

# Effects of E. coli on Zebrafish Development

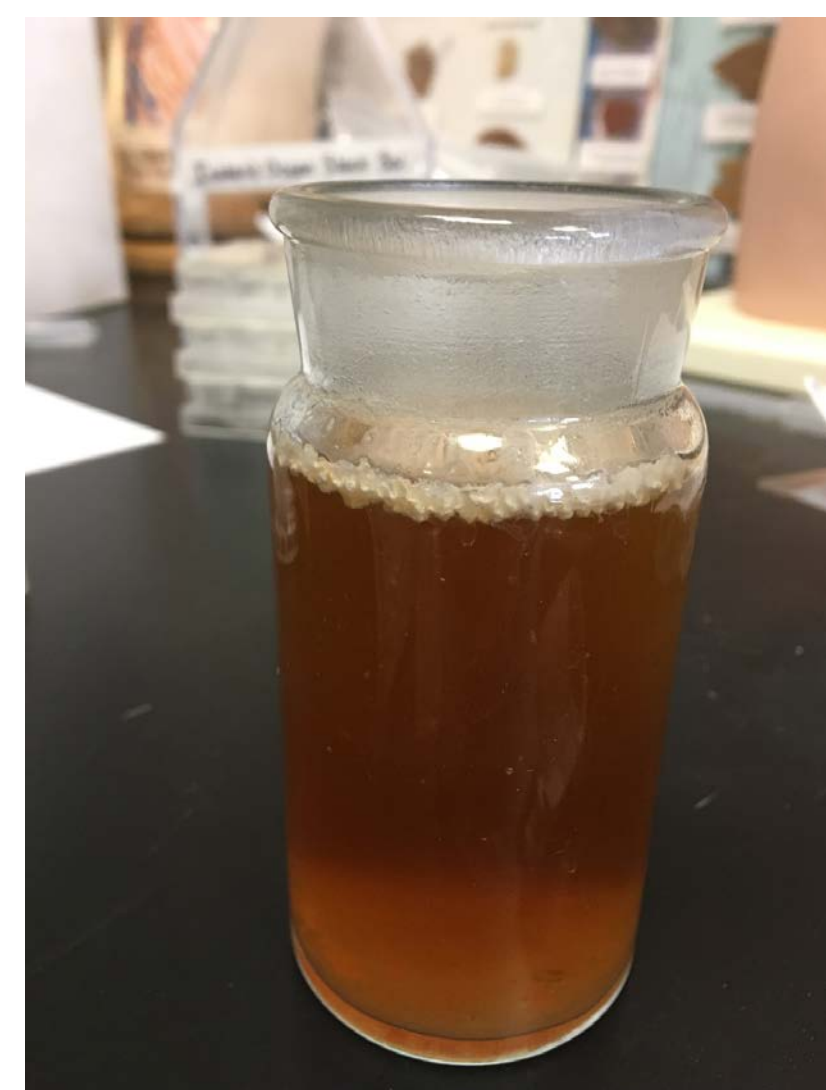
