The Effects of Caffeine Exposure Presented on Zebrafish Embryos

Abstract:
Providing a purpose in testing this experiment was to examine the different effects of caffeine exposure on zebrafish embryos. When controlling a constant concentration of 0.25mg/ml of caffeine was exposed to forty different embryos over an amount of seventy two hours post fertilization and then counted every twenty four hours prior to the solution. The results after tested, did not show any evidence in proving that the amount of caffeine presented was enough to be statistically significant. Although when comparing the concentration of 0.25mg/ml of caffeine to even higher concentrations, the results conclude as a higher impact when distinguishing the different amounts together. In relation to the zebrafish embryos, the overall results of caffeine concentration every twenty four hours allows scientists to examine the impacts affected, when comparing them to the actual human body in the first three major stages of life.

Introduction:
Known as a commonly found stimulant present in many of the craved beverages we humans drink today, caffeine has been found to be one of the main chemicals in leading to a widespread of major birth defects on animals. Found to be in the diets the majority of Americans consume, it is also expected that existing mothers may be drinking caffeine on a regular basis as well. The scientific research found based on the consumption of caffeine intake during pregnancy proved to have found numerous studies done on animals in relation to what has shown to be effective. (“Caffeine During Pregnancy - American Pregnancy2015”). In further studies shown to affect animals beyond birth defects, caffeine has also found to cause premature labor, preterm delivery, reduced fertility, lower birth weight, and other reproductive problems. Often questioning scientists in the process of whether or not the practices tested on humans would be effective, no other conclusive studies today have shown to be concluded. When applied more findings, other science researchers have tested and noticed that their zebrafish embryos had shown a sign of uncontrolled twitching and higher heart rates. (“Caffeine Effects on Zebrafish Embryo Development” compared to Normal Zebrafish Embryo Development2015). Distinguishing between the impact of results on the zebrafish embryos, it is made known that when exposing them to a higher concentration of 1.0mg/ml caffeine, the results would prove to be statistically significant. (“The Effects of Different Caffeine Concentrations on the Development of Zebrafish Embryos” 2015). Furthermore, I wrote my hypothesis stating, “If I expose a certain amount of zebrafish embryos to 0.25mg/ml of caffeine, then I believe the total concentration exposed over a total of 72 hours would increase the amount of deaths because of the higher expected chances of getting a birth defect”.

Method (Materials):
- 80 fertilized zebrafish embryos
- Caffeine stock solution of 0.25mg/ml
- Instant Ocean solution
- Blue antibacterial solution
- Beaker for dead embryos liquid disposal
- Plate with wells depression coverslip
- Disposable pipette (1.5mm for transporting eggs)
- Disposable pipette (1ml)
- 28.5 degrees Celsius incubator
- Label tape
- Sharpie marker
- Dissecting/Compound microscope

Method (Procedure):
Day 1:
1.) Obtain Materials
2.) Using a sharpie and some label tape, label your well plate coverslip with each a control and experimental, numbering 1-4 for each of the wells
3.) Place 10 embryos into each of the wells making sure they all are alive
4.) Replace the access instant ocean solution from each of the wells carefully and refill them back up each with 3 ml of the solution
5.) Place the 0.25mg/ml concentration into each of the four experimental wells, filling the amount of concentration up twice
6.) Obtaining the blue antibacterial solution, place two drops each into all of the filled wells
7.) Put the depression coverslip back on your well plate and place it into the 28.5 degrees Celsius incubator

Day 2:
1.) Remove your well plate from the incubator
2.) Slowly and carefully replace the dead embryos from each of the wells and place them into a waste beaker (if you’re not sure if the embryo is dead or alive, make sure to look at it underneath a microscope)
3.) Count the remaining embryos from each of the wells and record the number into your data table
4.) Remove the solutions from each of the wells and replace each old solution/concentration once and then using the same technique used the first day of experimenting

Day 3:
1.) Repeat day 2

Control Day 1: Figure #1
Experimental Day 1: Figure #2

Control Day 2: Figure #4
Experimental Day 2: Figure #5

Control Day 3: Figure #3
Experimental Day 3: Figure #6

References:
http://www.ncbi.nlm.nih.gov/pmc/articles/PMC3648459

Method (Materials):

Method (Procedure):

Control Day 1:

Experimental Day 1:

Control Day 2:

Experimental Day 2:

Control Day 3:

Experimental Day 3:

References:
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