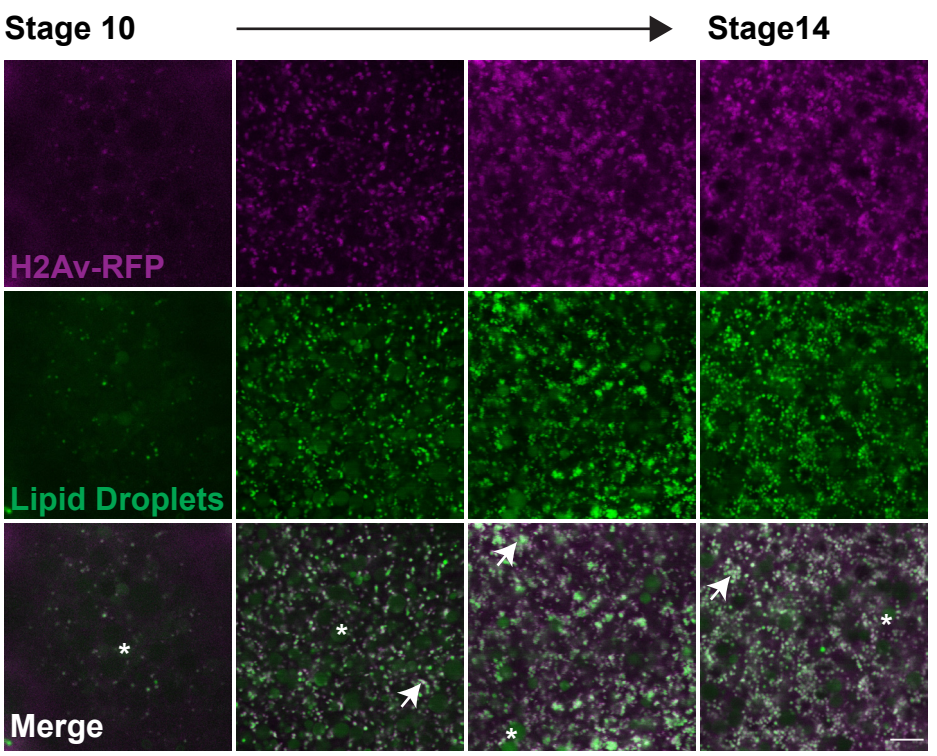
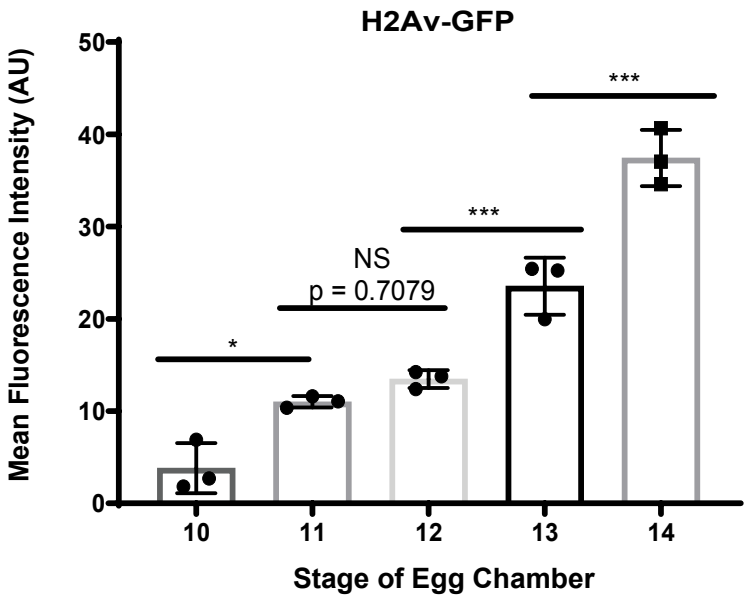


Fig. S1. Ooplasmic H2Av is present on LDs.

Labeled LDs (BODIPY, green) in ECs of flies expressing H2Av-RFP (magenta). Arrows: H2Av-LD colocalization. Yolk (large structures, asterisk) are visible due to autofluorescence. Scale bar: 10μm

