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Metabolic response to hypoxia in Gulf Toadfish (*Opsanus beta*) by ventilation amplitude and frequency

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Introduction

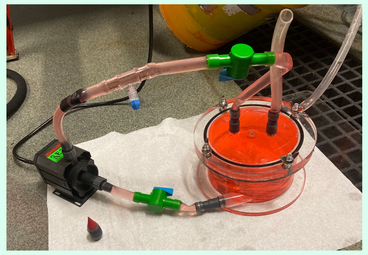
- Many coastal areas with dense vegetation, like seagrass meadows along the Florida coast, undergo natural intermittent periods of hypoxia in diurnal cycles
- This stems from high oxygen consumption during nighttime when photosynthetic activity is absent
- During this time, many benthic organisms that gain food and shelter from this vegetation undergo oxygen limitation stress and exhibit changes in metabolic behavior.
- The Gulf Toadfish (*Opsanus beta*) is a prime example of these adaptable benthic creatures - upon sensing decreasing oxygen levels, toadfish adjust their respiration to lower the energy-costs to preserve energy throughout the night without the aid of abundant O2.
- Unfortunately, there are limits. Near anoxia was suspected to be the cause of the 2022 Biscayne Bay mass fish kill that killed > 1,000 fish, many of which were toadfish

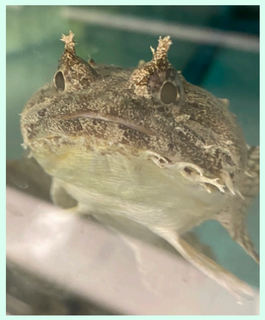
Researchers with Miami Waterkeeper, a nonprofit organization, sounded the alarm on Wednesday about a fish kill in Biscayne Bay.

- The culprit is likely two nearby discharge canals that, with the influx of freshwater from Hurricane Ian, facilitated further septic tank leakage and stormwater runoff into the nearby Bay area
- Anthropogenic activities that result in excess nutrient runoff (fertilizers, sewage, stormwater, etc.) → eutrophication events & algal blooms → algae dies after consuming excess nutrients → aerobic bacteria consumes algae & O2 → low DO → mass fish kills
- These fish kills events cause the mass mortality of even the hardiest of species. Understanding the minimum saturation level that the hardiest of fish can sustain without resorting to hyperventilation will further improve our understanding of the tolerance of hypoxia/near-anoxia events in relevant local ecosystems.
- These studies will provide the intel necessary to make informed environmental decisions regarding safe wastewater treatment and disposal, decreasing the severity and frequency of fish kills

Methods

The respirometry system entailed 2 large Loligo respirometry chambers 1080mL in volume and the use of two IR (and later a phone) cameras, funded by the SURGE and Linda Farmer Awards. One 50 - 80 gram mature wild-caught toadfish was placed in each chamber and acclimated overnight on an intermittent cycle that alternates between flushing with oxygenated UV water (240 seconds), a wait period (90s), and a measurement period (360s). During the measurement period, the water is only recirculating with no fresh inflow. The system is fully closed the next morning (~8am) until the fish becomes distressed (excrement, side-swimming, agitation) or until 5% oxygen saturation. A 30-minute blank was taken before and after the experiment to correct for background microbial respiration. The fish is then returned safely, and the system bleached and rinsed after each experiment.

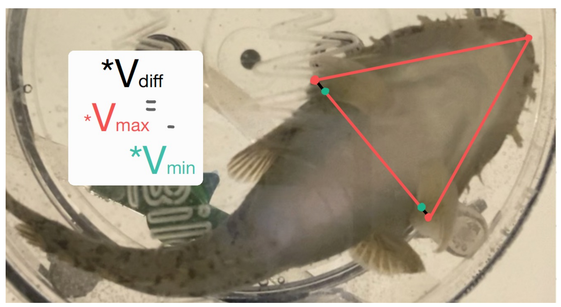




Video data: tracks underneath of fish (to track gill ventilation volume and frequency) - recorded using an IR camera/cell phone for 1 minute at each increment of 10% as well as 5%:

90%, 80%, … 20%, 10%, 5%

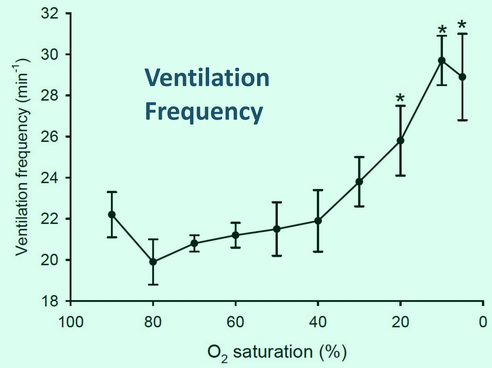
*[O2] never reaches 100% while fish are in the chamber

