

Investigating the Impacts of Arsenic Exposure on Chinese Mystery Snail Behavior and Gut Microbiome



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Introduction

- Many freshwater systems in the South Puget Sound area are contaminated with arsenic, a legacy of pollution from the former ASARCO smelter.
- The invasive Chinese Mystery Snail (CMS), commonly found in freshwater ecosystems, feeds on periphyton and can bioaccumulate arsenic through diet and water exposure.

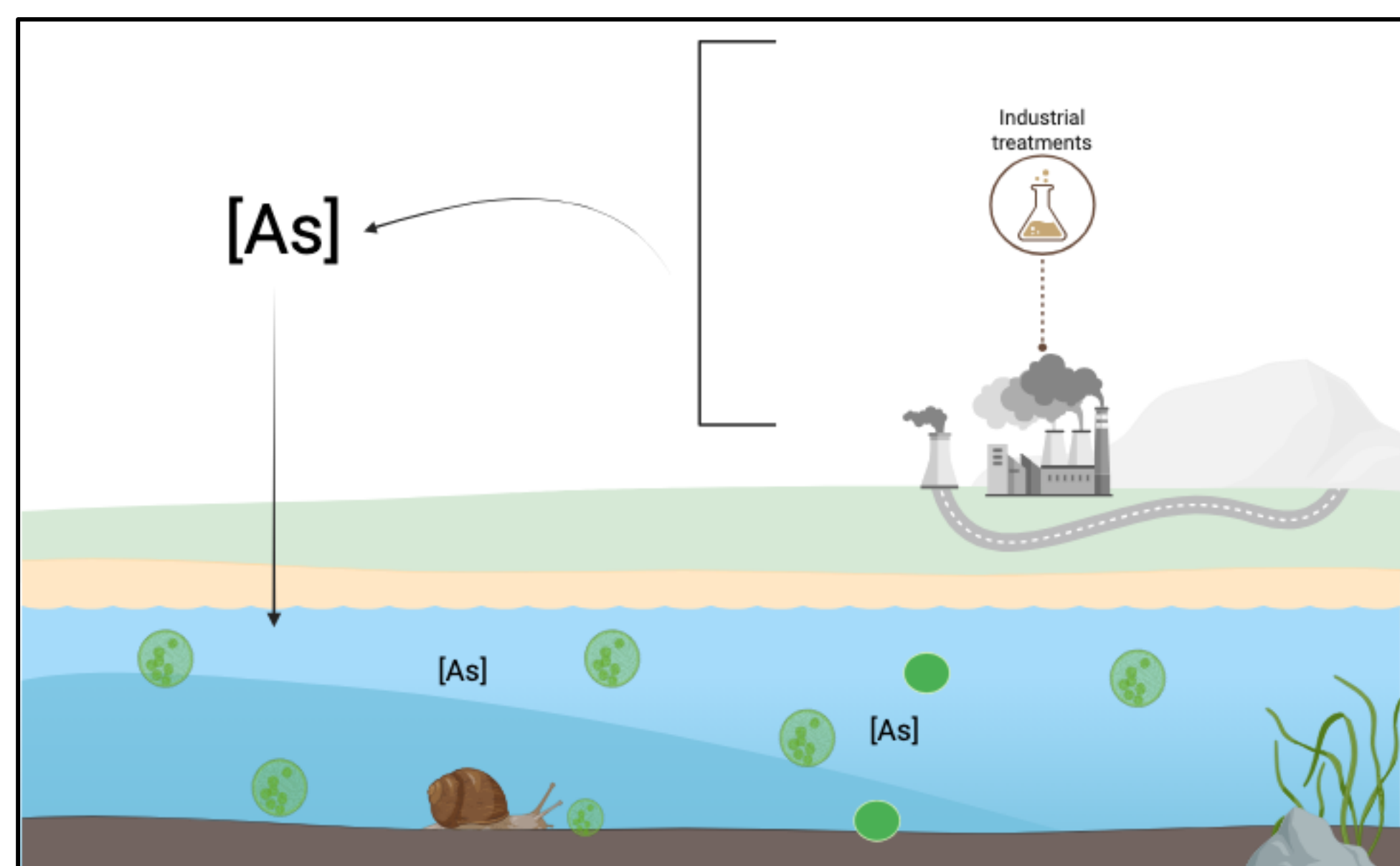


Figure 1: Bioaccumulation of Arsenic in Chinese Mystery Snails. Industrial activities such as factories and transportation can be potential sources that can release Arsenic in freshwater lake environment. This arsenic enters the freshwater system, where it sticks to algae and other tiny organisms that grow on underwater surfaces. Freshwater snails may ingest arsenic contaminated periphyton, causing the arsenic to build up in their bodies over time.

