Decoding the molecular mechanisms underlying sex-specific cell fate switches in *C. elegans*

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Introduction

Traditionally, development has been viewed as a unidirectional process where embryonic stem cells progressively lose plasticity as they commit to increasingly restrictive cell fates. However, switches in differentiated cell identity have been described in many different tissues and organisms ([1],[2],[3]) as part of their development in a process known as transdifferentiation ([4],[5]). Despite its prevalence, **the fundamental principles underlying *natural* transdifferentiation remain poorly understood.**

Dr. Poole's Lab has identified two sexually dimorphic glia-to-neuron transdifferentiation events in *C. elegans* ([6],[7]). The amphid socket glial cells (AMso) in the head asymmetrically divide to produce the mystery cells of the male (MCM) interneurons, and the pair of phasmid socket glial cells (PHso1) in the tail become Phasmid D (PHD) neurons through direct transdifferentiation (Figure 1).

Proneural genes and LIN-48 Transcription Factor

Proneural genes act as developmental switches that control neuronal fate, instructing stem cells to commit to neuronal differentiation[8] (Figure 2). Preliminary data from the Poole Lab shows that *hlh-14* is functionally required for the described glia-to-neuron fate switches.

The LIN-48 transcription factor is expressed in the AMso and PHso1 glial cells and the MCM and PHD neurons since they are born, but its expression is downregulated in adult animals. This expression pattern suggests *lin-48* could play an early role in the commitment of AMso and PHso1 into a neuronal fate. Furthermore, it raises the possibility that it could be activating the expression of *hlh-14* in these cells.

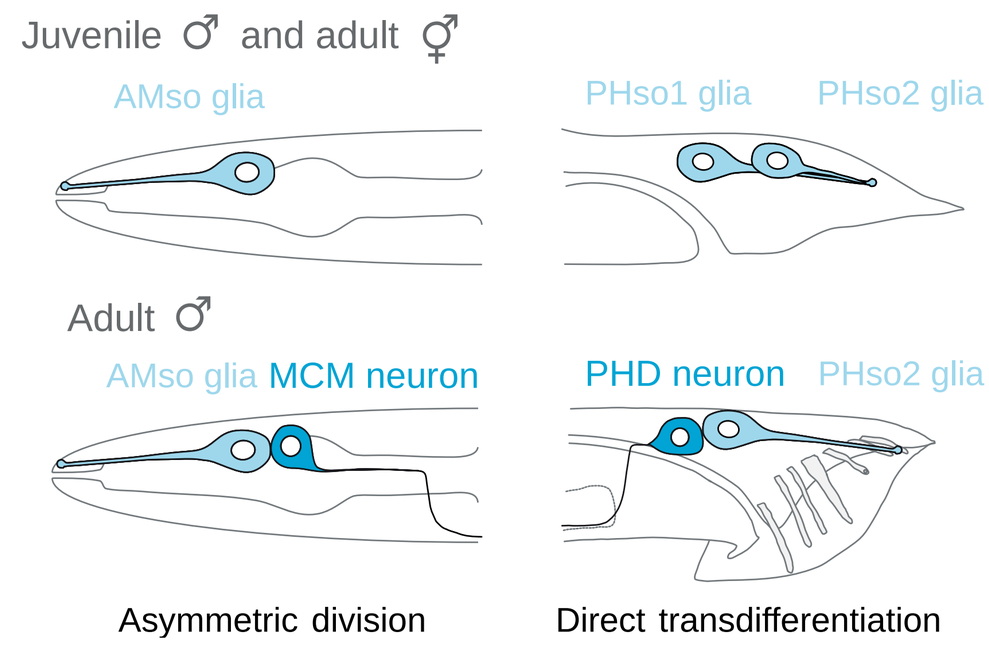

Figure 1. Glia-to-neuron transdifferentiation in *C. elegans* males.

