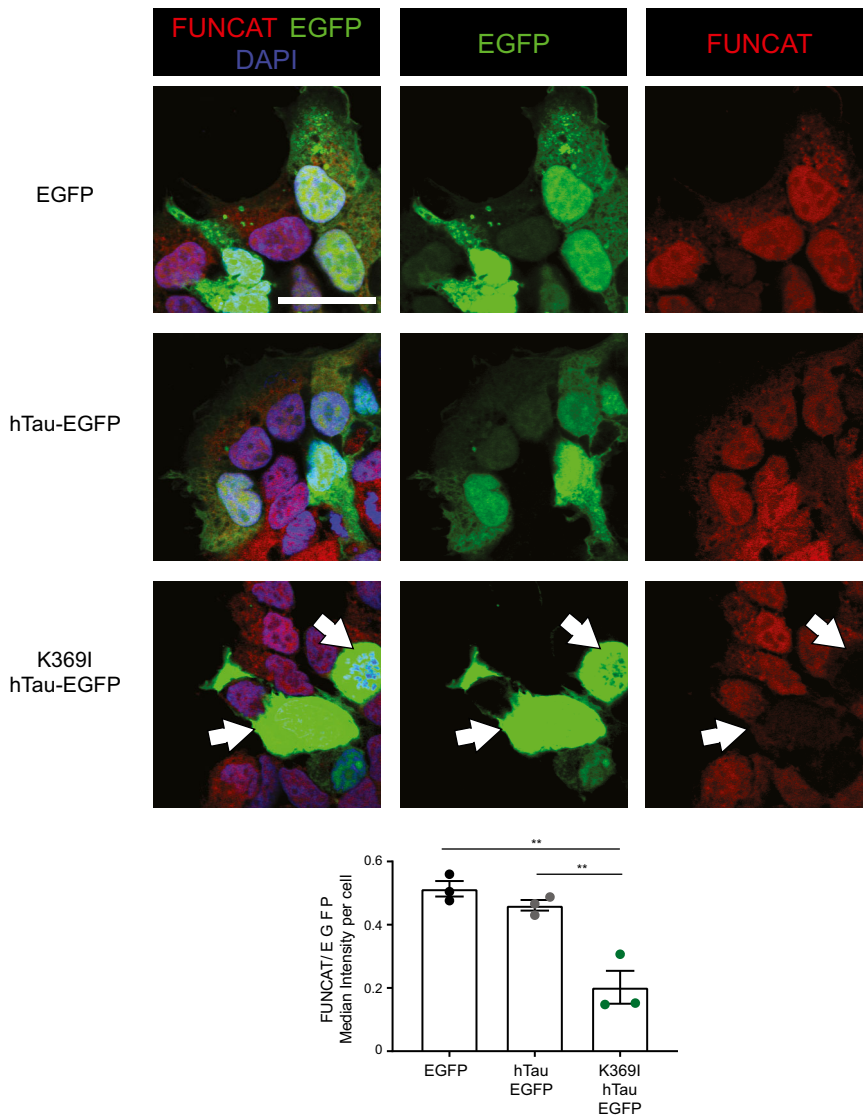


## Expanded View Figures

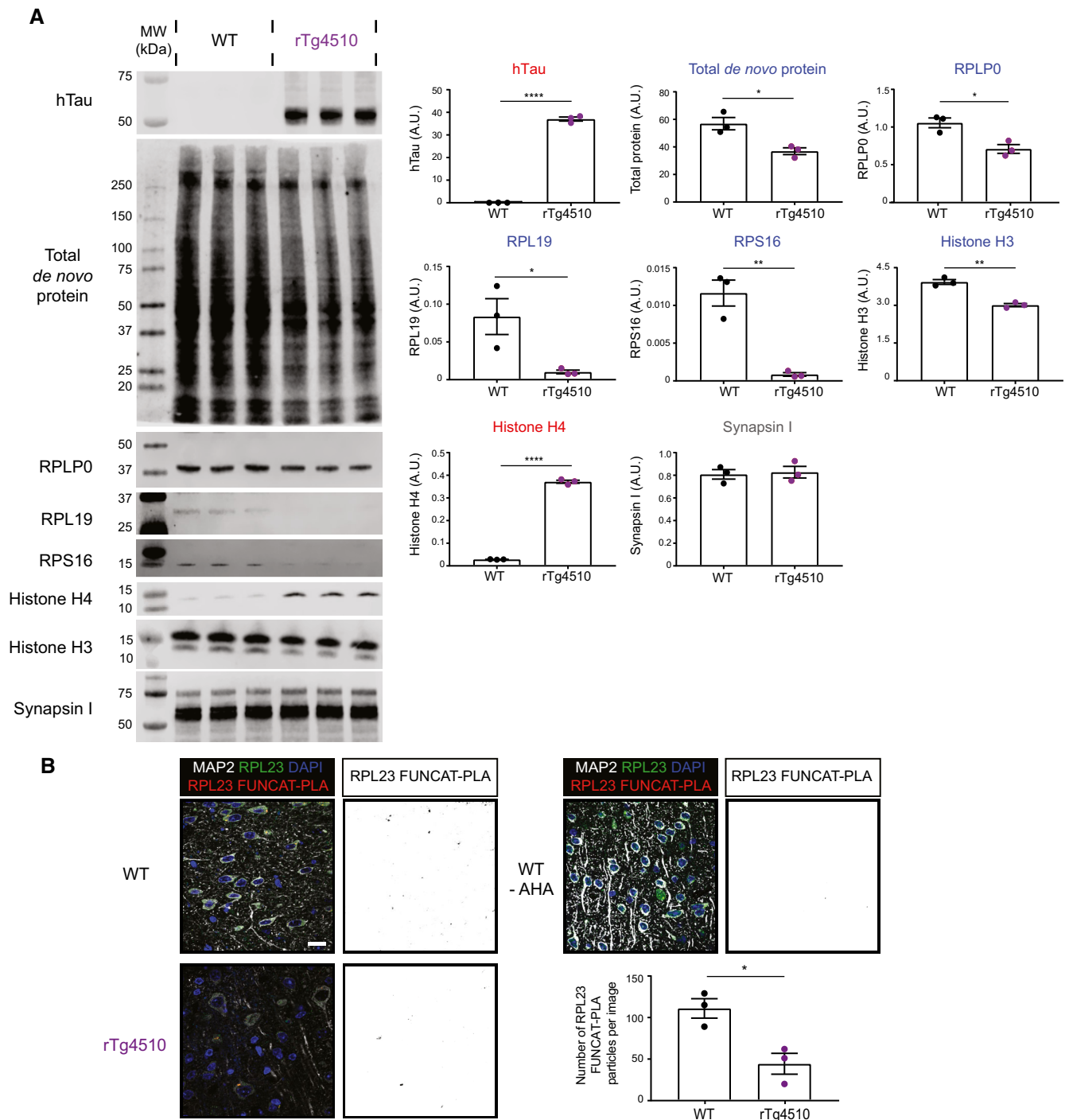
**Figure EV1. HEK293 cells transfected with K369I-hTau-EGFP have significantly decreased protein synthesis compared with EGFP or hTau-EGFP expressing cells.**

After treatment with AHA for 4 h, HEK293 cells transfected with K369I-hTau (1N4R)-EGFP showed a significantly lower FUNCAT signal normalised to the EGFP signal than cells transfected with hTau (1N4R)-EGFP or the EGFP vector alone, suggesting that protein synthesis is decreased in the mutant cells (one-way ANOVA, Tukey's multiple comparison test,  $n = 3$  replicates). Cells with K369I-hTau-EGFP and low FUNCAT signal are indicated by white filled arrows.

Data information: Mean  $\pm$  SEM.  $**P \leq 0.01$ ,  $***P \leq 0.001$ . Scale bar: 40  $\mu$ m.







