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Terra MH Hanks

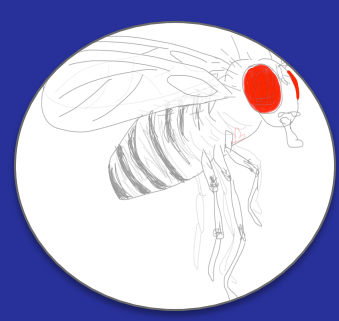
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Identifying octopamine receptor expressing neurons in the adult *Drosophila* male



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1-What is Octopamine?

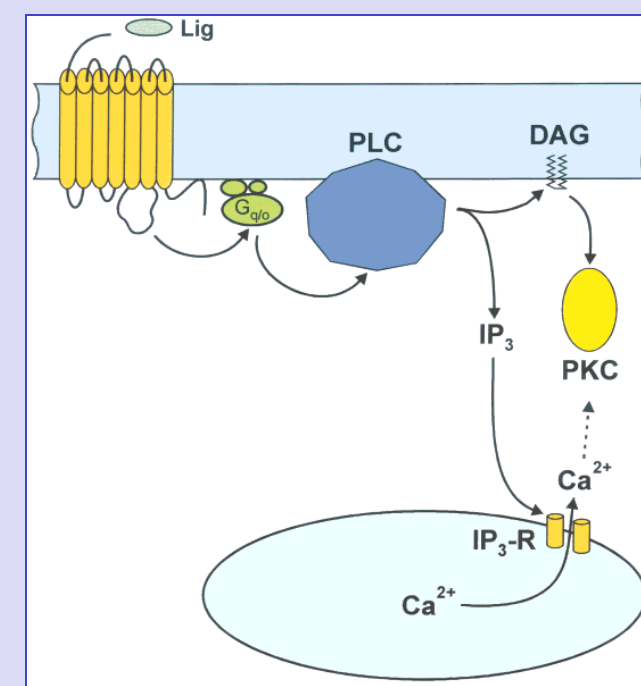
Octopamine (OA) is a biogenic amine equivalent to the human neuromodulator norepinephrine.

In insects OA acts functionally as a neuromodulator. It is important in many physiological processes.

Specifically in *Drosophila*, they are important for the generation of certain Behaviors including:

- Aggression
- Arousal
- Learning
- Memory

Drosophila have approximately 100 OA-expressing neurons. OA signals subsequent neurons by binding to specific G-protein coupled receptors.



