

Supplementary Table 1 | Ability of *Ilp6* expression in various cell types to reactivate quiescent neuroblasts during nutrient restriction

Cell type	Driver	Reactivation
Mushroom Body neuroblasts/neurons*	<i>OK107-GAL4</i>	+
Dopamine Receptor⁺ neurons	<i>DopR-GAL4</i>	–
Eagle⁺ neuroblasts/neurons	<i>eg-GAL4</i>	–
Ubx-expressing neurons & glia	<i>Ubx-GAL4</i>	++
Ilp6-expressing glia	<i>Ilp6-GAL4</i>	+
Midline glia/neurons	<i>Sim-GAL4</i>	–
Midline glia	<i>Slit-GAL4</i>	–
Subperineurial glia (BBB subset)	<i>Moody-GAL4</i>	–
Cortex glia	<i>NP577-GAL4</i>	++
Ensheathing glia	<i>NP6520-GAL4</i>	–
Glial clones (few & small)	<i>Repo-FLP,tub>>GAL4,UAS-GFP</i>	–
Trachea & glial subset	<i>btl-GAL4</i>	–
Segmentally repeated neuroblasts/ neurons	<i>wg-GAL4</i>	–
Segmentally repeated neuroblasts/ neurons	<i>en-GAL4</i>	+
Fat body	<i>Cg-GAL4</i>	–

* Used as MB driver but not restricted to MB neuroblasts

– No reactivation

+ Reactivation (central brain neuroblasts and few or no thoracic ones)

++ Strong reactivation (including many thoracic neuroblasts and few or no abdominal ones)

BBB Blood-brain barrier

Supplementary Table 2 | Ability of *Ilp* expression in mNSCs, neurons or glia to reactivate quiescent neuroblasts during nutrient restriction

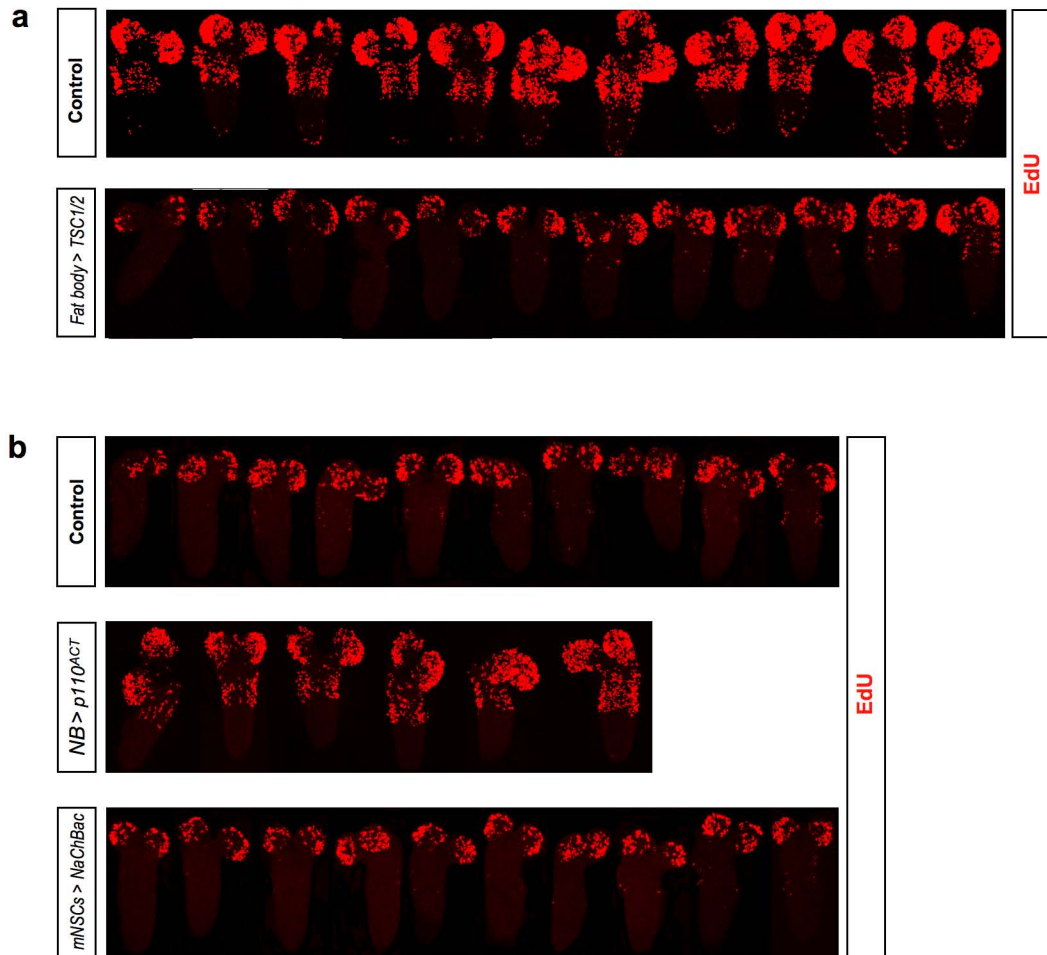
Cell type	Driver	<i>Ilp1</i>	<i>Ilp2</i> *	<i>Ilp3</i>	<i>Ilp4</i>	<i>Ilp5</i>	<i>Ilp6</i>	<i>Ilp7</i>
mNSCs	<i>Ilp2-GAL4</i>	–	–	–	–	–	–	–
Pan neuronal	<i>n-syb-GAL4</i>	–	+	+	++	++	++	–
Pan glial	<i>Repo-GAL4</i>	–	+	–	++	+	++	–

