Unraveling Internal Brain States by Interneuron Manipulation in *C. elegans*

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Introduction

C. elegans, a tiny roundworm known for its simple brain structure and behaviors, presents a unique opportunity to understand neural computation principles.

The internal brain state is believed to determine the worm's locomotive response to chemical stimuli. [1] As a proof, the brain state is manipulated by histamine silencing pairs of the second-layer interneurons in chemosensory network and the change in the rate of exhibiting a specific behavior is examined. Also, a control experiment is performed to confirm that the of recovery of the network is immediate.

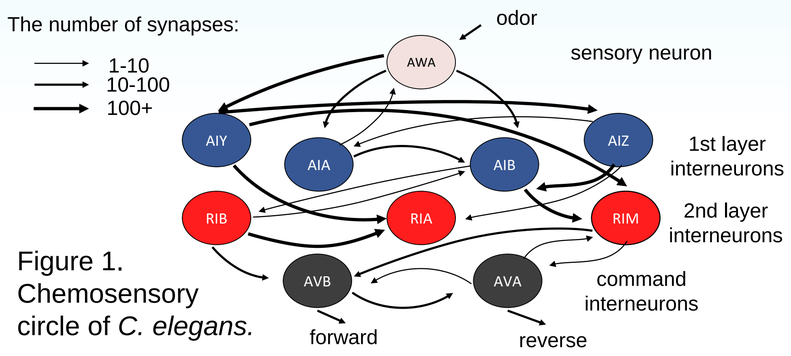


Figure 1. Chemosensory circle of *C. elegans.*

Methods

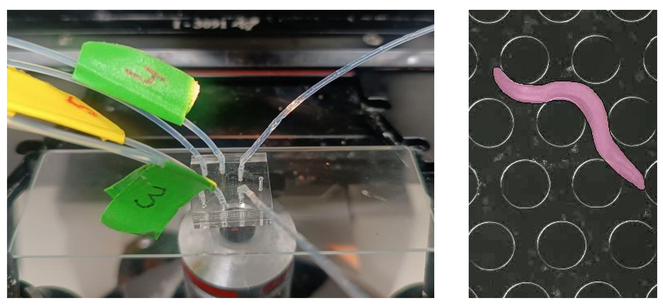


Fig. 2. Microfluidic chip used for experiments (left) and segmented image of the worm inside the chip (right).

Histamine Silencing: Transgenic worms engineered to express inhibitory histamine-gated chloride (HisCl) channels [2] in specific interneurons enable the controlled and reversible silencing of these neurons using histamine.

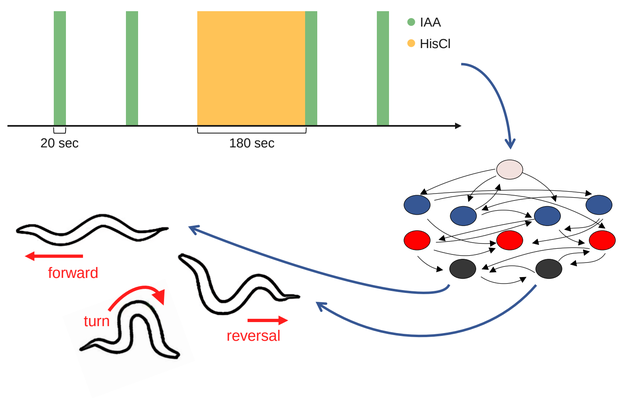
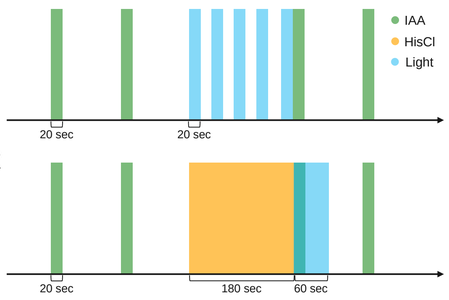


Fig. 3. Experimental procedure. Moving in a microfluidic arena, the worm receives histamine as well as pulses of IAA which acts as an attractive odor.The change in its locomotor behavior is examined.

Optogenetic Activation: Blue light activates interneurons expressing channelrhodopsine (ChR), a light-gated ion channel, facilitating precise control of neural activity. [3]

Fig. 4. Control experiment procedures with RIB::ChR mutant (up) and RIB::HisCl::ChR mutant (down).



Results

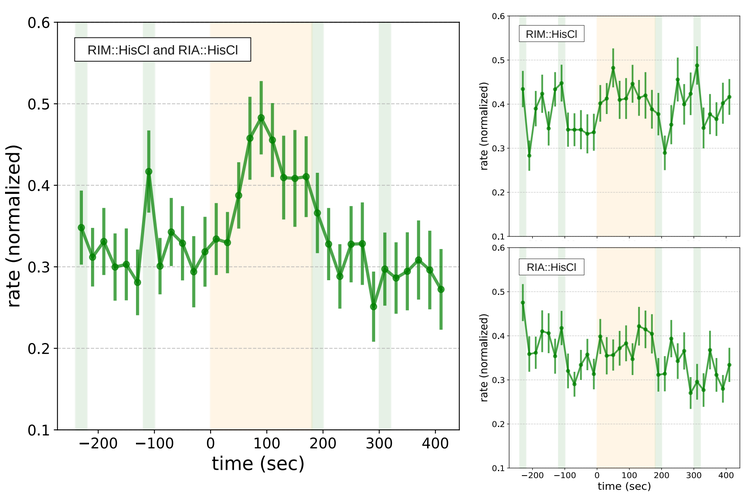


Fig. 5. Forward locomotion rate (normalized) of mutants expressing both RIM::HisCl and RIA::HisCl (left), and each of them (right). The activity is dominated by RIM in the crossed mutant.

