

The Effect of Caffeine on the Development of Zebrafish Embryos

By: Aurora Thomason

Abstract

This experiment tests the effect of caffeine on Danio Rerio (zebrafish) embryos. In this experiment zebrafish embryos were placed into a diluted caffeine solution (experimental group), and another set of embryos were placed into an instant ocean solution (control group). The embryos were observed every 24 hours for 72 hours. The results show that the embryos placed in the caffeine solution developed to have an abnormal curvature in their spines, unlike the embryos in the instant ocean solution, which had straight spines. This data is important because zebrafish are a model organism, which means that any effect zebrafish take on could be the same effect that a human would take on. If caffeine causes a curved spine in zebrafish embryos we can infer that it will cause a curved spine in a human fetus if the fetus is under the same circumstances.

Introduction

Zebrafish were chosen for this experiment because they are a model organism. This means that the buildup of this fish is similar to a human, so if zebrafish are experimented on it is inferable that whatever the outcome of the fish was (under an experimental environment) could also be the same for humans.

The variable tested was caffeine. The zebrafish embryos received a dilute caffeine solution, and were observed every 24 hours for 72 hours.

The purpose of this investigation was to see what would happen to a zebrafish embryo if it was put under certain circumstances. Since zebrafish are model organisms, there is a chance that the effect the zebrafish take on could be the same as a human child, if put under the same circumstances. (Petering, Berg, Tomasiewicz, et al., 2018).

Results

In this lab, 120 zebrafish embryos were placed in a 12 well plate. Half of the embryos were placed in a diluted caffeine solution (experimental group), and the other half were placed in an Instant Ocean solution (control group). The embryos were observed every 24 hours for four days. Evidence shows as the time that the zebrafish developed in the caffeine went up, the percent of the curvature in the spines went up as well. However, the zebrafish in the control group developed normally. In the experiment, it was found that at 48 hours, the control group had 0% curvature in spines, and the experimental group had 13% curvature in spines. At 72 hours, the control group had 2% curvature in spines and the experimental group had 93% curvature in spines.

Materials and Methods

The materials used were:

- Stock solutions of caffeine
- Beaker for dead embryos and liquid disposal
- 1 sharpie
- Instant ocean/embryo media solution
- Disposable pipette, minimum bore, 1.5 mm for transferring eggs to observation container and manipulating them into the container
- Disposable pipette
- Plate with wells
- 28.5C incubator
- Dissecting and compound microscope

(Petering, Berg, Tomasiewicz, et al., 2018).

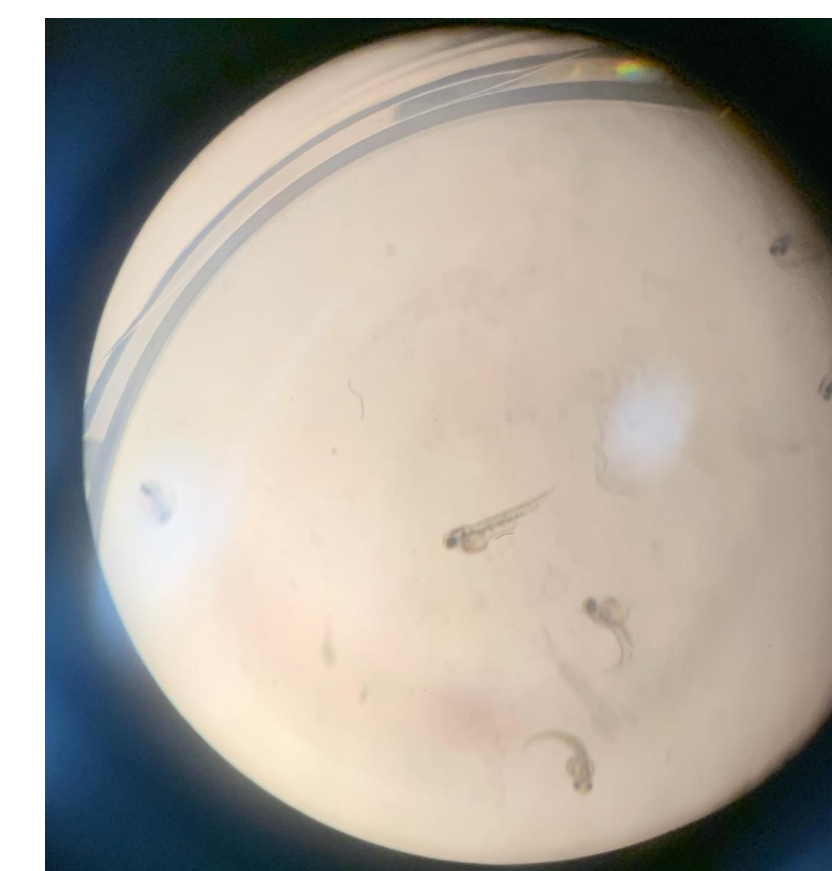
On the first day, we obtained rinsed embryos from our teacher. Then we filled the wells of the plate with 1 mL of Instant Ocean/Embryo Media solution using the disposable pipette. After that we filled the remaining wells with the diluted caffeine solution. The embryos were divided so there are approximately 10-15 embryos in each well. We then labeled the plate on the student data sheet. Exact numbers of live embryo were recorded on our data sheet. Then the plate was placed in the 28.5OC incubator overnight. On the second day we removed the plate from incubator and removed dead embryos from the plate using the transfer pipette. Then the dead embryos were placed into the waste beaker. Count remaining embryos, hatched fish, and record in data table. j. Remove caffeine stock solutions from each well of the plate. k. Add the appropriate fresh caffeine stock solution using a clean pipette each time. l. Place the plate under dissecting microscope and record observations on student data sheet. Note/describe any developmental markers and abnormalities. Repeat for all caffeine concentrations. m. Remove 1-2 embryos and place on the depression slide with cover slip. Observe the embryos using the compound microscope. Record observations on student data sheet. Repeat for all caffeine concentrations. n. Return the embryos to their well in the plate. o. Return the plate to the appropriate 28.5OC incubator (Petering, Berg, Tomasiewicz, et al., 2018).