* At 25 °C due to lethality at higher temperatures; for glia, *repo-GAL4* was used with *tub-GAL80^{ts}*

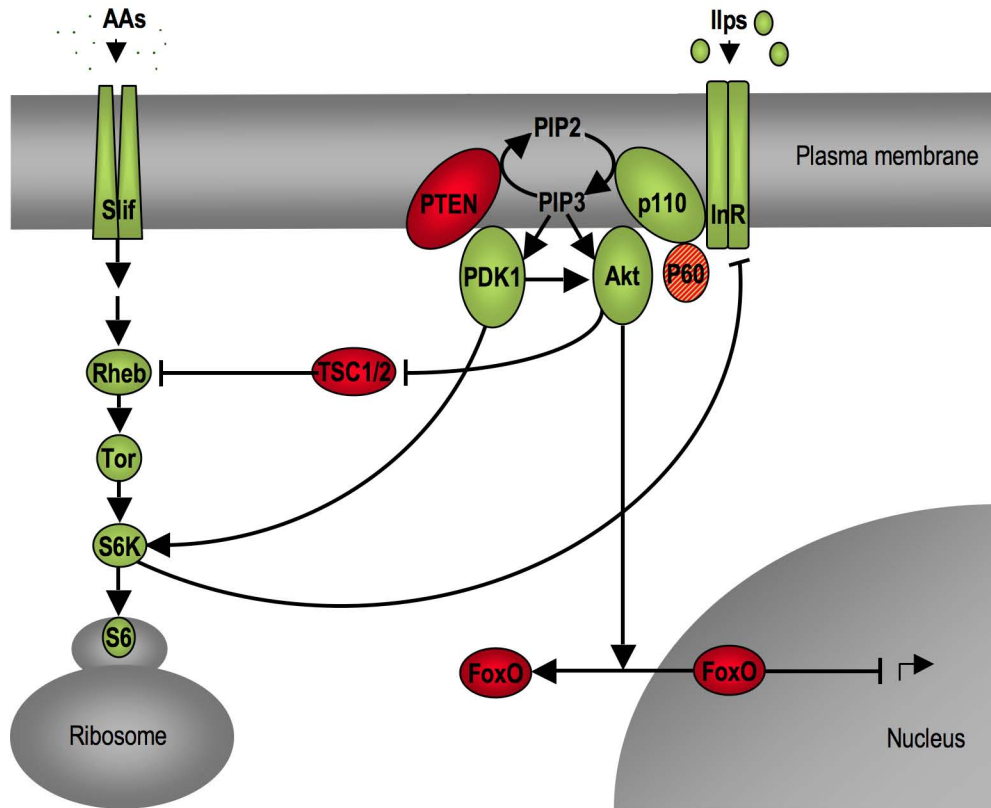
– No reactivation

+ Reactivation (central brain neuroblasts and few or none thoracic ones)

++ Strong reactivation (including many thoracic neuroblasts and few or none abdominal ones)



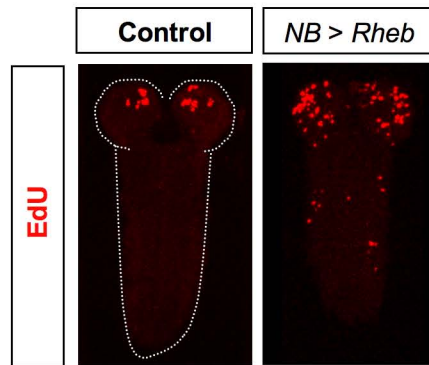
Supplementary Figure 1 | The EdU incorporation assay. **a**, Galleries of larval CNSs comparing a control genotype: *Cg-GAL4/+; TM6B,Sb,Dfd-YFP/+* (*Fat body* >) and a genotype impairing neuroblast reactivation: *Cg-GAL4/+; UAS-TSC1,UAS-TSC2/+* (*Fat body* > *TSC1/2*). **b**, Galleries of larval CNSs comparing a control genotype: *Ilp2-GAL4/CyO,Dfd-YFP*, a genotype resulting in precocious neuroblast reactivation: *nab-GAL4; UAS-p110^{ACT}* (*NB* > *p110^{ACT}*), and a genotype with no significant effect on reactivation: *Ilp2-GAL4/+; UAS-NaChBac/+* (*mNSCs* > *NaChBac*). EdU⁺/Repo⁺ double-positive cells represent less than 6% of total EdU⁺ cells in this study (data not shown).



