Supplementary Materials

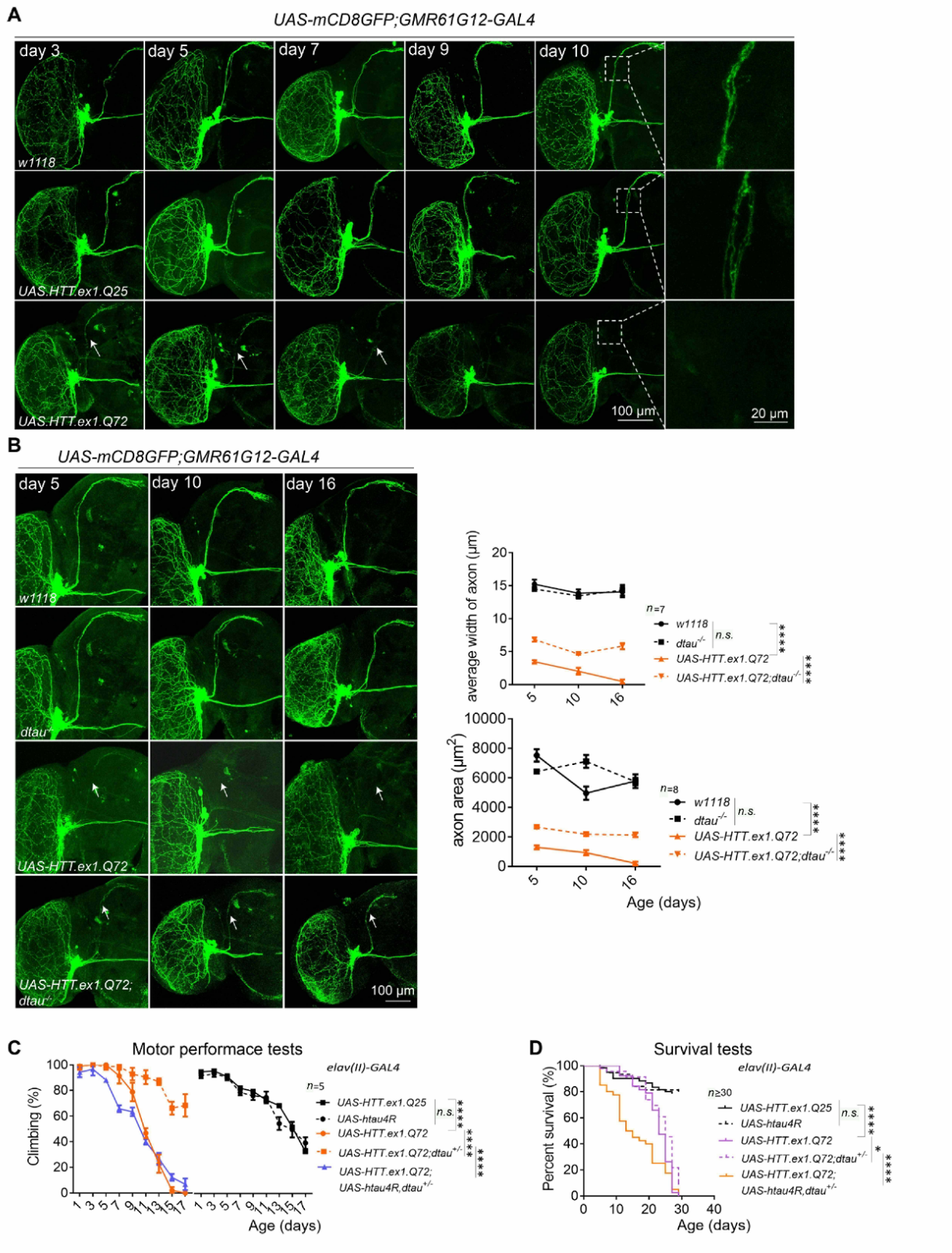

Fig. S1 ***Homozygous or heterozygous dtau-knockout*** **rescues neurodegeneration, motor function deficits, and shortened lifespan in HD flies.**

