Supplementary Figure 1

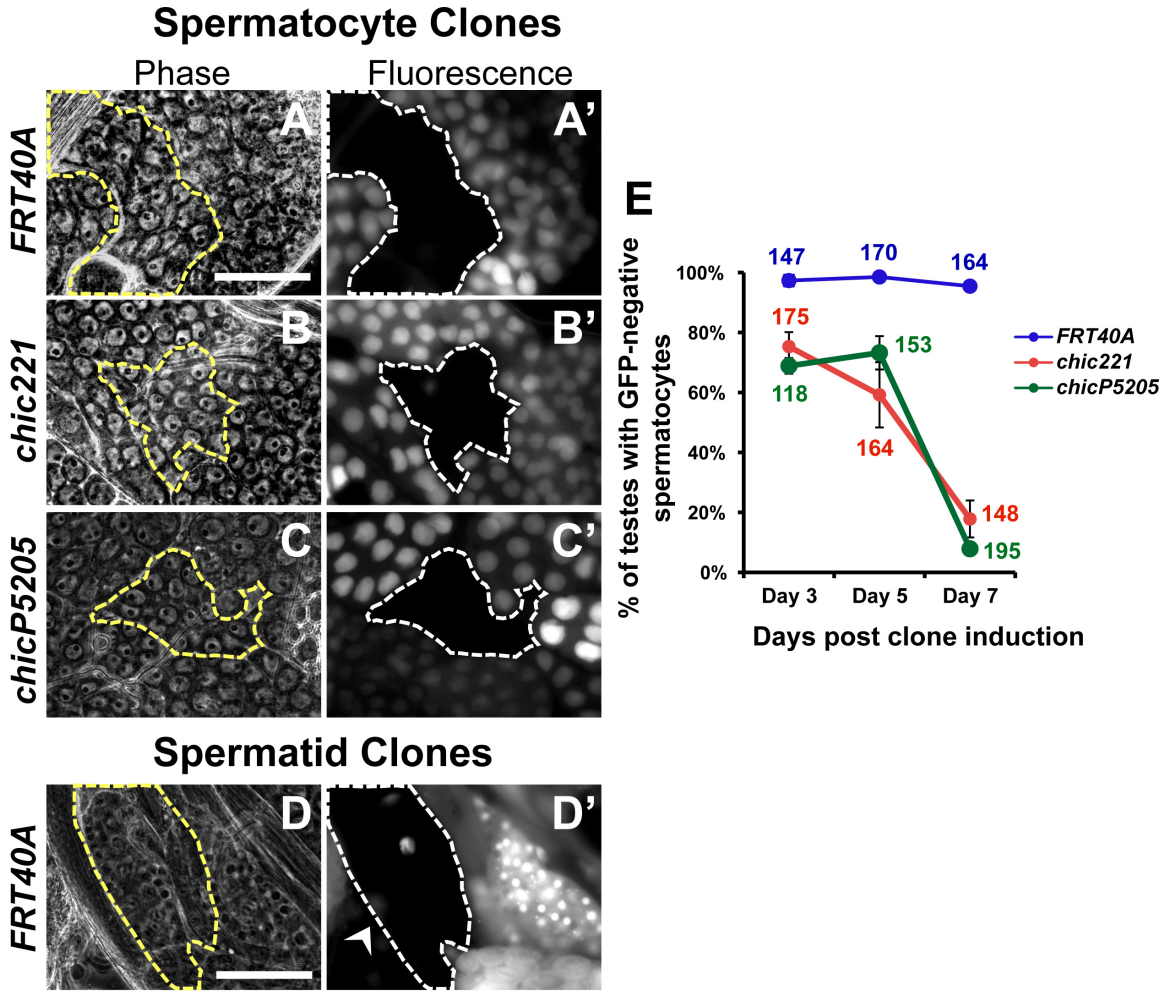

Fig. S1. *chic* homozygous null GSCs differentiated into mature spermatocytes but failed to execute spermatid differentiation.

(**A-D'**) Phase and fluorescence images of adult testes from (**A,A',D,D'**) control *FRT40A/GFP*, (**B,B'**) $chic^{221}$*/GFP*, and (**C,C'**) $chic^{P5205}$/GFP animals showing homozygous clones marked by loss of GFP. Dashed outline: GFP-negative clones. Arrowhead: spermatid clones present only in control *FRT40A* testes. Scale bars: 10 µm. (**E**) Percent of testes with one or more GFP-negative spermatocyte cysts in control *FRT40A* (blue), $chic^{221}$ (red) or $chic^{P5205}$ (green) over time after clone induction. Numbers

next to data points reflect the number of testes scored for each genotype at each time point. Error bars denote standard error of the mean.