A



B

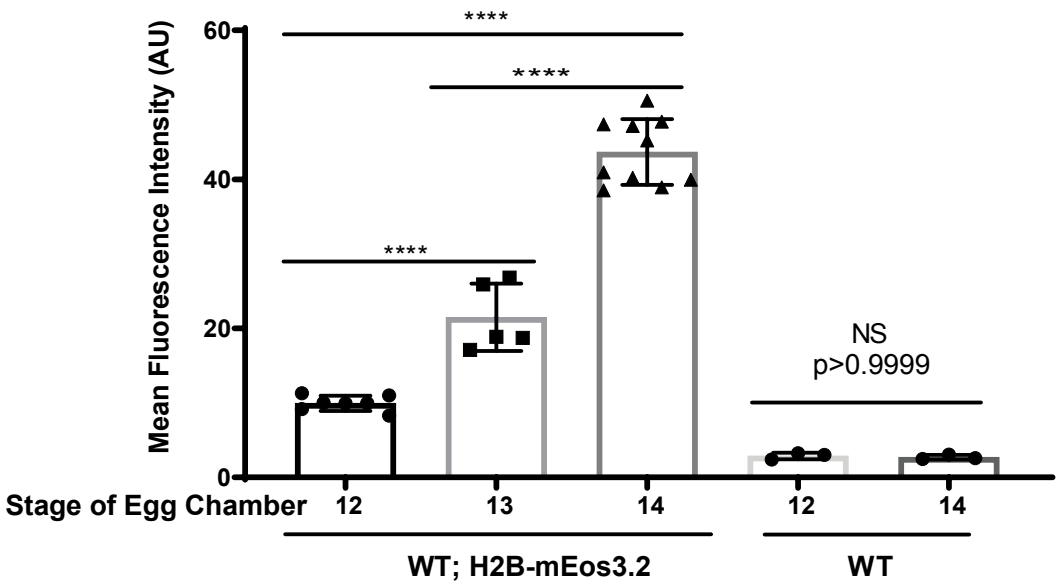


Fig. S2. H2Av and H2B levels increase during oogenesis.

(A) Quantitation of H2Av-GFP Mean Fluorescence Intensity (AU) in the ooplasm over developmental time. S10 vs S11, p=0.0243; S11 vs S12, p=0.7079; S12 vs S13, p=0.0026; S13 vs S14, p=0.0002; n=3. (B) Quantitation of H2B-mEos3.2 Mean Fluorescence Intensity (AU) in the ooplasm over developmental time. Intensity did not change significantly in ECs expressing no fluorescently-tagged H2B. S12 vs S13, p<0.001; S13 vs S14, p<0.001; S12 vs S14, p<0.001; WT S12 vs WT S14, p>0.9999. n=3-10. Statistical test: a one-way ANOVA followed by Tukey's test

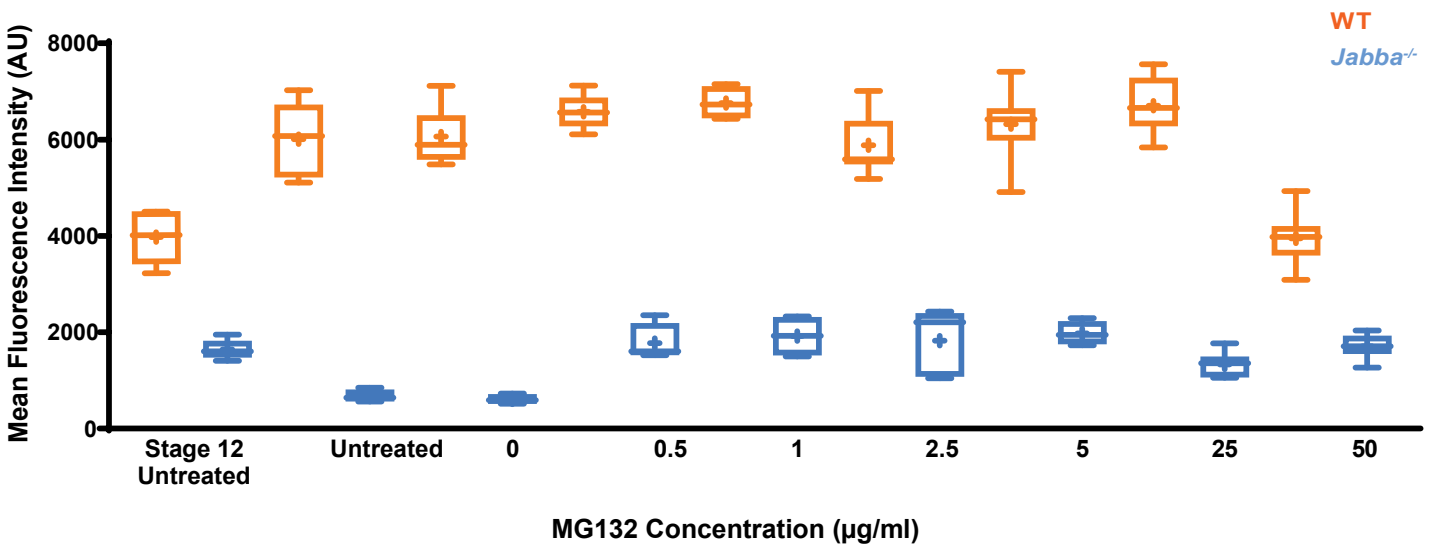


Fig. S3. H2Av degradation in the absence of Jabba

Mean fluorescence intensity (AU) in S14 WT (orange) or *Jabba*$^{-/-}$ (blue) ECs after IVEM. Length of box plot: 25th and 75th percentile. Line: median, cross: mean. Stage 12 Untreated = S12 ECs analyzed immediately after dissection; Untreated = cultured in IVEM media. Fig. 5B contains a subset of these data. n=4-8

