Analysis of Variation in Survival of Drosophila Exposed to Tetrodotoxin

Madelaine Rangel
Questions

- Do organisms not normally exposed to TTX display variation in response to the poison?
- If so, what is the genetic basis for the variation?
- If we can map the genes responsible, by what mechanism do they confer variable tolerance of TTX?
Research design

- 134 DGRP lines exposed to $1 \times 10^{-4}$ M TTX + 5% sucrose on Whatman paper
- 2 blocks of ~20 males and ~20 females separately exposed
- Number dead counted every hour till all died
- Use the R package survival to quantify survival statistics
- Block, Line, Sex,
- 0=female, 1=male

```
block line sex time.of.death.hours
1   26   1          1
1   26   1          1
1   26   1          1
2   38   1          1
2   38   0          1
1   40   0          1
```
TTX survival is similar between sexes
TTX survival is highly variable among lines
Genome-Wise Association Study (GWAS)

- searches the genome for SNPs
- to pinpoint genes that may contribute to a person’s risk
- Median Survival Time for males and females and pooled
- Input to DGRP2 web tool to obtain P-values, qq-plots, etc.
QQ Plot: MalePval highly significant

- compute the theoretically expected value for each data point

- deviation from the expected P-value distribution is evident only in the tail area
GWAS

- Top Hits for MalePval:
  - *Spalt-related (Salr)*: a zinc finger transcriptional repressor that, together with *salm*, mediates the *Decapentaplegic (Dpp)* signaling pathway in the wing imaginal disc. This pathway is also involved in the development of the gut and of fibrillar flight muscle.
  - *Timeless (Tim)*: a key component of the *Tim-per* complex, required for the production of circadian rhythms. It is involved in mating behavior, DNA replication and larval phototaxis. Na channels are critical for intercellular communication in the suprachiasmatic nucleus and for normal circadian rhythms.

Current Progress

- RNAi constructs to knockdown Spalt, Tim, Glu-RIB, Pdi, etc.
- Assess whether they cause hyper-sensitivity or greater resistance to TTX.
GAL4/UAS

- Gal4 is a transcriptional activator that binds to UAS enhancer
- Progeny express a specific gene shRNA in a specific tissue
- shRNA then decreases gene expression of targeted genes (Salr & Tim)
Acknowledgments

- Xiaoling Wong
- Grace Li
- Andy Ding
- Dr. Clark

The Clark Lab