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SnapShot: Fly piRNAs, PIWI Proteins, and the Ping-Pong Cycle

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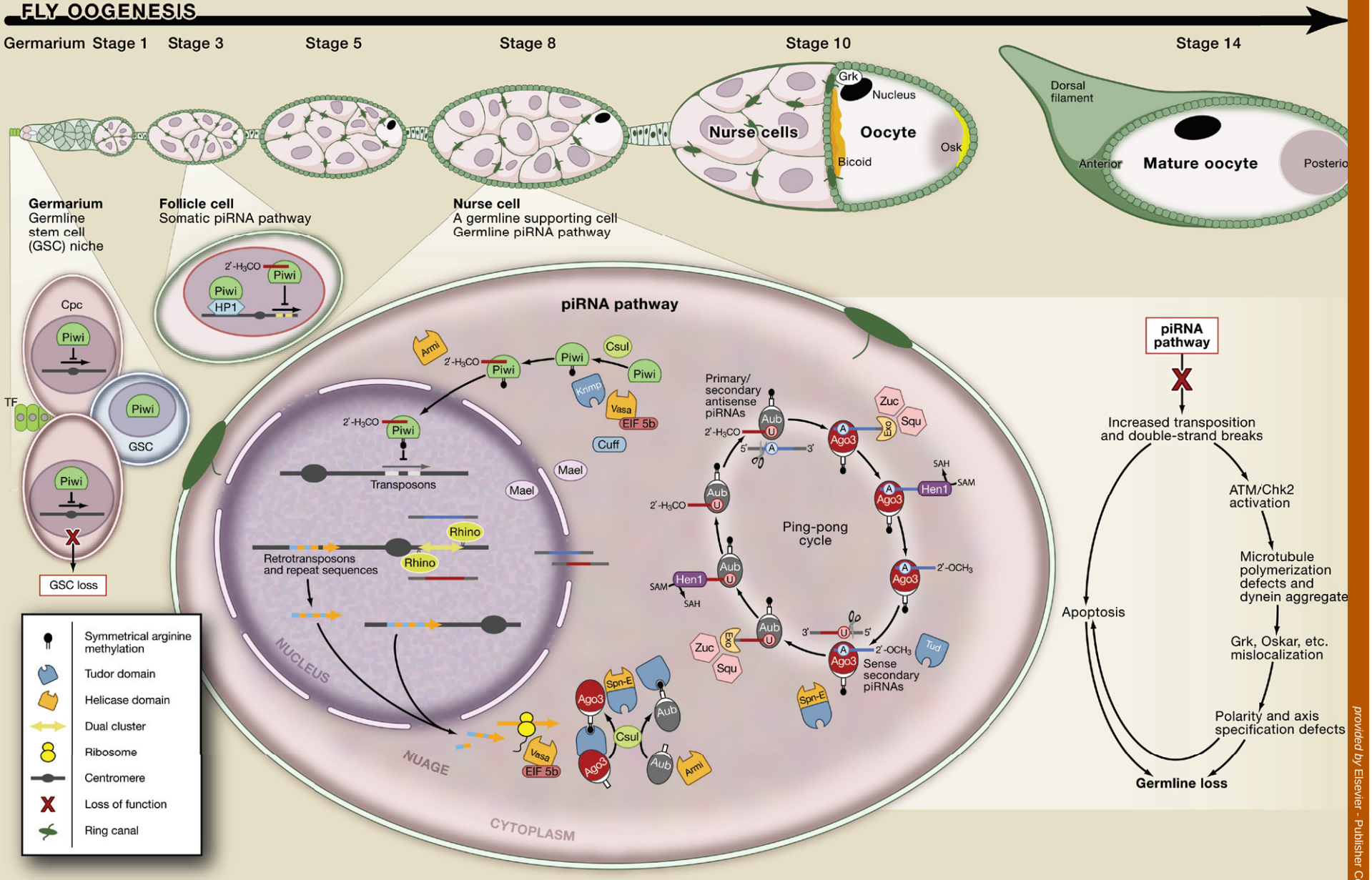

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In animals, PIWI-interacting RNAs (piRNAs) repress expression of transposons and other repetitive genomic sequences, ensuring faithful transmission of the genetic material from one generation to the next. These 23–30 nucleotide-long small RNAs are generally found in the germline. They bind to "PIWI" proteins, a specialized subclass of the Argonaute protein family, whose members use small RNA guides to silence gene expression. In zebrafish and *Drosophila*, piRNAs are required for fertility in both males and females, and loss of piRNAs results in male sterility in mice.

piRNAs in Fly Oogenesis

The three fly PIWI proteins—Piwi, Argonaute3 (Ago3), and Aubergine (Aub)—are expressed in both ovaries and testes. In ovaries, Piwi, which localizes to the nucleus, is found both in the germline and in two types of somatic cells: those that form the germline stem cell niche and the somatic follicle cells that surround the developing oocyte. Unlike Piwi, Ago3 and Aub reside in the nuage, a structure that rings the nucleus; Ago3 and Aub are restricted to the oocyte and its nurse cell sisters.

Drosophila piRNAs are produced in the nurse cells by a cycle of reciprocal, PIWI-protein catalyzed cleavage events that both amplify a small initial pool of primary piRNAs and bias the piRNA population toward antisense. This "ping-pong" cycle creates sense secondary piRNAs bound to Ago3. In turn, these sense piRNAs generate new antisense, Aub-bound piRNAs that can silence transposons and also create yet more sense piRNAs. It is not known if these events occur in the cytosol or the nuage.

Like other maternally deposited RNAs, piRNAs produced by ping-pong amplification are transported from the nurse cells to the oocyte via the ring canals, cytoplasmic bridges that link the 16 germline sister cells. Loss of piRNA pathway proteins such as Vasa, Aubergine, Spindle-E, Krimper, Armitage, Zucchini, Maelstrom—all of which are found in nuage—perturbs the embryonic axes. Remarkably, these polarity defects are a secondary consequence of activation of the double-stranded DNA break repair pathway, suggesting that piRNAs mainly function to block transposon mobilization in the oocyte.

In the somatic follicle cells, at least one source of piRNAs, the *flamenco* cluster, generates piRNAs directly and without ping-pong amplification. The unique organization of this locus ensures that nearly all of these piRNAs are antisense. How these piRNAs—which bind to Piwi rather than Aub or Ago3—are made is unknown.

Abbreviations

Cpc, cap cell; TF, terminal filament cell; GSC, germline stem cell; PIWI, P-element-induced wimpy testis; piRNA, PIWI-interacting RNAs; Mael, Maelstrom; eIF5b, eukaryotic translation initiation factor 5b; HP1, heterochromatin protein 1; Rhino, a HP1 homolog; RTs, retrotransposons; Krimp, Krimper; Csul, Capsuleen; Hen1, Hua enhancer 1; Armi, Armitage; Aub, Aubergine; Ago3, Argonaute3; Spn-E, Spindle-E; Zuc, Zucchini; Squ, Squash; Cuff, Cutoff; Grk, Gurken; Osk, Oskar; SAM, *S*-adenosylmethionine; SAH, *S*-adenosylhomocysteine; Tud, Tudor protein; ATM, Ataxia-telangiectasia gene; Chk2, Checkpoint kinase 2; MT, microtubule.

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