



Identifying Regulators of Rho Family GTPases Required for Collective Cell Migration in *Drosophila* Testis Nascent Myotubes

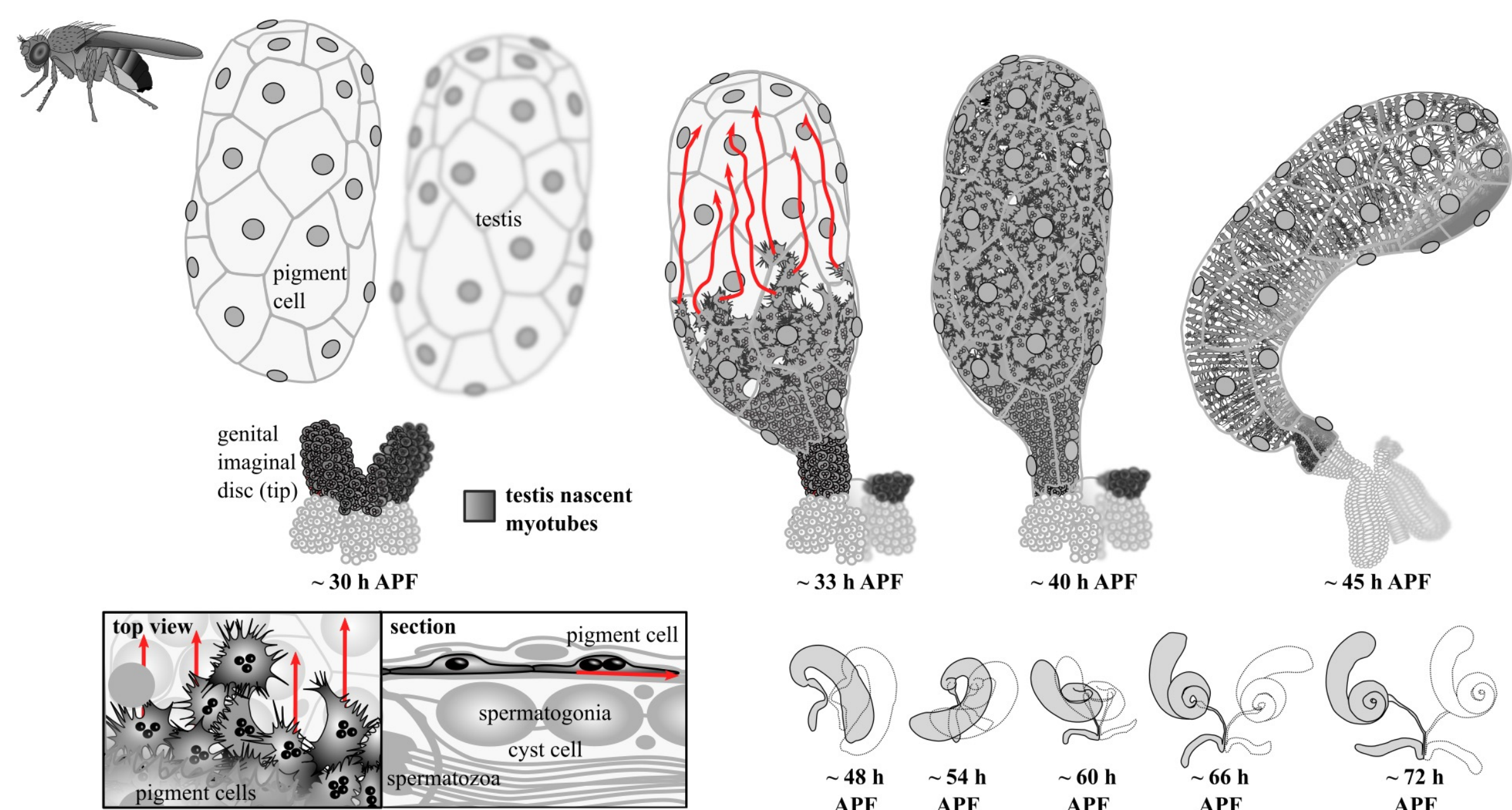
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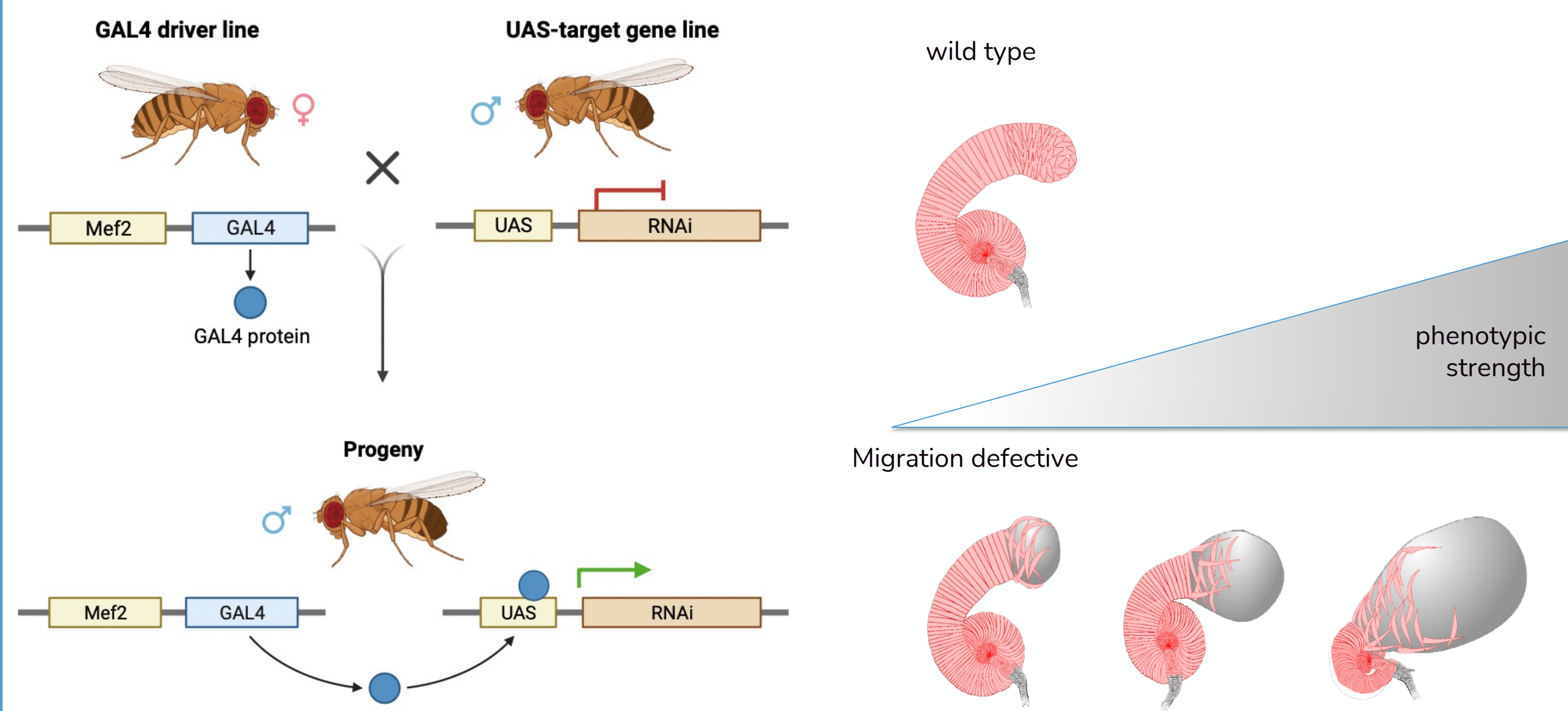


