

**Introduction:** Zebrafish are shallow-water fish that have been shown to tolerate pressures up to 100 atm (~1,000 m). Higher hydrostatic pressures have been shown to cause mortality, however, their tolerance varies among individuals. Hydrostatic pressure is the main limiting factor of marine organisms' ability to vertically traverse the ocean, and information regarding shallow-water fish's tolerance and response to this stress is sparse. Exposing zebrafish to short-term increases in hydrostatic has shown that they up-regulate and down-regulate gene expression in response to this stress. Prior research has shown that the brain has the least stable response to hydrostatic pressure while sexual organs present the most stable response.

**Research Questions:** We are investigating the plasticity of zebrafish, and which genes are affected when they are under hydrostatic pressure stress.

**Methods:** To further investigate this area of genetic adaptation, we are studying the genetic responses of adult zebrafish to increased hydrostatic pressure via bulk and single-cell RNA sequencing, which we will use kallisto to quantify. We will be collecting transcriptome data from tissue samples across the entire zebrafish. To understand how genes adapt to their surrounding environments, we will be studying adult zebrafish of both sexes. Fish will be stored at 25°C and starved for 1 day prior to pressurization experiments. We will be conducting pressurization experiments in a range from 0-4 atm with 0.5 increments with 10 fish per trial. Following these experiments, fish will be dissected and tissue samples will be flash frozen using liquid nitrogen. Specific RNA extraction kits have yet to be determined.

**Predictions/Expected Outcomes:** Prior research has shown that long-term adaptation has resulted in changes in membrane properties, intrinsic and extrinsic properties of proteins, and nucleic acid secondary structures. Hydrostatic pressure has been shown to reduce fluidity and increase the thickness of cell membranes, which are key features in healthy and productive cells in organisms. Proteins have shown that slight changes in atmospheric pressure can disrupt enzyme ligand binding and kinetics and protein assembly, which results in a decrease in stability. These results lead us to believe that our research will discover gene adaptations in cellular pathways related to membrane fluidity and protein stability.