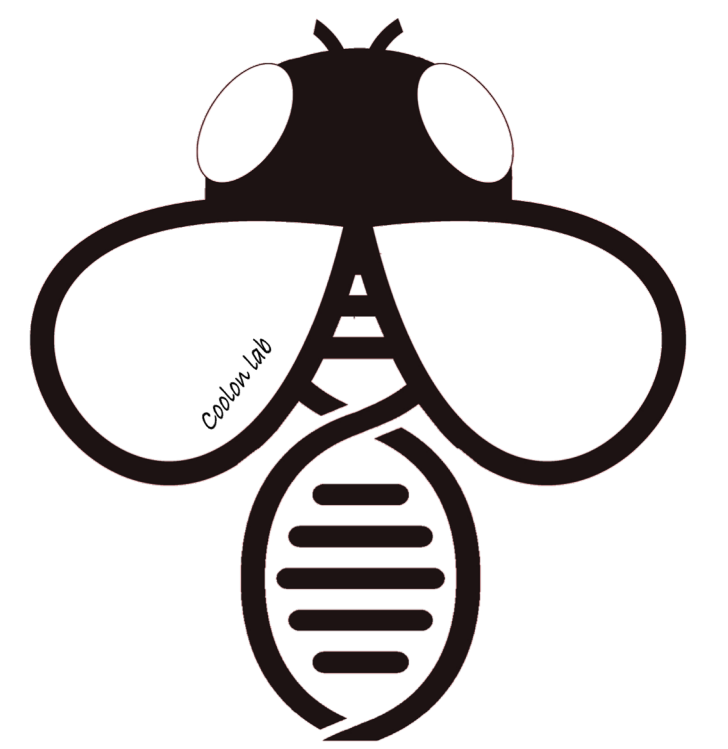# Genome-wide Effects of the GeneSwitch GAL4 System on *Drosophila melanogaster* Gene Expression

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Gene Expression Manipulation in *Drosophila*