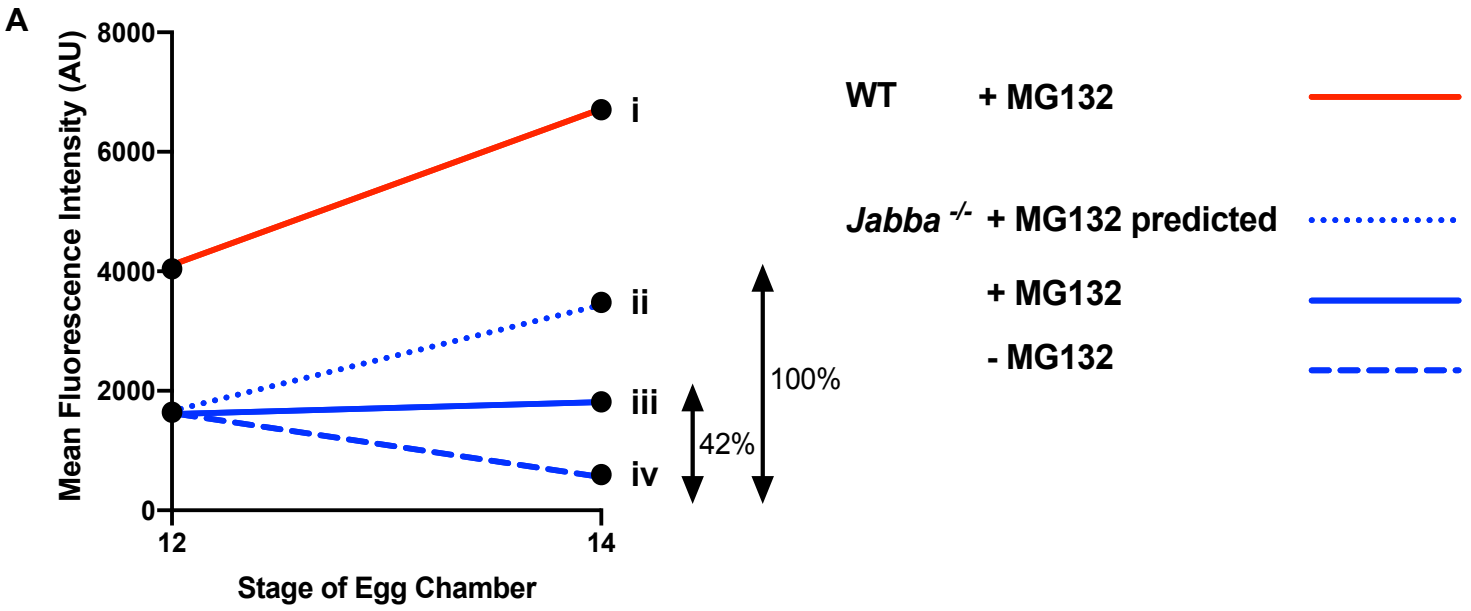


Fig. S4. Quantitative estimate of the drug effect on proteasome inhibition. (A) Analysis of the quantitative estimate of the effect on proteasome inhibition for 2.5 µg/ml of MG132. From S12 to S14, WT H2Av levels increase by 1839 AU (i, red line). Upon MG132 treatment, a similar rise in *Jabba*$^{-/-}$ ECs would lead to 3481 AU (ii,1642 AU in S12 + 1839 AU; blue, dotted line). With no drug, levels in *Jabba*$^{-/-}$ fall to 606 AU (iv; blue, dashed line). The observed value (iii) upon MG132 treatment represents 42% of the maximal possible effect (blue, solid line). (B) Estimated drug effect on proteasome inhibition for MG132 concentrations.

