The FASEB Journal / Volume 34, Issue S1 / p. 1-1

Physiology | Free Access

Effect of Chronic Temperature Acclimation on the Liver Proteome of Threespine Sticklebacks (*Gasterosteus aculeatus*)

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First published: 19 April 2020
https://doi.org/10.1096/fasebj.2020.34.s1.07128

Abstract

This study investigated the liver molecular phenotypes of two different populations of first generation lab-reared threespine sticklebacks (*Gasterosteus aculeatus*) from northern California: a fully plated population from the Klamath River in Klamath, CA and a low plated population from Big Lagoon in Trinidad, CA. Temperature is one of the most influential abiotic factors affecting fish and this study gives insight into the molecular changes used to offset temperature stress in the liver. We constructed an assay library for the liver that consisted of 1,715 proteins, 5,897 peptides, 6,995 precursors, and 40,999 transitions with Skyline (MacCoss Lab). The assay library was constructed with samples from fish (N=22) of both populations after chronic warm, chronic cold, acute warm, and acute cold (Klamath population only) temperature stress experiments using data dependent acquisition (DDA) shotgun proteomics. This assay library was then used to compare the molecular phenotypes of Klamath and Big Lagoon stickleback livers after a three-week exposure to either 7°C (cold), 15°C (control), or 25°C (warm). It will also be used to analyze acute temperature stress experiments in the future. Liver proteomes were extracted, trypsinized, and analyzed by gel-free quantitative proteomics using liquid chromatography-mass spectrometry (LC-MS/MS). There were only 6 proteins with significantly different abundances between the Klamath and Big Lagoon populations ($p < 0.05$, fold change ± 2), suggesting similar molecular phenotypes between the two groups. There were 8, 141, and 234 proteins with significantly different protein abundances for comparisons of 7°C and 15°C, 15°C and 25°C, and 7°C and 25°C respectively (across both populations combined). Our results suggest that gradual acclimation to a warmer temperature, as opposed to a colder temperature, results in larger changes in relative protein abundance and gives insight into potential liver adaptations to sustained warmer temperatures.

Support or Funding Information

This study was funded by a grant from NSF (IOS-1656371).