Hypothesis

We hypothesize that increased arsenate exposure would disrupt CMS gut microbiota composition, leading to behavioral changes related to movement and feeding activity.

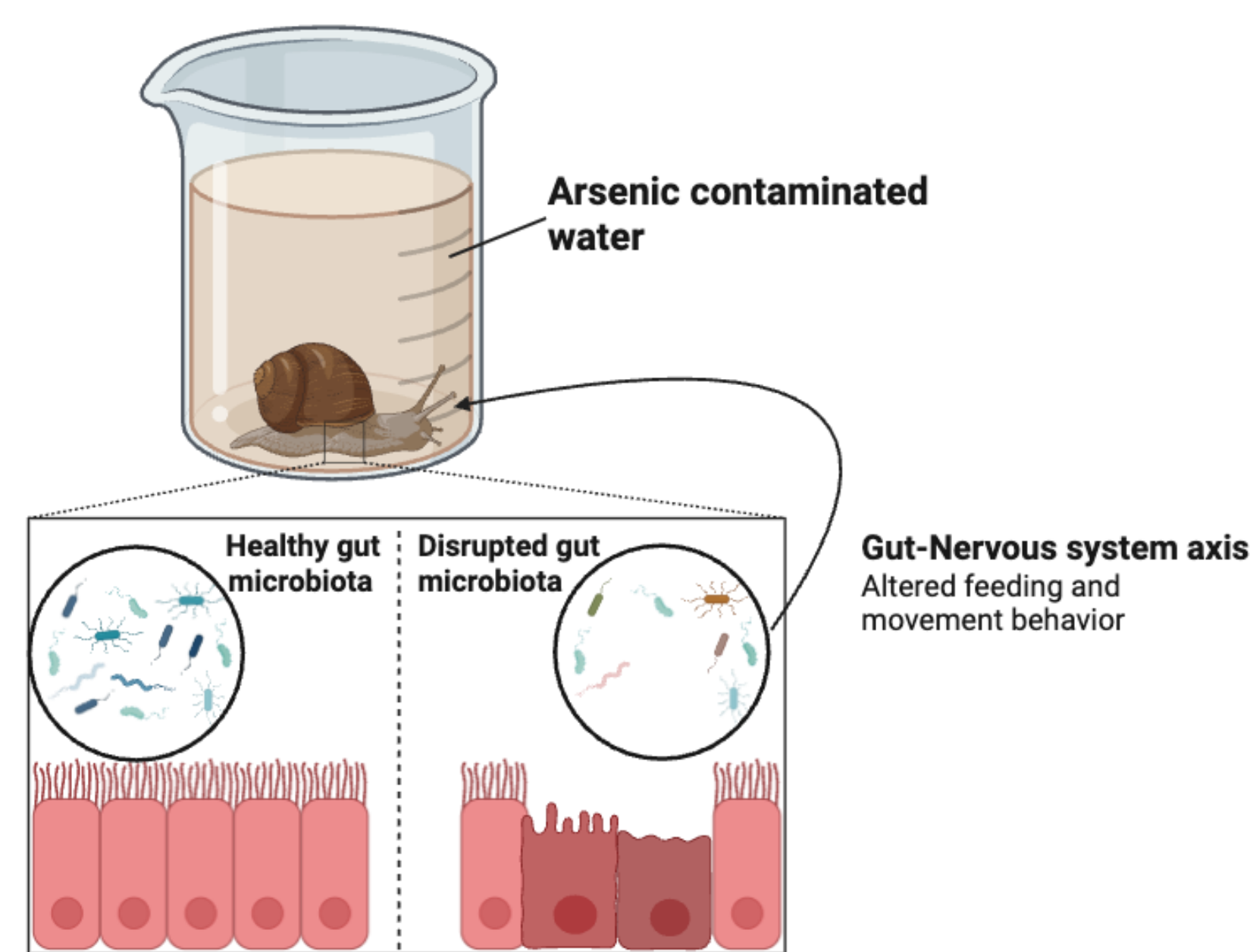


Figure 2: Proposed conceptual model illustrating how arsenic exposure disrupts the gut microbiota in Chinese Mystery Snails, leading to altered feeding and movement behaviors via the gut-nervous system axis.