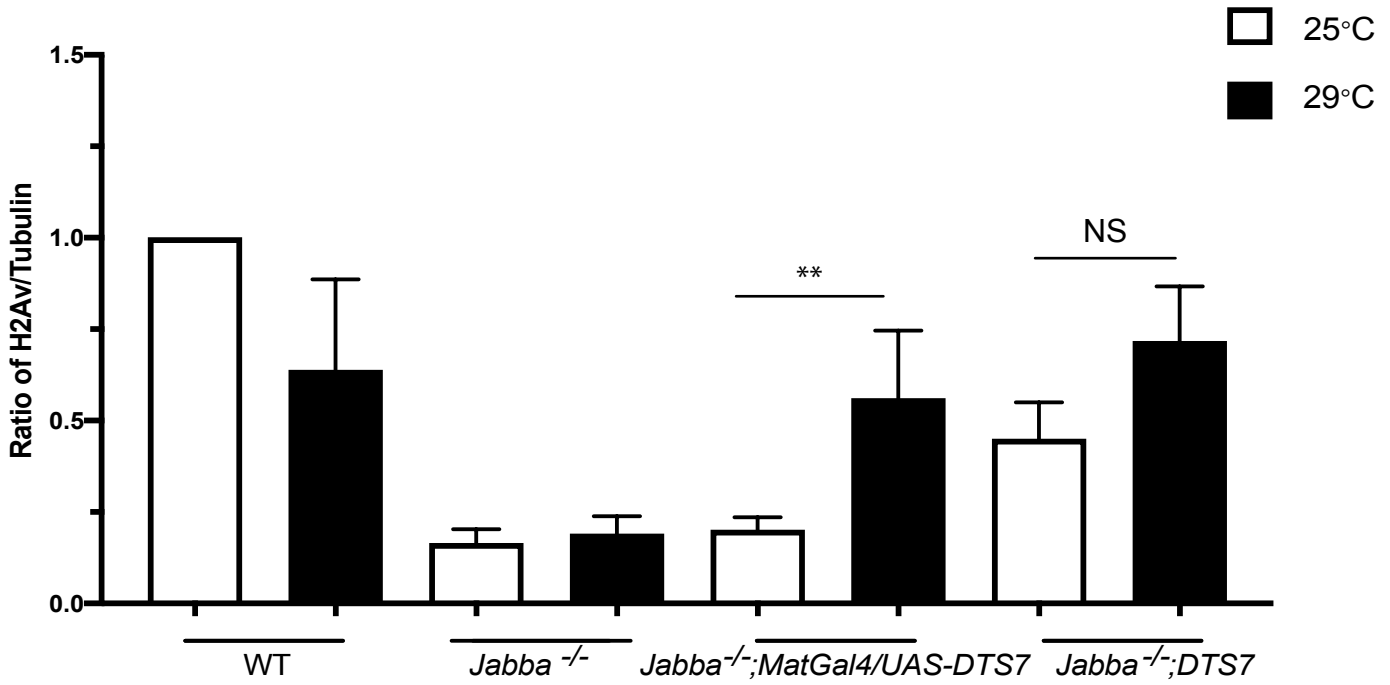
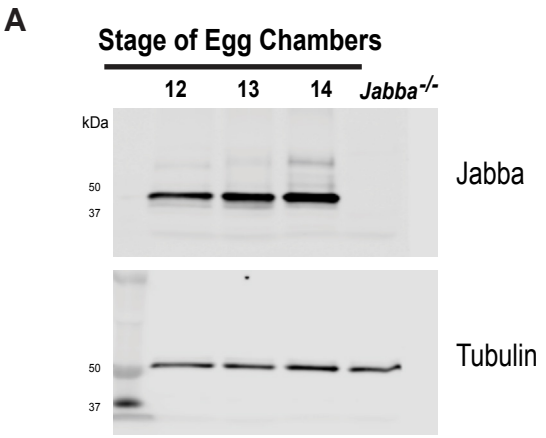
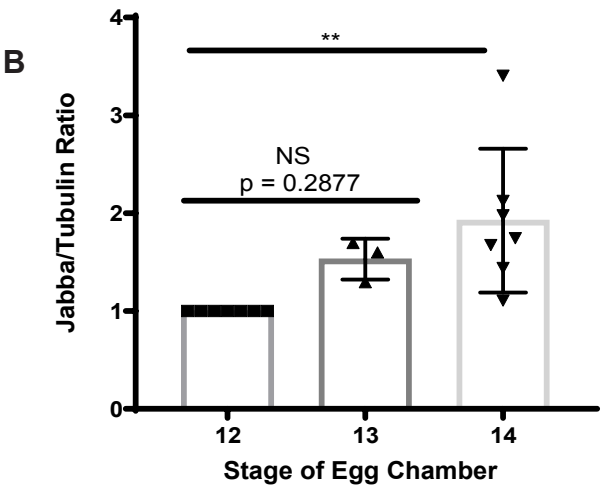
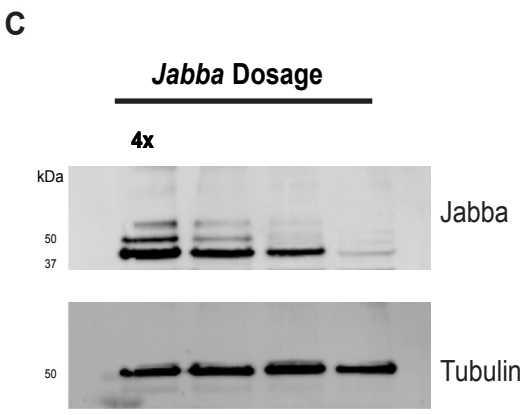
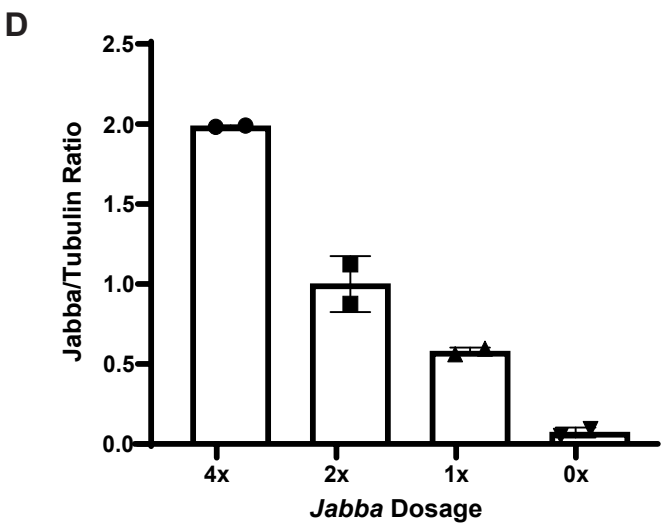


