

Assay	Figure	Cell type	Transfection/ Transduction	Time point(s)
β protein aggregation	Fig. 1A-B Fig. S1A-D	Neurons Neurons	Transfection Transduction	DIV 10+1 DIV 10+3, 10+4, 10+6
β protein toxicity	Fig. 1C-D, S5B-C Fig. S1E	Neurons Neurons	Transfection Transduction	DIV 10+1, 10+3 DIV 10+4 to 10+14
Sholl analysis	Fig. 1E-F	Neurons	Transfection	DIV 10+2
Cryo-ET	Fig. 2, 3, S3, S4 Fig. S3	Neurons HeLa	Transfection Transfection	DIV 6+1 24 hours after transfection
Lysotracker	Fig. 4 Fig. 8E-F	Neurons Mocha cells	Transfection None	DIV 6+1 Day 1 after plating
LC3 +/- chloroquine	Fig. 5A-C	HeLa	Transfection	48 hours after transfection
mCherry-GFP-LC3 reporter	Fig. 5D-G	Neurons	Transfection	DIV 10+3
Western blot, RT-PCR for autophagy markers	Fig. 5H-J	HeLa	Transfection	24 hours after transfection
β protein interactome and total proteome	Fig. 6A-H, S6, S7A-D, F	Neurons	Transduction	DIV 10+4
AP-3 μ 1 / β protein colocalization	Fig. 7A-B	Neurons	Transfection	DIV 10+2
Western blots for AP-3 μ 1 in soluble and pellet fraction	Fig. 7C-E	Neurons	Transduction	DIV 10+6
TauRD seeding	Fig. 7F-J	Neurons	Transduction	DIV 10+7
Western blot for other AP-3 subunits	Fig. 8A-B	Neurons	Transduction	DIV 10+6
β protein / AP-3 μ 1 co-IP	Fig. S7E	Neurons	Transduction	DIV 10+7
Lamp1 localization	Fig. 8C-D, S7G	Neurons	Transduction	DIV 10+4, 10+5
AP-3 subunits in mocha cells	Fig. S8A-C	Mocha cells	None	Day 1 after plating
Toxicity rescue by AP-3 μ 1	Fig. S8D-E	Neurons	Transfection	DIV 6+1

Data analyzed	Cell type	Condition	Number of experiments	Number of cells	Number of observations
Aggregates	Neurons	β 4-mCherry	3	9	21
	Neurons	β 23-mCherry	2	2	4
	HeLa	β 23-mCherry	4	7	23
Lysosomes analyzed for aggregate content	Neurons	β 4-mCherry	5	10	38
	Neurons	β 23-mCherry	3	5	31
Early autophagosomes	Neurons	mCherry	5	14	1
	Neurons	β 4-mCherry	5	15	2
	Neurons	β 23-mCherry	4	9	0