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## Supplementary Information for

The Role of Translationally Controlled Tumor Protein (TCTP) in Proliferation of *Drosophila* Intestinal Stem Cells

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Legend for Supplementary Dataset S1 Figs. S1 to S4 Dataset S1: Mass spectrometry results

### SUPPLEMENTARY FIGURES & LEGENDS



#### Figure S1. The role of Tctp in maintenance of ISCs and EBs in the Drosophila

**midgut.** A-B. Tctp immunostainings in midgut. *UAS-Tctp* (7) and *UAS-Tctpi* (7) were expressed with *esgts*. The genotype of control is *esgts*/+. Scale bar indicates 15 μm. C. Midgut images. *UAS-Tctp* and *UAS-Tctpi* was expressed with *esgts*. *Dl-nls-LacZ* line was used to visualize ISCs. After incubating flies at 29°C for X days, midguts were dissected and stained with anti-β-Galactosidase antibody. Scale bar indicates 50 μm.







Expression of rpr with *esg<sup>ts</sup>* reduced ISCs presumably due to induction of cell death. This reduction in cell number by expression of rpr was rescued by p35 expression. **B.** Images of midguts. Transgenes were expressed with *esg<sup>ts</sup>* for a total of 7 days, and bleomycin was fed while expressing transgenes for 2 days before analysis. Scale bar indicates 50 µm.



Figure S3. Tctp protein levels are not significantly altered by activation of insulin,

mTOR, Ras, and JAK/STAT signaling pathways. A-D. Tctp staining in midguts.

Transgenes were induced with  $esg^{ts}$  by shifting to 29°C. Tctp was stained with anti-Tctp antibody (7). *UAS-InR<sup>act</sup>* and *UAS-myr-Akt* were induced for 5 days (**A**), *UAS-Rheb* was induced for 8 days (**B**), *UAS-Ras<sup>V12</sup>* was expressed for 3 days (**C**), and *UAS-Hop<sup>Tum</sup>* was induced for 4 days (**D**).



**Figure S4. TCTP mRNA levels are unaltered by Yki<sup>3S/A</sup> expression.** RNA samples were prepared from midguts with or without Yki<sup>3S/A</sup> expression for 4 days. *Tctp, thread* (*th*), and *expanded* (*ex*) mRNA levels were measured by RT-qPCR. The primer sequences used for qPCR are the followings: *Tctp*-1, 5'-TGATCTACGAGGTGTACGGAAA-3', 5'-TCGGTGGTTAAGCACAACATC-3';

*Tctp*-2, 5'-ACCCTCTACCTGAAGGACTACA-3', 5'-GCCGTTGATTTCGCGGTATT-3'; *th*-1, 5'-GCTGGACTGGCTGGATAAAC-3', 5'-

GCCGCAGAAAAAGCATTTAACT-3'; *th*-2, 5'-GCGTGGAAATCGGTTGCTG-3', 5'-GATGCGATCTAATGCTTCGGC-3'; *ex*, 5'-TGTGCTCATAGATGGCGAGTA-3', 5'-CGCGAAAGTGTAGCTCCAGG-3'.



Figure S5. Tctp does not significantly alter Hpo signaling and JAK/STAT signaling.

A. Diap1-LacZ staining. Transgenes were induced with *esgts* for 5 days at 29°C. Hpo signaling was assessed by staining with anti- $\beta$ -Galactosidase antibody. Scale bar indicates 50 µm. **B.** STAT-GFP reporter assay. GFP is expressed under the control of 5 repeats of a genomic fragment of *Socs36E* promotor region, which contains two Stat92E binding sites. Scale bar indicates 50 µm.