A Representative immunofluorescence images of whole-mount brains of flies of indicated genotypes at indicated ages showing *in vivo* neurodegeneration. Small ventral lateral clock neurons are labeled by mCD8GFP protein driven by *GMR61G12-GAL4*. HTT.ex1.Q72 or HTT.ex1.Q25 expression is also driven by the same *GAL4*. Arrows indicate a major axon bundle that degenerated in flies expressing HTT.ex1.Q72, compared to those expressing HTT.ex1.Q25 or w1118 control flies. Scale bars, 100 μm and 20 μm.

B As in **A**), but with or without *dtau* knockout (KO) as indicated. Right: quantification (mean and SEM) of axon bundle width and total axon area based on the GFP images (n, number of individual flies; statistical analysis, two-way ANOVA and Turkey's post-hoc test; scale bar, 100 μm.

C Motor performance tests measuring the climbing ability of flies with indicated genotypes and ages. *(n*, number of independently tested vials, each containing 15 virgin females (statistical analysis, two-way ANOVA and Turkey's post-hoc test).

D Lifespan measurements of flies with indicated genotypes. *(n*, number of individual flies; statistical analysis, log-rank tests).

In all plots in **A**–**C**, error bars indicate the mean ± SEM; *n.s.* $P > 0.05$, *$P \leq 0.05$, ****$P \leq 0.0001$.

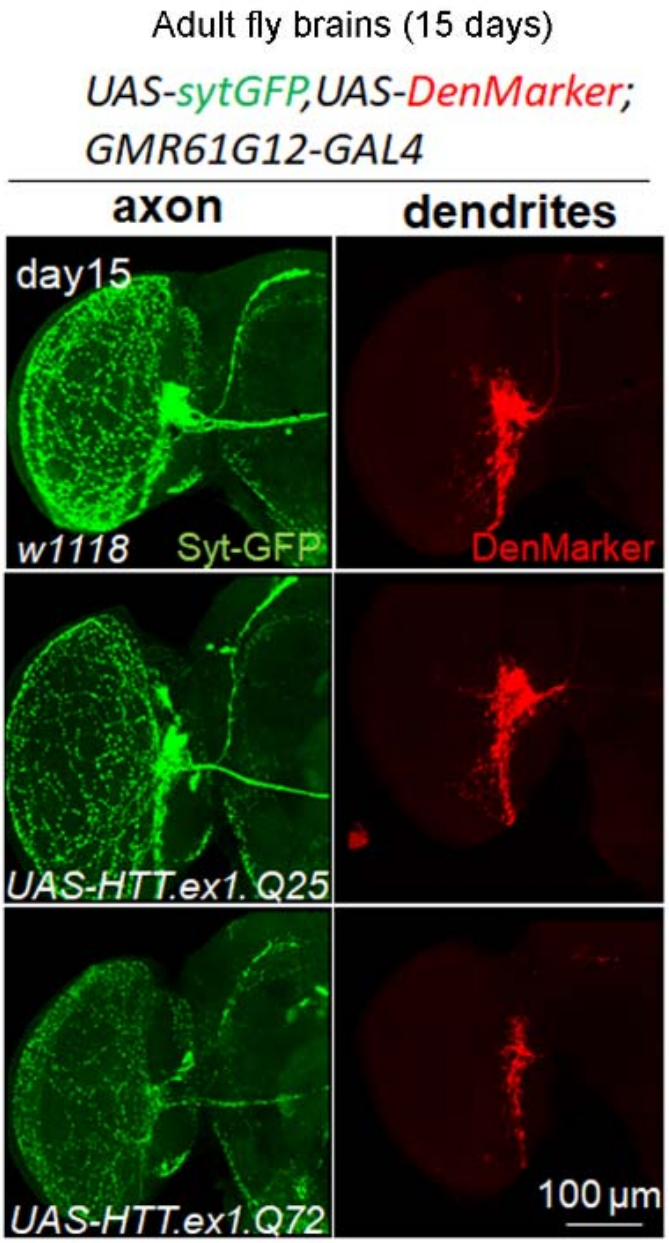

Fig. S2 Validation of neurodegeneration detection by axon and dendrite markers.