Methods

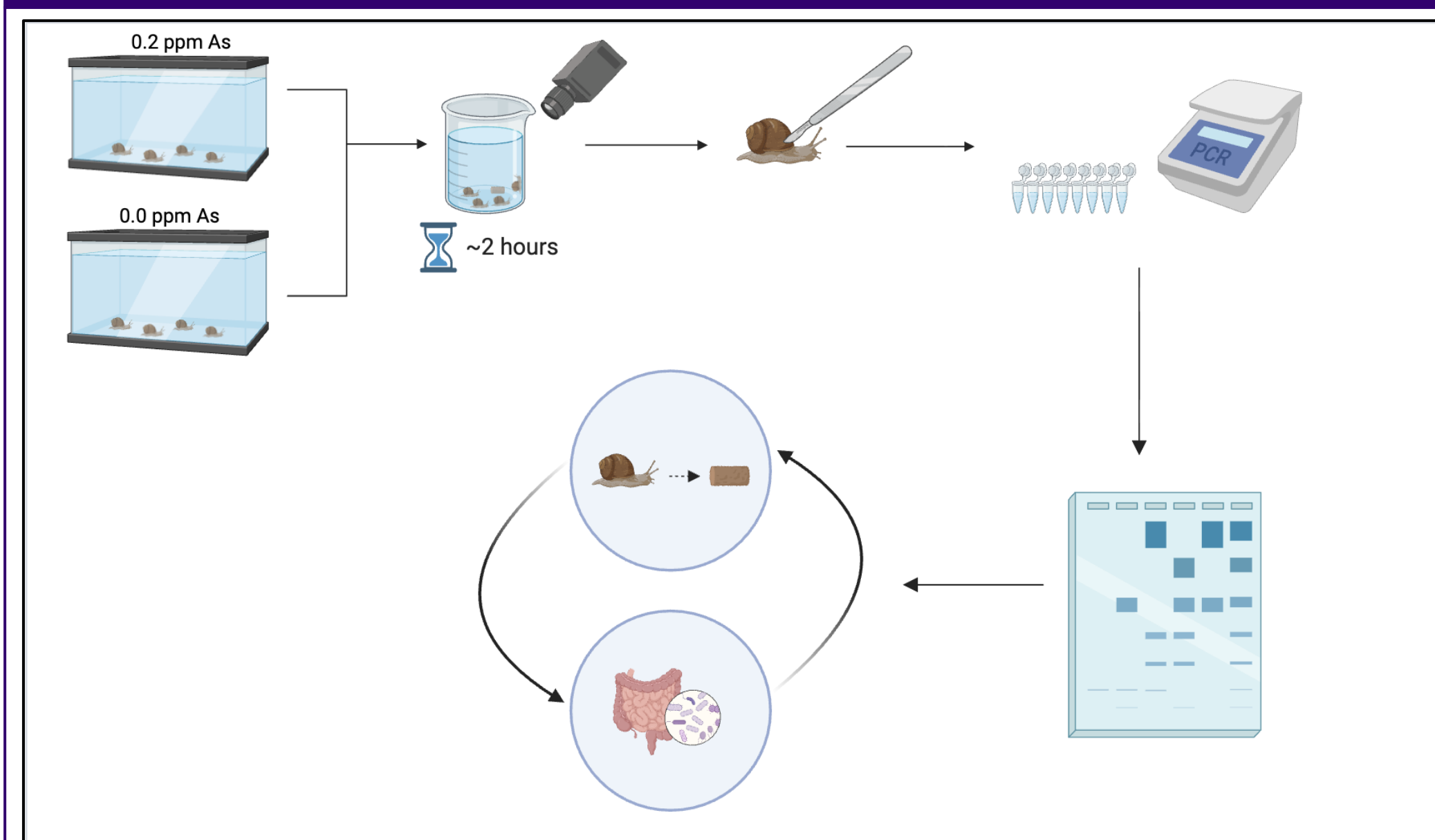


Figure 3: Experimental workflow for gut microbiome and behavioral analysis. Snails were divided into two treatment groups (0.0 ppm and 0.2 ppm arsenic) and observed in separate beakers for motility time and food contact. Post-observation, digestive tracts were dissected and analyzed via PCR and gel electrophoresis to detect *arsM* and *arsB* genes, aiming to link potential gut dysbiosis with behavioral changes in Chinese Mystery Snails.