A New System to Study In Vivo Cell Migration in *Drosophila*

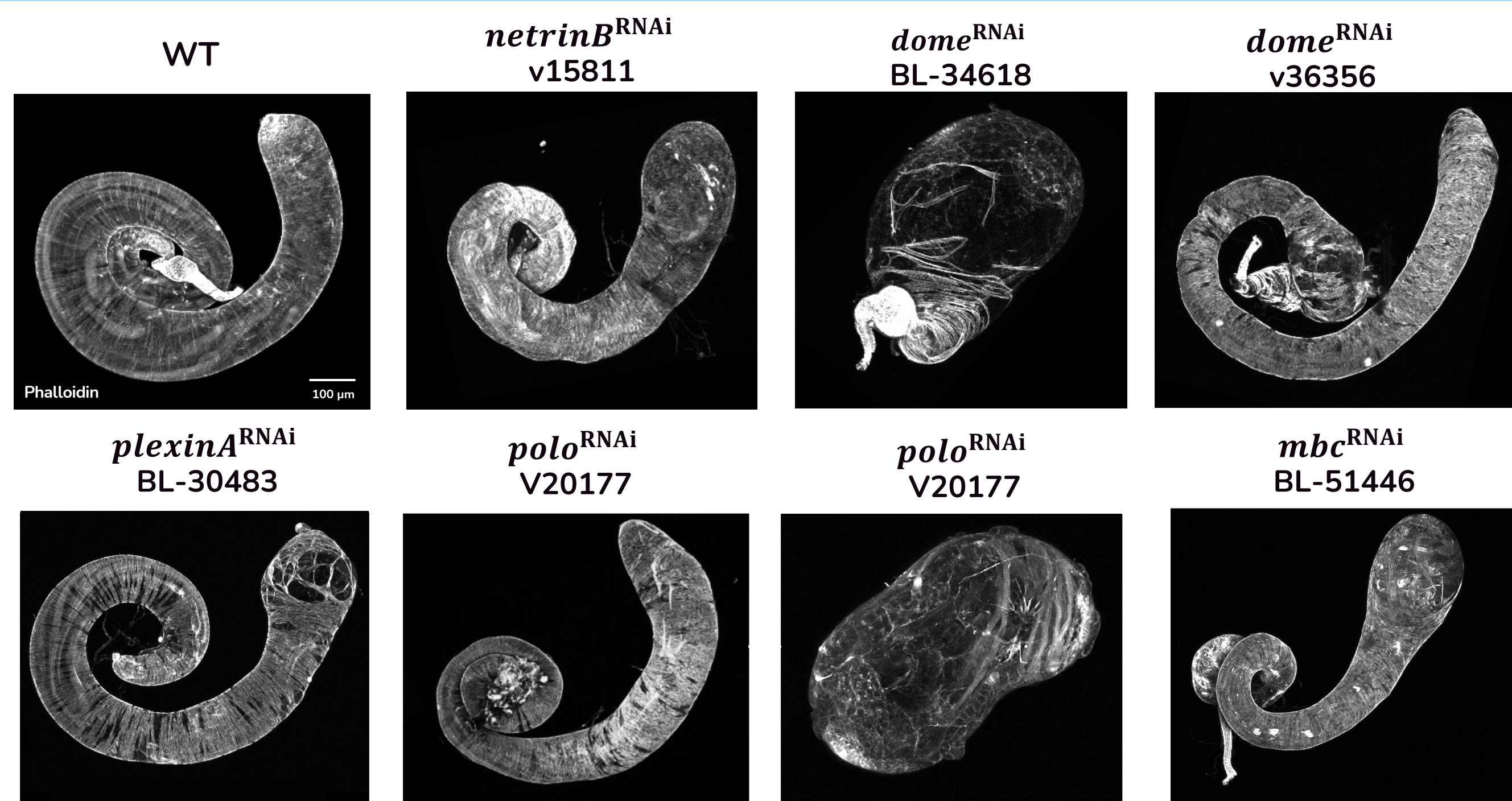
Testis myotubes use an interesting mechanism known as 'contact stimulation of migration,' migrating toward the free edge of the cell sheet



