UP, DOWN, & UP AGAIN: DAILY CYCLES IN WATER OXYGEN CONTENT

Oxygen is essential to respiration, the process of converting nutrients into the biochemical energy that fuels life. Sometimes oxygen is in short supply, making it tough for animals to meet their energy needs. Investigating how fish cope with such periods of low oxygen ("hypoxia") has inspired plenty of scientific studies. But, it can be tricky to connect textbook lab responses with the messy realities of the natural world. For example, we're experts in how fish cope with continuous hypoxia, but amateurs when it comes to the effects of daily cycles in oxygen level.

Diurnal hypoxia is a fact of life for many fishes. Oxygen level rises and falls with the tides in coastal waters. Intensive aquaculture operations often deal with hypoxia due to high stocking densities – fish can be, literally and figuratively, "packed to the gills." Many lakes and rivers also cycle between daytime oxygen booms and nighttime busts due to the interplay between photosynthesis (which adds oxygen) and respiration (which consumes it) in aquatic plants. In areas like Lake Erie and Chesapeake Bay, seasonal dead zones develop when oxygen level dips too low to support life.

Though notorious, diurnal hypoxia is understudied by fish researchers. Our aim was to better understand how fish cope with diurnal hypoxia. Do they respond to it like other hypoxia patterns? Or, do they have a special coping strategy? Like any proper group of physiologists, our first step was to pick the best animal for our study. Enter the killifish, *Fundulus heteroclitus*. Small and skittish, they rarely grace the glossy covers of magazines. Even their common name, mummichog, speaks to their habit of going in crowds. It's a bit like naming a bird for its *habit* of flying. But our humble mummichogs famously thrive in estuaries, where they encounter various patterns of hypoxia.

We exposed our hypoxia champs to continuous hypoxia or diurnal hypoxia, and compared them to fish in oxygen-rich conditions[1]. Killifish in continuous hypoxia boost their capacity to shuttle oxygen around their body by increasing the concentration of oxygen-carrying red cells in their blood. They also "turn down the pilot light"[2] of metabolism, cutting back on costly biological processes to reduce oxygen and energy demands. Gill structure changes to perhaps reduce the cost of maintaining ion balance. Skeletal muscle composition also shifts, likely reflecting less routine swimming activity. While not as dramatic as in animals like hibernating turtles, these and other responses help to stretch out limited energy supplies.

Diurnal hypoxia needs its own strategy. During low oxygen periods, killifish rely heavily on anaerobic glycolysis as an alternative pathway to meet energy needs. Glycolysis is great because it converts carbohydrate fuels like glucose into biochemical energy without using oxygen. Glycolysis is not-so-great because it's far less efficient than respiration, and can cause the build-up of nasty metabolic waste products. Killifish solve

[1] B.G. Borowiec et al., 2015.
[2] Famously coined by Kjell Johansen, and popularized by P.W. Hochachka et al., 1996, to describe the responses of cells to severe hypoxia.

these problems by exploiting “oxygen boom” periods, when they restock fuel stores to feed glycolysis and dispose of metabolic waste. This helps them to recover in time for the next hypoxia bout, making them masters of dealing with huge swings in oxygen level. Killifish are able to do all this by increasing the activity of key proteins involved in these processes.

These distinct coping strategies may have wider consequences. Since they avoid reducing routine energy demands, fish exposed to diurnal hypoxia *may* stay more active during hypoxia bouts. However, this coping strategy is likely limited by the ability to deal with wastes. The most sustainable approach is probably to reduce oxygen (and energy) demands to match the new reality of hypoxia. We know this pays off in some cases: killifish exposed to continuous hypoxia tolerate extremely low oxygen levels better than fish exposed to diurnal hypoxia. But this strategy probably has its own issues– a little killifish *not* firing on all cylinders is easy prey.

Cycles of hypoxia are a hot topic in medical research because they’re associated with obstructive sleep apnea, hypertension, and other conditions. As it turns out, hypoxia cycles also have unique effects on fish. With the rising global incidence of hypoxia due to human influences and climate change, understanding how fish cope will be key to predicting its future impacts on fish and fisheries.

WORDS: 698

REFERENCES

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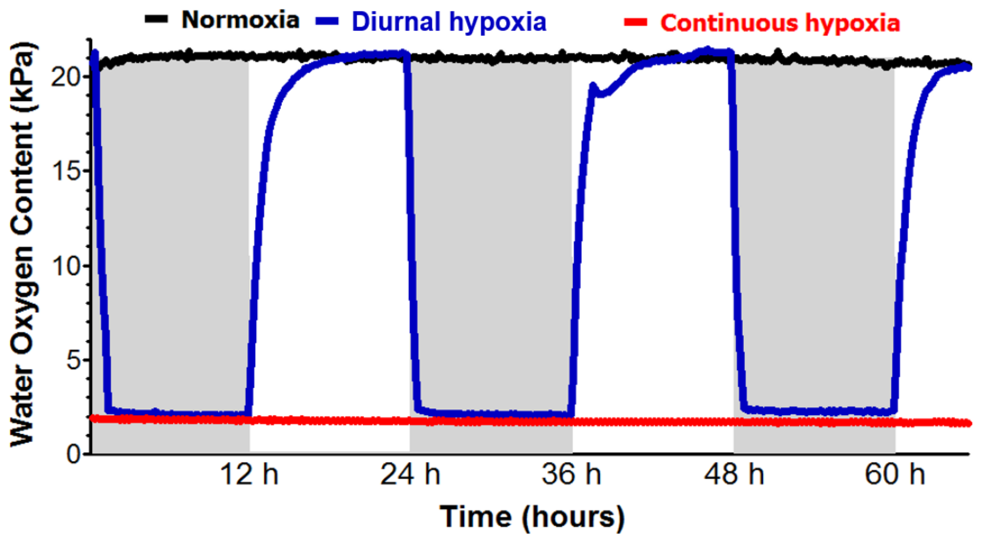
GRAPHICS & PHOTOS



We exposed killifish for 7 or 28 days to normoxia (fully-oxygenated, **black**), diurnal hypoxia (**blue**), or continuous hypoxia (**red**). The light cycle is also indicated (**gray** signifies night/low light).

A large mummichog (*Fundulus heteroclitus*) in a glass respirometry chamber. Photo taken at McMaster University.