Conclusion

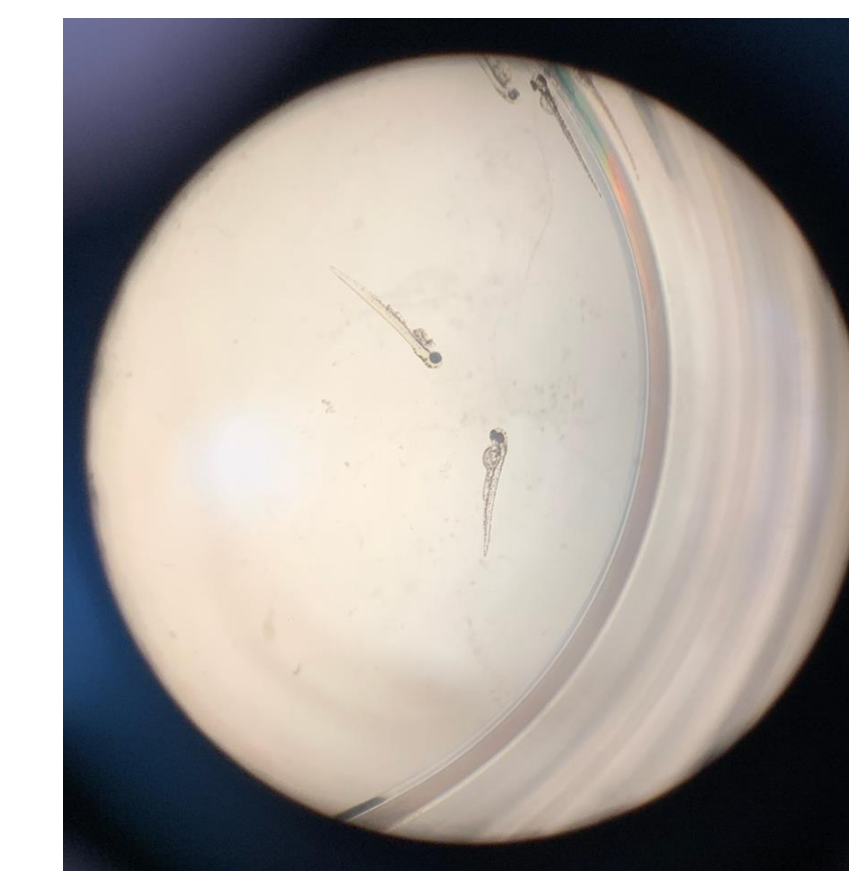
This topic is important because zebrafish are a model organism for humans, and it is inferable that whatever effects a zebrafish embryo may also affect a human fetus. For example, if caffeine causes a curvature in the zebrafish embryo's spine, then it is possible that a human child's spine may be curved if exposed to those amounts of caffeine pre-birth. That's why this experiment is important.

