

Figure S1. Tau P301L expression did not affect cell viability. HEK T-Rex cells were treated with 1.0 $\mu\text{g/mL}$ doxycycline to induce the expression of human tau P301L (Induced) or with the same volume of 10 mM PBS, pH 7.4 (Not-induced). Cell viability was recorded 1, 3, and 5 days after treatment, with (a) MTT, (b) LDH, and (c) Alamar blue assays. Data are expressed as values normalized on the mean of Not-induced cells from three technical replicates of three independent experiments.

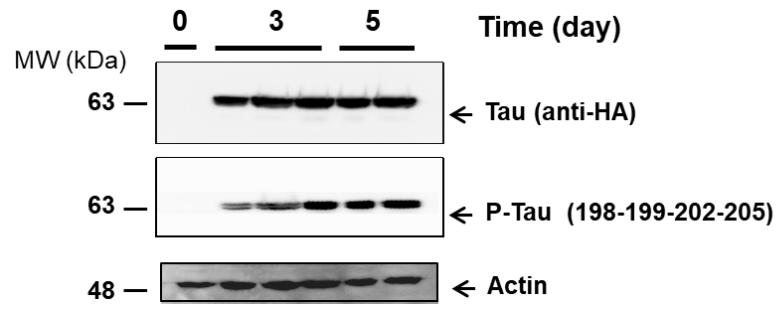


Figure S2. Tau P301L expression in HEK T-REx cells. Representative Western blot of total tau and phosphorylated tau (P-Tau) in lysates of HEK T-REx cells collected before (time 0) or 3 and 5 days after treatment with doxycycline. An equal amount of proteins were loaded in each gel lane (30 μ g) and immunoblotted with an anti-HA tag, anti-P-tau (198-199-202-205), or anti-actin antibody.

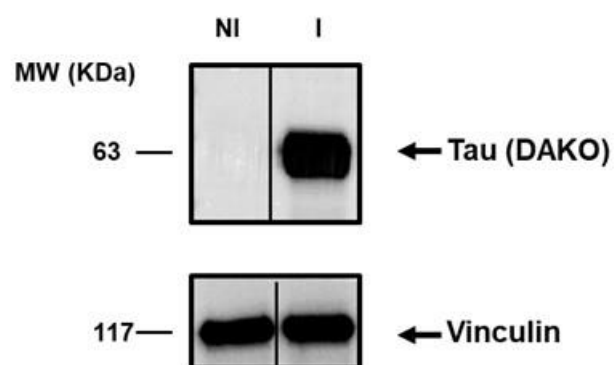


Figure S3. Western blot of cell lysates before immunoprecipitation and used for the input.

Cell lysates were prepared from HEK T-REx cells induced (I) or Not-induced (NI) for 5 days to express tau P301L. An equal amount of proteins (15 μ g) was loaded in each lane of gel and immunoblotted with an anti-tau antibody (DAKO) or anti-vinculin antibody.

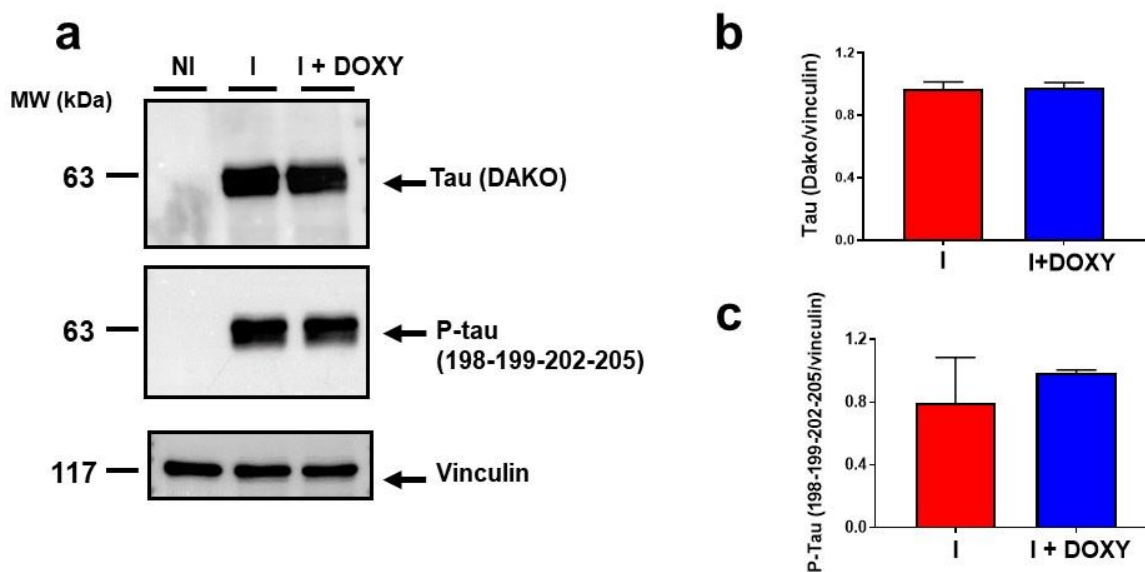


Figure S4. Effect of Doxy on tau expression. Representative Western blot of (a) total tau and phosphorylated tau (P-Tau) in lysates of HEK T-REx cells induced or not-induced (NI) for 5 days to express Tau_{P301L}. Cell lysates of Induced cells (30 µg) were incubated for 2 h with 50 µM Doxy (I+ DOXY) or the same volume of 10 mM PBS, pH 7.4 (I). Equal amounts of proteins (30 µg) were loaded in each gel lane and immunoblotted with (a) anti-tau antibody (DAKO), anti-P-tau (198-199-202-205) antibody, or anti-vinculin antibody. (b) Total tau and (c) P-tau quantification is expressed as the mean volume of the immunoreactive band/vinculin. Data are mean ± SD (N=3).