Be Wary of the Edge: Well Position Influences Larval Zebrafish Behavior

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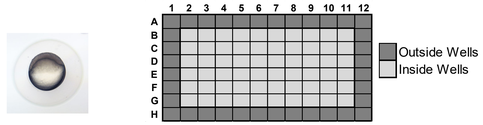
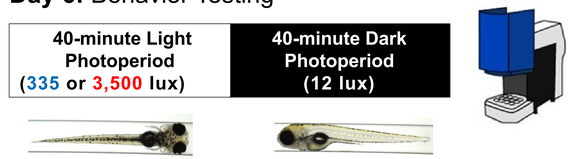
Larval zebrafish behavior is utilized as a higher throughput testing strategy to screen chemicals for developmental neurotoxicity potential. One challenge is that many different protocols are utilized.

We are investigating the confounding influence of method variables on locomotor activity and how these can contribute to the variability of results.

Days 1-6: Embryos reared in incubator in the same microtiter plate

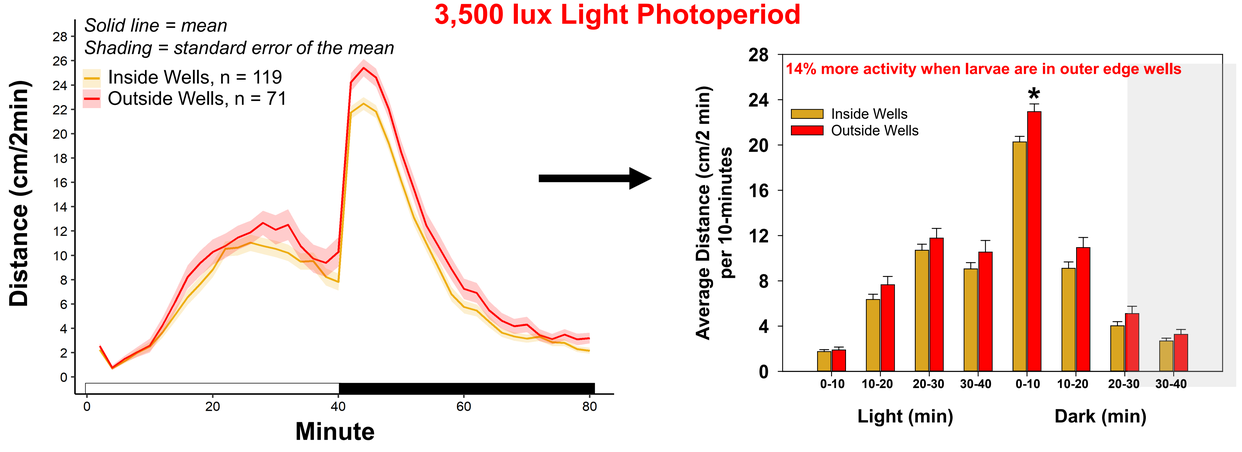


Day 6: Behavior Testing

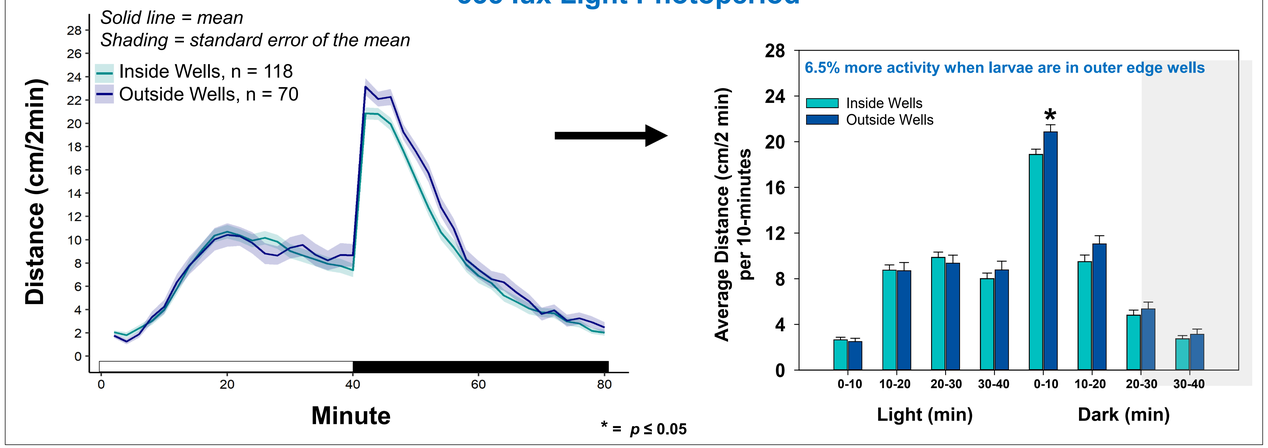


Well Location on Plate Influences Larval Zebrafish Behavior and Differs with Protocol Light Intensity

3,500 lux Light Photoperiod



335 lux Light Photoperiod



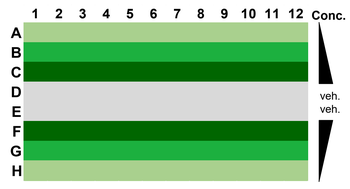
* = $p \leq 0.05$