Tools capable of manipulating gene expression in model organisms are of paramount importance in modern genetics for determining gene function. One such tool is the UAS-GAL4 system, where a fly line containing the yeast activator GAL4 is crossed with one containing a target gene just downstream of an Upstream Activation Sequence (UAS), resulting in the expression of the target gene[1]. Further refinement of this technique produced the GeneSwitch GAL4 system, described in the neighboring panel[2]. These techniques have numerous applications, including RNA interference (RNAi)[3] and overexpression experiments[4]. While the GeneSwitch GAL4 system is widely used, its consequences on genome-wide gene expression remain unknown. Here, we compare RNA-Sequencing data from control (w1118) and GeneSwitch line *Drosophila melanogaster* exposed to control or RU486-treated food to identify differential gene expression resulting from the GeneSwitch GAL4 system.
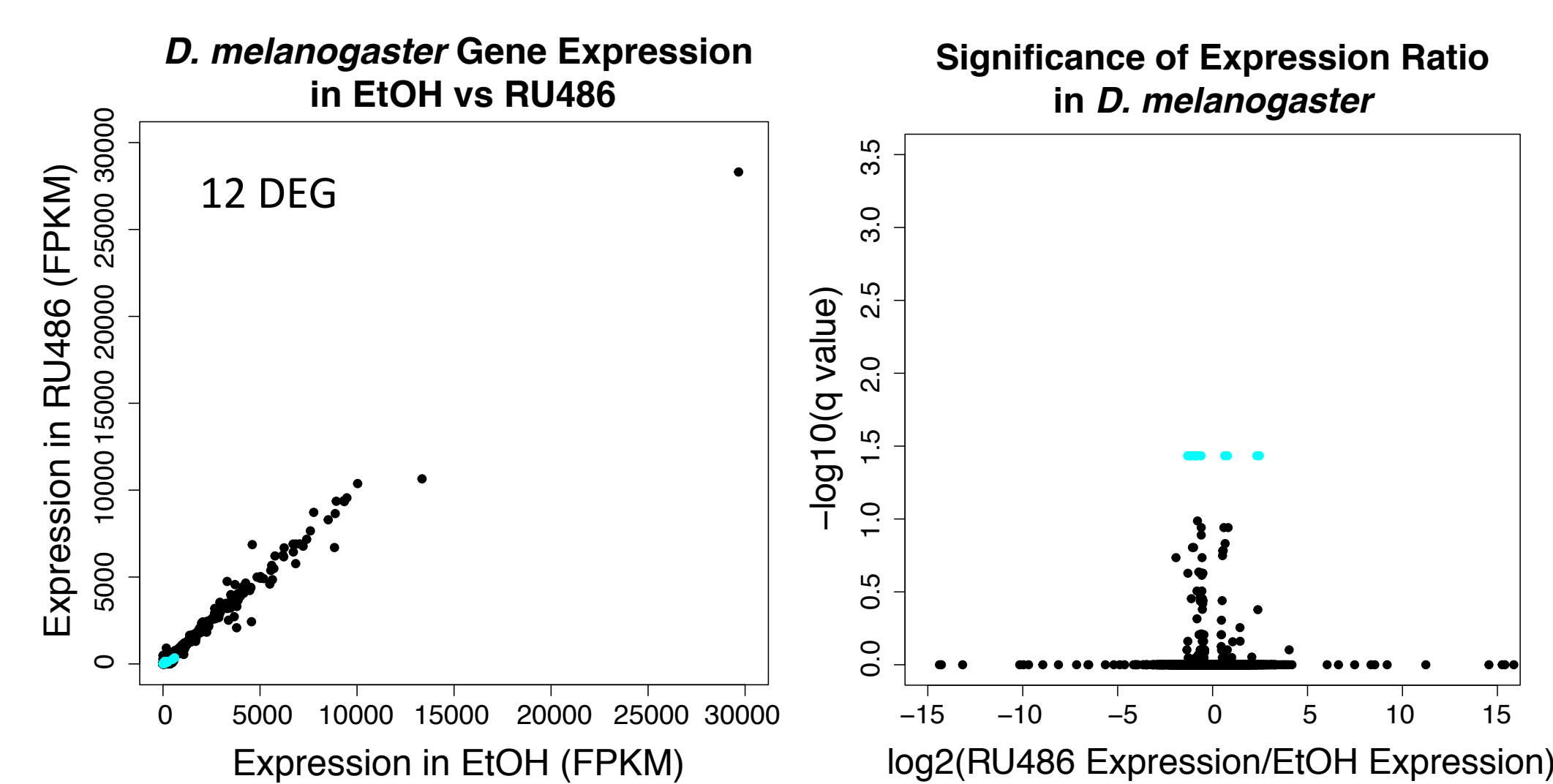