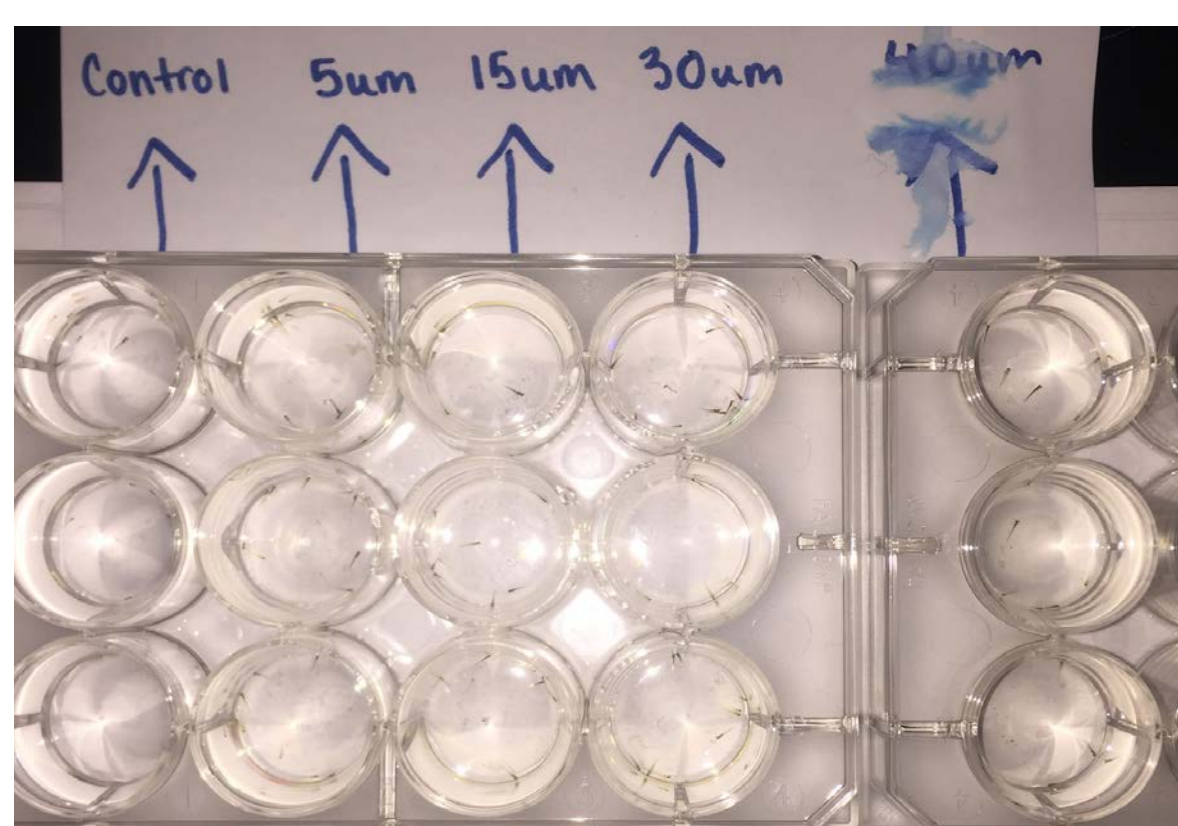
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## ABSTRACT