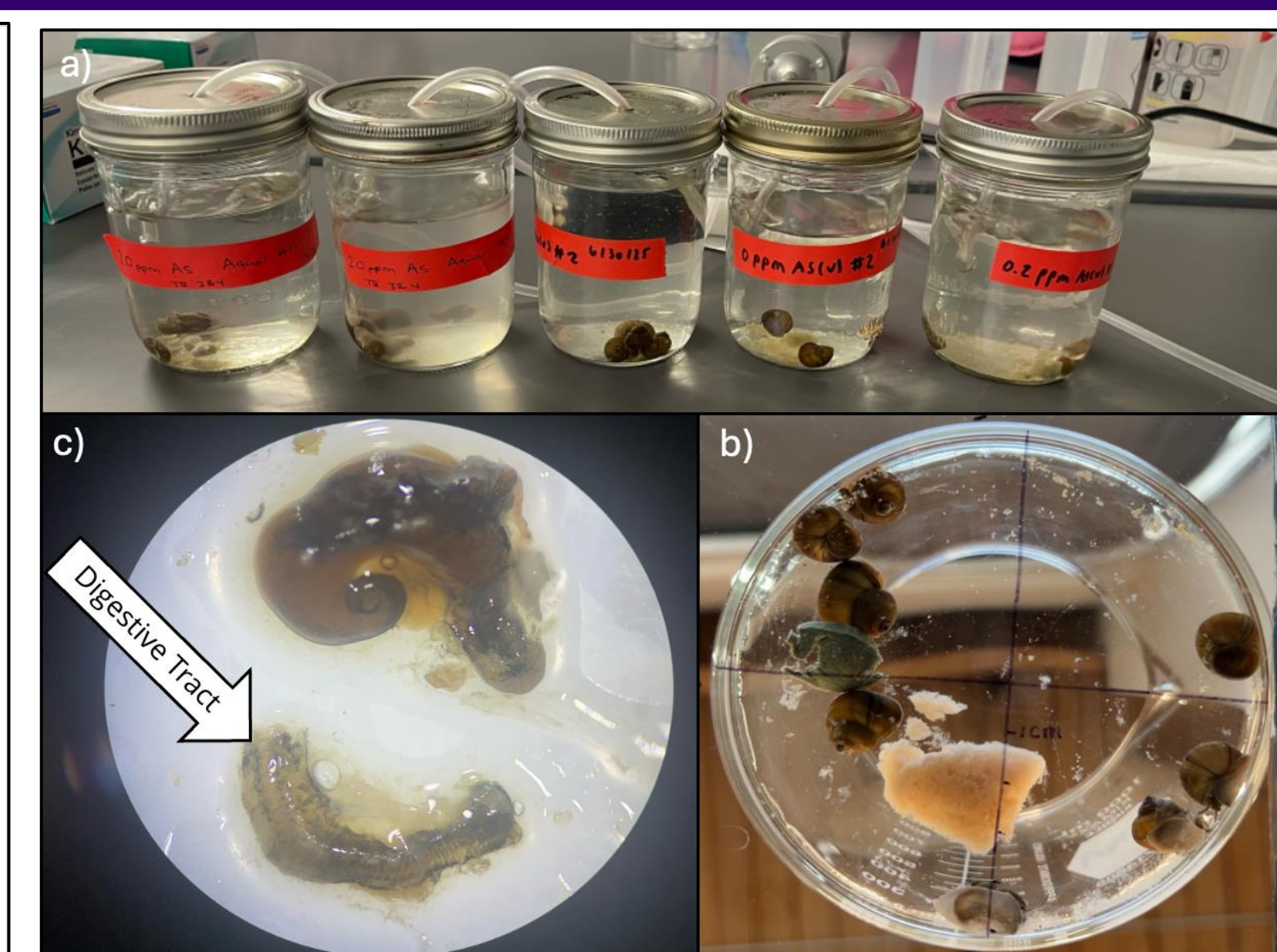


Figure 4: Images of the experimental setup and dissection. (a) Snails acclimating to 0, 0.2, or 2.0 ppm arsenic. (b) Underside view of the beakers used to observe motility and contact with two food options. (c) Macroscopic view of an extracted digestive tract (appearing as a clear tube with small black spots) collected after behavioral observations.