Representative immunofluorescence images of whole-mount brains from *Drosophila* of indicated genotypes. The expression of axon marker (syt.eGFP) or the dendrite marker (DenMarker) were driven by *GMR61G12-GAL4* in the sLNv clock neurons with HTT.ex1.Q25 or HTT.ex1.Q72 expression, or in the wild-type background (w1118).

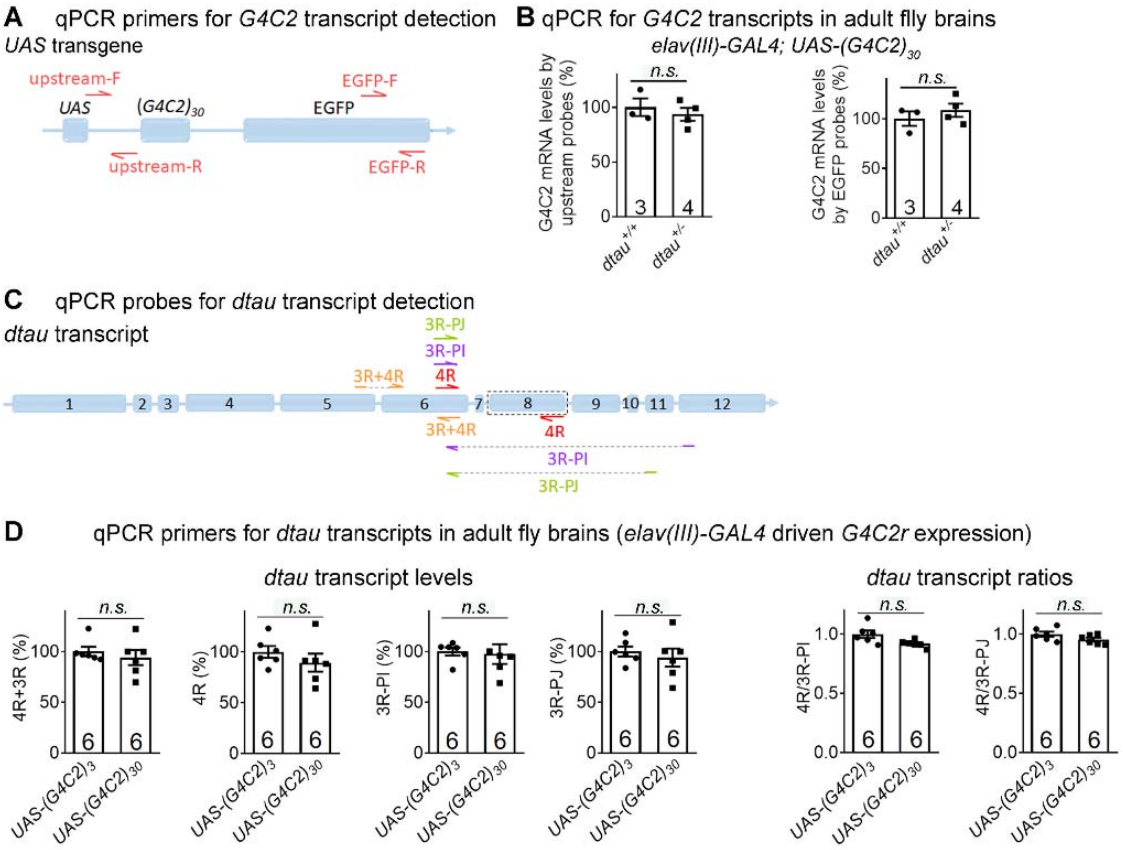

Fig. S3 dtau and expanded *G4C2r* do not influence each other's mRNA levels.

