Zebrafish Hatched and Living After Exposure to Round-up: Glyphosate

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ABSTRACT:

The purpose of this experiment is to the result of glyphosate on developing embryos. The method that was used to complete this experiment was placing embryos in a well plate with different chemical concentrations. The results ended up that 0.1 and 0.12 mg/mL concentrate of glyphosate kills zebrafish embryos, without even hatching. The results were statistically significant because every embryo that came in contact with the glyphosate died.

INTRODUCTION:

On December 12, an experiment was begun to determine the effects of glyphosate on zebrafish, which pertains to the topic of environmental health. There have already been studies on this topic, and all the experiments used a non-toxic amount (Industry Task Force on Glyphosate, 2013) and have not studied long term effects (Earth Open Source, 2012), but only found it to be harmful to the animal uterus (Engdahl, 2016). The predicted outcome of this experiment was for the control group to survive and develop healthily, the embryos exposed to 0.1 mg/mL concentrate will survive, but with defects, and the embryos exposed to a concentration of 0.12 mg/mL will not survive.

MATERIALS AND METHODS:

The zebrafish embryos and experiment materials were received on December, 12th 2016 with support from the UW-Milwaukee's Science Education Partnership Award (SEPA) program which is sponsored by the National Institutes of Health.

- **2 Round-up stock solution concentrations low 0.1mg/mL and medium 0.12mg/mL**
- **1 bottle instant ocean/Embryo media solution (concentration 60 mg/mL)**
- **4 wide bore transfer pipettes**
- **1 transfer pipette, minimum bore, 1.5 mm: (for transferring eggs to observation container and manipulating them in the container)**
- **1 sharpie**
- **1 beaker of dead embryos and liquid disposal**
- **1 83.3 degrees fahrenheit incubator**
- **1 multi-well plate**
- **1 depression slide and cover slip**
- **1 dissecting and compound microscope**

In this experiment a well plate was filled with three different chemical concentrations, then 10 embryos were placed in each well. Every day, after taking observations, the dead embryos, and/or the egg chorion were removed. After that the chemical mix was sucked out, and replaced with fresh mixture. For safety, gloves were worn so there was no chemical contact, as they could be harmful. The statistical t-test was used to compare the two groups, hatched and live, to see their correlation with each other using their means.

Treatment	Beginning number	Well 1	Well 2	Well 3	Well 4	Average	Probability	Result
Control	40	5	8	7	10	7.5	-	-
0.1mg/mL	40	0	0	0	0	0	p=0.0004	Statistically significant
0.12 mg/mL	40	0	0	0	0	0	p=0.0004	Statistically significant

Figure 1: Shows how many fish were alive on the last day of the experiment.

Treatment	Well 1	Well 2	Well 3	Well 4	Average	Probability	Result
Control	1	5	7	10	5.8	-	-
0.1mg/mL	0	0	0	0	0.0	p= 0.0226	Statistically sigificant
0.12mg/mL	0	0	0	0	0.0	p= 0.0226	Statistically significant

Figure 2: Shows how many fish hatched on the last day of the experiment.

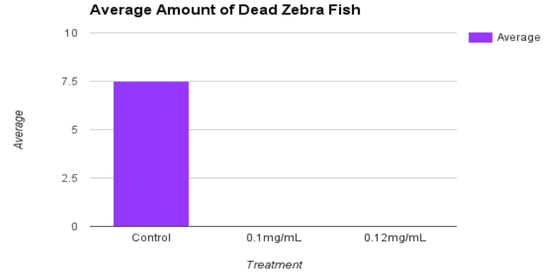

Figure 3: Overall amount of zebrafish deaths. All died except for the control group, which still lost a few.

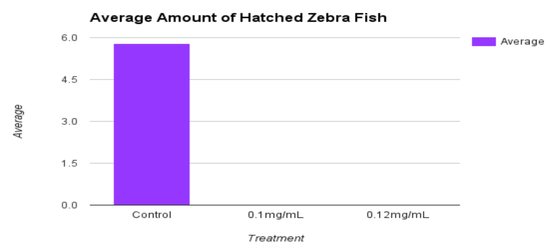

Figure 4: Overall amount of hatched zebrafish. Majority hatched in control group, otherwise no other ones hatched.

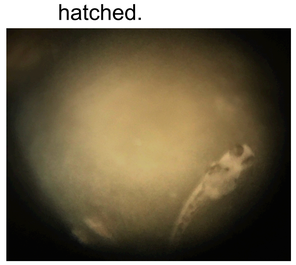

Figure 5: Concentration 0.12 mg/mL- dead, hatched, small curve of the spine.

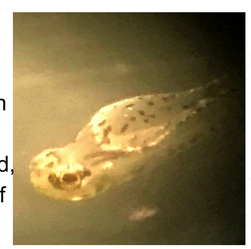

Figure 6: Control- Live, hatched, straight spine, blood flowing, swimming freely

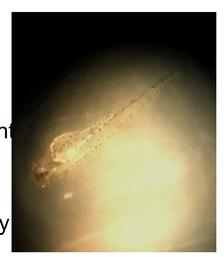

Figure 7: Control- Live, hatched, straight spine, blood flowing, swimming freely.

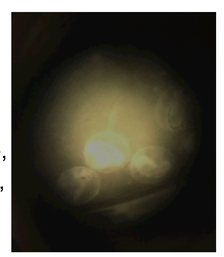

Figure 8: 0.1 mg/mL concentration- Dead, unhatched, curved spine, very little development.

RESULTS:

The conducted experiment to see how much Roundup an embryo can be exposed to, without it becoming toxic. The hypothesis was that the fish with the concentration 0.1 mg/mL would be fine, while the 0.12 mg/mL would all die. In the end, the control survived, and both of the concentrations died. Overall all of the zebrafish in the concentration 0.1 mg/mL or 0.12 mg/mL died before they hatched, dying after only one day.

This experiment turned out to be statistically significant. The amount of dead zebrafish was ultimately because of the glyphosate that was added, as all of the ones that were exposed to it, died.

DISCUSSION:

The main results of this experiment showed that a chemical mix with the concentrate 0.1 mg/mL and 0.12 mg/mL of glyphosate will kill zebrafish embryos, while still giving it curved spines. The results negate the hypothesis. The hypothesis was that the 0.1 mg/mL would survive with minor defects, but they had slight defects and they died. However, it was hypothesized that the 0.12 mg/mL would die, and they did.

There could always be different reasons for the deaths besides the chemicals, but since the results were significant, the odds are low. The results are relevant because we are narrowing down the range of the level of concentration that is not toxic. To further this research, one would need to do the same experiment, but lower the concentration levels.

REFERENCES:

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