**Title** – Pollution Tolerance of Crayfish Ectosymbionts (Branchiobdellidans)

**Program of Study** – Biomedical Sciences

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**Abstract:** Macroinvertebrates are commonly used to study pollution tolerance in aquatic systems. However, it is relatively unknown how common pollutants (e.g. pesticides from run off) impact the symbiotic relationships of macroinvertebrates. In this study, two of the most commonly applied pesticides which make their way into streams, Glyphosate(GLY) and Atrazine(ATZ), are examined for effect on crayfish ectosymbionts (Branchiobdellidans). Branchiobdellidans were exposed to ATZ and GLY at 50 and 500 ug/L for 48-hours in single and combined treatments (10 Branchiobdellidans/treatment). ATZ and GLY did not result in mortality when exposed as a single pollutant at 50 and 500 ug/L. Interestingly, the combined effect of ATZ and GLY at 50 and 500 ug/L resulted in 100% mortality after 48-hours. The surviving Branchiobdellidans from the ATZ and GLY exposure were analyzed histologically to assess for sub-lethal effects (tissue degradation in the form of cell counts/total area) from the pollutants. Using ImageJ, cell counts (at 10X) were conducted on the surviving worms (control, ATZ 50 ug/L, and GLY 50 ug/L) by preparing the tissue with a cryostat and fluorescent DAPI staining. A one-way analysis of variance (ANOVA) was conducted on cell counts/total area (0.0 ug/L, ATZ 50 ug/L, and GLY 50 ug/L). The
analysis was significant, $F(2,19)=26.80$, $p=2.95\times10^{-6}$. A follow up comparison with the control (0.0 ug/L) revealed that the ATZ 50 ug/L cell count/total area was not significantly different ($df=14$, $p=0.12$), but the GLY 50 ug/L cell count/total area did have a significantly lower cell count/total ($df=10$, $p=0.0002$). These results of mortality and sub-lethal effects indicate the potential damaging effects of common pollutants on freshwater symbionts and provide helpful information for freshwater conservation.