

NO DISERNABLE DIFFERENCE IN BODY LENGTH OF CHICK EMBRYOS TREATED WITH ALCOHOL AND CONTROL (NO ALCOHOL)

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Introduction:

This report has to deal with the effects of alcohol on embryonic development and more specifically the effects of fetal alcohol syndrome. Past research has shown that embryos treated with alcohol are much smaller and less advanced in their development when compared with other embryos at the same stage, not treated with alcohol (Ahlgren, 2002). It is also believed that developmental flaws in the neural tube effect the gene expression of the sonic hedgehog-signaling cascade, which thereby significantly alters the rest of the embryo's development (Yamanda, 2005). This is an important subject to study because it is an issue strictly confined to the human species and one that is also easily avoidable. The more research and literature is available for public consumption, the better the chances of reducing cases FES in the future. In this study, we tested the effects of embryonic exposure to alcohol on chicks to test the hypothesis that the addition of alcohol into three day old, explanted, fertilized, chick embryos will result in smaller body lengths than the control chicks that were not exposed to alcohol.

Materials and Methods:

Materials needed to complete this lab included a lab notebook, instruction sheets from *A Laboratory Text for Developmental Biology* (by P.B. Armstrong et al. Kendall/Hunt Publishing Co. 1994.), and other materials as described in the P.B. Armstrong protocol.

Begin by following the Chick-in A-Boat procedure given in the handout by Armstrong *et al.* Note the following differences. Did not use Betadine solution. Held the egg low over sterile fixed Weigh-boat dish (not in a plastic wrap hammock).

Used the tips of forceps, not a dissecting needle, to make a hole in the small end of the egg. Then labeled dishes before placing them together in the incubator. Then labeled the lid of the petri dish around the periphery in order to see the embryo through the lid. Then labeled the bottom of the dish along the vertical side of the dish bottom to tell which embryo is which even when the lid is off the dish.

Ten-day chick embryos (*Gallus gallus*) were explanted from their shells as per the method of Morris (2015) and Armstrong, et al., (1994) except that a sterile weigh boat was used instead of a sterile hammock described by Armstrong. Ethyl alcohol was administered to each embryo to achieve a final concentration on-the-cells of 0.0 % (for control), or 0.002% (for low ethanol dose), or 0.02% (for high ethanol dose). It is important that a control substance was used in order to have the same level of disturbance in each of the three embryo treatments. Penicillin/streptomycin was also added to each embryo to achieve a final concentration on-the-cells of 100 U mg/ml.

I opened the door of the big incubator as infrequently as possible. Then removed the embryos from the incubator to do measurements, measured the pulse first. As the embryos cool, their heart beat rate dropped. Doing this measurement when the embryos are warmest helped ensure consistent data.

I then measured other features as described in class using a millimeter ruler and the ocular units on the microscope, and recorded quantitative and qualitative results in lab notebook. Data sheets helped record quantitative results. Measurements include: area vasculosa diameter, body length, wing bud length, leg bud length, and allantois width. The regular lab notebook was used to record qualitative results and any conclusions reached during measurement. I replaced embryos in incubator, but did not stack dishes. I placed dishes beside each other on the shelves. (Stacking the dishes causes condensation that obscures the view.)

Results:

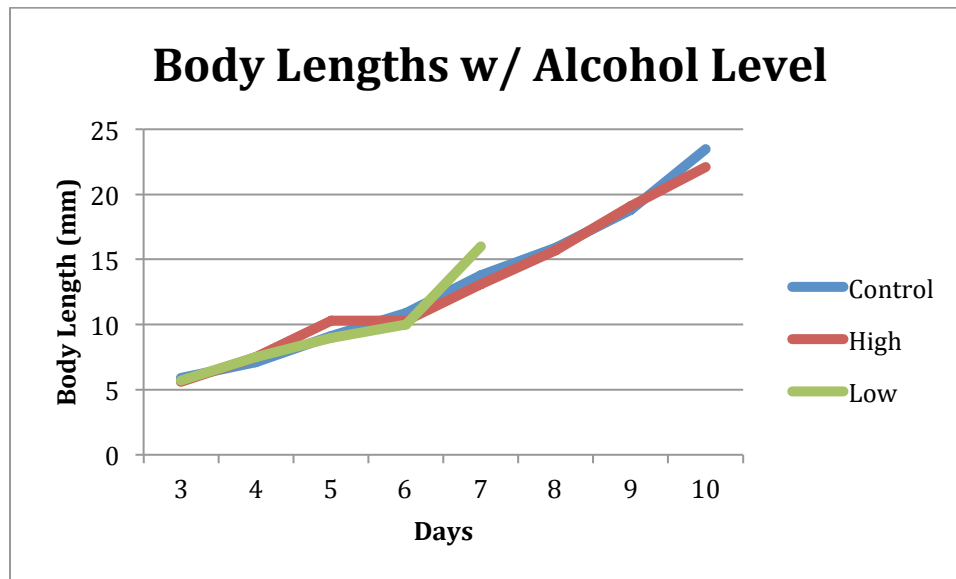


Figure 1: Data for day 3 represent 11 control embryos, 3 low, and 12 high alcohol embryo treatments. Data for day 4 represents 13 controls, 2 low, and 12 high. Data for day 5 represent 13 controls, 1 low, and 11 high. Data for day 6 represent 8 controls, 1 low, and 11 high. Data from day 7 represent 8 controls, 1 low, and 11 high. Data for day 8 represent 7 controls, 1 low, and 10 high. Data from day 9 represents 4 controls, 0 low, and 9 high. Data from day 10 represents 4 controls, 0 low, and 8 high.

As can be seen in figure 1, the control embryos have a slightly higher average body length than the high alcohol concentration embryos, and a significantly higher average than the low-level alcohol embryos. It is important to note the text and data in the figure legend as it explains the relative “power” for each data point. Because of the decreased number of observable embryos at later dates in the experiment, (due to dead embryos), the data from those days have significantly more variability and less quantitative reliability because of their decreased sample sizes. However, it does not appear that there is a significant difference between the three treatment levels on wing bud length. They all follow a similar upward curve and often crisscrossing each other at various points along the graph. There are very few spikes in the data that would indicate outliers or inaccurate observation/calculations (growth should be relatively constant).

Discussion:

From the data collected in this study, it can be concluded that there is not much of a difference between the low, high and control treatments of alcohol in regards to body length of developing chick embryos. While the final point on the graph for the control line is slightly higher than that of the high treatment line, it is not sufficient enough to separate the two. The three lines also essentially mirror each other as they increase across

the graph. All of this in conjunction with each other leads me to reject my hypothesis that alcohol treated embryos will have smaller body lengths than the control embryos due to insufficient evidence. These were not the results I expected because in all of the research that I came across there was an emphasis that embryos with fetal alcohol syndrome tend to be small and underdeveloped (Yamanda, 2005). The underdeveloped aspect of the FES embryos may still be true and observable in other parts of the chick, such as in the wing/tail bud length or in the allantois width, but those would be separate experiments to be done in the future.

References:

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