A Schematic of the qPCR primers for measurement of *(G4C2)30* transcript levels.

B qPCR measurements of *(G4C2)30* transcripts in brain tissues from flies of indicated genotypes using the indicated qPCR primers (*n*, number of individual flies; statistical analysis, two-tailed unpaired t test).

C Schematic of the qPCR primers for measurement of endogenous *dtau* transcript levels. Different *dtau* transcripts were detected by different sets of primers.

D qPCR measurements of the indicated *dtau* transcripts in brains from flies of the indicated genotypes (statistical analysis, two-tailed unpaired t test).

In all plots, bars indicate the mean ± SEM; $*P \leq 0.05$, $**P \leq 0.01$, $***P \leq 0.001$, $****P \leq 0.0001$.

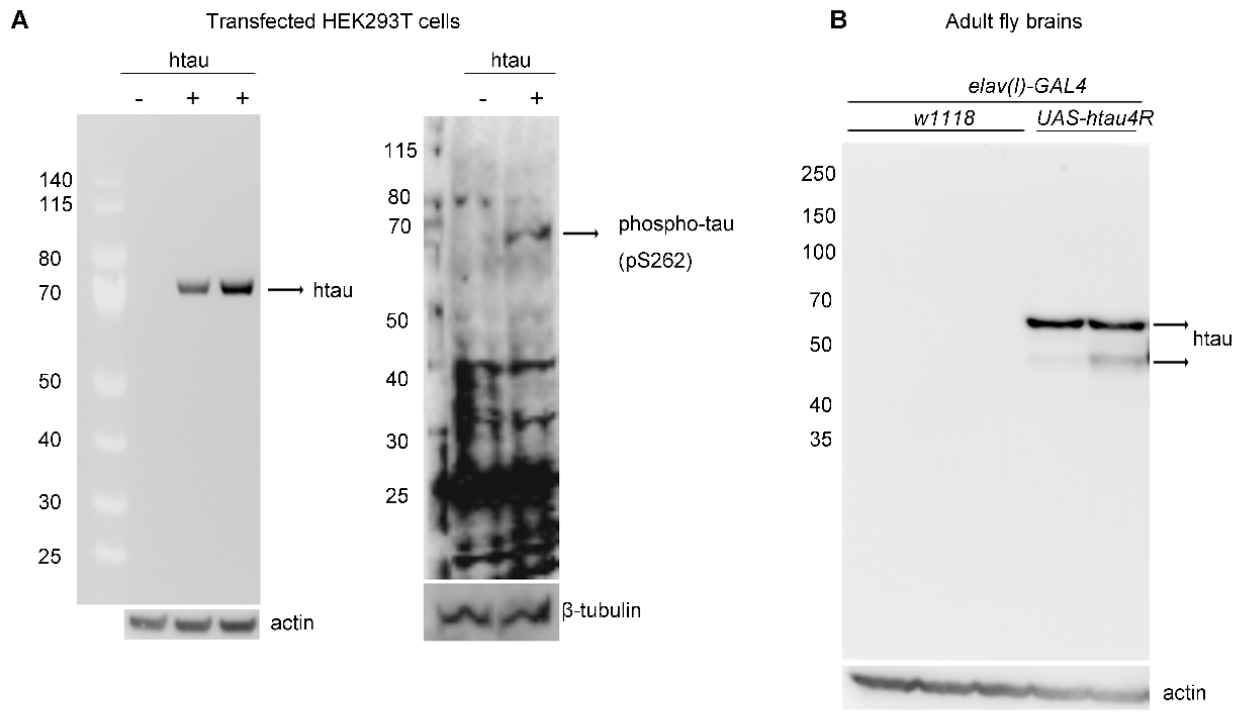

Fig. S4 Validation of specificity of major antibodies.

A, **B** Representative western blots of HEK293T and fly samples demonstrating the specificity of different tau antibodies (arrows, specific tau or phospho-tau bands). The specificity of AT8 and pS199 is shown in Fig. 3**A**, and thus is not presented here.