

1 **Supplementary Figure 1. CtBP limits eye growth and is required for Tgi-induced growth**
2 **inhibition.**

3 (A) Adult female *D. melanogaster* heads, anterior is to the right. Tissues were predominantly
4 comprised of cells that were homozygous for the indicated genotypes, which were generated by
5 eyFLP-driven mitotic recombination over a FRT82B cell lethal chromosome.

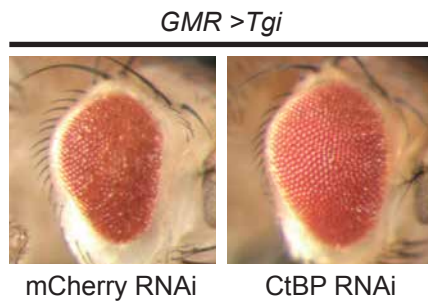
6 (B) Adult male *D. melanogaster* heads from the indicated genotypes, anterior is to the right. CtBP
7 RNAi line: *BSC#32889*.

Vissers Figure S1

A



B



8 **Supplementary Figure 2. Pits is required by Tgi suppress eye growth.**

9 (A) A third instar larval wing imaginal disc that expresses a Pits RNAi transgene in the posterior

10 compartment under the control of *en-Gal4*. Pits expression is in red, GFP (green) marks the

11 posterior compartment. The third panel is a merge of Pits and GFP, whilst DAPI (blue) marks

12 nuclei in the panel on the right.

13 (B) Heads of adult flies harbouring *tubulin-Gal4* and crossed to the indicated RNAi transgenes were

14 subjected to western blot analysis using the indicated antibodies. Molecular mass markers (kDa) are

15 indicated.

16 (C) Schematic diagram of the *pits* locus. Locations of P element insertions *P(EP)PITS^{EP1313}* (EP),

17 *P(SUPor-P)PITS^{KG07818}* (KG) and CRISPR/Cas9-generated frameshift mutations (SK) are indicated

18 relative to *pits* transcript variant D, which is the longest ORF of three *pits* transcript variants.

19 (D) Heads of adult flies of the indicated genotypes were subjected to western blot analysis using the

20 indicated antibodies. Molecular mass markers (kDa) are indicated.

21 (E) The same third instar larval eye imaginal disc as in Figure 3C, anterior is to the right. Pits

22 expression is in red, GFP (green) marks wild-type tissue, whilst GFP-negative tissue harbours the

23 *pits^{SK2}* allele. The third panel is a merge of Pits and GFP, whilst DAPI (blue) marks nuclei in the

24 panel on the right.

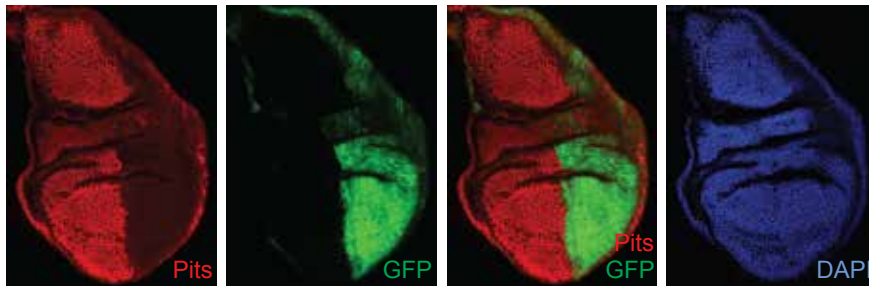
25 (F) Adult male *D. melanogaster* heads, anterior is to the right. Flies expressed *GMR-Gal4* and *UAS-*

26 *HA-Tgi* in either a control background (*w¹¹¹⁸*), or in backgrounds that were hemizygous for either

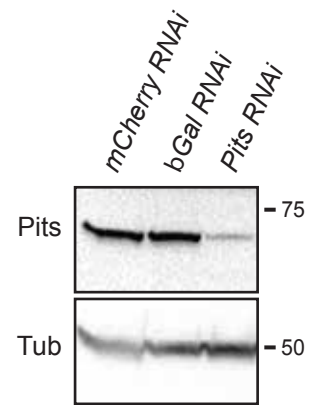
27 *pits^{SK5}* or *pits^{KG07818}*.