Since the data shows that caffeine causes a curvature in the zebrafish embryos spine, we can infer that a human fetus may take on the same effects. This way, we can inform pregnant women to not drink caffeine, and that may cause the baby to be healthier when it is born.

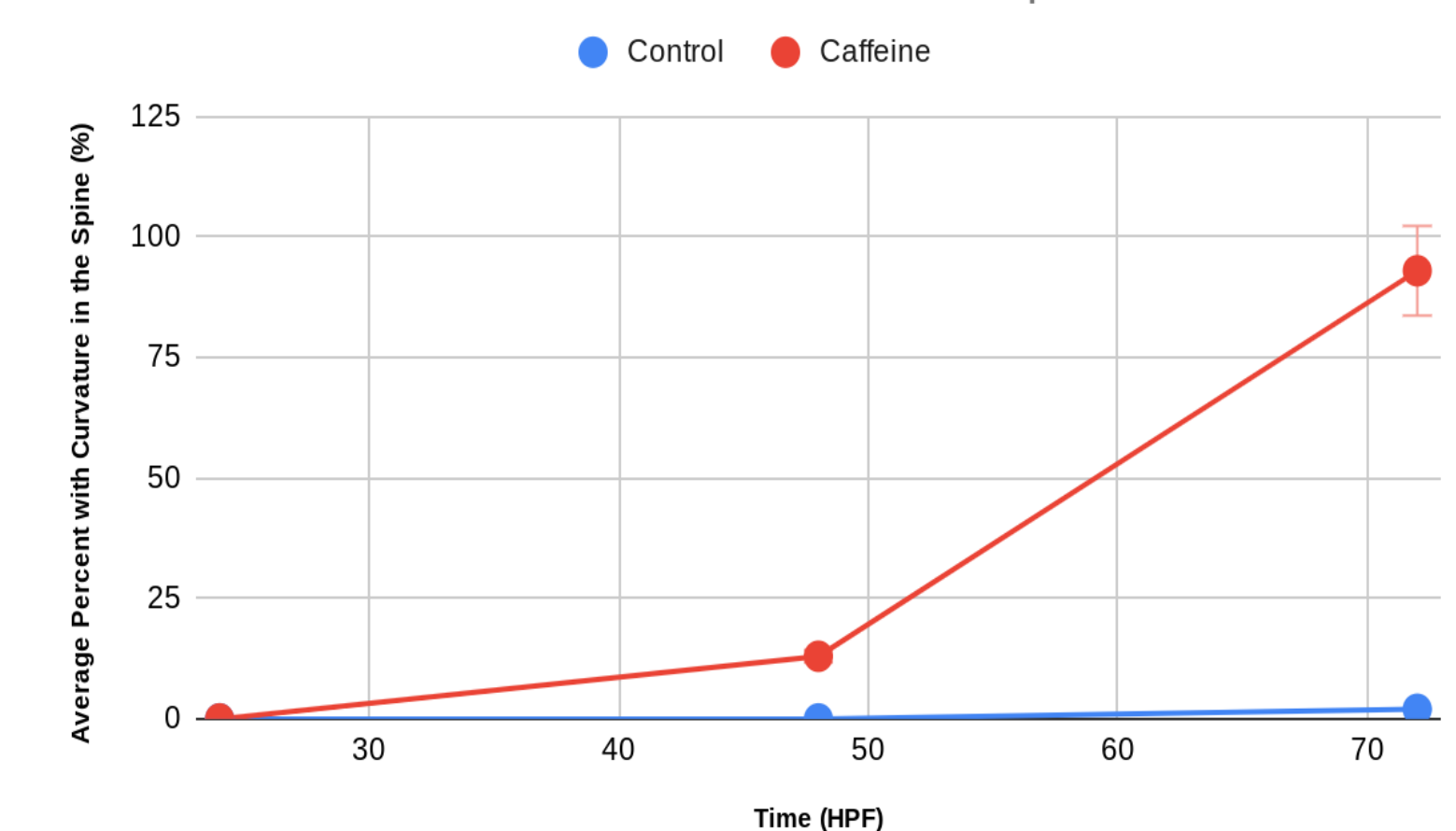
Experimental Group



Control Group



The Effect of Caffeine on Curvature of the Spine in Zebrafish



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