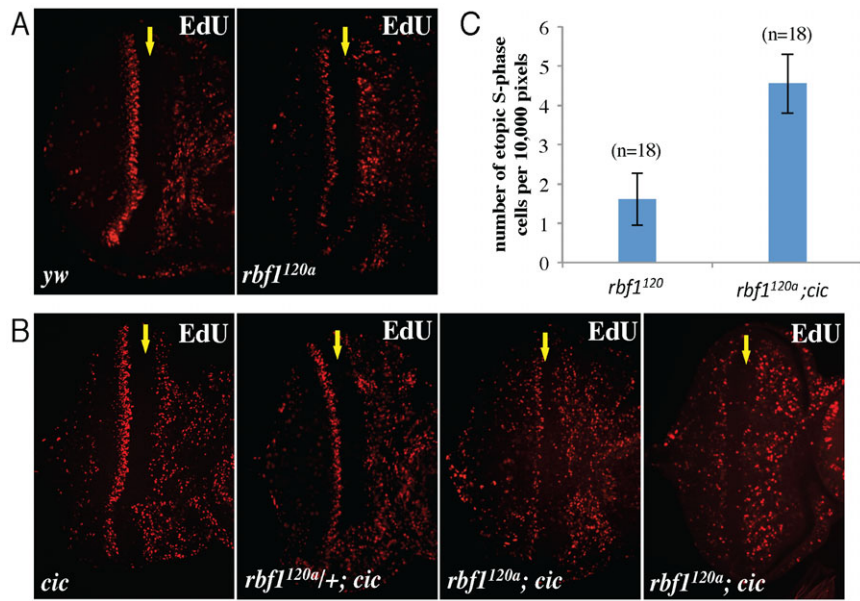
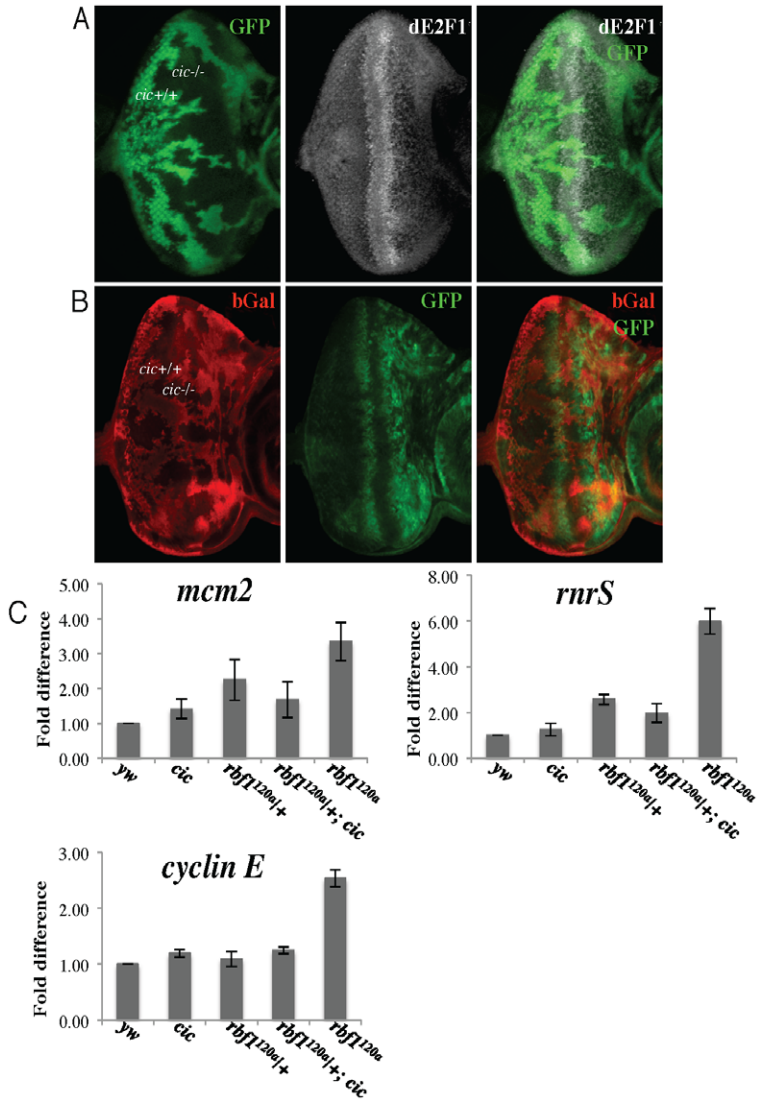