Visser's Figure S2

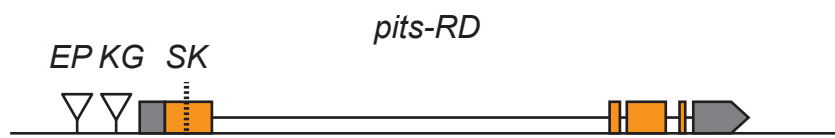
A



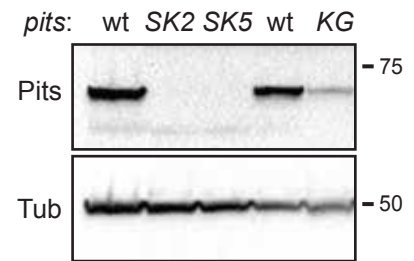
B



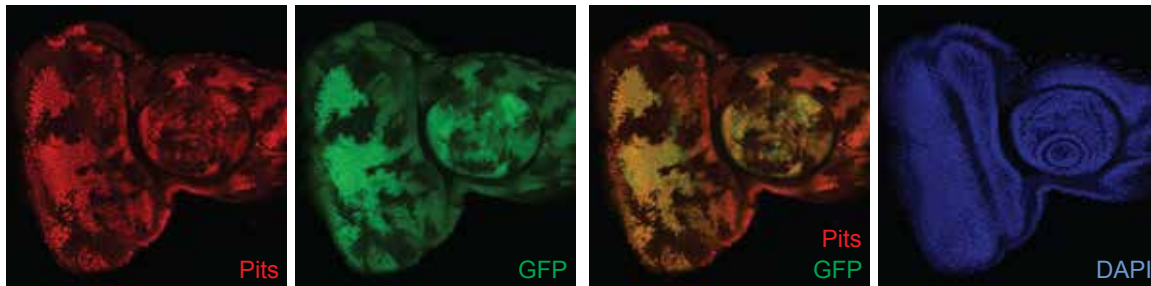
C



D

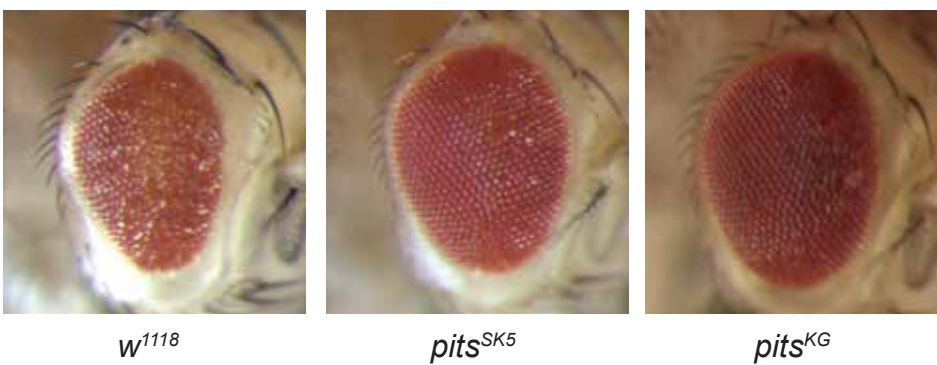


E



F

GMR >>Tgi



28 **Supplementary Figure 3. Cooperative effects of Tgi and Pits overexpression on eye growth**
29 **and *D. melanogaster* survival.**

30 (A) Heads of adult flies harbouring *GMR-Gal4* and crossed to the indicated transgenes were
31 subjected to western blot analysis using the indicated antibodies. Molecular mass markers (kDa) are
32 indicated.