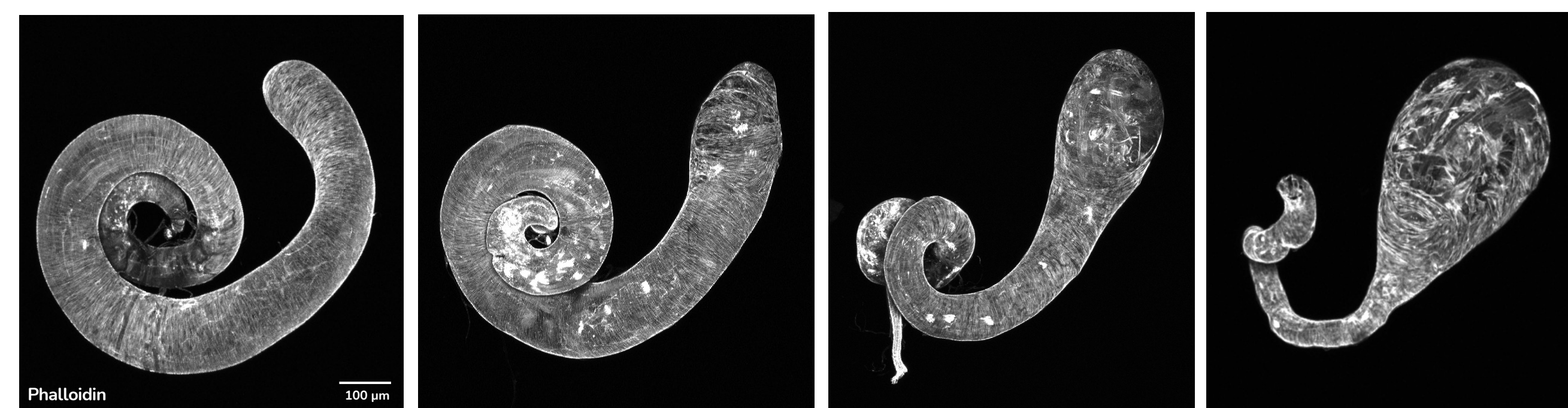
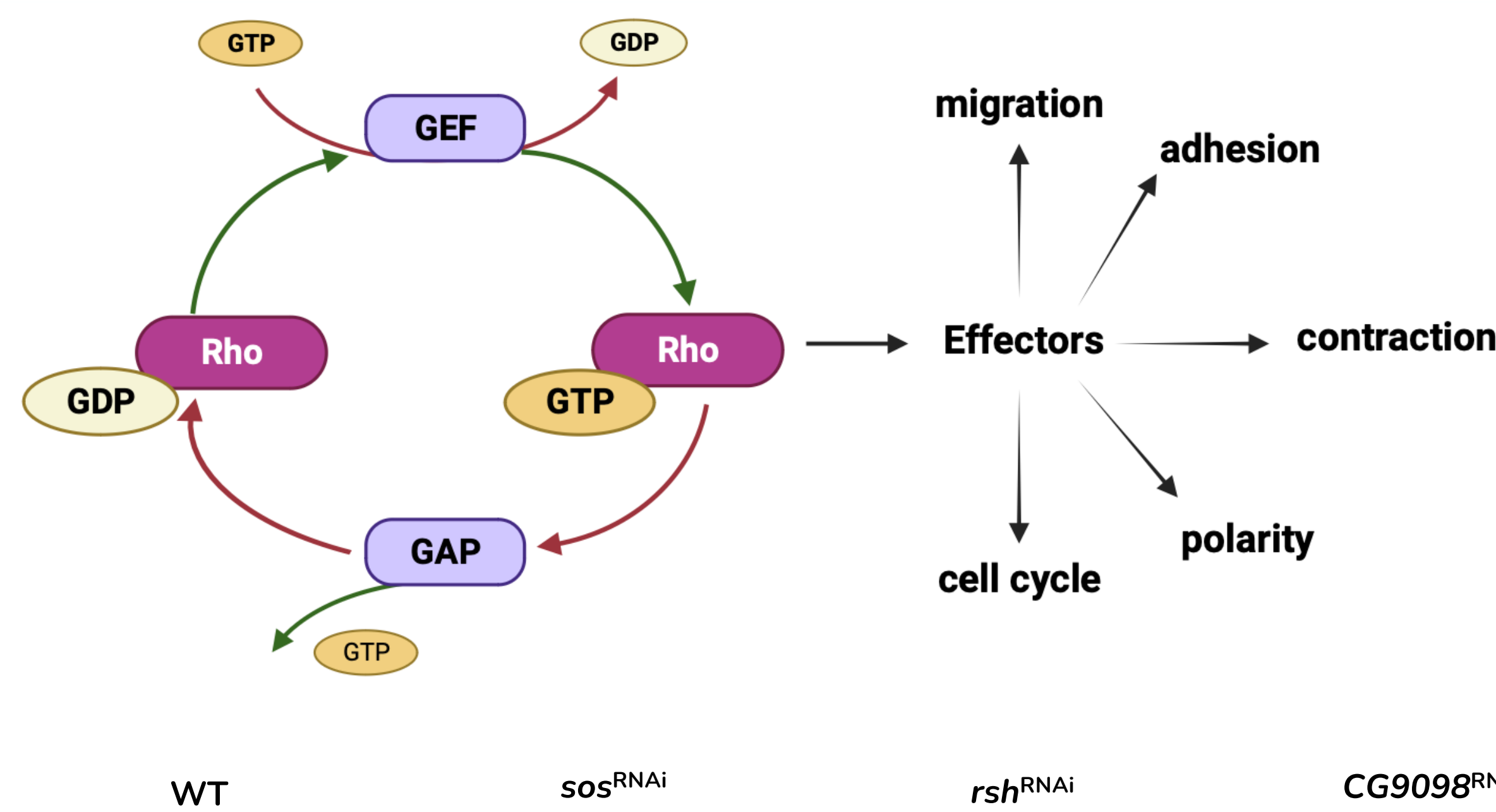
Our goal is to assess which receptors and ligands are important for cell migration and what proteins act downstream. To understand the 'social behavior' of these cells, we have knocked-down expression of genes in the migrating myotubes using the Gal4-UAS System and assessed adult testis