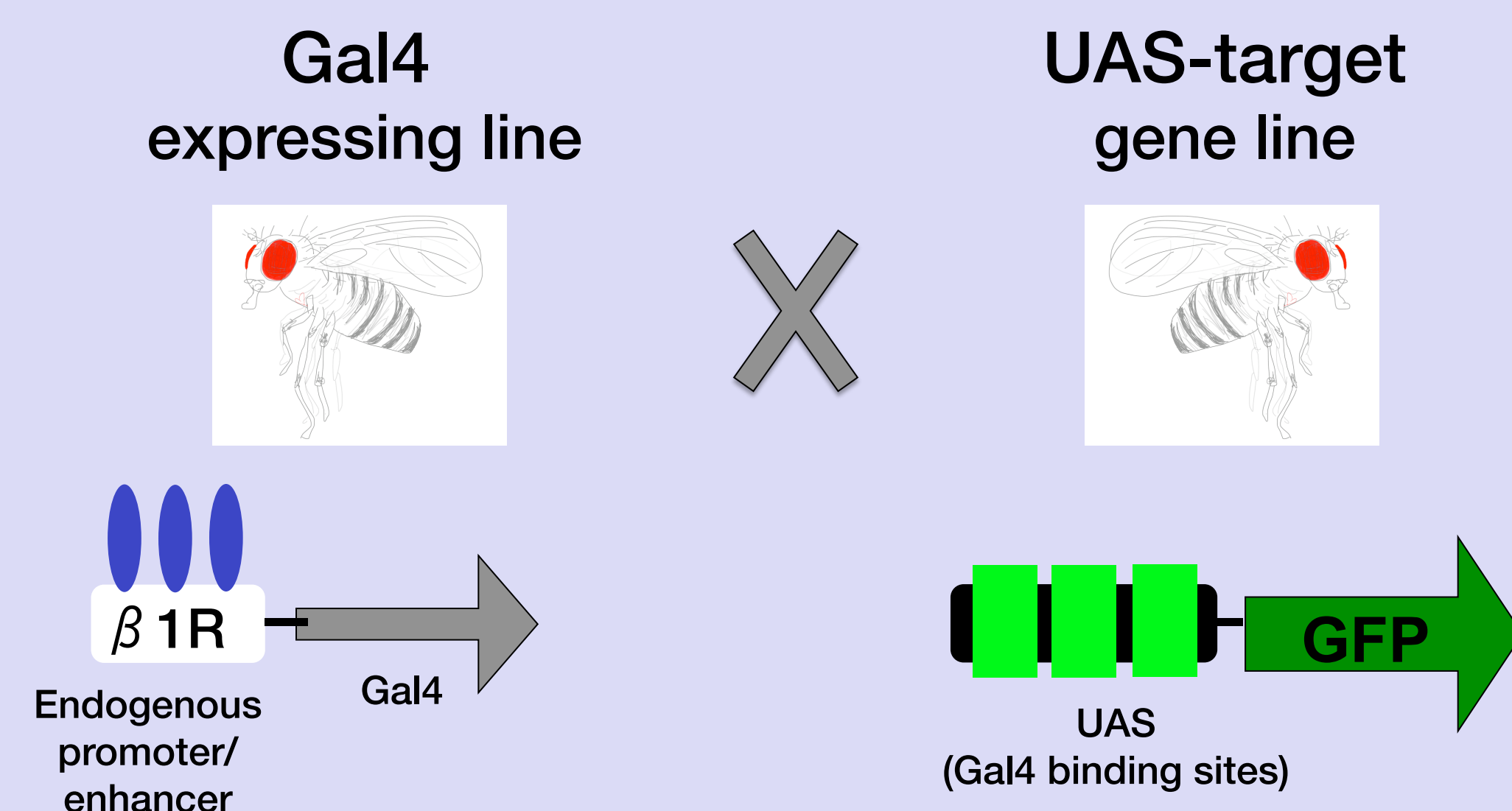
My focus is primarily on Octopamine $\beta 1$ Adrenergic-like Receptors (OA $\beta 1$ R).

2-Research Goals:

It is my aim to quantify the number of OA $\beta 1$ R-expressing neurons and to map the morphology of said neurons within the male *Drosophila* nervous system.

3-Genetic tools to illustrate OA $\beta 1$ R-expressing neurons

Gal4/UAS System (Brand and Perrimon, 1983)