Lowest Observable Effect Concentration Differs Depending On Plate Location. **Randomizing the Treatment Groups in Plate Designs Will Enable Even Distribution to Minimize the Edge Effects.**

Below, we modeled whether the location of controls and lowest treatment concentration influenced our Lowest Observable Effect Concentration by using the same data set and the percent activity of outer edge wells: (A) includes treatment in only the outer wells and controls only in the inside wells, versus (B) a randomized plate design with an even distribution of controls and treated larvae.

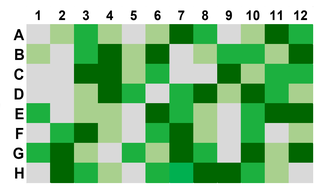
A

Example of plate design with four different chemical concentrations (colors) plated by hand per row.

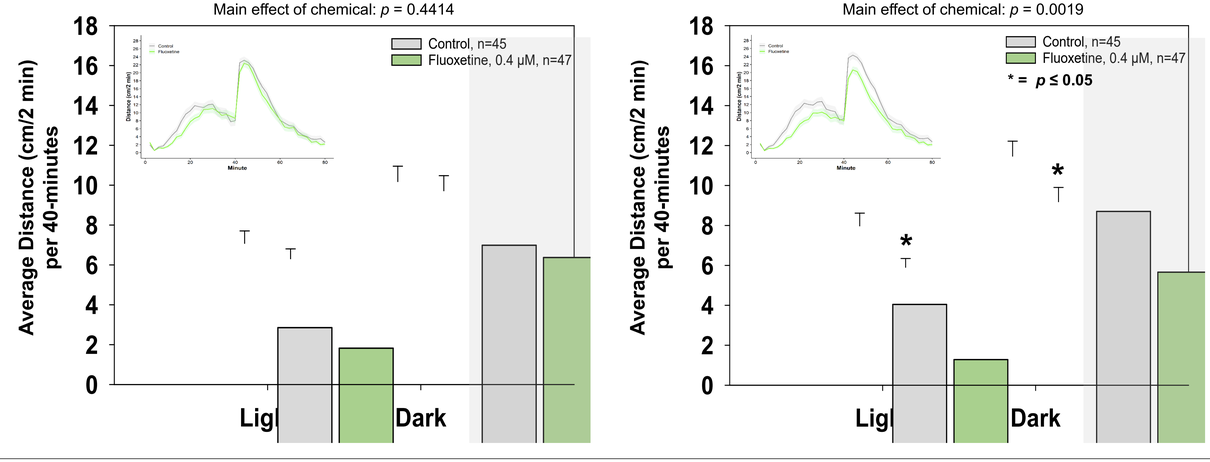


B

Example of a randomly distributed plate design, containing four different chemical concentrations (colors), using a randomization Excel function and dispensed via ECHO 550 liquid handling system.



3,500 lux Light Photoperiod



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