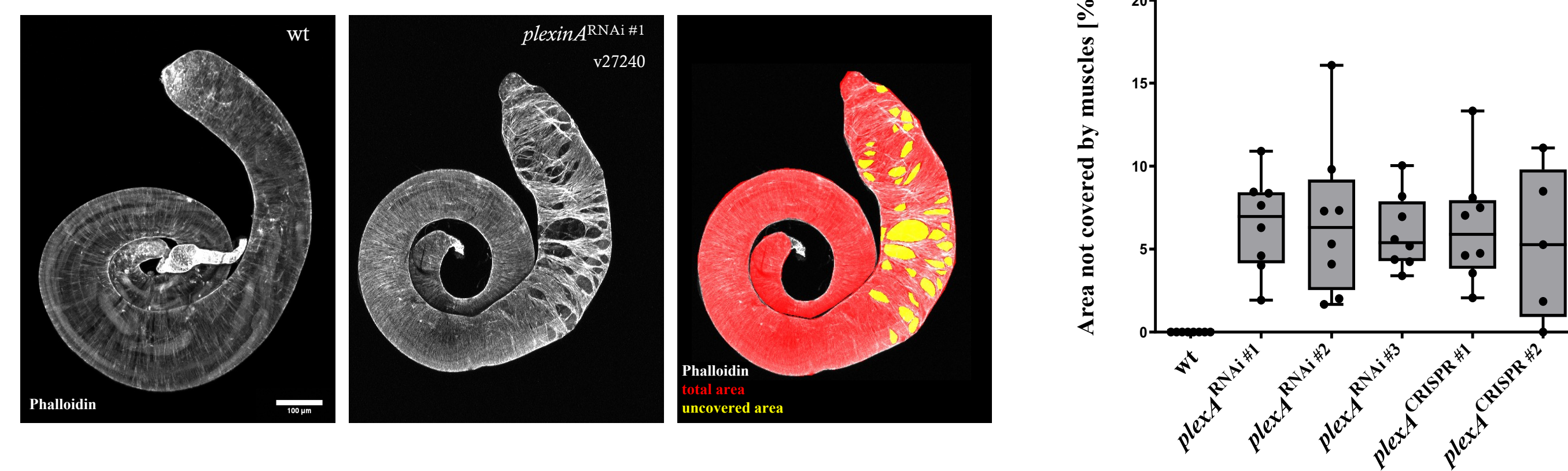
We performed two independent screens. In the first screen, myotube specific RNA-seq was used to identify potential ligands, receptors and regulators