Results

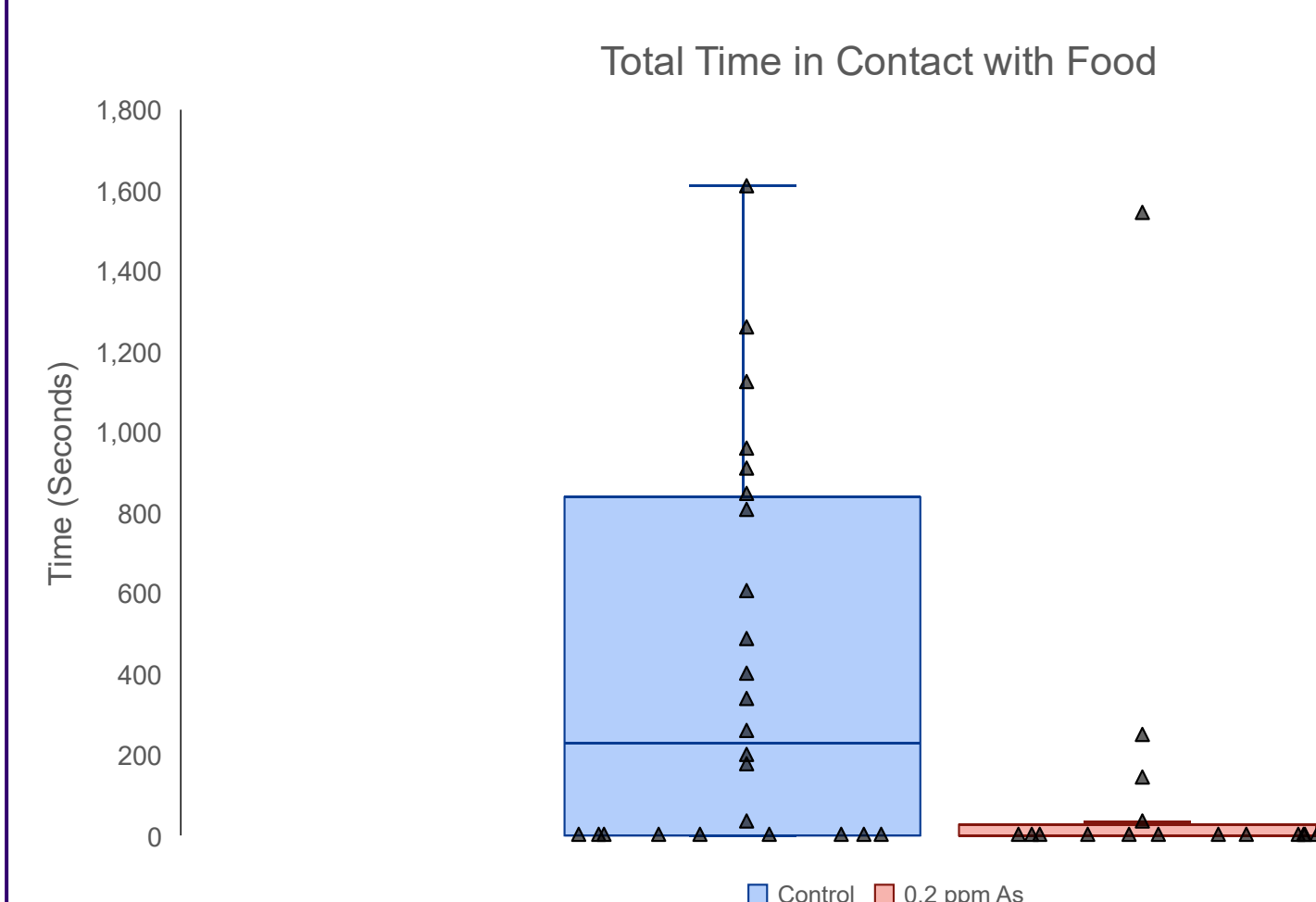


Figure 5: Arsenic exposure significantly reduced feeding behavior in Chinese Mystery Snails. Boxplots show the median, interquartile range (IQR; Q1-Q3), and min/max whiskers. Control (blue, N=24): median = 229s, IQR = 5375 (Q1=0s, Q3=5375s). 0.2 ppm arsenate (orange, N=16): median = 0s, IQR = 25 (Q1=0s, Q3=25s). The reduction is statistically significant ($p=0.02$; Mann-Whitney U, two-tailed, $p < 0.05$).