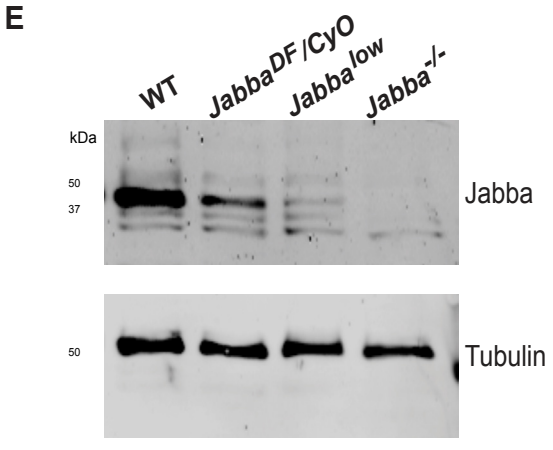
Fig. S5. Quantitation of H2Av levels upon genetic proteasome inhibition. We measured H2Av and tubulin levels (by Western) in S14 oocytes at 25°C and 29°C. H2Av/tubulin ratio was normalized to WT at 25°C. ECs from *Jabba$^{-/-}$; MatGAL4/UAS-DTS7* and *Jabba$^{-/-}$; DTS7* flies kept at 29°C have higher H2Av levels compared to controls. Black: Incubated at 29°C, White: Incubated at 25°C. *Jabba$^{-/-}$; MatGAL4/UAS-DTS7* 25°C vs 29°C, p=0.0073; *Jabba$^{-/-}$; DTS7* 25°C vs 29°C, p=0.1784; a one-way ANOVA followed by Tukey's test. n=5.











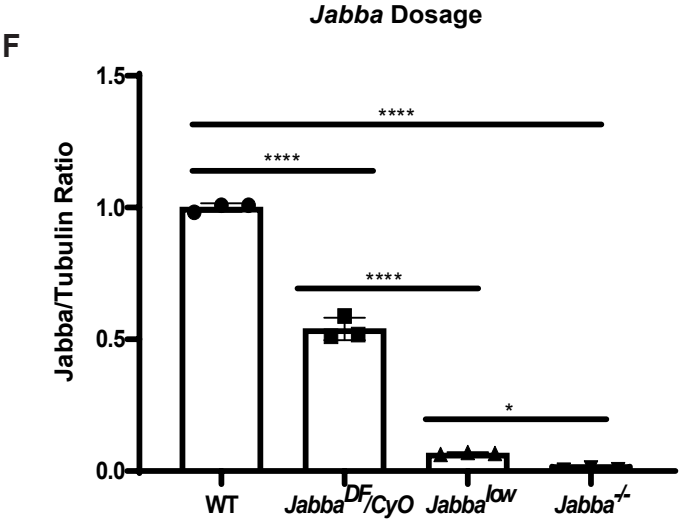


Fig. S6. Analysis of Jabba levels during oogenesis and embryogenesis

(A) Western analysis of Jabba levels in ECs from S12-S14. (B) Quantitation of (A) expressed as the Jabba/tubulin ratio normalized to S12. S12 vs S13, p=0.2877; S12 vs S14, p=0.0085. n=3-7. (C) Jabba expression scales with *Jabba* dosage. Western analysis of Jabba levels in S14 ECs of flies expressing varying *Jabba* dosages. (D) Quantitation of (C) expressed as the Jabba/tubulin ratio normalized to *2x Jabba*. n=2. (E) Anti-Jabba Western of embryos from wild-type and various *Jabba* genotypes. $Jabba^{low}$ expresses low levels of wild-type *Jabba*. (F) Quantitation of (E) expressed as the Jabba/tubulin ratio normalized to wild type. In $Jabba^{DF}/CyO$, Jabba is detected at roughly half the wild type level. Jabba protein is decreased in $Jabba^{low}$. n=3. WT vs $Jabba^{-/-}$, p<0.0001; WT vs $Jabba^{DF}/CyO$, p<0.0001; $Jabba^{DF}/CyO$ vs $Jabba^{low}$, p<0.0001; $Jabba^{low}$ vs $Jabba^{-/-}$, p=0.0485. n=3. Statistical test: one-way ANOVA followed by Tukey's test.