Supplementary Figure 2

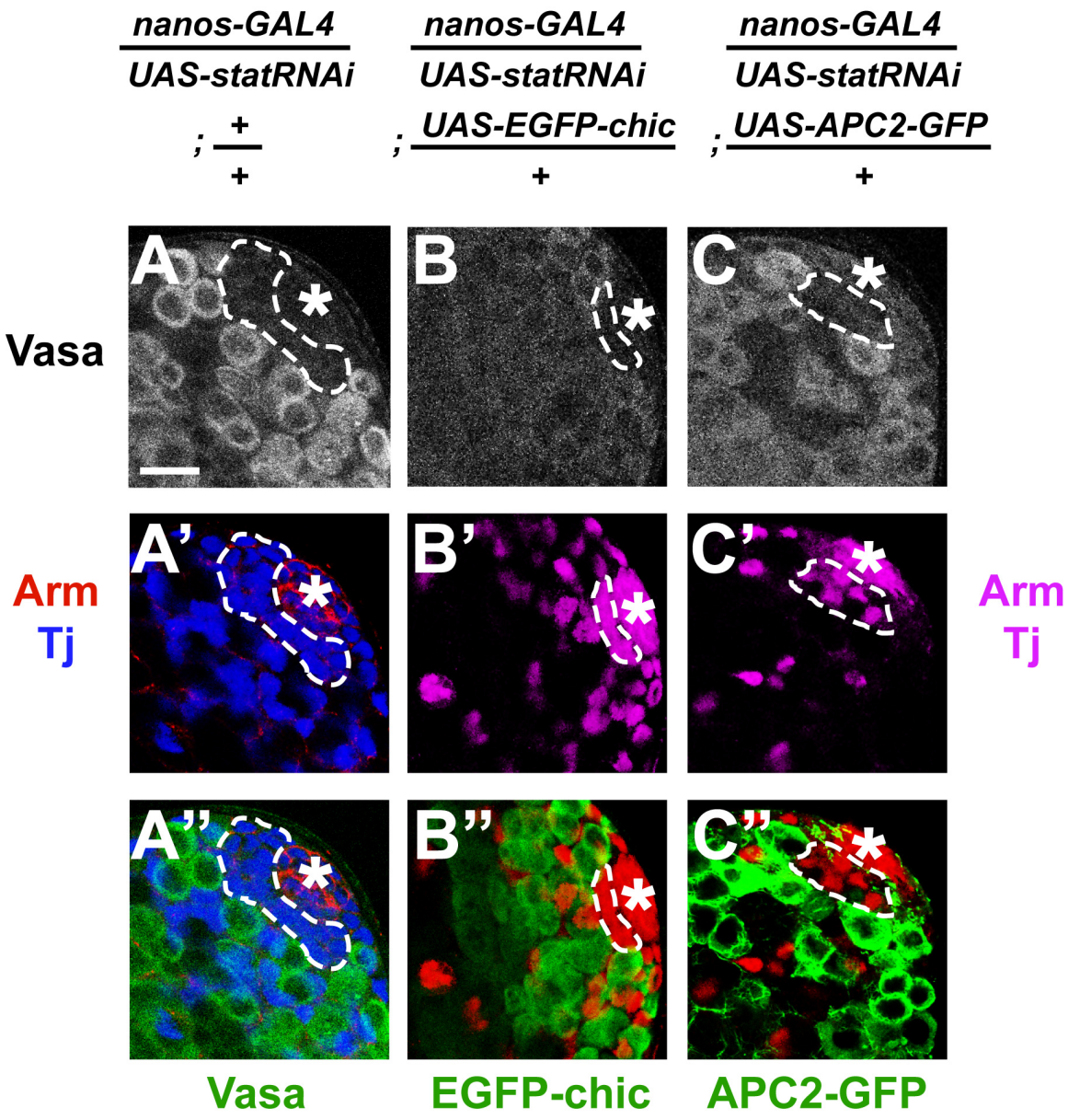

Fig. S2. Over-expression of chickadee or APC2 in *stat*-depleted GSCs failed to rescue GSC attachment to the hub.