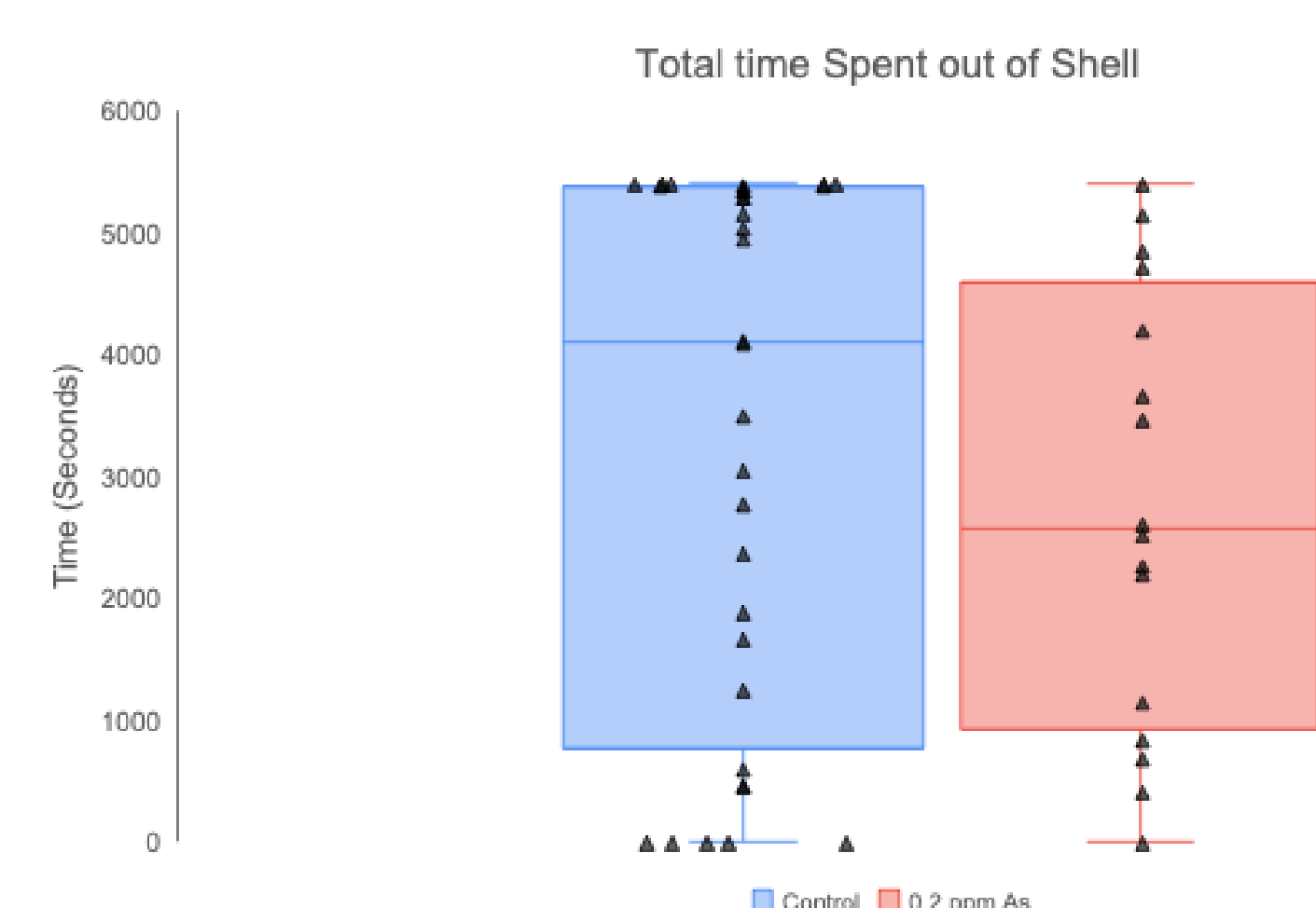


Figure 6: Arsenic exposure shows a trend toward reduced shell emergence behavior in Chinese Mystery Snails. Boxplots show the median, interquartile range (IQR; Q1-Q3), and min/max whiskers. Control (blue, N=33): median = 4101s, IQR = 4608 (Q1=767s, Q3=5375s). 0.2 ppm arsenate (orange, N=16): median = 2565s, IQR = 3666 (Q1=923s, Q3=4589s). This trend was not statistically significant ($p=0.25$; Mann-Whitney U, two-tailed, $p < 0.05$).