## Supplementary Material

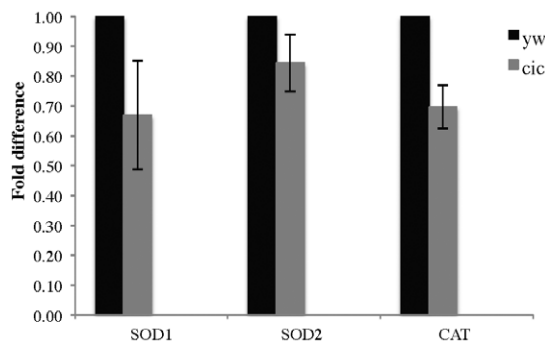
Kate Krivy et al. doi: 10.1242/bio.20123277



**Fig. S1. *cic* mutations cooperate with *rbf1* mutation to promote proliferation.** (A) Control (*yw*) and *rbf1<sup>120a</sup>* eye discs were fluorescently labelled with EdU to mark S-phase cells (red). (B) Eye discs composed entirely of *cic* mutant cells in control, *rbf1<sup>120a</sup>* heterozygous, and *rbf1<sup>120a</sup>* homozygous mutant backgrounds were labelled with EdU (red) to visualise S-phase cells. The position of the MF is indicated by a yellow arrow. Note the abundant presence of ectopic S-phase cells at the MF in the two eye discs composed of *rbf1 cic* double mutant cells. (C) Numbers of ectopic EdU positive cells within the MF of *rbf1<sup>120a</sup>* single and *rbf1 cic* double mutant eyes were counted and normalised by the size of the MF. The MF size was determined by the numbers of pixels that encompass the region between the first and second mitotic waves using the lasso tool and histogram function in Photoshop. Eighteen confocal images taken at the same magnification were used for each genotype. The error bars indicate standard deviation.



**Fig. S2. Cic does not regulate dE2F1 expression.** (A) *cic* mutant clones (absence of GFP) were generated in eye discs using *eyFLP* and immunostained with anti-dE2F1. Note that the dE2F1 expression pattern as well as the level of dE2F1 expression is unaffected by *cic* mutations. (B) *cic* mutant clones (absence of bGal) were generated in eye discs that express GFP under the control of *PCNA* promoter whose activity is regulated by dE2F1. The pattern and level of GFP expression are not affected by the *cic* mutation. (C) Quantitative real-time PCR is used to compare the transcript levels of *mcm2*, *rnrS* and *cyclin E* in control (*yw*) and *cic* mutant eye discs generated in either the control or the *rbf1<sup>120a</sup>* heterozygous (*rbf1<sup>120a</sup>/+*) background. The average fold difference of three independent triplicated experiments is presented. *rbf1<sup>120a</sup>* eye discs are used as a positive control where the expression of E2F target genes is known to be upregulated. The error bars indicate the standard error of three independent experiments.



**Fig. S3. Cic does not regulate scavenger enzyme expression.** Quantitative real-time PCR is used to compare the level of *sod1*, *sod2* and catalase (*cat*) expression in control (*yw*) and *cic* mutant eye discs. The average fold difference of three independent triplicated experiments is presented. The error bars indicate the standard error of three independent experiments.