(**A-C"**) Immunofluorescence images of adult testis apical tips from *nanos-GAL4/UAS-statRNAi* animals in which function of *stat* had been specifically knocked down in germ cells. Testes from (A-A") a *nanos-GAL4/UAS-statRNAi; +/+* animal containing *stat-*

depleted GSCs, (B-B") a *nanos-GAL4/UAS-statRNAi; UAS-EGFP-chic/+* animal containing *stat*-depleted GSCs which ectopically express profilin/chickadee, (C-C") a *nanos-GAL4/UAS-statRNAi; UAS-APC2-GFP/+* animal containing *stat*-depleted GSCs which ectopically express APC2. Anti-Vasa (green) to mark germ cells. Anti-Arm (red or magenta) to outline hub cells. Anti-Tj (blue or magenta) to mark the nuclei of somatic cyst cells and hub cells. Anti-GFP (green) to highlight expression of the GFP-tagged transgenes. Asterisk: the apical hub. White dotted area: region of somatic cells surrounding the hub and displacing *stat*-depleted GSCs. Scale bar: 20 μm.

Supplementary Figure 3

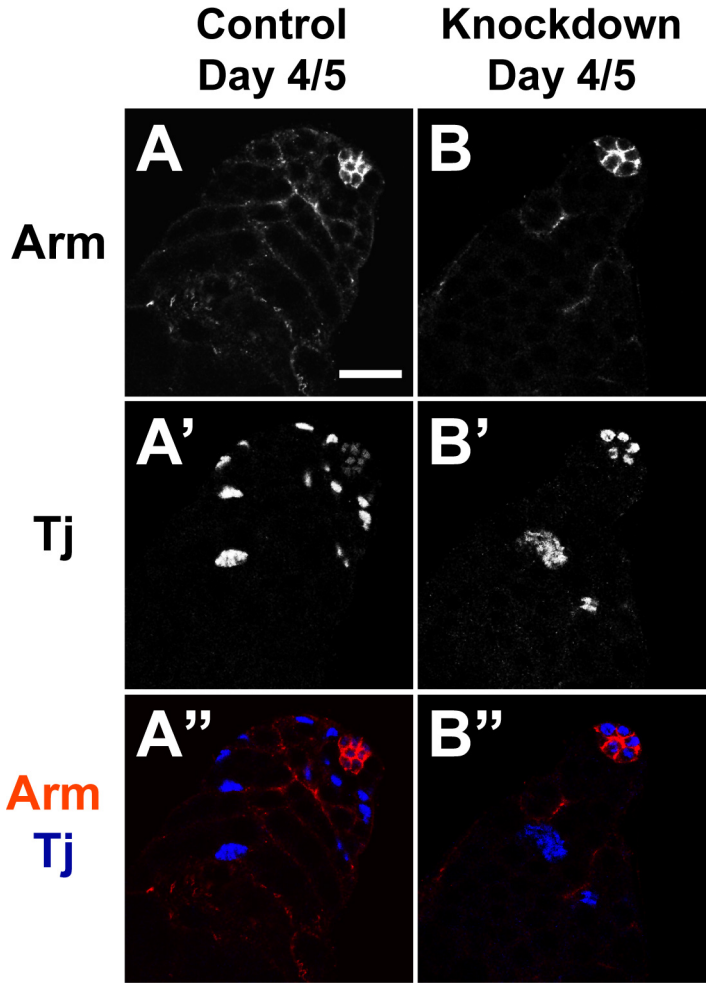

Fig. S3. Somatic cells lacking *chic* function fail to maintain cytoplasmic extensions. Immunofluorescence images of testis apical tips from (**A-A''**) *c587-GAL4;+* controls and (**B-B''**) *c587-GAL4; UAS-chicRNAi* animals. (**A,B**) anti-Arm to outline the cell membranes of somatic cyst cells, (**A',B'**) anti-Tj to label early cyst cell nuclei, (**A'',B''**) Merge. Scale bar: 20 µm.

