

Figure S1. The B-boxes are dispensable for Brat-dependent translational repression of *hunchback* mRNA during embryogenesis.

(A-G) Over-expression of *brat* but not *brat*^{hB-boxes} restores the generation of neurons in*brat*null mutant brains. The Brat transgenic protein is detected by the expression of the Myc epitode. The dotted line separates the brain from the optic lobe where both are visible in the optical section. Immature neurons (Dpn Pros⁺). The scale bar, 20 mm.</sup>

(H-L) Over-expression of full-length *brat*, *brat*^{ΔB -boxes} or *brat*^{ΔC -coil} rescues the abdominal cuticular defect in *brat* mutant embryos, but over-expression of *brat*^{ΔNHL} did not.

(M) The frequency of rescuing the abdominal defect in in *brat* mutant embryos by the expression of various *brat* transgenes.





Figure S2. *Apc2* is a specific genetic modifier of the supernumerary neuroblast phenotype in brat mutant brains (A) Quantification of total type II neuroblasts per brain lobe in *brat* mutant brains heterozygous for *Apc2*, *Apc1*, or both *Apc2* and *Apc1*. Error bars indicate s.d.

(B-H) Reduced function of *Apc2* enhanced the supernumerary neuroblast phenotype in *brat* mutant brains. The scale bar: 20 mm.



Figure S3. Defects in asymmetrically segregating Brat do not perturb neuroblast proliferation

(A-F) Neuroblasts over-expressing *brat*^{ΔC -coll} or *brat*^{$\partial NHL}</sup> generate a similar number of progeny as control neuroblasts.$ $(G-H) The total number of INPs or neurons per clone in the larval brain of the genotype indicated. Error bars indicate s.d. Larvae carrying <math>\beta$ -Gal or GFP-marked type II neuroblast lineage clones (outlined in the yellow dotted line) were aged for 48 hours after clone induction, and brains were stained for the markers indicated. Type II neuroblast (Dpn⁺Ase⁻Elav⁻); Ase⁺ immature INP (Dpn⁻Ase⁺Elav⁻); INP (Dpn⁺Ase⁺Elav⁻); immature neurons (Dpn⁻Ase⁻Elav⁺)</sup>





Figure S4. Brat specifies INP identity independently of Numb

(A-C) Over-expression of *brat* efficiently rescues the supernumerary neuroblast phenotype in *brat* mutant clones, but over-expression of *numb* does not.

(D-E) The expression and the asymmetric segregation of Numb is not effected in *brat* mutant brains.

(G-I) Over-expression of *numb* efficiently rescues the supernumerary neuroblast phenotype in *numb* mutant clones, but over-expression of *brat* does not.

(J-K) The expression and the localization of Apc2 is not effected in *numb* mutant clones.

Larvae carrying GFP-positive type II neuroblast mosaic clones (outlined in the yellow dotted line) were aged for 72 hours after clone induction, and brains were stained for the markers indicated.