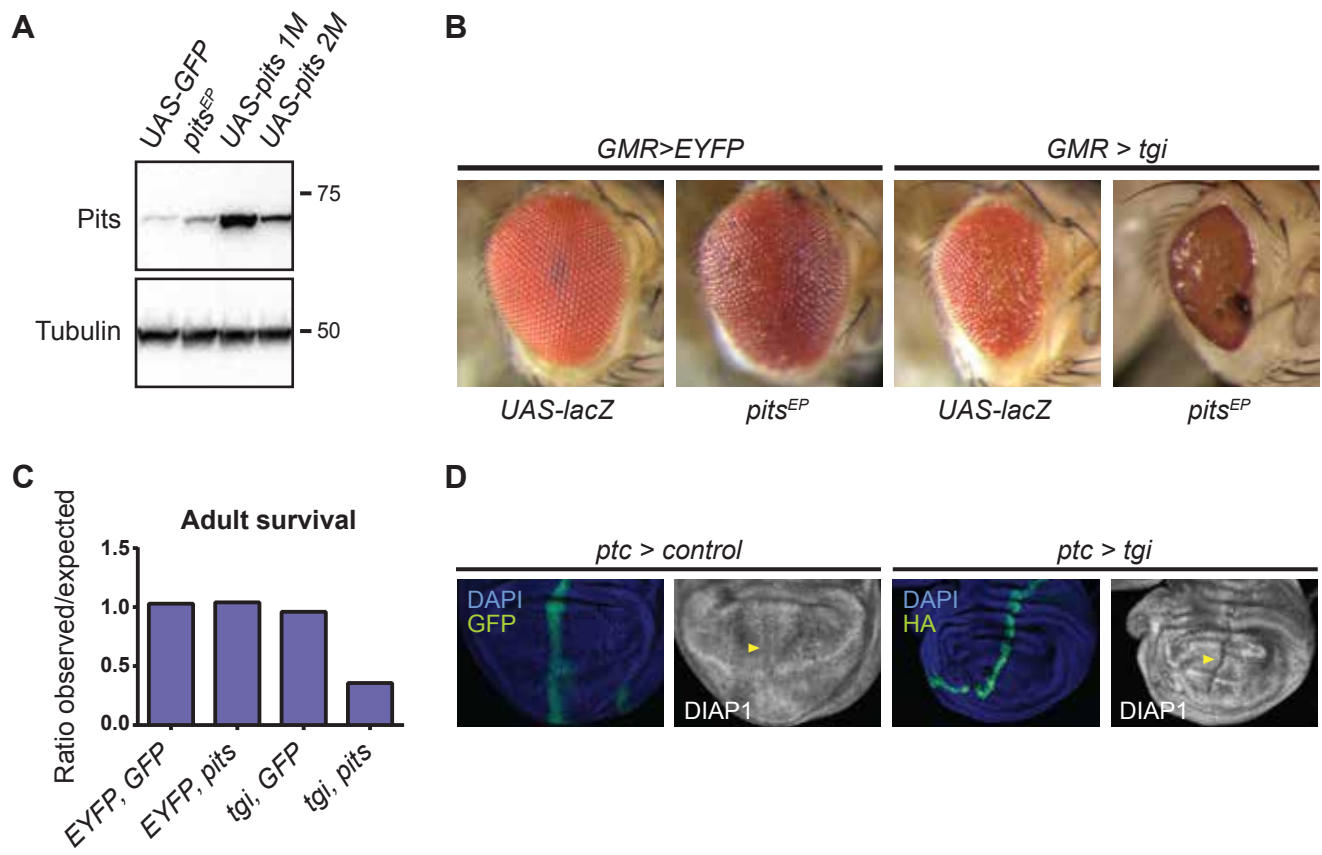
33 (B) Adult male *D. melanogaster* heads, anterior is to the right. Flies expressed *GMR-Gal4* and
34 either *UAS-EYFP* or *UAS-HA-Tgi*, and either *UAS-lacZ* (control) or *pits*^{EP1313}.

35 (C) Relative amounts of viable adult *D. melanogaster* after *GMR-Gal4* driven expression of the
36 indicated transgenes. The numbers of *D. melanogaster* counted were (relevant genotype/total):
37 157/305 (*EYFP*, *GFP*), 258/496 (*EYFP*, *pits*), 161/335 (*Tgi*, *GFP*), 45/505 (*Tgi*, *pits*).

38 (D) Third instar larval wing imaginal discs of the indicated genotypes. DIAP1 protein is in
39 greyscale. DAPI (blue) marks nuclei and GFP (green) marks the expression domain of the indicated
40 transgenes. Yellow arrowheads indicate the expression domain of control or *tgi* transgenes. Note the
41 same control tissue is displayed here as in the related figure, Figure 3D.

42

Visser's Figure S3



43 **Supplementary Table S1. Mass spectrometry data for Tgi-SBP purification.**

44 *Drosophila* proteins identified in Tgi-SBP ('TGI-C') and control ('EV-C') pulldowns (minimum 2
45 unique peptides and intensity value 1000). The peptide intensity ratio of proteins in Tgi-
46 SBP/control (TGI/EV) identifies proteins enriched in Tgi-SBP pulldowns. Proteins of interest were
47 additionally selected by comparison with a common contaminant list (Alexey Veraksa, personal
48 communication).