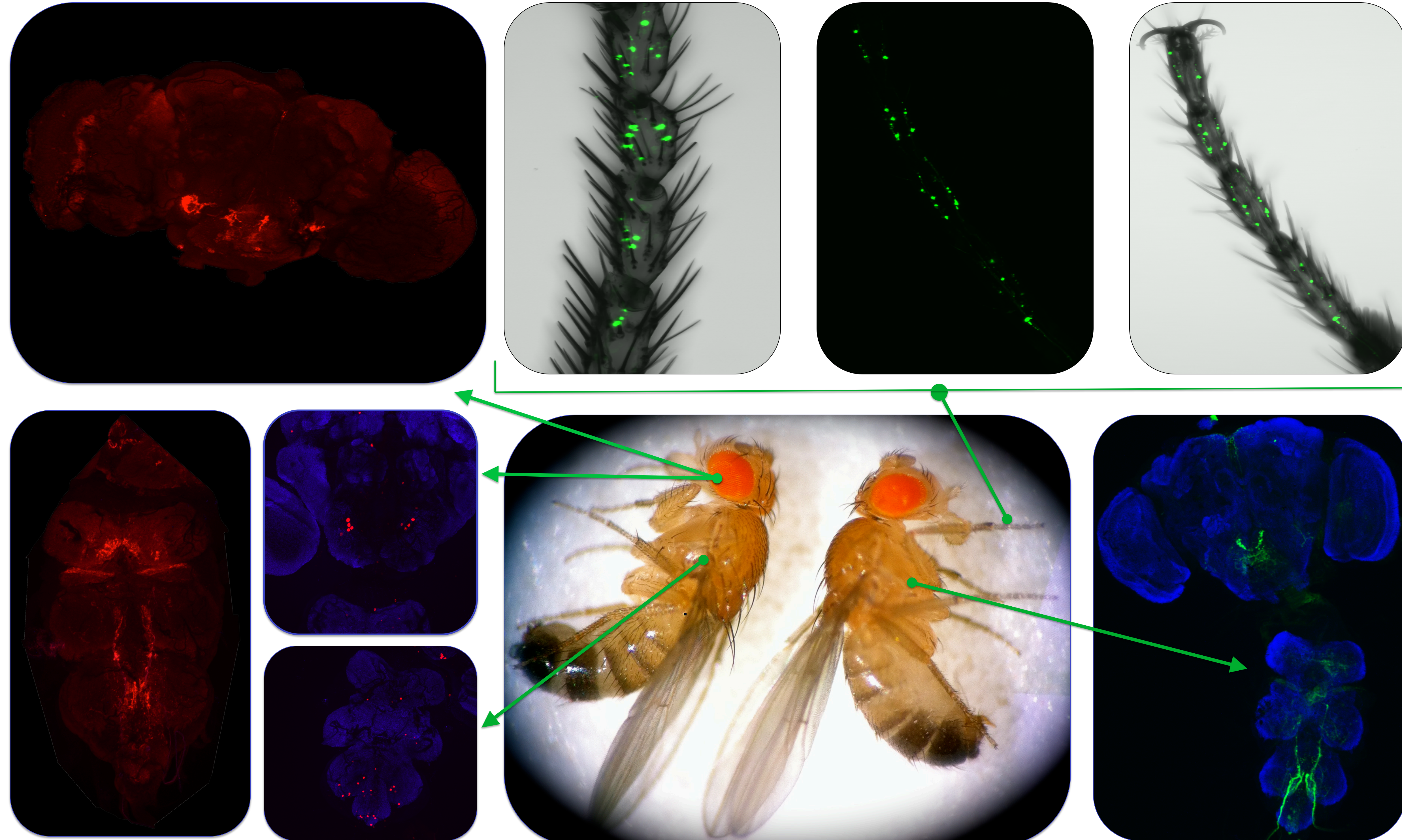
4-Dissection Methods

- Crossed genetically suitable strains
- Dissected progeny in PBS over ice
- Fixed brains and central nerve cord in 4% PFA
- Washed in a series of PBS, PBT, & blocking solution
- Incubated brains and central nerve cord in primary antibodies overnight
- Washed again
- Incubated in secondary antibodies for 2 hours
- Washed again
- Mounted on 25 x 77 mm slide
- Examined under confocal microscope
- Prepared images



Drosophila brain dissection

5-Expression patterns of OA $\beta 1$ R-expressing neurons in the legs, ventral nerve cord, & brain



6-My research identified the following:

Legs

The green fluorescent protein (GFP) in the forelimbs indicates the OA $\beta 1$ R-expressing neurons. The pattern is congruent in multiple legs allowing me to count the number of such neurons.

Ventral Nerve Cord

Fiber tracts and cell bodies are visible throughout the ventral nerve cord. By isolating the expression of a red fluorescent protein (RFP) to the nucleus of the OA $\beta 1$ R-expressing neurons, the specific cell count can be observed. The majority of these neurons appear to be concentrated within the last segment.

Brain & Subesophageal Ganglion (SOG)

Using the same genetic tools as in the ventral nerve cord and legs, cell count and fiber tracts can be established throughout the brain. Synaptic terminals can be identified by labeling the protein synaptotagmin. The SOG, a region that receives pheromone input, appears to have bilateral symmetry.

7-Conclusions

*I observed between eighteen and twenty-one OA $\beta 1$ R-expressing neurons in the forelimbs.

*I located synaptic terminals and thirty-four OA $\beta 1$ R-Gal4 GFP-expressing neurons in the ventral nerve cord.

*I identified morphology and location of nine GFP-expressing neurons through the OA $\beta 1$ R-Gal4 line in the SOG.

This research has provided a foundation for further analysis of octopamine neurons in *Drosophila melanogaster*.

Acknowledgements:

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