GeneSwitch GAL4 System
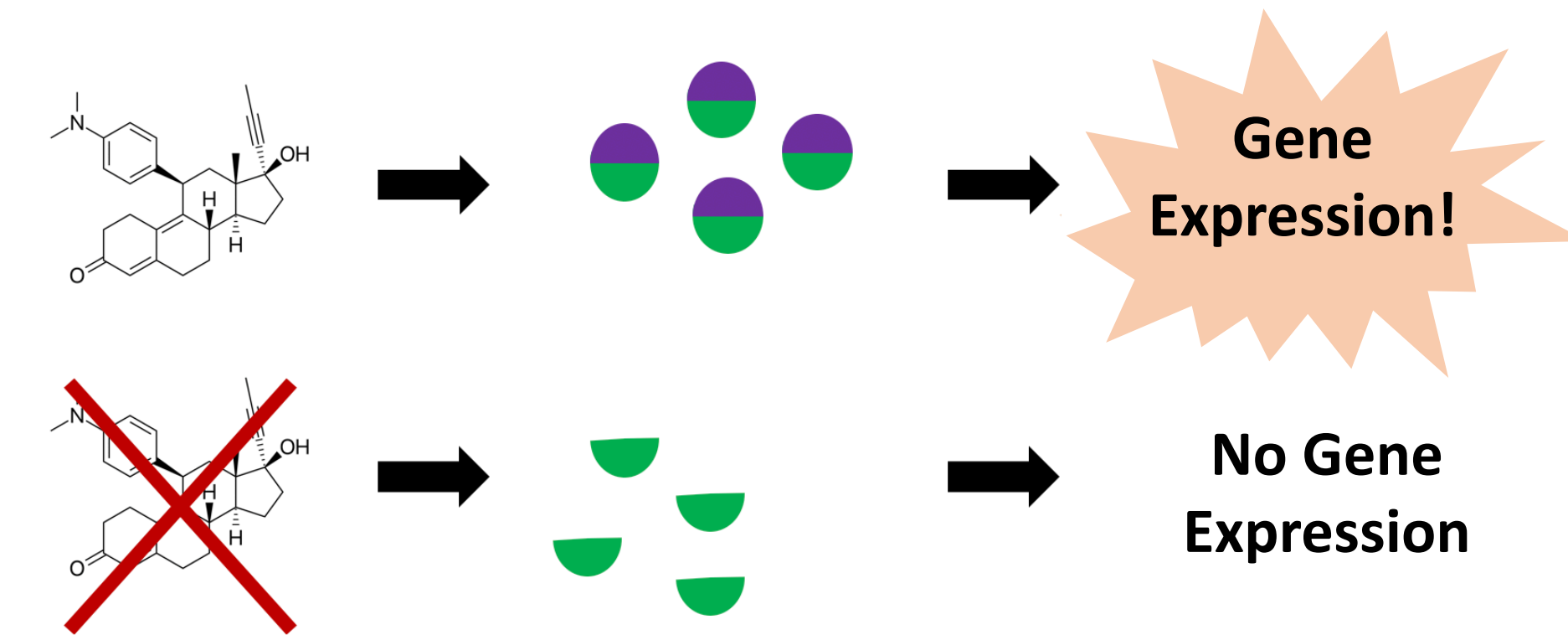
- GeneSwitch GAL4 is a modified GAL4 protein fused to progesterone receptor activated by RU486
- Dosage dependent: more RU486 → more expression of target gene
- Allows for more spatial and temporal control of target gene than UAS-GAL4 system
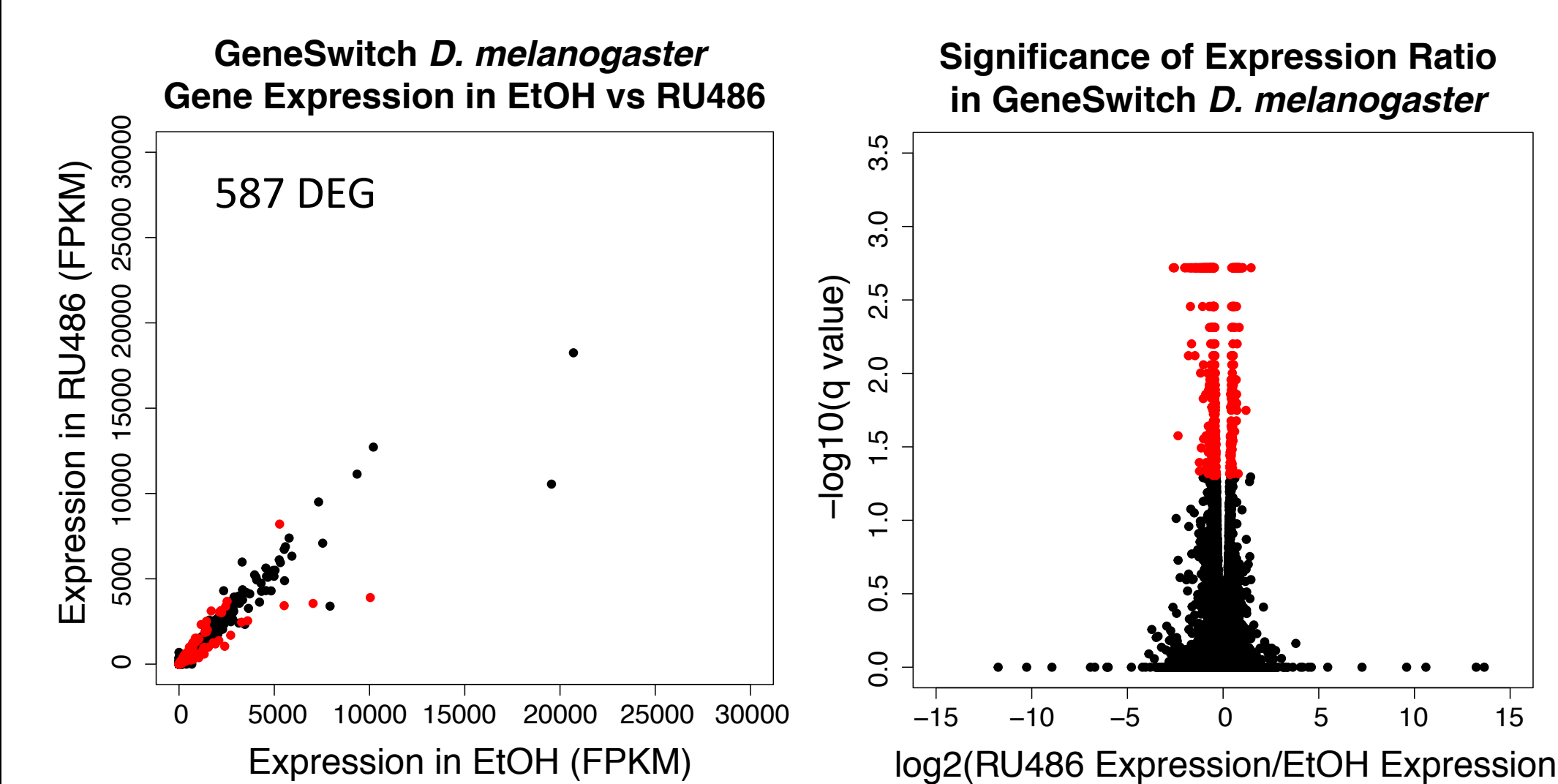
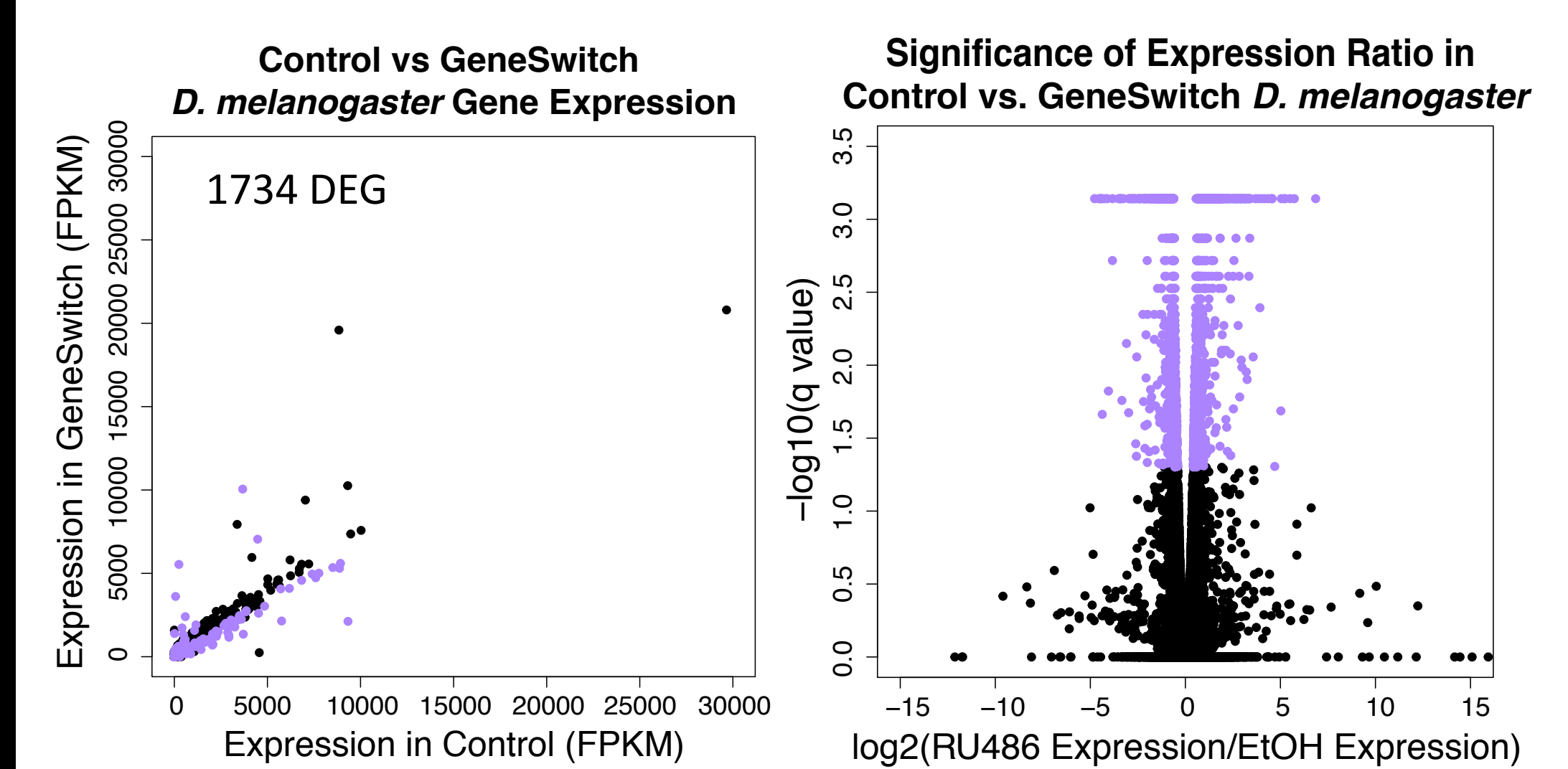## Experimental Design
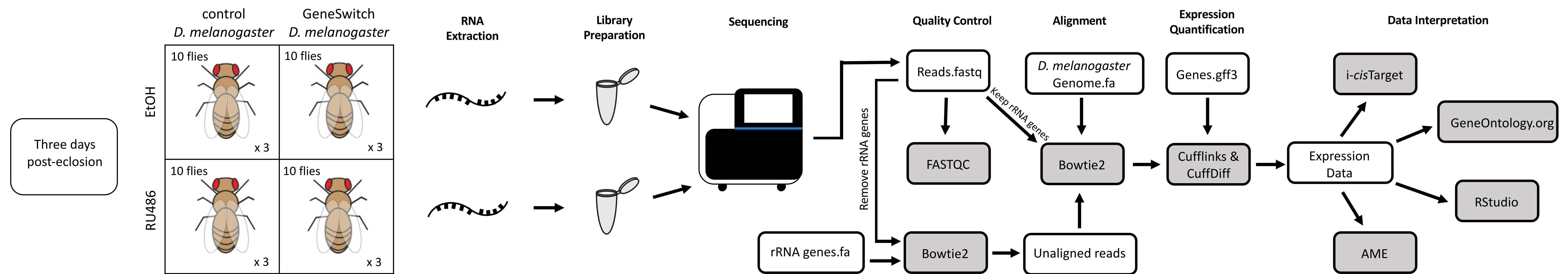
Control with vs. without RU486