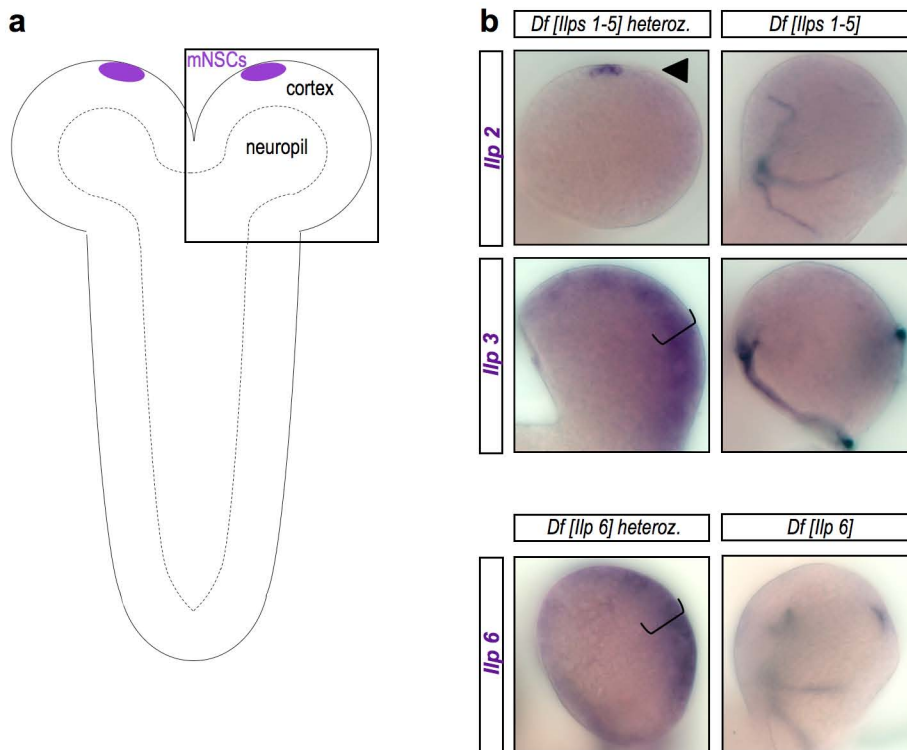
Positive
 Negative

} Regulators of the pathway and of growth and proliferation

Supplementary Figure 2 | The Tor and PI3K signalling network. Simplified schematic of the interconnected Slif/TOR and InR/PI3K pathways in *Drosophila*, depicting the proteins genetically manipulated in this study. AA, aminoacids; Slif, Slimfast; TSC1/2, Tuberous Sclerosis Complex 1 and 2; Rheb, Ras homologue enriched in brain; S6K, S6 Kinase; S6, Ribosomal protein small subunit 6; Ilps, Insulin-like peptides; InR, Insulin receptor; PIP2, Phosphatidylinositol (4,5)-bisphosphate; PIP3, Phosphatidylinositol (3,4,5)-triphosphate; p110, catalytic subunit of the phosphatidylinositol 3-kinase (PI3K); p60, adaptor subunit of PI3K (green and red hatching as it acts as a dominant-negative when overexpressed); PTEN, Phosphatase and Tensin homologue; PDK1, Phosphoinositide-dependent kinase 1; Akt, AKR mouse T-cell lymphoma-inducing Serine-Threonine kinase; FoxO, Forkhead box subgroup O transcription factor.



Supplementary Figure 3 | Hyperactivation of TOR signalling increases neural proliferation at larval hatching. EdU-labelled CNSs from newly hatched L1 larvae showing that Rheb overexpression in neuroblasts (*NB > Rheb*) inhibits early larval quiescence. The neuroblast driver used (*nab-GAL4*) is only expressed from late embryonic stage 14 onwards.



Supplementary Figure 4 | *Ilp2*, *Ilp3* and *Ilp6* expression in the early L2 CNS. **a**, Schematic of early larval CNS, highlighting the cortex (containing most soma) and neuropil (containing most axons), the position of mNSCs and the brain-lobe region shown stained in **b** (square inset) are indicated. **b**, Panels show *in situ* hybridizations for *Ilp2*, *Ilp3* or *Ilp6* mRNA in the brain lobes of early L2 larvae heterozygous (left) or homozygous (right) for a deficiency removing the *Ilp1-5* gene cluster, *Df*[*Ilps1-5*] or removing *Ilp6*, *Df*[*Ilp6*]. Brackets highlight the cortex expressing *Ilp 3* and *Ilp 6*. Note that the strong background staining (seen in heterozygotes and homozygotes) corresponds to tracheal branches associated with the CNS.