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Question 1: The Effects of Environment and Genetics on Bluefin Killifish Sensitivity to Blue Light

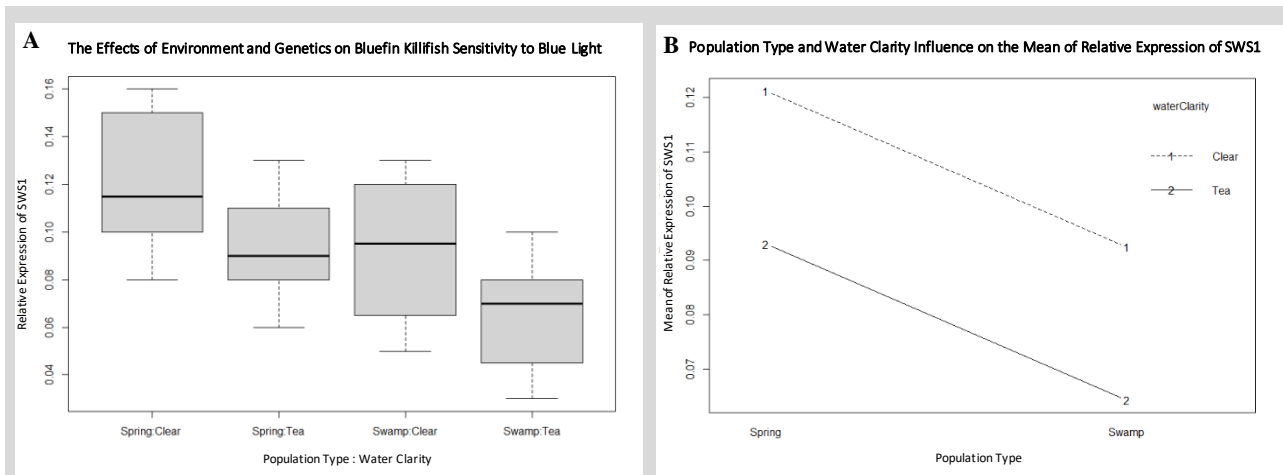


Figure 1: Showing analysis of the effects of Environment and Genetics on Bluefin Killifish Sensitivity to Blue light with (A): A boxplot displaying how the environment (either tea or clear) and the genetic background (swamp or spring) can effect SWSI gene expression. The boxplot indicates the median values with a thick horizontal line and quartile values with thin horizontal lines. (B) An interaction plot showing the relationship between environment (tea or clear) and genetic background (spring or swamp). The clear (dotted) and tea (solid) lines lying parallel to one another demonstrates the lack of interaction between the environment and genetic background.

The Assumptions of a 2 factor General Linear Model are as follows; Firstly subjects must be randomly selected. Secondly, data within treatments must be independent from one another and normally distributed. Finally, there must be equal variance between treatments.

The fish were randomly selected from each population and each fish was randomly assigned to each treatment. Each data point was independent of each other and visual assessment of a Q-Q plot confirmed normal distribution whilst residual vs. fitted plot indicated equal variance. This means that all the assumptions of the 2-Factor General Linear Model were met and so the test was deemed appropriate. The data however was unbalanced, so the Anova function was used to consider this, and contrasts for both water clarity and population were stipulated.

The data provides moderate evidence that both population type (2-Factor GLM, $F_{1,29} = 8.9875$, $p = 0.005528$) and water clarity (2-Factor GLM, $F_{1,29} = 8.6774$, $p = 0.006293$), have an effect on the relative expression of the SWS1 gene. However, the data does not provide evidence that the interaction between population type and water clarity ($F_{1,29} = 0.00$, $p = 0.998526$) has a significant impact on expression of the SWS1 gene. This means that though the genetic background and water clarity impact the relative expression of the SWS1 gene separately, the data does not provide evidence that there is any relationship between genetic background and water clarity interacting to affect the expression of SWS1.

Further analysis of the data using estimated marginal means found moderate evidence that there was a difference in relative SWS1 gene expression between spring and swamp populations in both the clear water environment ($p = 0.0440$) and in the tea water environment ($p = 0.0414$). Furthermore, SWS1 gene expression in the Spring population varies between individuals in the clear and tea environment ($p = 0.0374$), but does not seem to vary in individuals in the swamp population between environments ($p = 0.0553$). Individuals in the spring population living in clear water showed the largest relative SWS1 gene expression (mean \pm SE = 0.1212 ± 0.00965), with the tea environment of the same population showing reduced expression (mean \pm SE = 0.0930 ± 0.00863). Similarly in the swamp population, individuals in clear water showed a higher relative gene expression (mean \pm SE = 0.0925 ± 0.00965) than in the tea (mean \pm SE = 0.0643 ± 0.01032).

