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Expression of Olfactory-Induced Anxiety Behaviors Following Hypoxia Treatment in Adult Zebrafish

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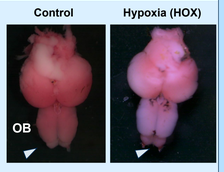
Background

Olfaction and Hypoxia

Olfaction assists with identifying danger, locating food, and social interaction (1). Hypoxia, or decreased oxygen in tissue, decreases brain activity, while prolonged hypoxia can lead to neuronal cell death (4).

Our group reported that hypoxia reduces cell metabolism in the olfactory bulbs (OBs)



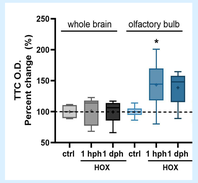


Fig. 1. Hypoxia reduces metabolism of cells in the OB, demonstrated by TTC staining.

Anxiety responses to cadaverine

Decay cues (e.g. cadaverine) elicit anxiety responses in zebrafish, such as changes in velocity, increased erratic swimming, decreased exploration,increased freezing, and negative vertical displacement (6,7,8)

Our goal was to study olfactory responses to cadaverine following hypoxic exposure in zebrafish

Hypothesis

We hypothesize that zebrafish exposed to hypoxia will not exhibit olfactory-induced anxiety behaviors as a response to cadaverine

Materials and Methods

Hypoxic exposure

-Adult fish were individually placed in a hypoxic chamber with container with a dissolved oxygen concentration between 0.6 and 0.8 mg/L for 15 minutes (ctrl: 5-7 mg/L)

Experimental chamber and cadaverine exposure

-½ gallon tanks with 1.5 L of water Fish were acclimated in silence and 1mL of cadaverine solution was injected into the tank

-Video recordings were taken 30 sec before (pre) and 30 secs after (post) cadaverine.

Fig. 2. Testing apparatus.

Results

Hypoxia reduces freezing time after cadaverine exposure

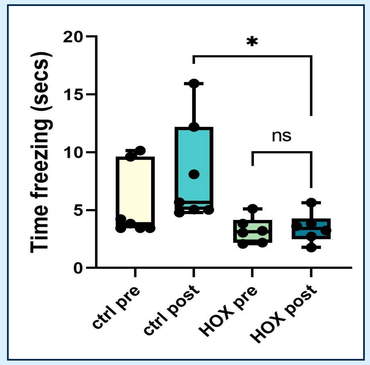


Fig. 3. Hypoxia significantly impacts freezing time after the administration of cadaverine. * p< 0.05

Hypoxia reduces cadaverine-induced erratic swimming

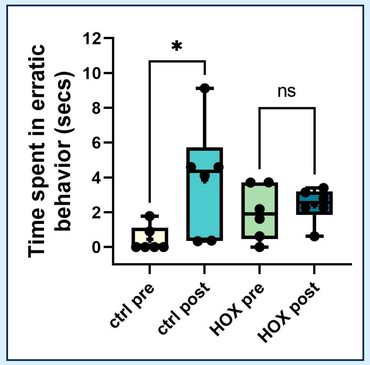


Fig. 4. Cadaverine exposure caused zebrafish to behave significantly more erratically in control groups than treatment.* p< 0.05

Hypoxia did not affect other swimming behaviors such as vertical exploration, tank exploration, and sinking, before and after cadaverine

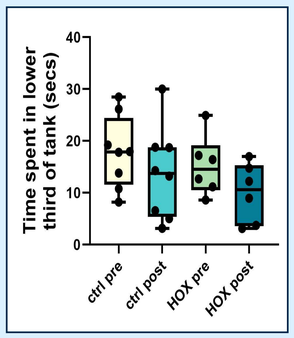


Fig. 5. Sinking behaviors were not impacted by exposure to hypoxia.

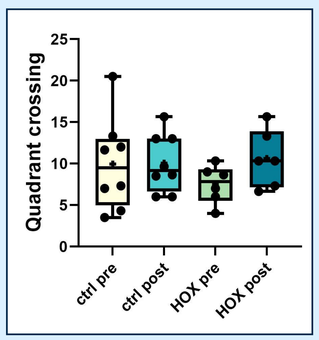


Fig. 6. Hypoxia did not alter tank exploration

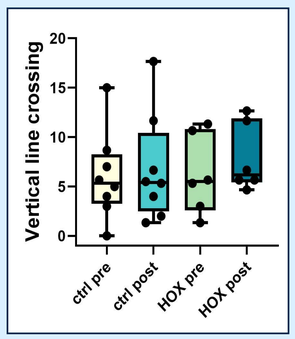


Fig. 7. Hypoxia did not affect vertical tank exploration

Fig 8. Zebrafish will sink to the bottom of the tank if there are swimming impairments

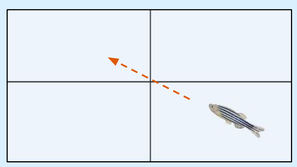


Fig. 9. Zebrafish were analyzed by dividing up the tank into four quadrants and marking how many times they crossed a quadrant line

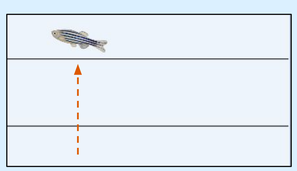


Fig. 10. Vertical displacement was analyzed by dividing up the tank into three sections and marking how many times they crossed a line

Discussion

Hypoxia reduces freezing time and erratic swimming after cadaverine exposure. This supports our hypothesis of decreased olfactory response to cadaverine. Our results support previous findings as hypoxia suppresses olfactory-induced anxiety responses (5, 6).

There were no significant differences between control and treatment fish in tank exploration or time spent in the lower third of the tank. This indicates that there are not significant swimming impairments due to hypoxic exposure, and that lower third lingering is not manifested as an anxiety behavior.

These results also contradict previous findings, as exploratory behavior decreases during an anxious response (6,7,8). We predicted that hypoxia would increase exploratory behavior before and after cadaverine.

Future Work

-Future studies may utilize a larger sample, as well as investigate other behaviors such as eating, mating, and memory tasks.

-We can study physical damage of the olfactory system in the context of the future aim to study neurogenesis (neuron regeneration) following hypoxia.

References

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Acknowledgements

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