Wing bud development based on varying concentrations of ethanol exposure

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Introduction:

In this study we observed fetal alcohol syndrome as it presented in explanted chick embryos starting at day three of development post fertilization. The observation period continued through day ten. Johnson et al. (2007) also tested the effects of early alcohol exposure by looking at the consequences of ethanol exposure on mice, their results showed a positive correlation of ethanol exposure to birth defects. Knowing more about the mutagen effects of alcohol on developing embryos will help to promote general pre-natal health knowledge with a better understanding of the consequences of embryonic alcohol exposure. This information can be applied to both human and non-human animal development. In this study, we tested the effects of embryonic exposure to alcohol on Gallus gallus to test the hypothesis that if alcohol is present in the developing embryo environment, then there will be a decrease in the length of the wing buds.

Materials and Methods:

Three-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). Penicillin/streptomycin was also added to each embryo to achieve a final concentration on-the-cells of 100 U mg/ml. Cells were kept in a 37°C incubator.

Heart rate was measured immediately after removing the embryo from the incubator each day. A microscope and ocular ruler were used to measure the allantois, embryo body length, diameter of the area vasculosa, wing bud and leg bud length. A control group with 0% ethanol was used in order to determine if the resulting effects were a result of the alcohol exposure or an extraneous variable. For this hypothesis I analyzed wing bud length. Average length values for each concentration are shown in figure 1.

Results:

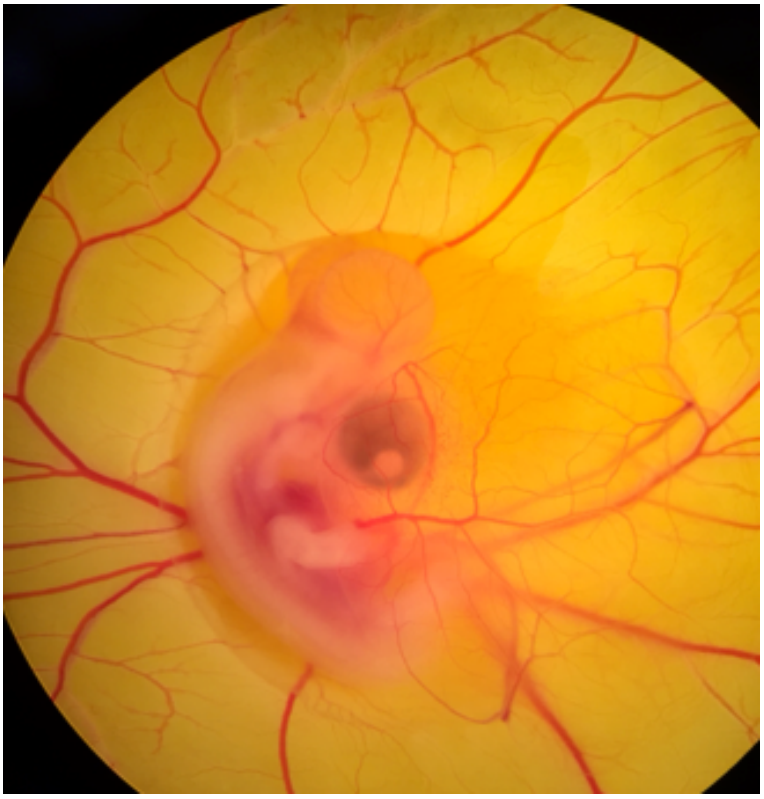

Figure 1. This image shows a typical Gallus gallus embryo (explanted into a sterile boat) on day 7 of development (Sweet, W., 2015).

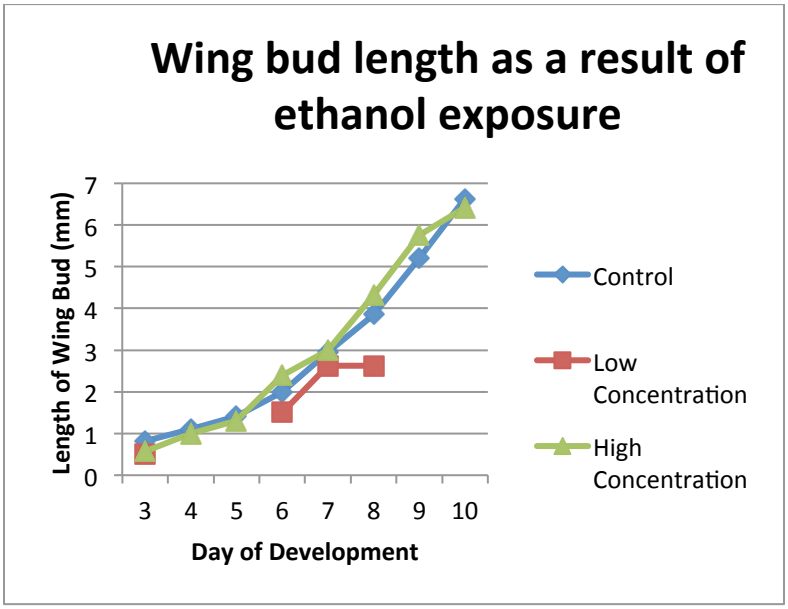

Figure 1. This graph shows the average measurement (in mm) of the surviving chicks in the three different testing groups, low concentration (0.002% ethanol), high concentration (0.02% ethanol) and control (0% ethanol). Measurements were taken every day for seven days. The numbers provided are based on only the surviving animals for each day.

These data showed an ideal growth curve for the Control embryos. The low concentration sample had a very high mortality rate (100% after day 7) which made it difficult to make conclusions based on the available results. The high concentration sample showed a growth pattern very similar to the control; however the main difference was the rate of development at the beginning and end of the observation period. There was slow initial growth (days 3-5) and slowed growth rate in the last 24 hours of observation. Survival rate for the control embryos through day 10 was 45%. The high concentration sample had a survival rate of 88%. The survival rates were based on only the embryos that survived from explanation to the first measurement.

Discussion:

Analysis of the data showed a variable growth rate among the high alcohol concentration sample. These chicks showed slowed limb development for the first three days and decreased growth rate at the end of development. Limb growth for the control group however was consistent and even throughout the observed period. The control embryos ended the observation period with longer wing buds on average. The final length of wing buds between testing groups was not significant. The data do show variability in growth patterns across the samples, meaning that the alcohol exposure disrupted the growth process but not the final wing bud length. The low concentration sample had a 100% mortality rate by day seven post-fertilization; the cause for this is unclear.

The data presented refute my hypothesis that alcohol present in the development environment of the chick embryo causes decreased wing bud growth. Though there was some observed variability in rate and a slightly smaller final length average, but it was not statistically significant to prove the teratogenic effect in this case.

A study done by Johnson et al. (2007) showed that alcohol exposure in developing mice caused postaxial ectrodactyly. This finding was aided by a 2005 study (Bell et al., 2005) which looked at the effect of sonic hedgehog function in mouse limb development. Future experimentation should have a larger sample size. This would help to gain a more accurate value for wing bud growth for different concentrations as well as produce more data points for the low concentration sample. Analysis should also focus additionally on digit development rather than only wing bud length.

References

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