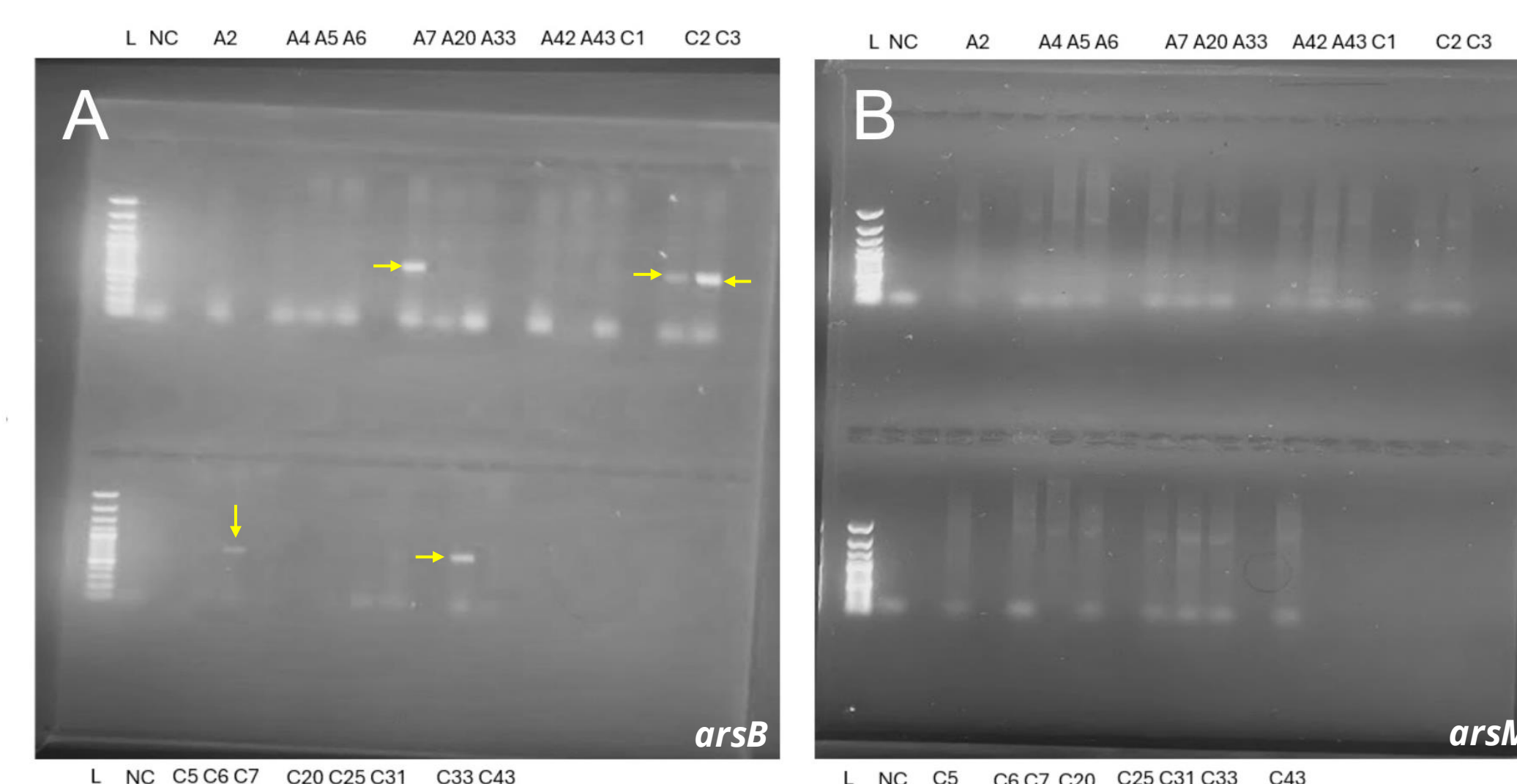


Figure 7: *arsB* was detected in the gut microbiome of select Chinese Mystery Snails in both arsenic-exposed and control groups. (A) Gel electrophoresis using the *arsB* primer shows distinct amplicons (yellow arrows) in sample A7, C2, C3, C6 and C33, with no amplification in the negative control (NC). (B) Gel electrophoresis using the *arsM* primer shows numerous non-specific amplicons, yielding inconclusive results. L = ladder; NC = negative control.

Discussion

- Behavioral impacts:** Snails exposed to 0.2 ppm arsenate showed a statistically significant decrease in feeding behavior compared to control snails, while no significant difference in motility was detected between groups.
- Exposure conclusion:** These findings confirm that non-lethal levels of arsenate measurably alter snail feeding behavior.
- ArsB gene analysis:** Gel electrophoresis for the arsenic-metabolizing gene *arsB* detected an amplicon in 1 out of 9 exposed samples, compared to 4 out of 11 controls.
- ArsM gene analysis:** Gel results for the *arsM* gene were inconclusive.
- Future directions:** Subsequent studies should examine gut bacteria colony-forming units between control and arsenic exposed CMS, to compare bacterial survival and arsenic tolerance.