*E. coli* is a gram-negative bacteria and contains amyloid these fibers are found in the brains of those affected by Alzheimer's. Zebrafish were exposed to varying levels of *E. coli*, 0um, 5 um/4 mL, 15um/4 mL, 30um/4 mL, 40um/4 mL. The embryonic development of the fish were observed for 96 hours. It was found the fish exposed to higher quantities of *E. coli* had a higher mortality rate the first day, did not have the same reflexes, had smaller body frames, and did not naturally swim around as the control group did. The results suggest exposure to *E. coli* could lead to potential problems with embryonic development and brain activity.

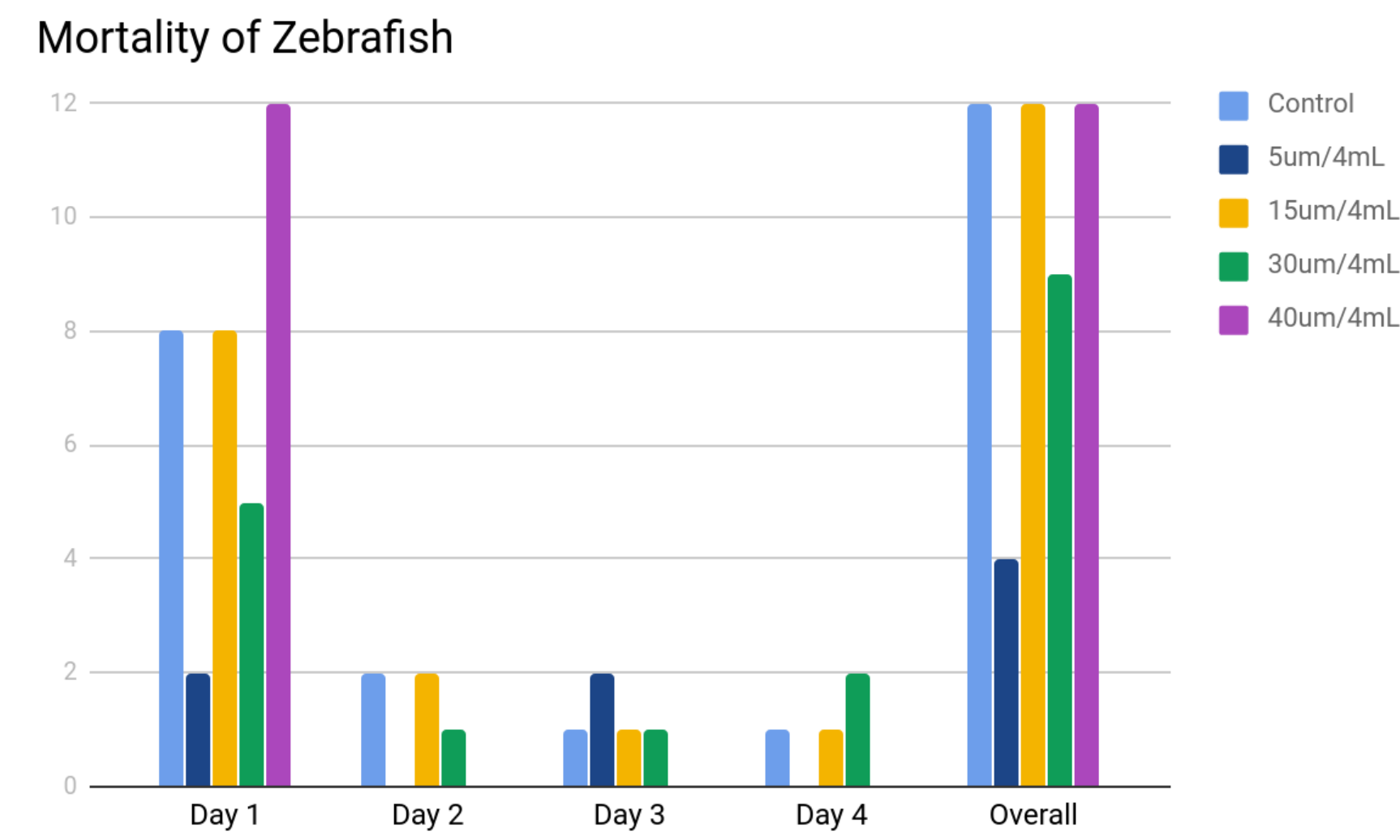
## INTRODUCTION

*E. coli* enter aquatic systems through feces, which can hurt animals and humans ("E. coli in Our Lake" 2014). *E. coli* is categorized as a gram-negative bacteria, which causes them to reject antibiotics and other immune defenses. It has been found that certain strains of *E. coli* produce amyloid fibers. Amyloid fibers, when combined into clusters, form amyloid plaques ("E. coli Bacteria Make Alzheimer's-Linked Fibers," 2002). These plaques are similarly found in the brain of a person with Alzheimer's disease. Amyloid production in *E. coli* occurs in relation to two proteins: CsgA and CsgB. CsgA is released by the bacteria, whereas CsgB is embedded inside the wall of the bacteria and exposed to the outside. The CsgB protein contains a nucleus that triggers precipitation of dissolved CsgA proteins, which combine into amyloid fibers attached to the CsgB protein. The plaque formation is thought to be due to either production of the amyloid fibers or triggering precipitation of previously present amyloid precursor proteins. These amyloid plaques lead people to believe that *E. coli* bacteria may be considered a possible cause of Alzheimer's disease. The recent discoveries of digestive bacteria, such as *E. coli*, relating to Alzheimer's disease is crucial. Alzheimer's is a disease in which amyloid plaques build up and change the structure of the brain. The brain shrinks in certain areas and spreads out in others. This results in people becoming confused, agitated, emotional, and to acquire great deals of memory loss.