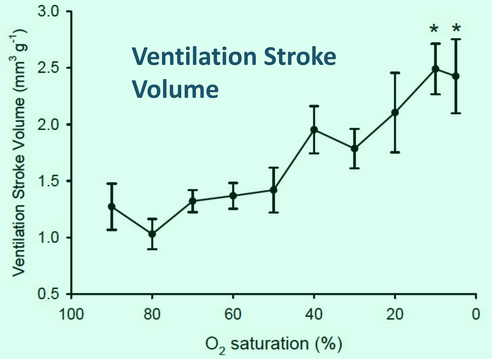


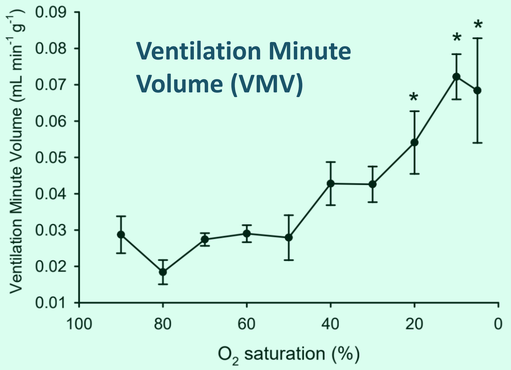
Objective

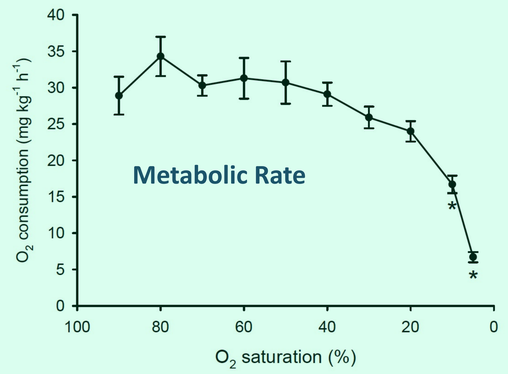
Quantify change in metabolism by measuring oxygen consumption through respirometry and recording their gill ventilation, measuring stroke volume and frequency of opercular movement through video monitoring.

Results









Results

One Way Repeated Measures ANOVA Analysis of Variance using 90% saturation as control (n = 11)

- Frequency: Fish in 5% ($p < .001$), 10% ($p < .001$), and 20% ($p = .022$) oxygen saturation had significantly higher ventilation frequencies (in beats per minute) than the control.
- Stroke Volume: 5% ($p < .001$) and 10% ($p < .001$) (20% was $p = .05$) oxygen saturation had significantly higher ventilation stroke volume (mm3) than the control.
- Ventilation Minute Volume: Fish in 5% ($p < .001$), 10% ($p < .001$), and 20% ($p = .020$) oxygen saturation had significantly higher VMV than the control.
- MO2: Fish in 5% ($p = .003$) and 10% ($p < .001$) oxygen saturation had significantly lower metabolic rates than the control.

Significant values have an asterisk *.

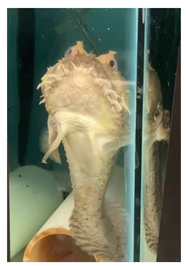
Conclusions

- Toadfish do not resort to altering ventilation until ~20% oxygen saturation. Cardiovascular adjustments may explain how MO2 remains constant at O2 saturations from 80% - 20% where there is no change in ventilation. During these lower O2 saturations, venous blood will have a lower [O2] which will help diffusion from water to the blood. (McDonald et al.). Upon reaching ~10%, they can no longer sustain themselves in severe hypoxia and VMV starts to decrease.
- Understanding such a hardy species' hypoxia coping mechanisms sheds some light on the impact of hypoxia on marine fishes. Toadfish are sturdy animals and can handle incredible amounts of stress, but dissolved oxygen levels near anoxia (2022 Biscayne fish kill .07 mg/L on seafloor and 0.22mg/L at surface (Zangroniz, Ana)) are unsustainable for any marine organism.
- Further understanding these species and how they are affected will guide conservation efforts and environmental awareness in Miami-Dade County, especially in wastewater treatment.

References

McDonald, M. D., Gilmour, K. M., Walsh, P. J. & Perry, S. F. Cardiovascular and respiratory reflexes of the gulf toadfish (*Opsanus beta*) during acute hypoxia. Respiratory Physiology & Neurobiology 170, 59-66

Zangroniz, Ana. "October 2022 Biscayne Bay Fish Kill." UF/IFAS Extension Miami-Dade County, 28 Oct. 2022



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