Supplementary Information

Merlin is required for coordinating proliferation of two stem cell lineages in the *Drosophila* testis

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Figure S1. The effect of *Mer* **mutation/knockdown on CySCs.** A) The number of Zfh-1⁺ cells/testis in *Mer*^{ts1} mutant and *c587-gal4>UAS-Mer*^{RNAi} testes. B, C) Examples of mitotic CySCs (arrows) in control (B) and *Merts1* mutant (C, 29°C) testes, where CySC mitoses were limited to next to the hub. Bar 10µm.



Figure S2. *Mer* **mutant cyst cells differentiate normally.** Control (A) and *Mer*^{ts1} (B) testes stained with anti-Tj, -Eya, and -Vasa antibodies, demonstrating that CCs differentiate normally into Eya-positive cells.



Figure S3. Neither Yki nor Hippo regulates Tj+ cyst cell number.

Yki, its nonphosphorylatable forms, and two independent RNAi constructs were expressed under the control of *c587-gal4* and combined with *tubulin-gal80ts*. The flies were cultured at 18°C until eclosion to avoid lethality during development, then transferred to 29°C for 7 days to allow transgene expression before analysis. Expression of neither *yki* nor *hpo^{RNAi}* affected either GSC number (A) or Tj⁺ cyst cell number (B).



Figure S4. Unchanged overall tissue architecture of testes upon overexpression of *Hh*, *Dl*, *Wg* or *Spi* in *Mer*^{ts1} mutant background.

Blue: Vasa, Red: FasIII and Add



Figure S5. Dpp-Tkv signaling in somatic CySCs/CCs regulates CySC proliferation.

A) *Mer*^{ts1} *nos-gal4>UAS-tkv** shows spermatogonial overproliferation (double-headed arrow) but not tumorous expansion of CCs.

B, C) Apical tip of the testes in control (B) and *c587-gal4>UAS-tkv** (C). Expression of Tkv* is sufficient to cause CC overproliferation.