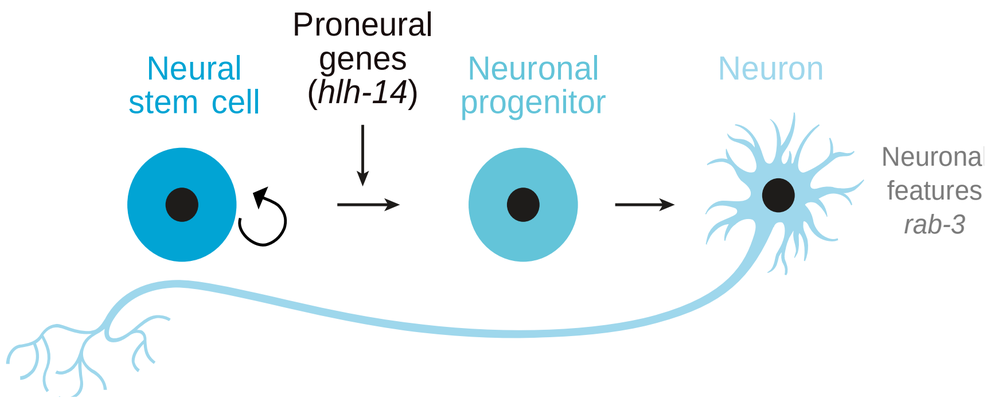

Figure 2. Schematic of proneural gene action during development.

Question:
What are the molecular mechanisms that regulate the AMso/MCM and PHso1/PHD cell fate switches?

Aim: Explore the role of *lin-48* as early regulator of the cell fate switch of AMso/MCM and PHso1/PHD, possibly acting through the *hlh-14* proneural gene, using *C. elegans* as a tool.

Generation of mutant strains

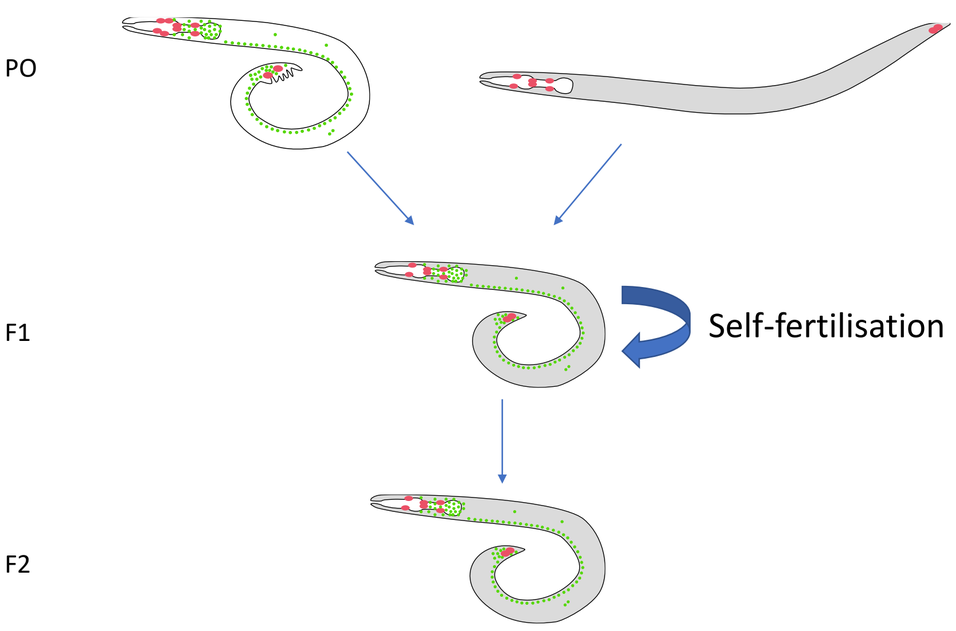

Figure 3. Cross design for *lin-48(sa469)* mutants with a fluorescent reporter for *rab-3* panneuronal reporter. The same cross was set to build the strain with the *hlh-14* proneural gene reporter. **Legend:**

- **White body –** Wild type worm.
- **Grey body –** Mutant worm
- **Red dots –** *drpIs3 (lin-48::tdtomato)* glial subtype reporter.
- **Green dots –** *otIs291 (rab-3::YFP)* panneuronal reporter.