Future Works

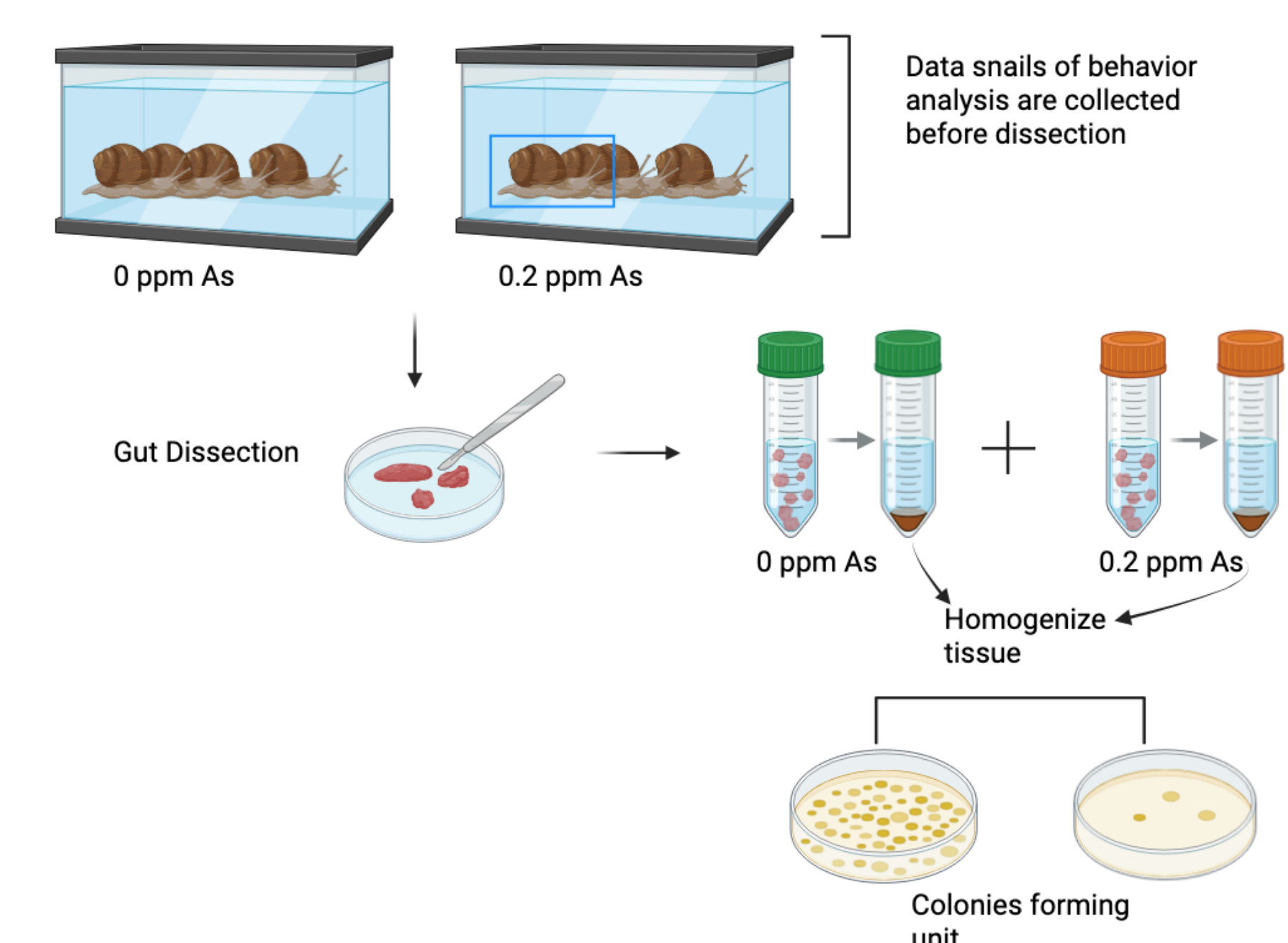


Figure 8: Gut bacteria survival and arsenic(V) tolerance in Chinese Mystery Snails. CMS guts will be dissected and homogenized for each arsenic(V) treatment group (0 ppm and 0.2 ppm). The homogenates will be plated on agar to allow colonies to form. Colony-forming units (CFUs) will then be counted to compare gut bacterial survival between arsenic-exposed and control snails

Acknowledgements

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References

