Arsenic-Induced Gut Dysbiosis and Behavioral Activity in Chinese Mystery Snails: Exploring the Microbiome–Nervous System Link

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Background

- Gut dysbiosis can serve as a biomarker for poor health outcomes across multiple species. The gut-nervous system signaling network in invertebrates can serve as a gateway to deepen our understanding of the gut microbiome and its influence on the nervous system, analogous to the vertebrate gut-brain axis.
- Our research used Chinese Mystery Snails (CMS), an invasive species commonly found in freshwater systems in the PNW, to examine how arsenic exposure disrupts the CMS gut microbiome and how it could affect the nervous system, evaluated through their feeding and mobility behaviors as well as the presence of arsenic-metabolizing bacterial genes in the snail

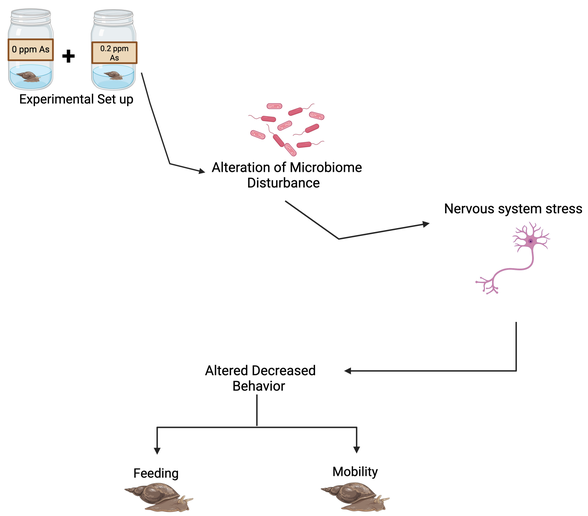

Figure 1: Introducing Chinese Mystery Snails Study. Disruption of the Chinese Mystery Snails, gut microbiome caused by exposure to Arsenic (V), leading to nervous system stress and decreased feeding and mobility behavior.

Hypothesis

We hypothesized that if the CMS gut microbiota influences neuronal behavior, then increased arsenic exposure will adversely alter the microbiome composition, leading to measurable decreases in the snail's feeding and movement activity.

Methods section

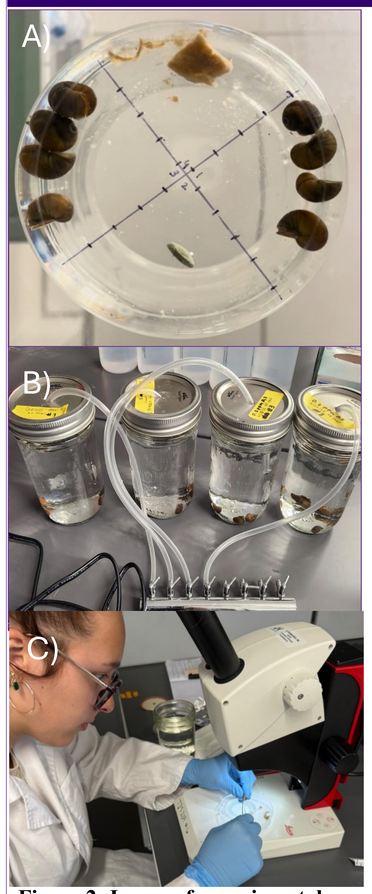

Figure 2: Image of experimental setup. A) Beaker divided into 4 quadrant, eight snails total. With algae wafer, and "snello" as food source. B) CMS acclimating in 0, 0.2, and 2.0 ppm arsenic. C) Dissections of CMS by Alejandra Gonzalez.