Blue points are significant (q < 0.05)
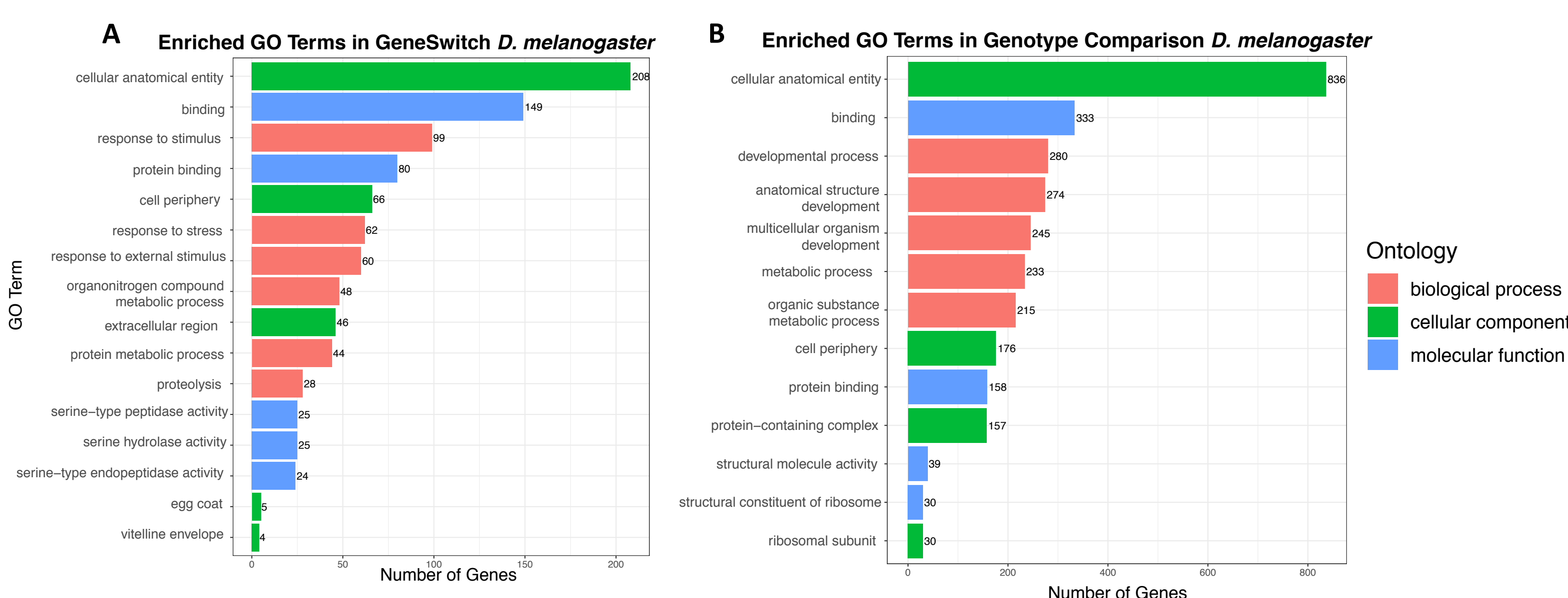
GeneSwitch with vs. without RU486