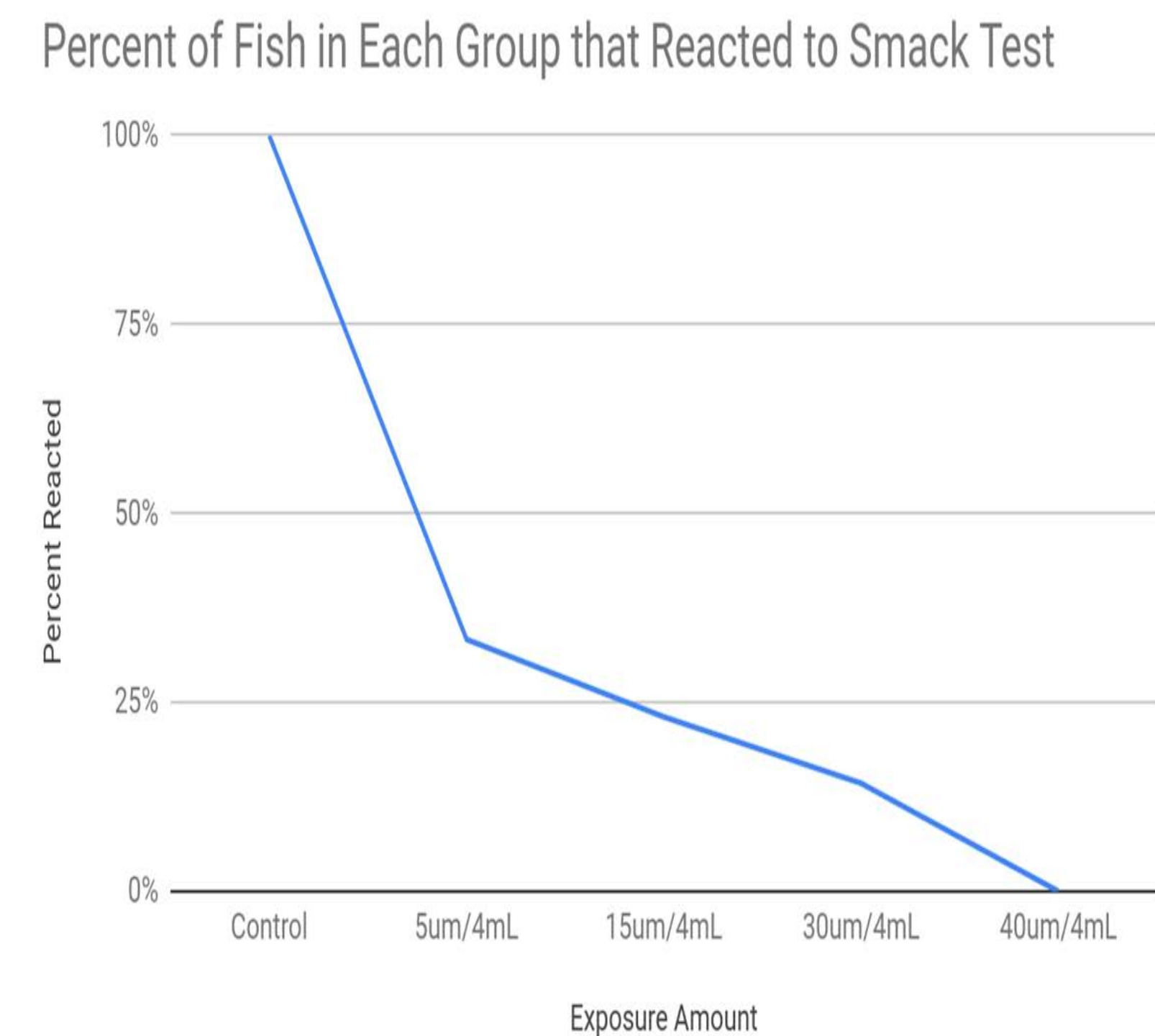


## PROCEDURE

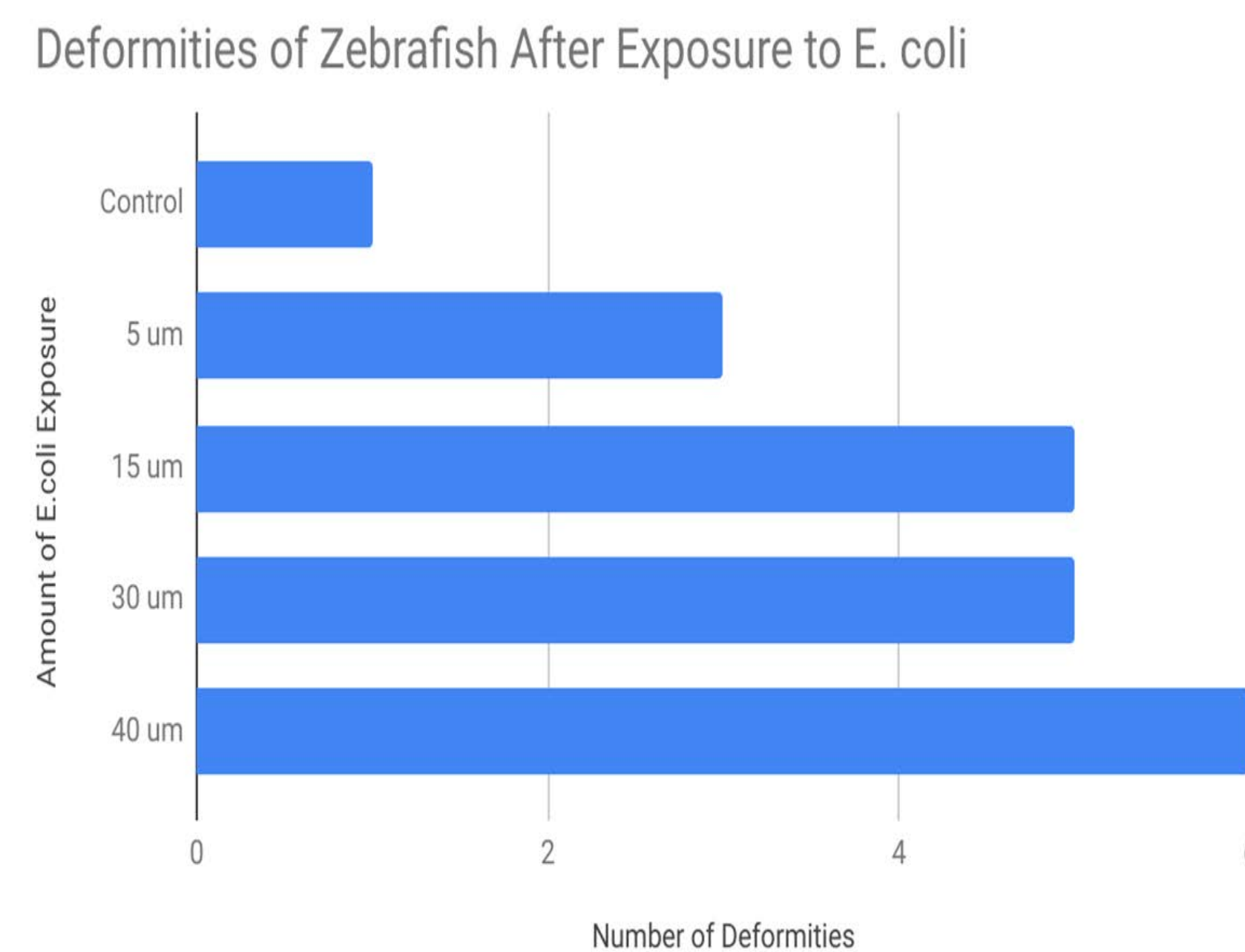
1. Make a slurry of *E. coli* and beef broth
2. Make instant ocean of put 4 ml in each culture
3. Place the desired amount of *E. coli* slurry in each culture
4. Place 10 fish in each culture, having 30 fish for each test group
5. Place fish in an incubator overnight
6. Count how many fish have died, hatched, and any noticeable deformities
7. Replace the fishes water (step 2-4) and then return them to the incubator
8. Each day mortality rates were measured, deformities were noted, and response time to sound vibrations were taken.



