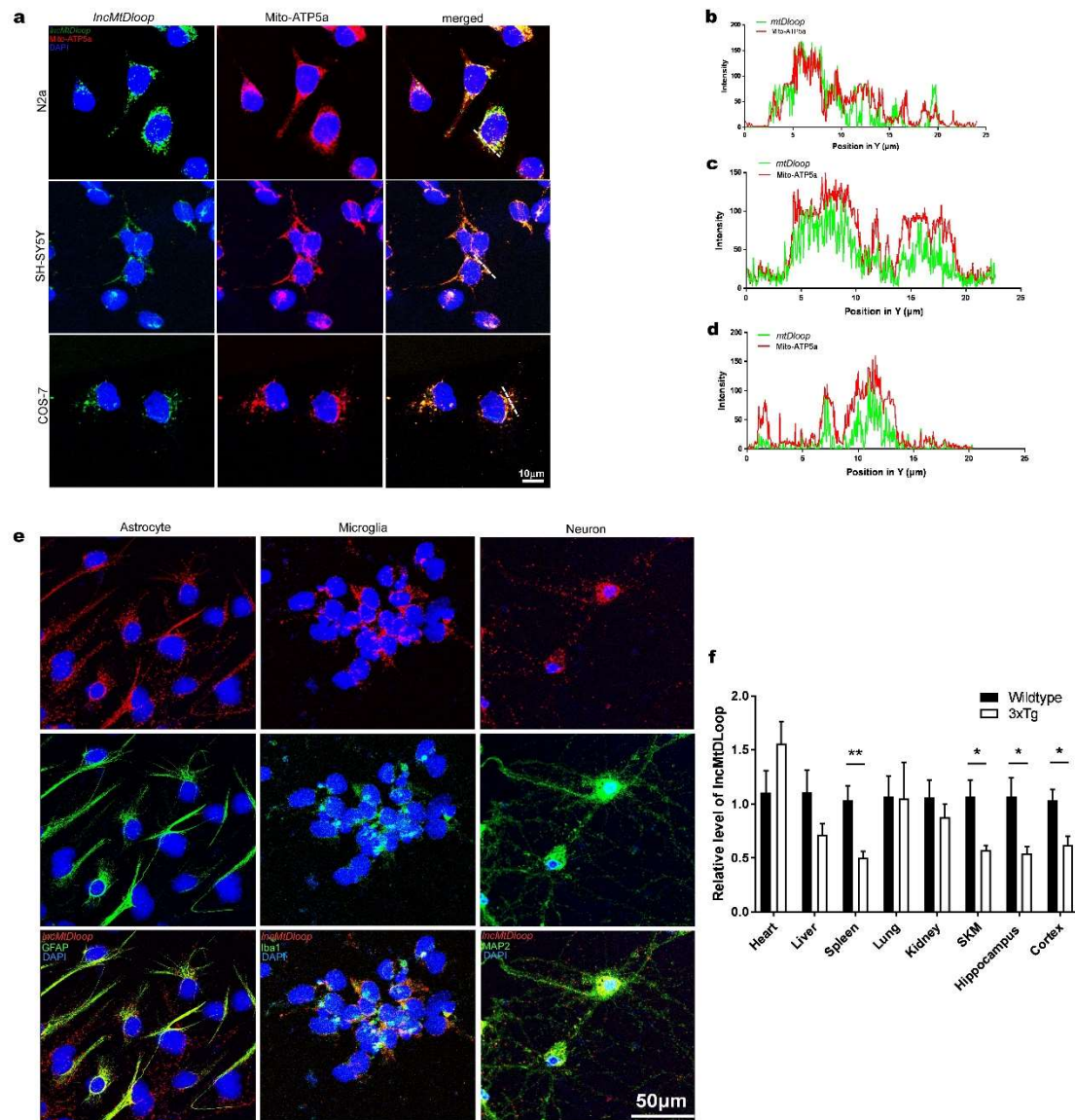
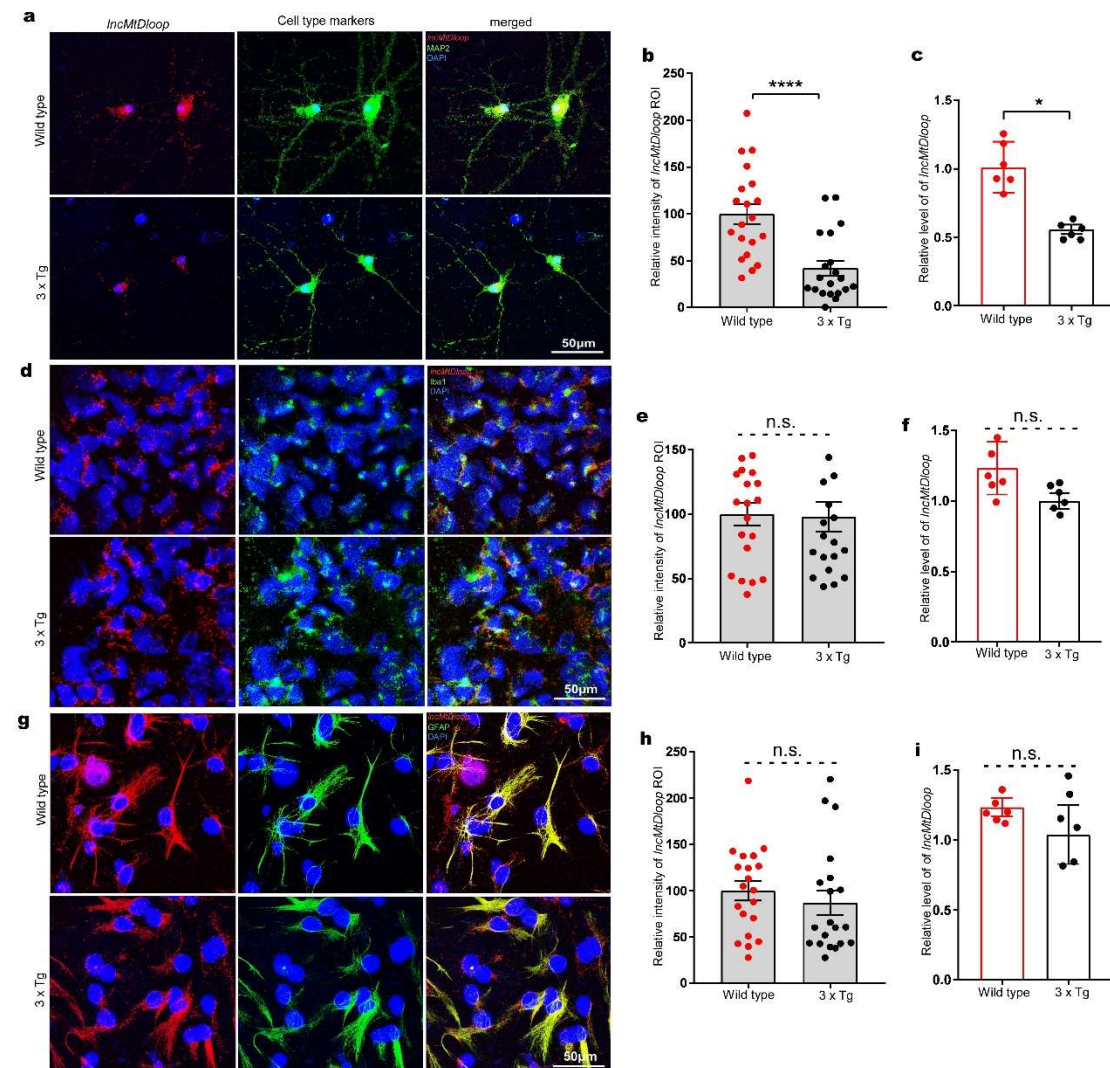


Supplementary Figures



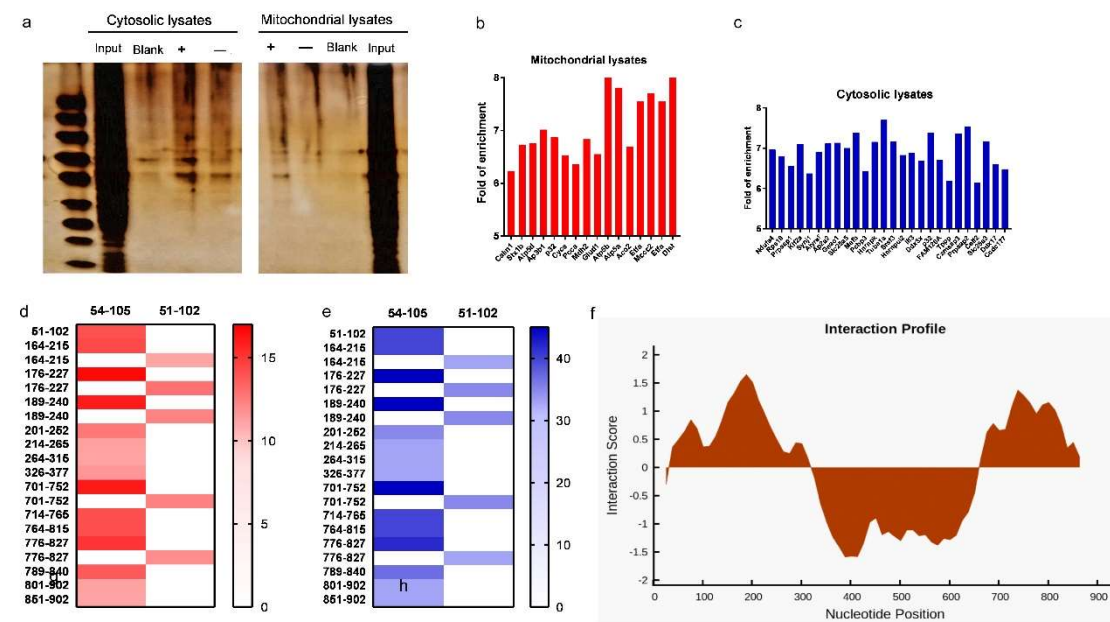
Supplementary Fig. 1 | *LncMtDloop* universally and differentially expresses in distinct cell types. (a) Representative images of RNAscope ISH and immunostaining showing the location of *LncMtDloop* (green) and mitochondria (red) in different cell lines, including N2a (mouse), SH-SY5Y (human) and Cos-7 (monkey). (b-d) Relative intensities of fluorescence signals of *LncMtDloop* and mitochondria markers illustrated in white dotted line in E) were quantified by Image J. (e) Representative images of RNAscope ISH and immunostaining showing the *LncMtDloop* expression (red) in different neural cell lines, including astrocytes (GFAP, green), microglia (Iba1, green) and neurons (MAP2, green). (f) ssRT-qPCR analysis shows changes in *LncMtDloop* expression in multiple organs of 12-month-old 3xTg mice compared to that of wild type

mcie. Bars indicate mean $\pm$ SEM. $n = 6-8$ per group. * $P < 0.05$, ** $P < 0.01$ by unpaired t -test. SKM, skeleton muscle.



