Behavioral and MRI neuroimaging effects of short-term intermittent hypoxia and psilocybin treatment in awake mice

R. AHUJA[1,2], K. BALAJI[1,2], A. KOGANOV[1,2], E. BENNET[1,2], T. NELSON[1,2], L. WILLIAMSON[2], M. A. GITCHO[3], S. HATFIELD[2], P. P. KULKARNI[1,2], C. F. FERRIS[1,2]

[1]Ctr. for Translational NeuroImaging, Boston, MA; [2]Northeastern Univ., Boston, MA; [3]Biol. Sci., Delaware State Univ., Dover, DE

Introduction

Intermittent hypoxia (IH), a hallmark of obstructive sleep apnea, is linked to neuroinflammation, white matter injury, and increased **neurodegeneration** risk. IH can increase **brain injury and dementia risk** through disruption in hippocampal function, blood-brain barrier integrity, and brain connectivity. This study investigates whether short-term IH produces early behavioral and neurobiological changes in mice. It also explores whether **psilocybin**, a psychedelic natural tryptamine alkaloid associated with enhanced neuroplasticity, can **reverse IH-related effects**. This work aims to establish a **translational model** of the early brain effects of IH and identify potential neuroprotective interventions.



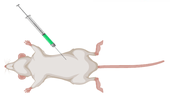
Experimental Design

4 Cohorts of C57BL/6J mice, 90 days old:

1. Normoxia + Saline Control
2. Normoxia + Psilocybin
3. Intermittent Hypoxia + Saline Control
4. Intermittent Hypoxia + Psilocybin

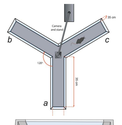
Exposure (Days 1–7)

- Normoxia: 21% Oxygen for 6 hrs
- IH: 9% to 21% Oxygen in 14 min intervals for 6 hrs



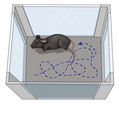
Dosing (Day 7)

- Intraperitoneal Injection:
- 0.9% Buffered Saline Control
- Psilocybin: 3mg/kg in 0.5 mg/mL saline solution



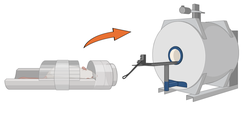
Behavioral Assays (Days 8-10)

- Open Field Test: Measures locomotor activity, exploration, and anxiety-like behavior.
- Novel Object Recognition: Assesses recognition memory and preference for novel objects.
- Y-Maze: Measures spatial working memory through spontaneous alternation behavior.



Anesthetized Scan (Day 11)

- T2 Weighted Anatomy : Measures brain structure and regional volumetric changes.
- DWI: Measures microstructural brain changes through water diffusion.



Acclimations + Awake Scans (Days 12-16)

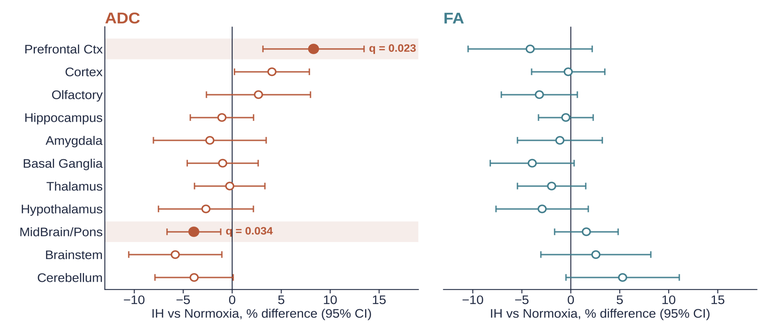
- 4-Day habituation to MRI restraint and scanner noise.
- Resting state functional connectivity: Measures spontaneous brain activity and connectivity between regions.



Tissue Collection and Histology (Day 17)

- Brain extraction

Diffusion Weighted Imaging



+8.3%
Prefrontal ADC, IH vs normoxia (g = 0.83, q = 0.023)

−11%
Prelimbic FA, IH vs normoxia (p = 0.013, uncorrected)

0 of 11
brain systems changed by psilocybin (FA or ADC)

Figure 1. IH effect on ADC and FA in 11 brain systems (normalized to whole brain; IH n = 32 vs normoxia n = 25, treatment groups pooled). Filled symbols: FDR q < 0.05. The prefrontal ADC increase survives adjustment for scan date (mixed model q = 0.028); the MidBrain/Pons decrease does not (q = 0.099).

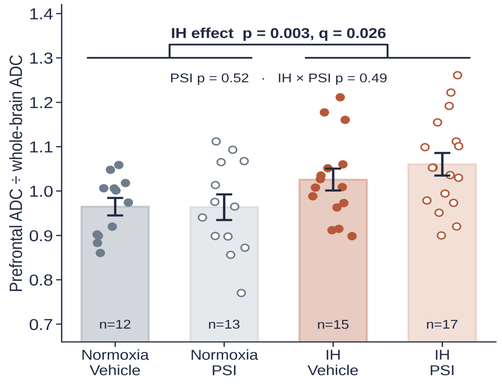


Figure 2. Prefrontal ADC was higher in IH mice with or without PSI; PSI had no effect. Mean ± SEM.