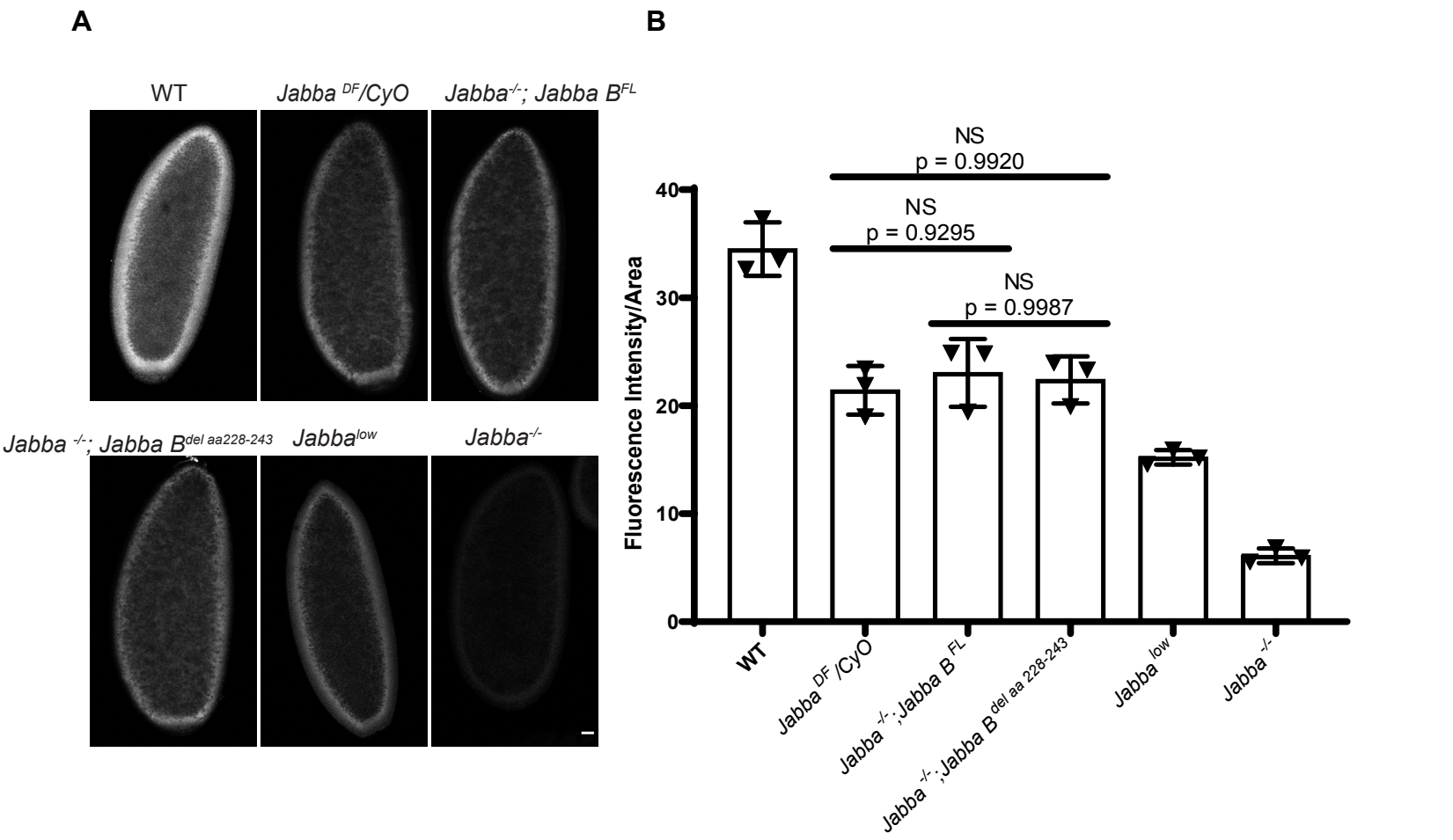


Fig. S7. Jabba B^{FL} and Jabba $B^{del\ aa228\text{-}243}$ expression levels are comparable to that of one copy of Jabba.

(A) Anti-Jabba (white) immunostaining of embryos of *Jabba* genotypes. Scale bar: 25µm. (B) Quantitation of Fluorescence Intensity/Area for (A). The Fluorescence Intensity/Area for *Jabba*$^{-/-}$*; Jabba* B^{FL} and *Jabba*$^{-/-}$*; Jabba* $B^{del\ aa\ 228\text{-}243}$ embryos are comparable to *Jabba*^{DF}*/CyO* embryos (expressing 1x *Jabba*). WT vs *Jabba*$^{-/-}$*; Jabba* B^{FL}, p=0.0003; *Jabba*^{DF}*/CyO* vs *Jabba*$^{-/-}$*; Jabba* B^{FL}, p=0.9295; *Jabba*^{DF}*/CyO* vs *Jabba*$^{-/-}$*; Jabba* $B^{del\ aa228\text{-}243}$; p=0.9920; *Jabba*$^{-/-}$*; Jabba* B^{FL} vs *Jabba*$^{-/-}$*; Jabba* $B^{del\ aa228\text{-}243}$; p=0.9987; WT vs *Jabba*^{DF}*/CyO*, p<0.0001; Statistical test: one-way ANOVA followed by Tukey's test. n=3.