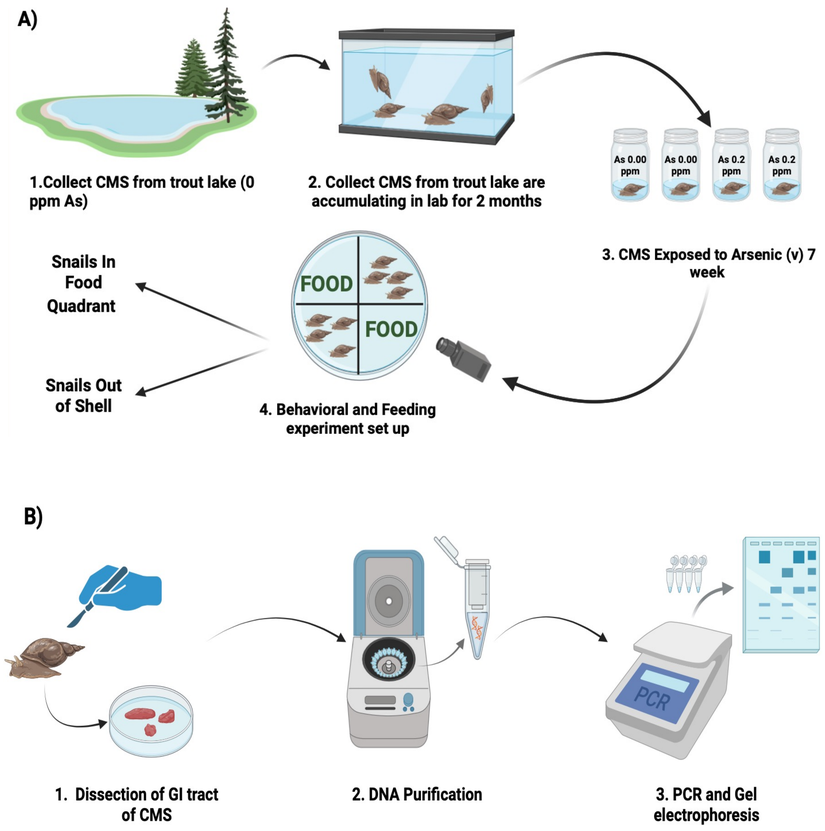

Figure 3. A) Experimental Setup of Behavioral and Feeding Assays of CMS. CMS are collected from Trout lake (0 ppm As) and accumulated in lab for 2 month. Then exposed to As (0 ppm and 0.2 ppm) for 7 weeks. Feeding and Behavioral assay is done and calculated snails in food quadrant and snails out of shell. **B) PCR and Gel electrophoresis of CMS Gut Microbiome.** CMS from Behavioral and Feeding assays are dissected for DNA extraction, PCR amplification and sequencing. Low DNA yield led to inconclusive results but arsenic metabolic genes were amplified.

Results

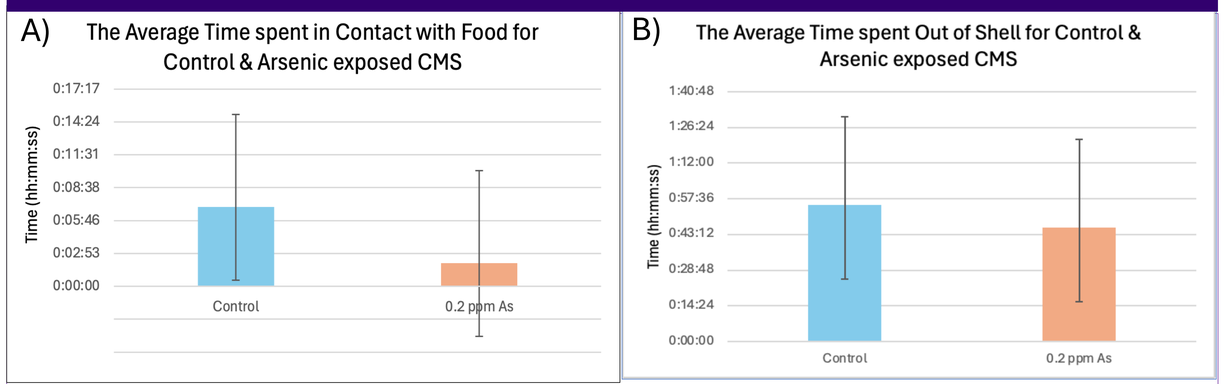

Figure 4. CMS Times in Contact With Food and Time spend Out of Shell. A) This graph shows the average time (hh:mm:ss) the snails spend in contact with food for both the control and 0.2 ppm As CMS. Blue bar represents Control group (0 ppm As) and the orange bar show arsenic exposed snails (0.2 ppm As). With error bars representing the standard deviation over the observation period of 90 min. Using Mann Whitney U Test, two-tail, and significant level set at $p < 0.05$ to calculate p value= 0.02. The arsenic exposed CMS spent less time in contact with food when compared to control. **B**) Graph shows the average time (hh:mm:ss), the snails spend out of shell for both the control and 0.2 ppm As CMS. Blue bar represents Control group (0 ppm As) and the orange bar show arsenic exposed snails (0.2 ppm As). With the error bars representing standard deviation. Mann Whitney U Test, two-tail, and significant level set at $p < 0.05$ to calculate p value = 0.25. The control CMS snails spend more time out of shell then Arsenic exposed snails.

Conclusion

Our experiment suggests an exposure to 0.2 ppm arsenic greatly reduces the time CMS spent in contact with food ($p<0.05$), indicating reduced feeding behaviors. It is important to note that this did not reduce overall mobility, suggesting an effect on the CMS nervous system. However, because we did not obtain measurable material on the effect of the CMS microbiome due to time restraints, we cannot determine whether these behavioral changes were correlated with the CMS microbiome.

Future Steps

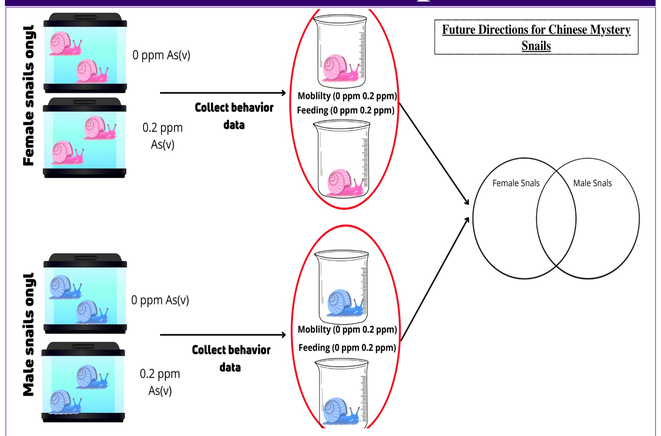

Figure 5. Comparison of behavioral response between female and male Chinese Mystery Snails. CMS that have been exposed to 0 ppm As and 0.2 ppm As are separated based of gender. Behavioral and feeding analysis is conducted to determine of sex acts as an intrinsic determinant of behavioral variation in Chinese mystery snails (CMS), modulating locomotor activity and feeding dynamics with and without Arsenic exposure through sex-specific physiological or neurobehavioral pathways

Acknowledgements

Thank you to Dr. Sarah Alaei and Dr. Marc Nahmani for their amazing support and guidance during the research and to my 14 peers in TBIOMD 495 for their incredible work.