The 2-factor GLM and subsequent post hoc test provides evidence that suggests both water clarity and genetic background impact relative SWI expression, and therefore blue light sensitivity separately. The effects of genetic background and water clarity do not interact and are independent of one another.

Question 2: The Effect of Treatment on Hair Follicle Density

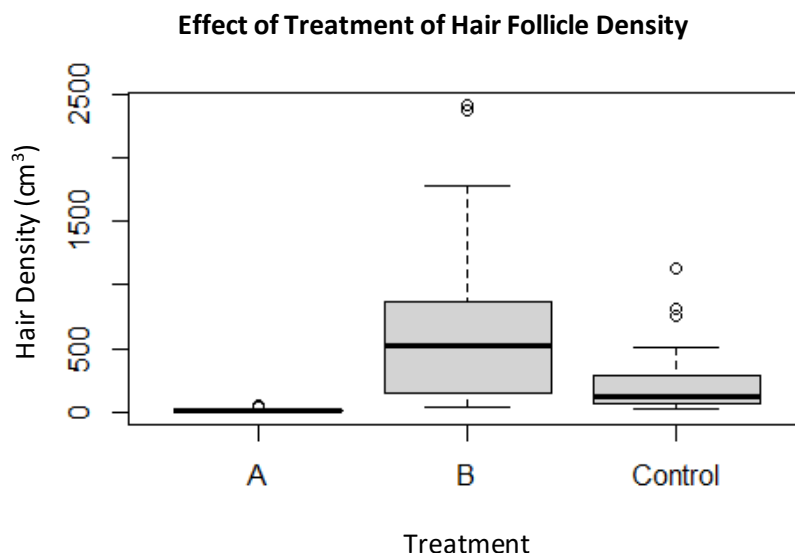


Figure 2: Boxplot showing how treatment (A, B and Control) effects hair follicle density. The boxplot indicates the median values with a thick horizontal line and quartile values with thin horizontal lines.

The assumptions of a 1-Factor General Linear Model are as follows; Firstly subjects must be randomly selected. Secondly, data within treatments must be independent from one another and normally distributed. Finally, there must be equal variance between treatments.

Subjects were randomly assigned to one of the three treatments and each data point was independent from one another. However, a visual assessment of the Q-Q plot showed that the residual vs. fitted plot indicated that the data was not normally distributed and did not have variance. In order to satisfy the assumptions of a 1- factor GLM the data was log transformed, after which the data was both normally distributed and had equal variance. Therefore the data met the assumptions of a 1-Factor GLM and the test was deemed appropriate.

Analysis of the data provided strong evidence that there was a significant difference between the log hair follicle densities for each of the treatments A, B and Control (1 Factor GLM, $F_{2, 117} = 167.94$, $p = 2.2e-16$). After back transformation, further analysis was carried out which found that treatment A produced a lower hair follicle density (mean \pm SE, 95% CIs: 5.85 ± 0.989 , 4.19-8.18) compared with the control and Treatment B produced a higher hair follicle density (mean \pm SE, 95% CIs: 405.15 ± 68.489 , 289.88-566.25) when compared with the control. This suggests that Treatment A has a reducing effect on hair follicle density, where as Treatment B has an increasing effect when compared with the untreated control.

Post hoc tests were carried out to determine the difference between Treatment A and Treatment B ($p = <0.001$), control and Treatment A ($p = <0.001$) and control and Team B ($p = <0.001$), which determined they were strongly significant. This is concurrent with the previous data analysis and provides strong evidence that both Treatment A and Treatment B have an impact on follicle density.

Question 3

The conclusions are not justified as the 1 factor GLM assumption of independence has been violated because pseudo replication has occurred. There were only 4 mice used per treatment, however there are 12 data points per box in the boxplot, suggesting the scientists repeated the experiment multiple times per mouse.

Question 4

The figure indicates that the 1g treatment had the smallest effect compared to the control, with the 95% confidence interval dipping below zero line on the y-axis. This means it has values in its range that overlap with the control demonstrating how small the effect of it is. The 5g and 10g treatments have similar means, however the range produced by the 10g treatments is much larger than the 5g. This means that the 5g treatment actually produced values that were consistently higher than the 10g, however the 10g treatment produced values with the largest difference compared to the control in its data set. Overall it can be said that the 10g treatment has the potential to cause the biggest effect to respiration, however it has a large range and its effect is less precise than the 5g treatment which would give a consistent increase in respiration rate compared to the control.