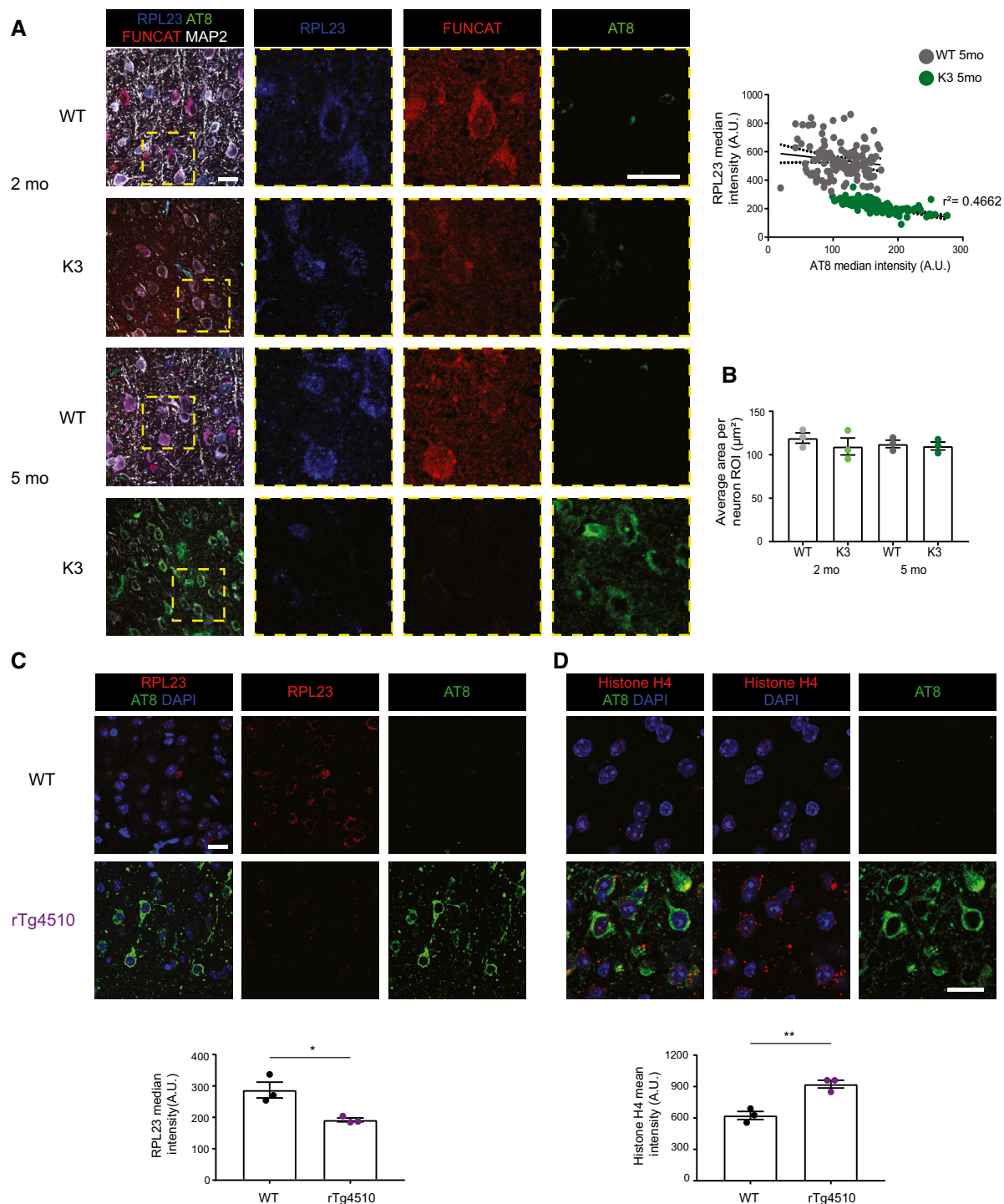
**Figure EV3. The synthesis of selected ribosomal subunits is decreased in rTg4510.**

A Western blot analysis of *de novo* synthesised proteins reveals altered protein synthesis in rTg4510 mice compared with WT controls. AHA-labelled proteins from rTg4510 and WT mice were purified using BONCAT and probed via Western blotting. The synthesis of the ribosomal proteins RPLP0, RPL19 and RPS16, as well as histone H3, was significantly decreased in rTg4510 mice, whereas that of histone H4 was increased, and that of synapsin I was unaltered (Student's *t*-test,  $n = 3$  animals).

B Synthesis of RPL23 is decreased in rTg4510 mice. Compared to WT mice, significantly lower numbers of RPL23 FUNCAT-PLA punctae are detected in layer 2/3 cortical neurons of rTg4510 mice (Student's *t*-test,  $n = 3$  animals). A PBS-treated WT control is also shown as a negative control.

Data information: Mean  $\pm$  SEM. \* $P \leq 0.05$ , \*\* $P \leq 0.01$ , \*\*\*\* $P \leq 0.0001$ . Scale bar: 40  $\mu$ m.

Source data are available online for this figure.



**Figure EV4. Total abundance of RPL23 and histone H4 is altered in K3 and rTg4510 mice.**

A RPL23 levels are negatively correlated with AT8 phospho-tau in the cortex of 5-month-old K3 mice (Pearson's correlation,  $r^2 = 0.4662$ , standard error of correlation represented by dotted black lines,  $n = 3$  animals,  $\geq 30$  neurons per animal). AT8-positive neurons were not detected in 2-month-old K3 mice.

B The average area of regions of interest drawn around individual neurons using MAP2 labelling does not differ between K3 and WT mice (two-way ANOVA, Sidak's multiple comparison test,  $n = 3$  animals,  $\geq 30$  neurons per animal).

C The total abundance of RPL23 is decreased in rTg4510 mice compared with WT controls (Student's  $t$ -test,  $n = 3$  animals).

D Histone H4 levels are increased in layer 2/3 of the cortex of rTg4510 mice compared with WT mice (Student's  $t$ -test,  $n = 3$  animals).

Data information: Mean  $\pm$  SEM. \* $P \leq 0.05$ , \*\* $P \leq 0.01$ . Scale bar: 40  $\mu\text{m}$ .