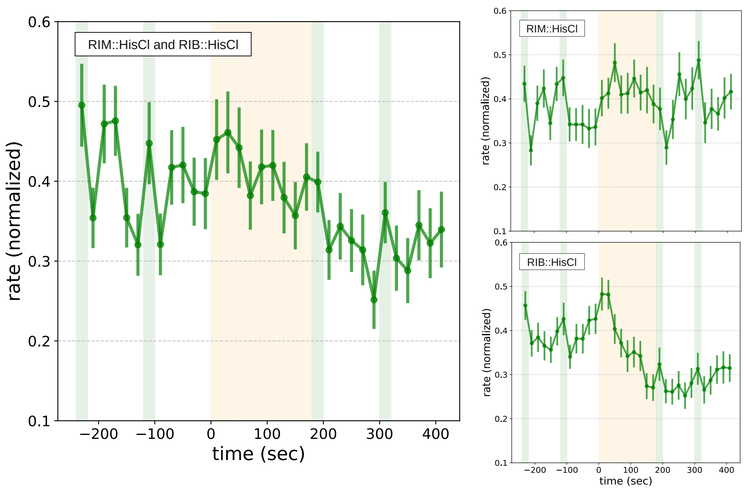


Fig. 6: Forward locomotion rate (normalized) of mutants expressing both RIM::HisCl and RIB::HisCl (left), and each of them (right). The behavior of the crossed mutant is in between.

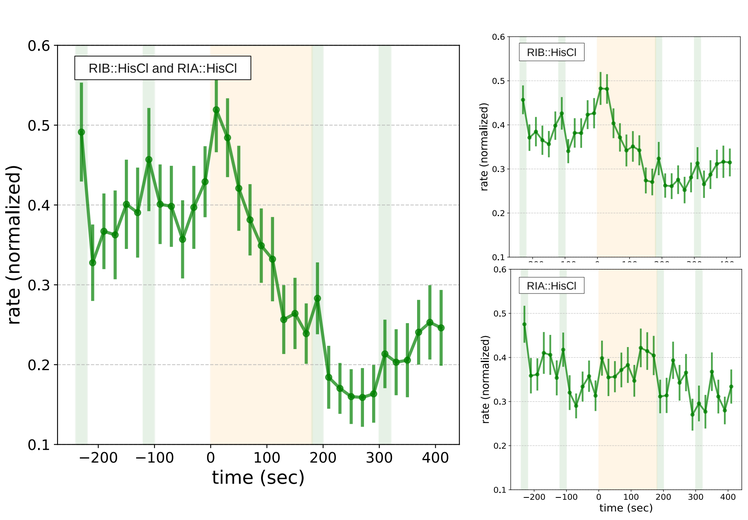


Fig. 7: Forward locomotion rate (normalized) of mutants expressing both RIB::HisCl and RIA::HisCl (left), and each of them (right). The activity is decreased in the crossed mutant due to dominancy of RIB.

Control Experiment

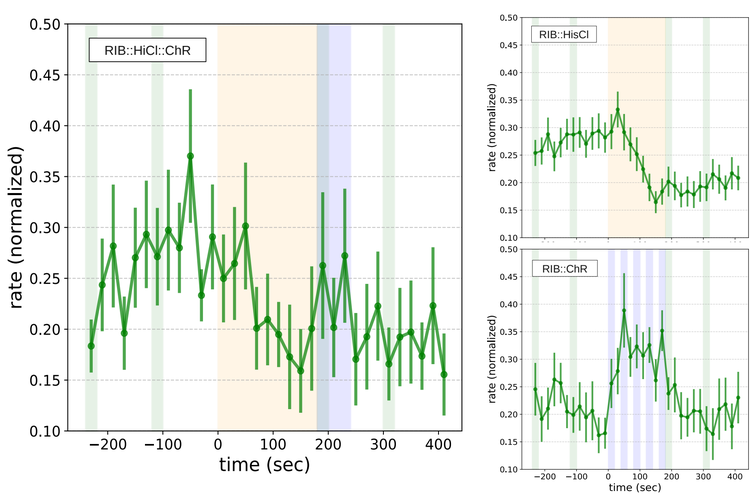


Fig. 8. Forward locomotion speed (normalized) of mutants expressing RIB::HisCl::ChR (left), and each of them (right) The normal activity is retrieved during the light pulse.

Conclusion

- Silencing both RIM and RIA mimics the effect of silencing RIM alone, indicating RIM's dominance.
- When both RIB and RIA are silenced, behavior is intermediate between the effects of silencing each individually.
- Silencing both RIM and RIB closely matches the effect of silencing RIB alone, as predicted by the network.
- Silencing RIB alone does not restore normal activity, suggesting the delay is due to an altered brain state.
- A control experiment rescuing RIB function confirms behavioral changes were due to neural inhibition, as normal behavior was restored with light exposure.
- This project, as a part of a larger plan, helps to link the structural connectome to behavior.

References

[1] Flavell, S. W., et al. (2022). The emergence and influence of internal states. Neuron, 110(16), 2545–2570.

[2] Pokala, N., et al. (2014). Inducible and titratable silencing of Caenorhabditis elegans neurons in vivo with histamine-gated chloride channels. Proc Natl Acad Sci , 111(7), 2770–2775.

[3] Larsch, J., et al. (2015). A Circuit for Gradient Climbing in C. elegans Chemotaxis. Cell Reports, 12(11), 1748–1760.