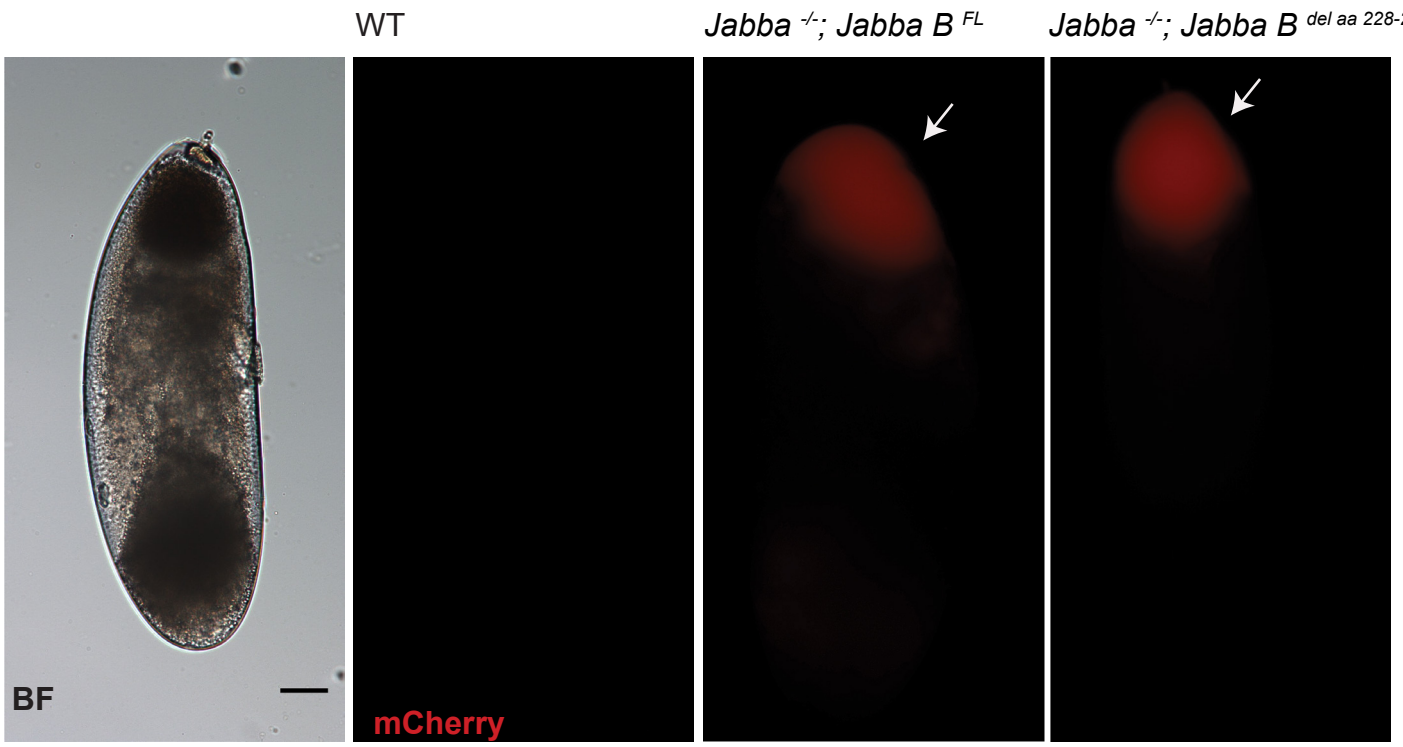


Fig. S8. Jabba B^{FL} and Jabba B$^{del\ aa\ 228-243}$ can associate with LDs.

mCherry (red) is detected in LD layers (arrow) of centrifuged embryos expressing the *Jabba* transgenes, but not in wild-type embryos. BF: Brightfield image of centrifuged embryo. Scale bar: 50µm.