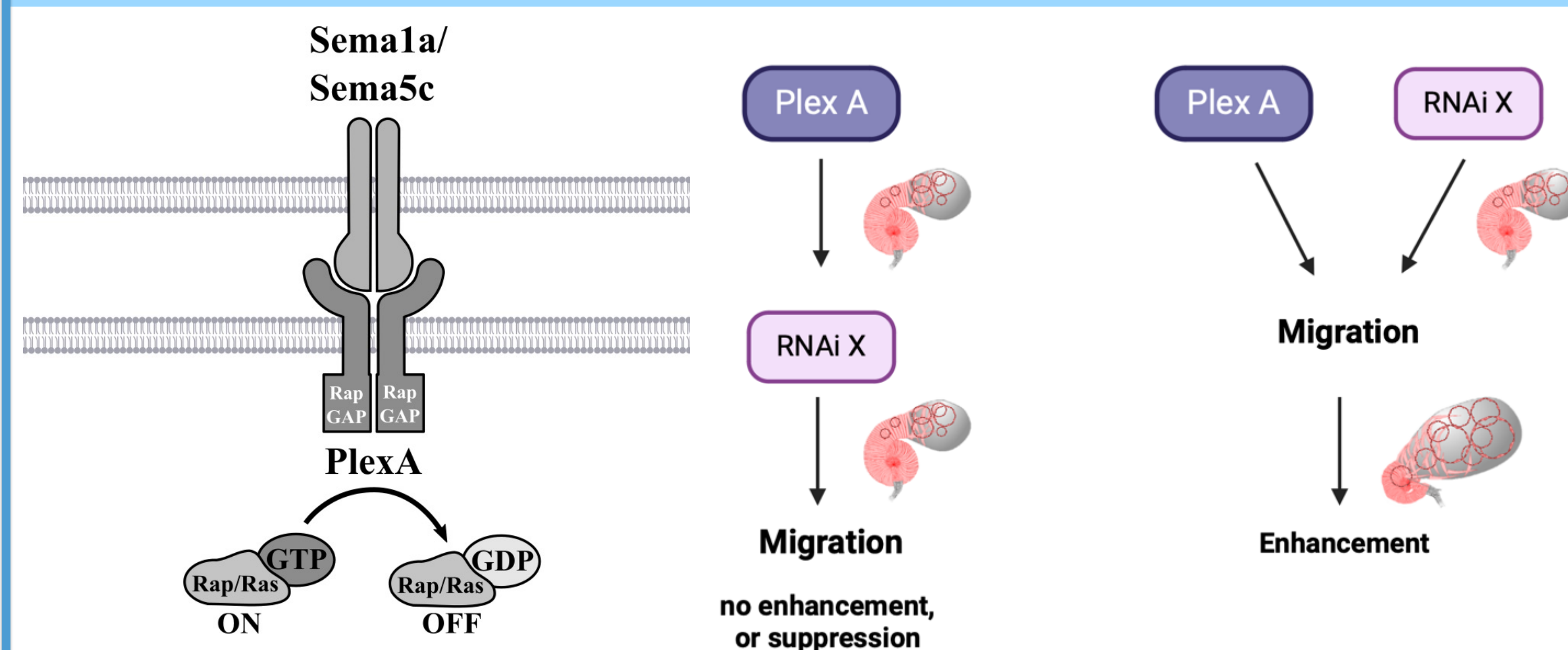
In the second screen, we knocked-down all Rho Family GAP and GEF regulators. Rho-family GTPases appear to act in different places during collective cell migration, and we identified several possible regulators