Supplementary Figure 4

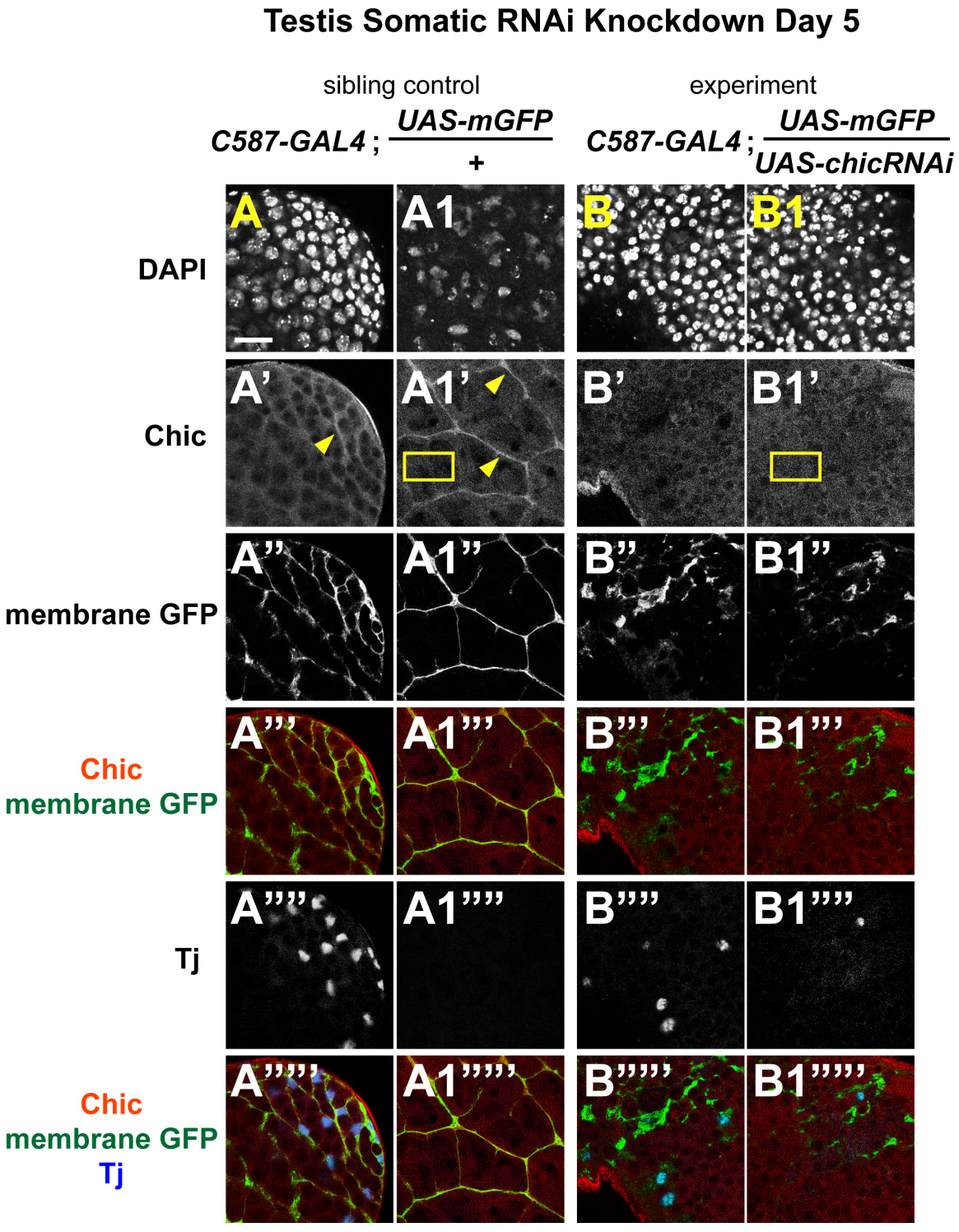

Fig. S4. RNAi knockdown of *chic* in testis somatic cells was specific and efficient.

(**A-B””””**) Immunofluorescence images of adult testis apical tips from (**A-A1””””**) a *c587-GAL; UAS-mGFP/+* control and (**B-B1””””**) a *c587-GAL4; UAS-mGFP/UAS-chicRNAi* animal in which function of *chic* had been specifically knocked down in somatic cells of the testis. (**A,A1,B,B1**) DAPI to mark cell nuclei. (**A’,A1’,B’,B1’**) anti-Chic to mark profilin/chickadee protein expression. (**A”,A1”,B”,B1”**) anti-GFP to highlight membrane-bound GFP expressed in the cytoplasmic membranes of somatic cyst cells. (**A””,A1””,B””,B1””**) anti-Tj to mark the nuclei of somatic cyst cells. Yellow arrowheads: chickadee protein expressed in the cytoplasmic membranes of somatic cyst cells. Yellow boxes: chickadee protein expressed in germ cells. Scale bar: 20 μm.