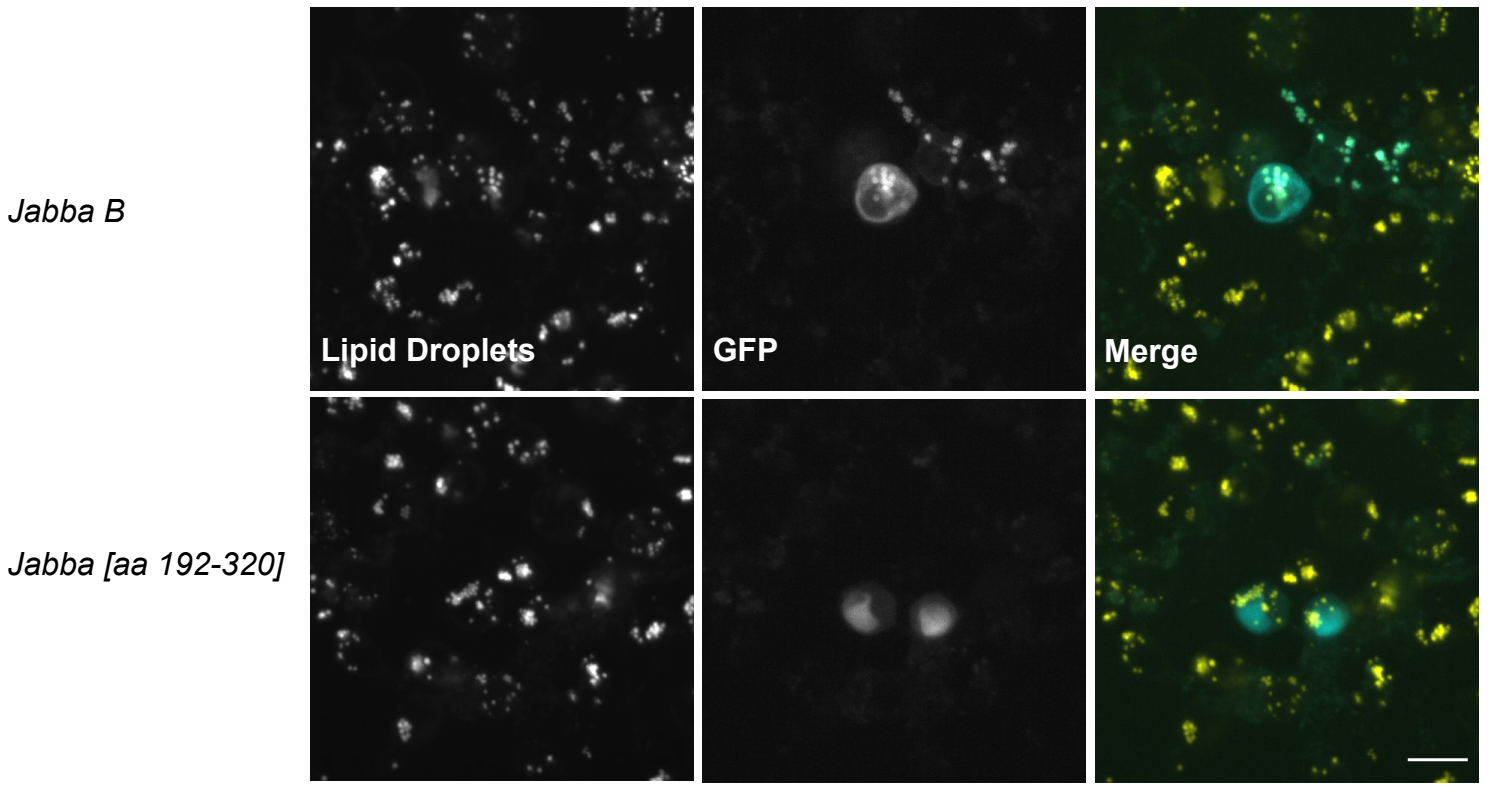


Fig. S9. Localization of Jabba [aa192-321] in S2R+ cells.

Jabba B (cyan, top) is present on LDs (yellow). Jabba [aa192-321] (cyan, bottom) is absent from LDs.

LDs are stained with C12 BODIPY558/568. Scale bar: 10μm

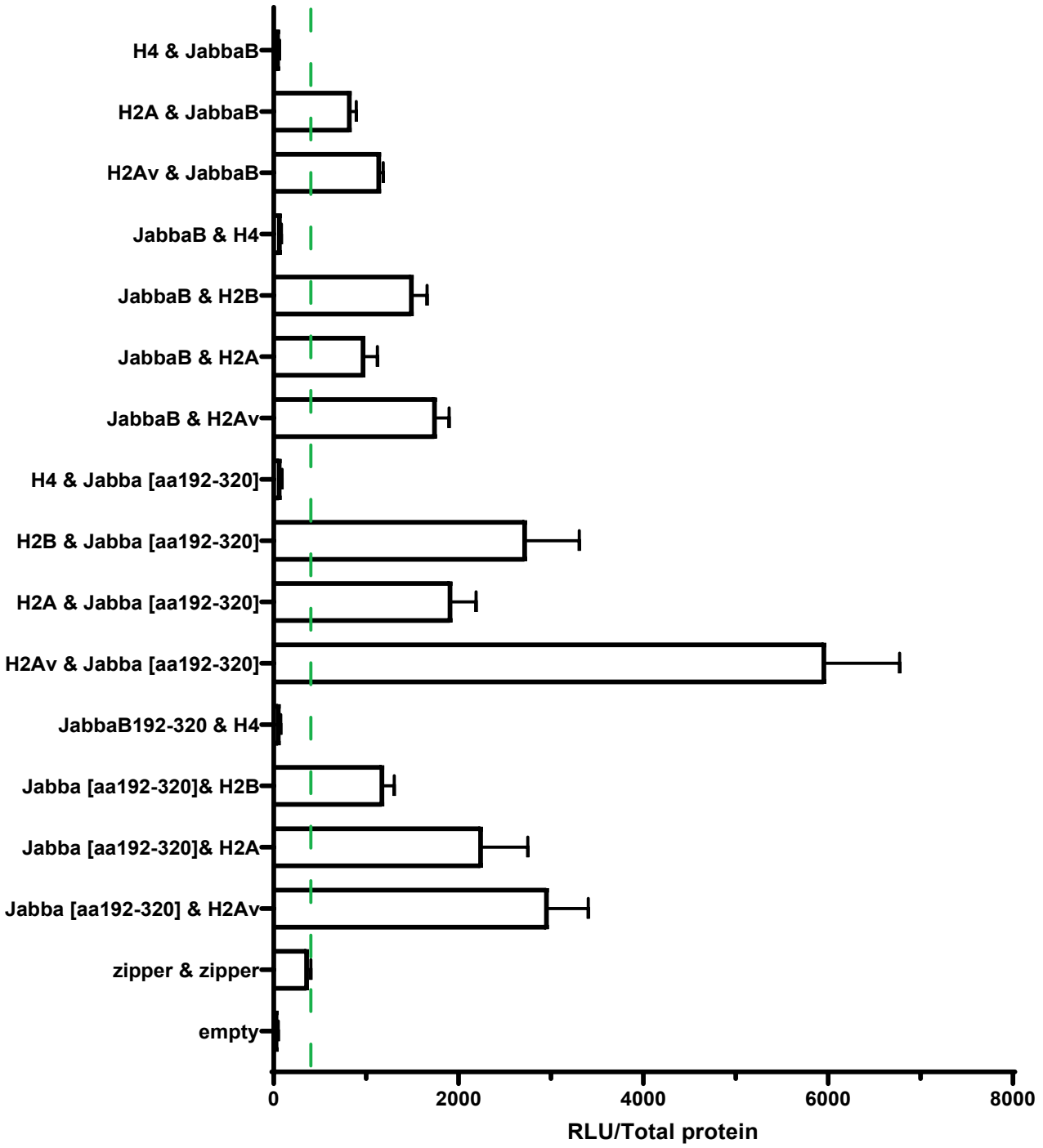


Fig. S10. Split luciferase complementation assay showing results for the indicated proteins.

Luciferase complementation readings are expressed as relative light units (RLU) per µg total protein. Green line: threshold for positive interactions. Fig. 8B contains a subset of these data. n=3. Data are mean±s.d.