**Figure 1: Results:**  
The graph indicates the deaths for the four day test period. The 40 um group had the most amount of death on the first day. No group had more than twelve out of the total thirty (in each group) die. The chi square test was not significant, therefore mortality rates were not affected by *E. coli* exposure.



**Figure 2 Results:**  
As the amount of exposure to *E. coli* bacteria increased, the number of fish that reacted to the sound vibrations test decreased. In the control group, every fish reacted to the vibrations, showing good reflexes. However, the 40 um group none of the fish react. The remaining groups in between demonstrated a gradual decrease. The chi square test was significant at .05, meaning that reflexes were affected by the *E. coli* exposure.



**Figure 3 Results:**  
The number of deformities in the fish increased as the exposure to *E. coli* increased. The 15 um group and the 30 um group had the same number of deformities. However, the control group had one fish with a deformity whereas the 40 um group had six. The data supported the hypothesis; the chi square was significant at .05. The fish had different deformities in their overall size, eyes, and spinal cords.

## DISCUSSION

- In the control group, only one of the zebrafish showed physical deformities and all fish passed the vibrations test.
- In the 5 um group showed that 33.333% of the fish reacted to the smack test and 11.53% of them showed physical deformities.
- The highest group, 40 um did not react to the slap test at all. They did, however, show the highest amount of deformities out of all five groups, at 33.333%.
- The groups in between showed an incremental decrease in reflexes and an increase in deformities
- Mortality rates were not significantly affected by the *E. coli* exposure.

Errors that could have occurred:

- Since the *E. coli* was made in a slurry, there is no exact bacteria count in each sample taken
- The *E. coli* continued to grow in the dishes, meaning the fish could have been exposed to a higher amount of *E. coli* than estimated
- The *E. coli* used is not the same *E. coli* found in our stomach, therefore it is not insured that the *E. coli* found in our intestinal tract will behave the same as the bacteria tested.



30um



40um



Control



5um

## CONCLUSION

The data supported the hypothesis that higher amount of *E. coli* will have an effect on physical deformities and reflexes. It did not support that *E. coli* affected mortality rates. As seen on the figure 1, the control fish did not experience any less deaths than the fish in the 40 um/4mL. This shows that being exposed to *E. coli* does not have a strong enough impact to kill the fish. In figure 2 it shows how the *E. coli* impacts reflexes. The 5um/ 4mL group had 33.3% react, 66.7% less of the group react to the test conduct. From there the 15um/ 4mL dropped down to 23.08% react, 30um/4 mL had 14.29% of the group react and the 40 um/4mL didn't react at all. There was also an increase in deformities, 5.5% of the control had deformities, it jumped up to 11.53% of the 15um/ 4mL, then to 27.7% of the 15um/ 4mL group. The percentage of deformities dropped down in the 30um/4 mL group, from 27.7% to 23.8%. Then it raised back up to the 40um/ 4mL group showing 33.3% of them had a physical deformity. This research suggests that if women are exposed to *E. coli*, during pregnancy, it could lead to both mental and physical set back and deformities.