Analysis

This experiment involved three different assays to investigate how behavior can be impacted by micro plastic exposure. The raw data for all of the assays can be found in Appendix B.

Locomotion Assay Results

Locomotive behavior was quantified by recording how many flies climbed up above the threshold every 0 second interval for a total of 120 seconds (See Appendix B for raw data). From a first glance, Figure 1 shows that more flies reached the threshold line than the other experimental groups. Specifically, at the 20 second mark, the control starts to separate from the other groups; then by the 30 second mark, the



Table 1: Z-test results for locomotion assay. The table below compares the average of the number of flies above the threshold line. This data utilizes the number of flies from the 120 second mark.

	Control vs 50 µg	Control vs 100 µg	Control vs 200 μg
<i>p</i> -value	0.001161	0.042317	0.186815

control has a visibly greater average number of *Drosophilae* above the threshold line. Table 1 depicts the *p*-values that were calculated utilizing the data from the 120 second mark, and it is seen that the results from the 50 μ g/mL and 100 μ g/mL resulted in statistically significant data, but the 200 μ g/mL did not. The error bars shown in Figure 1 weaken the correlation as the 200 μ g/mL error bars stretch up to the control data. The 200 μ g/mL have the largest error bars due to the wide range of data (see Appendix B), while the control data has the smallest error bars. The control vs 50 μ g/mL resulted in a *p*-value less than 0.01 while the control vs 100 μ g/mL resulted in a *p*-value less than 0.05. As shown in Figure 2, a linear regression analysis was also conducted with the data from the 120 second mark to quantify the relationship between an increased

concentration and a more significant change. The linear regression model suggests that there is a negative correlation between the number of flies that pass the threshold line at the 120 second mark and the concentration of exposure. However, the R² value for the model ended up being very low at 0.0838. This means that only 8.38% of the dependent variable—number of flies—is explained by the independent variable—the concentration of microplastics the *Drosophilae* were exposed to.

Table 2, the 50 µg/mL has a *p*-value less than 0.05, but when looking at just the blue solution

Feeding Assay

Feeding behavior was quantified by placing *Drosophilae* into centrifuge tubes where they only had access to capillary tubes filled with a sucrose solution. The *p*-values were calculated with a student's t test by compaing the final heights' of the capillary tubes. All capillary tubes were filled to the 5 μ L line, this corresponded to a height of 2.7 cm. Since they were all filled to the same height at the beginning of the assay, I only analyzed the final heights of the capillary tubes. In



Figure 3: Average Final Liquid Heights of all Capillary Tubes Across Experimental Groups. This data compared the final heights of the control the concentrations.

Table 2: *T-test results for feeding assay*. The table below compares the final liquid heights of the control to the experimental groups.

	Control vs 50 µg	Control vs 100 µg	Control vs 200 μg
<i>p</i> -value All Tubes	0.03342659	0.06151085	0.06958714
p-value Just Blue	0.00002353	0.00001033	0.00000202
p-value Just Red	0.12307883	0.09397269	0.05177601

tubes, all three conentrations resulted in *p*-values less than 0.001. In addition none of the *p*-values for the red solution are stastically

Table 3: *T-test results for feeding assay*. The table below compares the final liquid heights of the evaporation to all experimental groups.

	Evaporation vs. Control	Evaporation vs. 50 μg	Evaporation vs. 100 μg	Evaporation vs. 200 μg
<i>p</i> -value All Tubes	0.02251954	0.31611670	0.20222857	0.19089361
<i>p</i> -value Just Blue	0.0000063	0.06055869	0.00052745	0.00000022
p-value Just Red	0.46999097	0.42314631	0.31073727	0.35062686

significant. I saw qualitatively (see Figure 2) that the capillary tubes filled with the red solution were much higher than the capillary tubes filled with the blue solution, so I decided to run additional t-tests to look at just the data from a certain solution. This allowed me to conclude whether or not a factor aside from evaporation decreased the height of the liquid in the capillary tubes. Table 3 highlights these *p*-values. When looking at all capillary tubes,



Figure 4: Average Number of Flies Alive at the End of the Feeding Assay Across Experimental Groups. This data graphs the average number of flies alive.

Table 4: *T-test results for feeding assay*. The table below compares the number of flies alive after the 24-hour period in the control to the experimental groups.

	Control vs 50 µg	Control vs 100 µg	Control vs 200 µg
<i>p</i> -value	0.0014395	0.0000196	0.0000129

it can be seen that only the control produced data that was statistically significant from the evaporation, but the concentration groups were not. When looking at just the blue solution, all experimental groups resulted in statistically significant results; and again, the data from the red solution is not statistically significant. Additionally, after the 24-hour feeding period, I noticed that the *Drosophilae* exposed to microplastics had experienced high mortality rates (shown in Figure 4), so I decided to run a t-test on the number of flies alive as all centrifuge tubes started out with eight *Drosophilae*. As seen in Table 4, the 50 μ g/mL has a *p*-value less than 0.01 while the 100 μ g/mL and 200 μ g/mL concentrations have a *p*-value less than 0.001. Even with the error bars shown in Figure 4, the control on average had many more *Drosophila* survive through the entirety of the assay.

Social Space Assay

The social space assay quantified social behavior by looking at the distance between organisms. This assay was limited as I ran into unexpected mortality so I had to limit the assay just to the control, 100 μ g/mL, and 200 μ g/mL concentrations. In Figure 5, the data from each experimental group is close in number and each group has a large error bar. A t-test was run to com pare the control to the concentration groups. As seen in Table 5, only the 200 μ g/mL resulted in a *p*-value less than 0.05



Figure 5: Average Distance Between Organisms Across Experimental Groups. This data contains the average distance between a fly and its nearest neighbor.

Table 5: *T-test results for social space assay*. The table below compares the number distance between flies from the control to the experimental groups.

	Control vs 100 µg	Control vs 200 µg
<i>p</i> -value	0.580716	0.040195