Red points are significant (q < 0.05)

Control vs. GeneSwitch

Purple points are significant (q < 0.05)

Gene Ontology Term Enrichment

Conclusions

- There are significant differences in gene expression between control (w1118) and GeneSwitch flies
- RU486 induces changes in expression in genes other than the gene of interest
- There may be cryptic GAL4 binding sites in the *Drosophila* genome
- Gene Ontology annotation reveals that the GeneSwitch GAL4 system affects expression of genes pertaining to binding and metabolism but does not affect most other functional groups
- When using the GeneSwitch GAL4 system, control experiments should be carefully designed

References

1. Brand, A. H., and N. Perrimon. "Targeted Gene Expression as a Means of Altering Cell Fates and Generating Dominant Phenotypes." *Development (Cambridge, England)*, vol. 118, no. 2, June 1993, pp. 401–15.
2. Osterwalder, T., et al. "A Conditional Tissue-Specific Transgene Expression System Using Inducible GAL4." *Proceedings of the National Academy of Sciences of the United States of America*, vol. 98, no. 22, Oct. 2001, pp. 12596–601. *PubMed*, doi:10.1073/pnas.221303298.
3. Nicholson, Louise, et al. "Spatial and Temporal Control of Gene Expression in Drosophila Using the Inducible GeneSwitch GAL4 System. I. Screen for Larval Nervous System Drivers." *Genetics*, vol. 178, no. 1, Jan. 2008, pp. 215–34. *PubMed Central*, doi:10.1534/genetics.107.081968.
4. Kim, Gye-Hyeong, et al. "Overexpression of Malic Enzyme in the Larval Stage Extends Drosophila Lifespan." *Biochemical and Biophysical Research Communications*, vol. 456, no. 2, Jan. 2015, pp. 676–82. *ScienceDirect*, doi:10.1016/j.bbrc.2014.12.020.

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