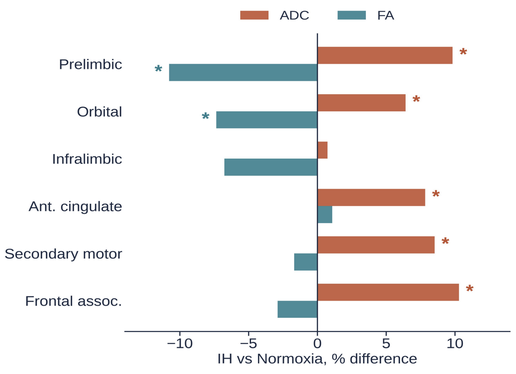
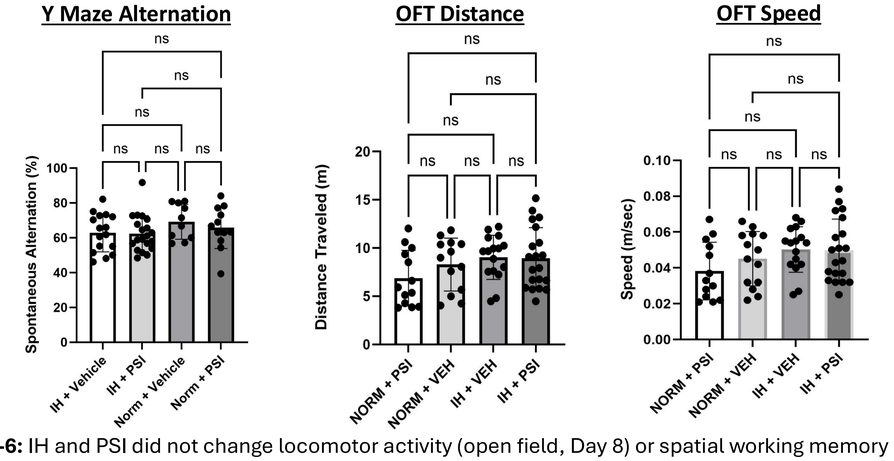


Figure 3. Prefrontal subregions: IH raised ADC and lowered FA, most consistently in prelimbic and orbital cortex. * p < 0.05, uncorrected.

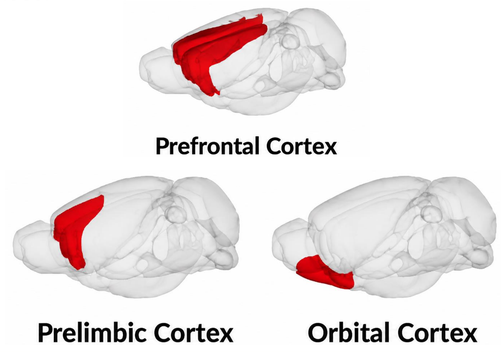
Y-Maze and Open Field Behavioral Tests



Figures 4-6: IH and PSI did not change locomotor activity (open field, Day 8) or spatial working memory (Y-maze spontaneous alternation, Day 10). All pairwise comparisons n.s.

Conclusions

- One week of IH selectively increased prefrontal water diffusivity (ADC), with FA decreases in prelimbic and orbital cortex.
- These changes occurred without deficits in locomotion or working memory: a silent neuroradiological signature, as seen after repetitive mild head injury.
- A single post-exposure dose of PSI (3 mg/kg) did not alter IH-induced changes.
- Higher ADC with lower FA suggests more extracellular water or loss of tissue microstructure; T2 mapping and histology (Iba1, GFAP, myelin) will distinguish edema from tissue loss.



Future Directions

- In head injury PSI was given after each impact; here mice received one dose after 7 days of IH. **Repeated dosing** during IH will be tested.
- Awake resting-state **functional connectivity** (Day 16) is being analyzed.
- Perform **tissue-based biomarker analyses** to evaluate mechanisms underlying early IH effects, including:
  - **Blood-brain barrier disruption**: IgG / albumin leakage
  - **Hypoxia signaling**: HIF-1$^{\alpha}$
  - **Neuroinflammation / glial activation**: GFAP, Iba1
- Extend this pilot into a larger **longitudinal model** to determine whether early IH-induced imaging and behavioral changes predict later more severe cognitive decline or persistent circuit dysfunction.

Acknowledgements:

- **Psilocybin**: National Institute on Drug Abuse
- **Project Funding**: EKAM Imaging

Stay connected! ahuja.re@northeastern.edu