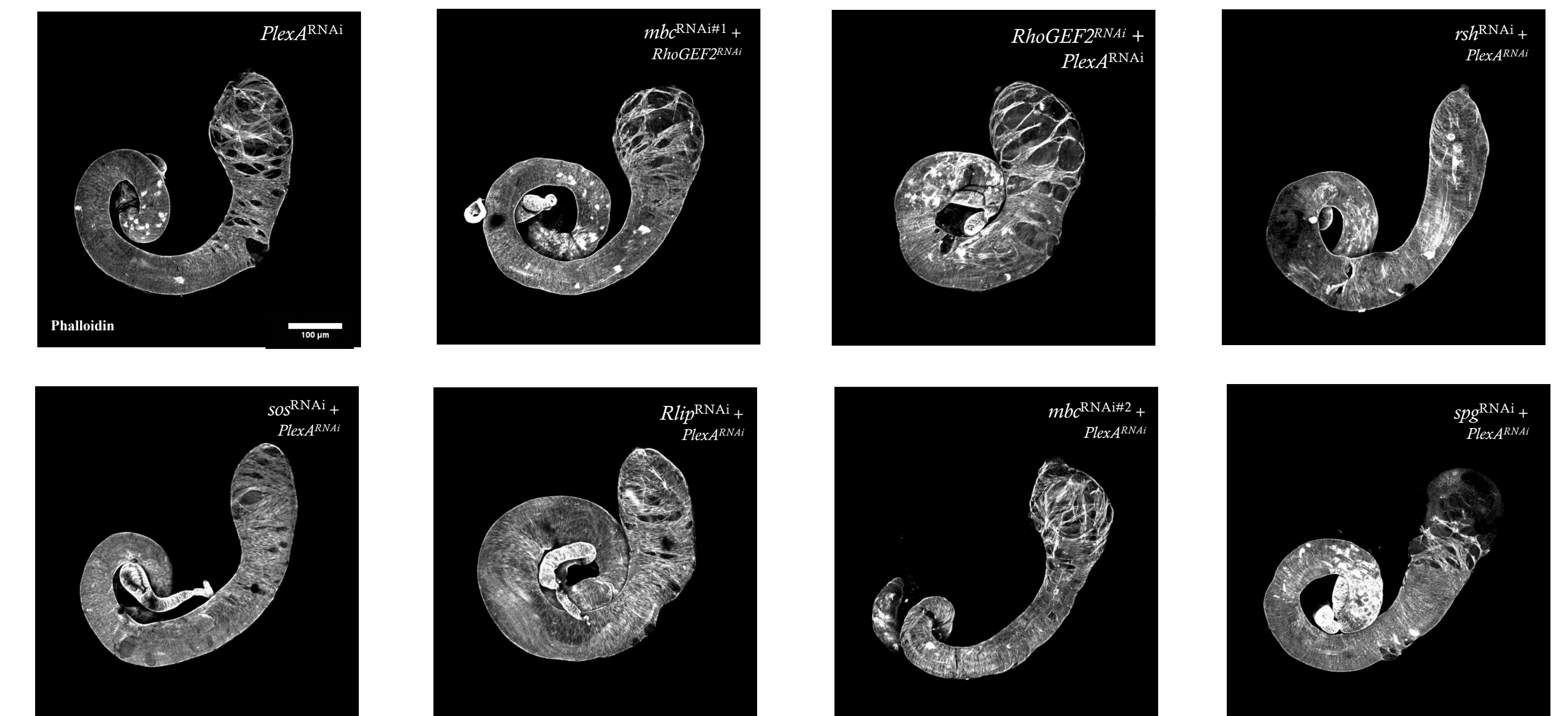
We're currently focusing on PlexA, a receptor known to regulate the migration of axons in the nervous system. We can quantify the strength of its phenotype by quantifying gaps in muscles