Lin-48 controls neurogenesis in the MCM neuron but not in the PHD neuron

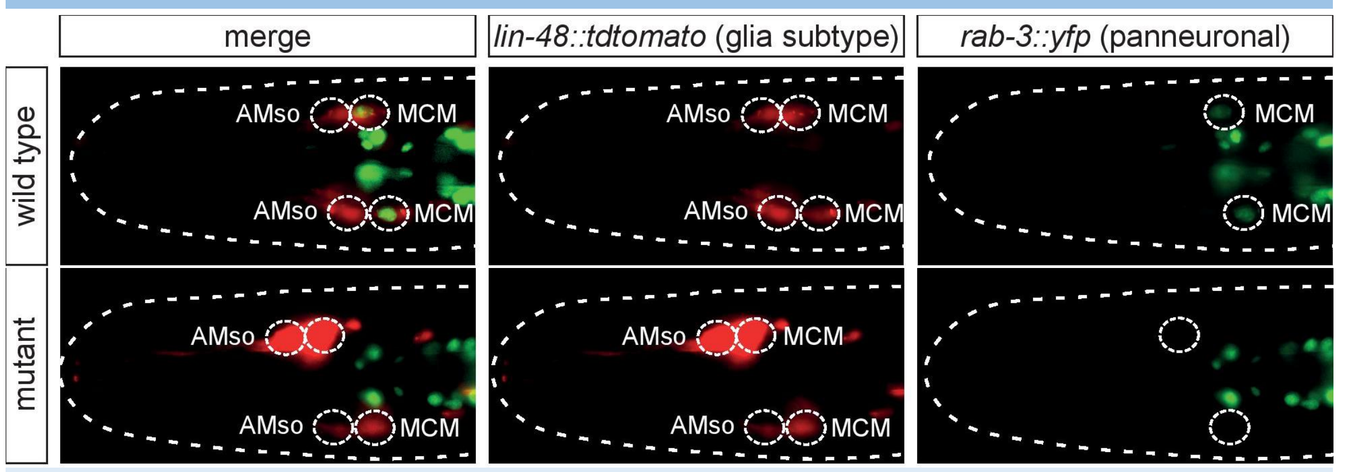

Figure 4. *Lin-48 (sa469)* affects *rab-3::YFP* expression in the MCM neuron, but not in the PHD neuron. **Legend:** (Red: *drpIs3 (lin-48::tdtomato),* Green: *otIs291 (rab-3::YFP*)

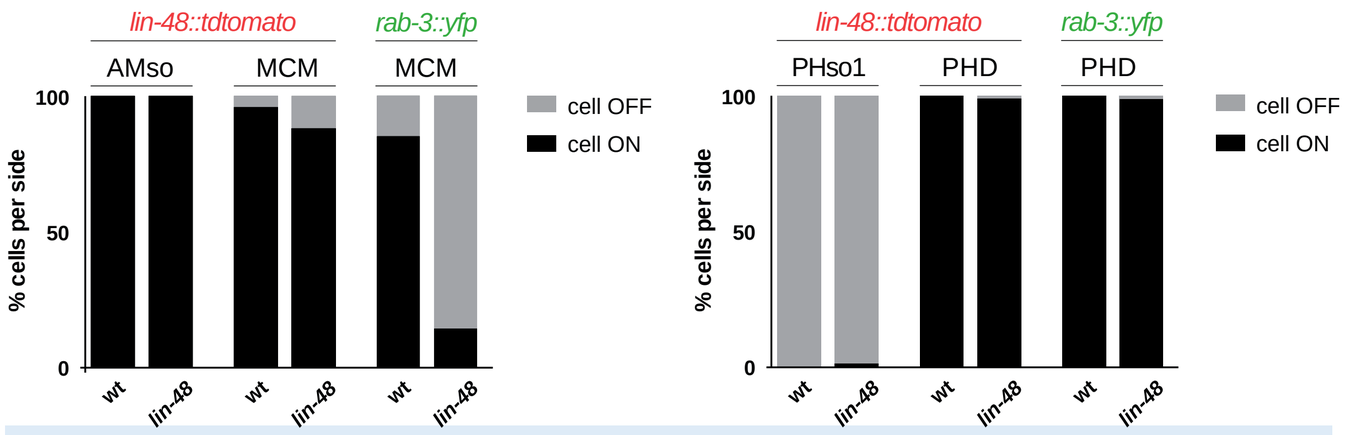

Figure 5. Quantification (n=80) of *otIs291 (rab-3::YFP)* and *drpIs3 (lin-48::tdtomato)* expression in *lin-48 (sa469)* mutant background. **The MCMs in some *lin-48* mutants failed to acquire their neuronal fate (no *rab-3::YFP* expression). (p=0.0001).**

Conclusions

1. *Lin-48* mutant strains carrying the *rab-3*panneuronal reporter and the *hlh-14* proneural gene reporter have been successfully generated.
2. *Lin-48* regulates MCM neurogenesis *(rab-3::YFP* expression), but not PHD neurogenesis, indicating that different molecular mechanisms regulate cell fate switch in MCM and PHD neurons.
3. The role of *lin-48* in *hlh-14* proneuronal gene expression remains to be determined.
4. A small percentage of AMso cells do not divide in *lin-48* mutants, suggesting an additional earlier role of the transcription factor in neuronal specification.
5. *Lin-48* mutants show two additional socket-like cells in the head, meaning *lin-48* likely acts upon other glial lineages.

Bibliography

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