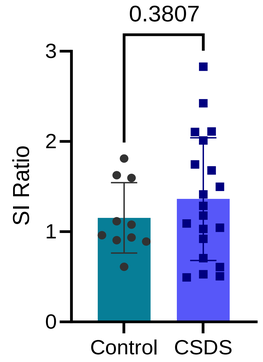



**Fig. 1B** There was no significant difference between control and CSDS mice in SI ratios (unpaired T-test; t (28) = 0.8906, p =0.3807).

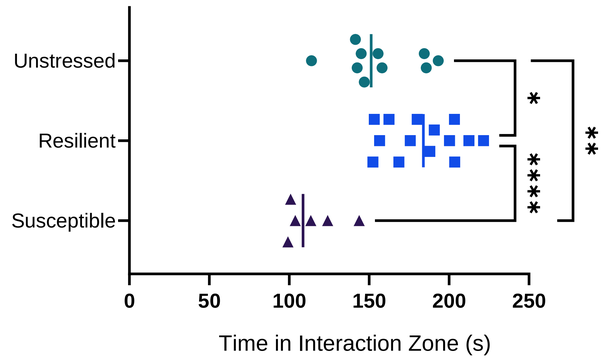



**Fig. 1C** Susceptible mice (*n*=6) spent significantly less time in the interaction zone compared to resilient (*n*=14) and control mice (*n*=10). A one-way ANOVA was performed to compare interaction times between groups, F (2, 27) = 20.05; p <0.0001.

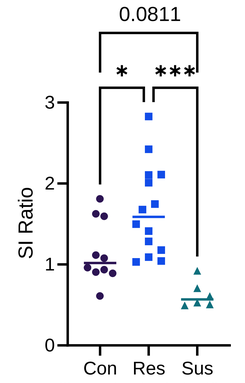



**Fig. 1D** Resilient mice had significantly higher SI ratios compared to control and susceptible mice. Susceptible mice had significantly lower SI ratios compared to resilient mice. A one-way ANOVA was conducted to compare groups, F (2, 27) = 11.94, p = 0.0002

### Detecting Fos Expression within VP-Projecting Neurons in the Nucleus Accumbens and Basolateral Amygdala

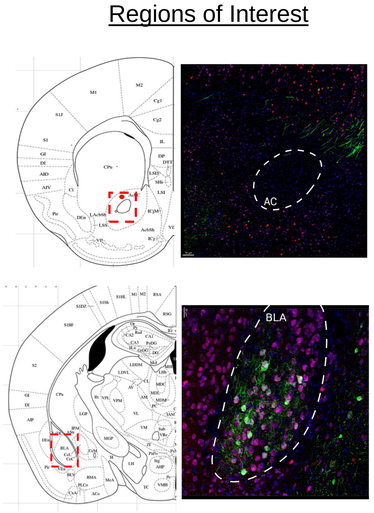



**Fig. 2A** Drawing from the mouse brain atlas with corresponding labeled sections of the NAc (top) and BLA (bottom)

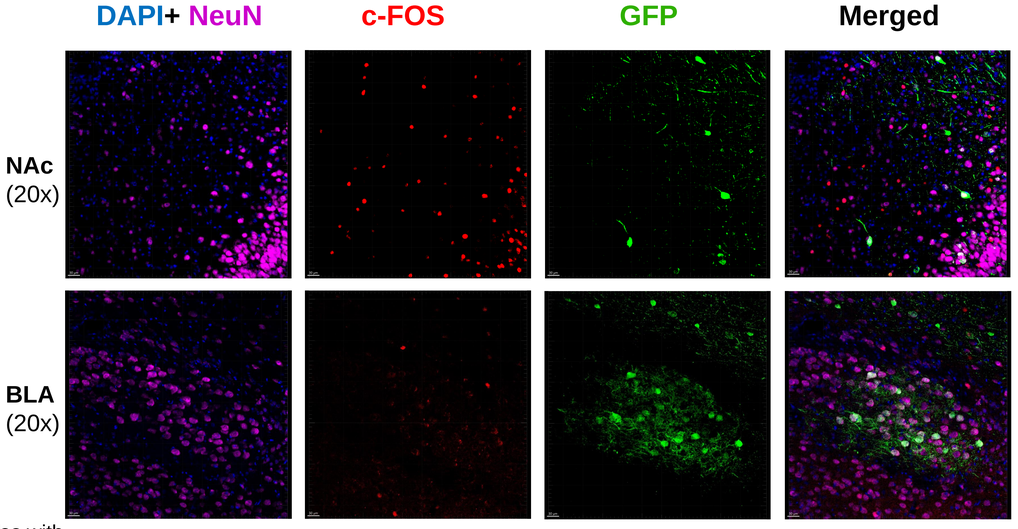



**Fig. 2B** Representative images of GFP, cFOS, neuronal marker NeuN expression with nuclear DAPI stain in the NAc (top; scale bar 30 μm) and BLA (bottom; scale bar 30 μm).

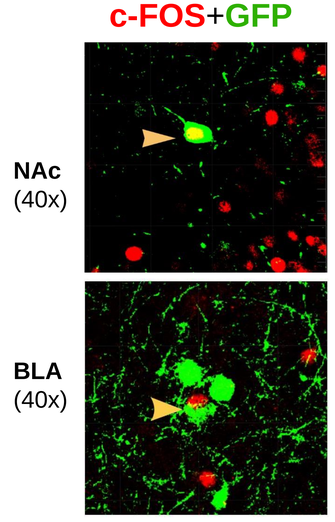



**Fig. 2C** Higher magnification of Rg-GFP (green) and c-FOS (red) expression in the BLA and NAc.

## Conclusions & Future Directions

- Continue analysis to identify Fos expression differences between control, resilient and susceptible mice.
- Higher c-FOS expression within the VP-projecting BLA neurons may indicate increased activity within this circuit.
- Characterization of cell populations in the neural circuits using RNAscope to identify specific markers (vGluT2, GAD).

## References

**1.** Wulff AB, Tooley J, Marconi LJ, Creed MC. Ventral pallidal modulation of aversion processing. *Brain Res*. 2019;1713:62-69. doi:10.1016/j.brainres.2018.10.010 **2**. Fox ME, Lobo MK. The molecular and cellular mechanisms of depression: a focus on reward circuitry. *Mol Psychiatry*. 2019;24(12):1798-1815. doi:10.1038/s41380-019-0415-3 **3**. Martinez M, Calvo-Torrent A, Herbert J. Mapping brain response to social stress in rodents with c-fos expression: a review. *Stress*. 2002;5(1):3-13. doi:10.1080/10253890290012369 **4.** Franco D, Wulff AB, Lobo MK, Fox ME. Chronic Physical and Vicarious Psychosocial Stress Alter Fentanyl Consumption and Nucleus Accumbens Rho GTPases in Male and Female C57BL/6 Mice. *Front Behav Neurosci*. 2022;16:821080. Published 2022 Feb 10. doi:10.3389/fnbeh.2022.821080