Liver Anaerobic Metabolic Capacity Does Not Increase in Response to Hypoxia in Antarctic and Related Cold-Temperate Fishes

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INTRODUCTION

- Due to climate change, temperature is increasing, dissolved oxygen is decreasing, and the frequency of hypoxic events is increasing in the ocean (2,7).

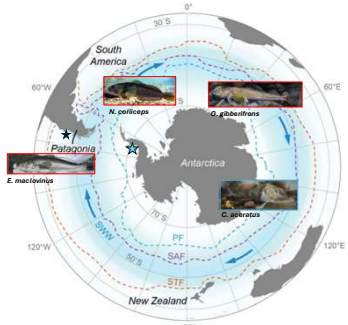

Figure 1. Map of Antarctica with fishes used in this study and their location.

- Antarctic notothenioid evolved in oxygen-rich waters south of the Antarctic circumpolar current, and evolved unique physiological traits, including the loss of hemoglobin in one family (Channicthyidae), potentially making them extremely vulnerable to hypoxia (3, 1).

- Lactate dehydrogenase (LDH) is a key glycolytic enzyme crucial for sustaining anaerobic metabolism in hypoxia by regenerating NAD+ and ensuring glycolysis continues (4).

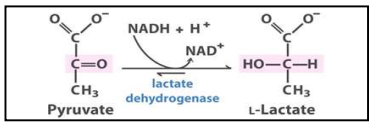

Figure 2. Reaction catalyzed by LDH

HYPOTHESES

- **(1) Lactate dehydrogenase activity in Antarctic fishes will be lower than in the cold-temperate *E. maclovinus*. (2) LDH activity will increase in response to hypoxia in all species.**

- We measured the maximal activity of LDH in liver tissue samples from fish exposed to normoxia and hypoxia.

MATERIAL & METHODS

- Antarctic fishes; *G. gibberifrons, N coriiceps, C. aceratus,* and their basal relative *E. maclovinus* were exposed to normoxia and hypoxia (15% above the O_2 critical level for 48h; n=8 per treatment).

- Liver tissues were collected, stored at -80° C. Tissue samples were homogenized in a buffer (40 mM HEPES, 1 mM EDTA, 2 mM $MgCl_2$) [pH=7.8 at 5° C], and centrifuged (4° C for 5 mins).

- LDH activity was quantified using spectrophotometric assay, monitoring the oxidation of NADH at 340 nm.

- Significant difference among species and in response to hypoxia were determined using a 2-way ANOVA (p<0.05) with Prism VX.

RESULTS

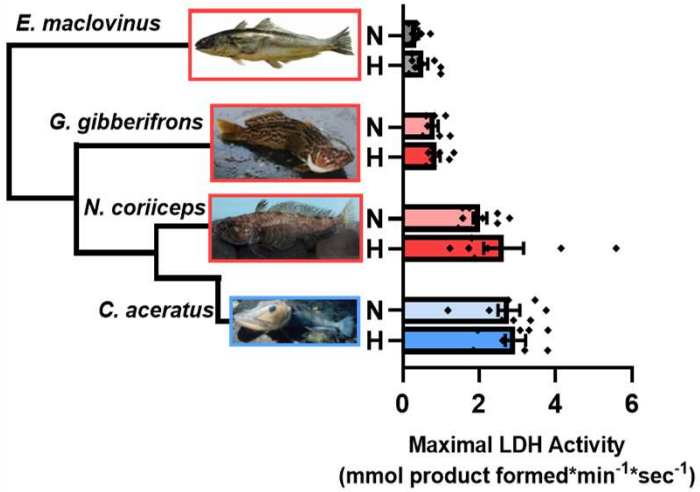

Figure 3. Phylogenetic tree of fishes used in this study and corresponding liver LDH activity. LDH activity was measured in liver tissue from animals held normoxia (N) and hypoxia (H) [15% above species-specific O_2 crit]. Error bars represent mean ± SEM (n=8 per treatment).

- **LDH activity did not change in response to hypoxia in any species (p=0.68).**

- **LDH activity was higher in *C. aceratus* and *N. coriiceps* compared with *G. gibberifrons* and *E. maclovinus* (p<0.0001).**

- **The overall trend indicates that LDH activity is higher in more derived notothenioids compared with the basal species.**

DISCUSSION

- **LDH activity did not increase in response to hypoxia,** suggesting notothenioids may have a limited capacity to increase anaerobic metabolism in response to hypoxia.

- **LDH activity differed significantly among the species,** suggesting that the differences in anaerobic metabolic capacity reflect phylogeny rather than hypoxia tolerance.

SOURCES

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DISCUSSION CONTINUED

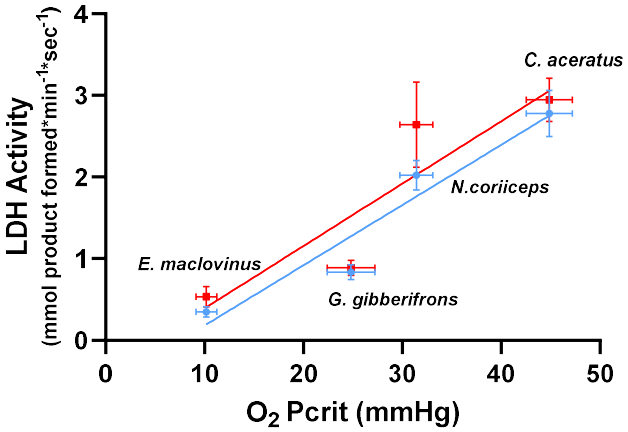

Figure 4. Relationship between O_2 Pcrit and lactate dehydrogenase activity across species. The mean maximal LDH activity in liver tissues of fish exposed to normoxia (blue) and hypoxia (red) is plotted against species-specific O_2 Pcrit values, which is the measure of hypoxia tolerance. LDH was positively correlated with O_2 Pcrit (linear regression, p<0.0001, r= 0.720; p<0.0001, r= 0.865). Each point represents the species' mean.

- The **positive correlation between O_2 crit and liver LDH activity** suggests that notothenioids rely on mechanisms other than anaerobic metabolism to cope with hypoxia. Other potential mechanisms include **suppressing metabolism, and increasing ventilation, heart rate, blood flow, and hematocrit in red-blooded species** (5,6).

- The lack of increase in LDH activity suggests a limited ability to increase anaerobic metabolism under hypoxia. With the ongoing climate change, species with low anaerobic metabolic capacity may be particularly vulnerable to hypoxia.

FUTURE DIRECTIONS

- Normalize LDH activity to protein concentration to account for differences in tissue composition and improve cross-species comparisons.

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