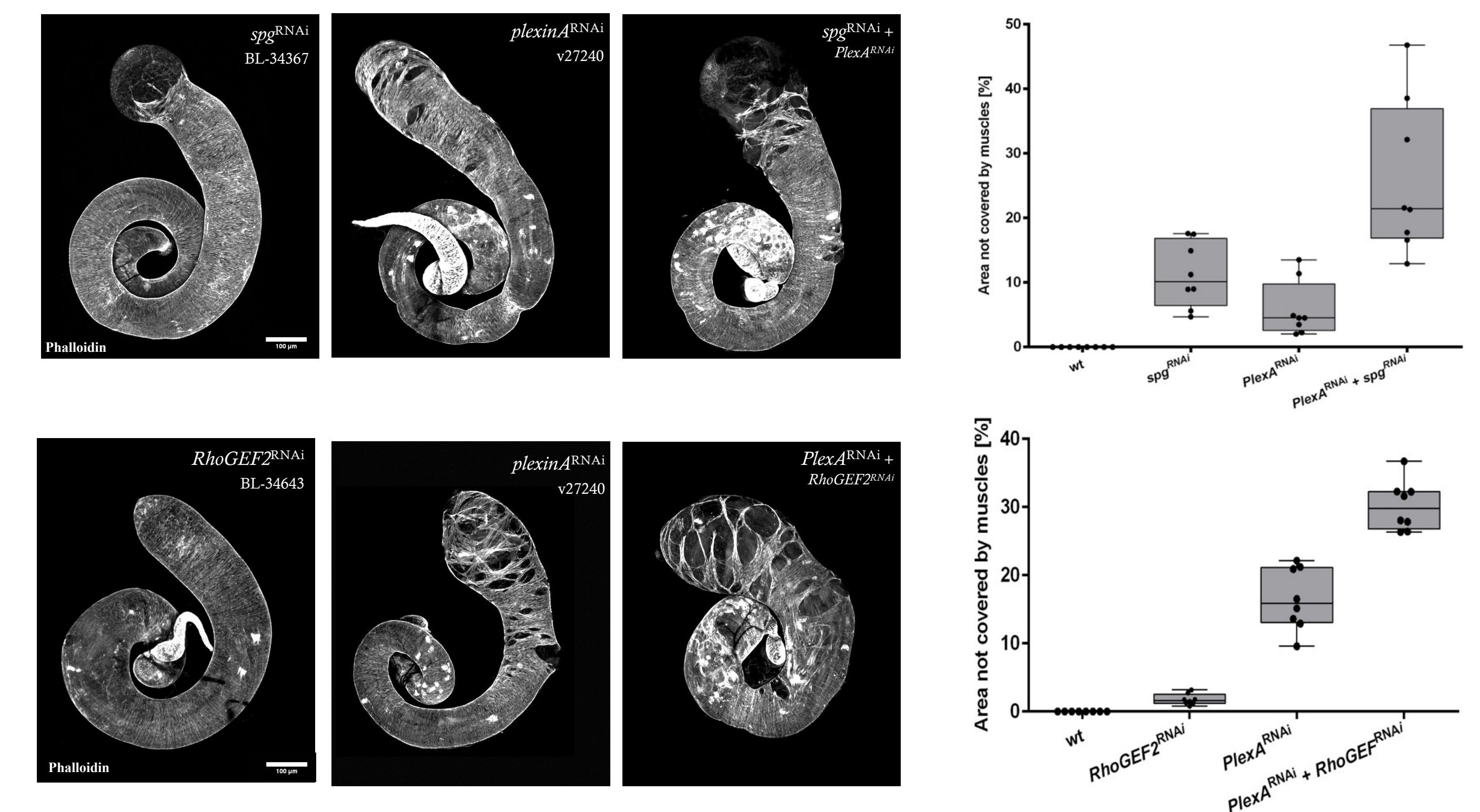
My project seeks to identify regulators acting downstream of PlexA, or in parallel pathways. If a regulator acts in the same pathway, double knockdown of PlexA and the regulator should NOT enhance the phenotype. However, if they act in different pathways, it might



I carried out double knockdown of PlexA and selected Rho-family regulators



Double knockdown of Plex A and two GEFs, Sponge and RhoGEF2, led to enhanced phenotype. This suggests both Sponge and RhoGEF2 are GEFs that act in parallel to the PlexA pathway



Moving Forward

- Complete single RNAi experiments for additional regulators: *Sos* RNAi, *mbc* RNAi#1, *rlip* RNAi, *rsh* RNAi, *mbc* RNAi#2
- Test more Rho Family Regulators
- Compare migration using live imaging after single and double knockdowns

Acknowledgements and Citations

I am beyond grateful for everyone in the Peifer Lab, specifically Maik for allowing me to be a part of his project, Jenna for guiding throughout the process, and Dr. Peifer for his constant advice and support. Our lab is supported by NIH R35 GM118096. Bischoff, M.C., Lieb, S., Renkawitz-Pohl, R., and Bogdan, S. (2021). Filopodia-based contact stimulation of cell migration drives tissue morphogenesis. Nat. Commun. 12, 1–18. Hu, S., and Zhu, L. (2018). Semaphorins and Their Receptors: From Axonal Guidance to Atherosclerosis. Front. Physiol. 9. Lawson, C.D., and Ridley, A.J. (2018). Rho GTPase signaling complexes in cell migration and invasion. J. Cell Biol. 217, 447–457. Rothenbusch-Fender, S., Fritzen, K., Bischoff, M.C., Buttgerit, D., Oenel, S.F., and Renkawitz-Pohl, R. (2017). Myotube migration to cover and shape the testis of *Drosophila* depends on Heartless, Cadherin/Catenin, and myosin II. Biol. Open 6, 1876–1888.