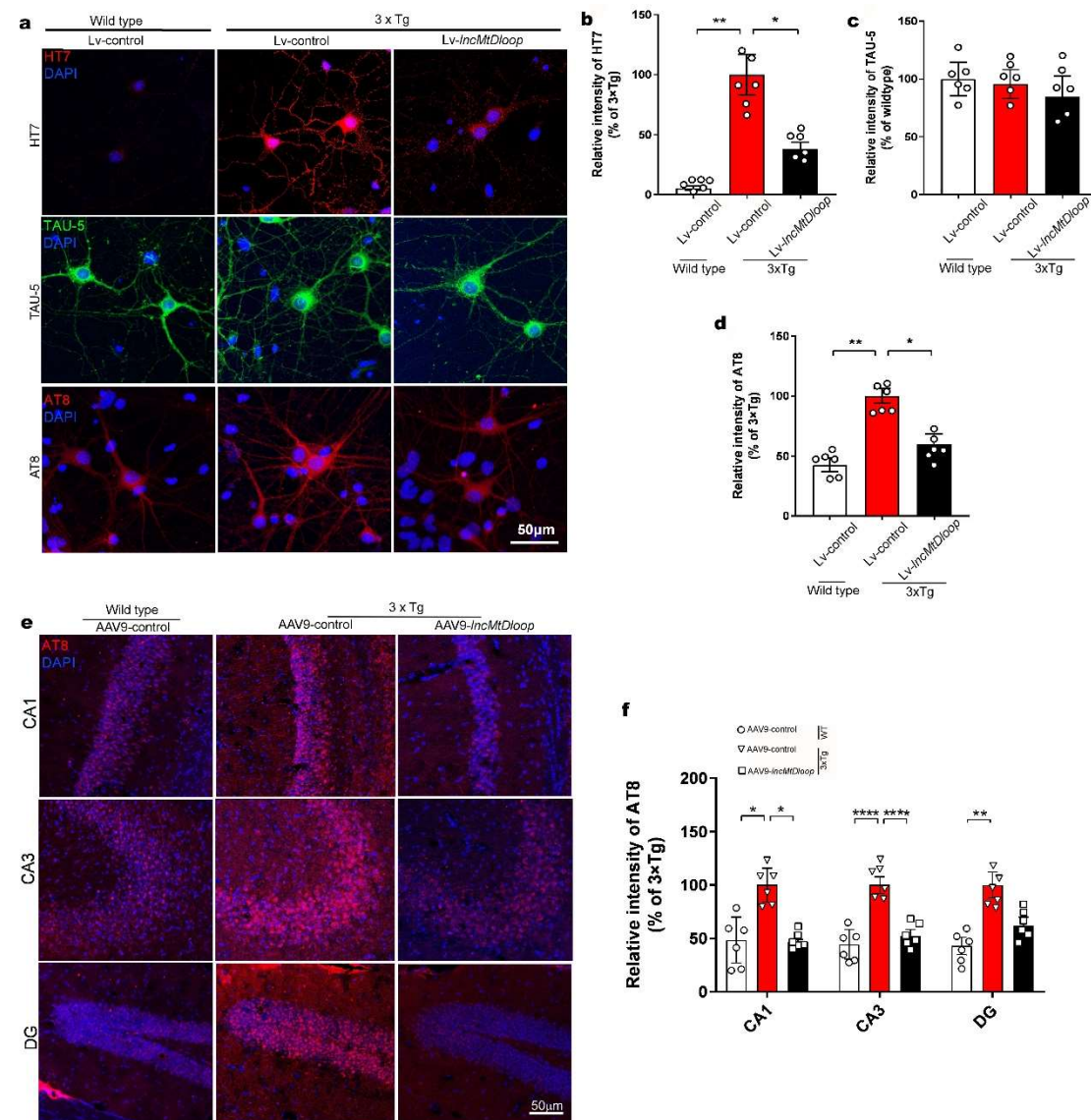
Supplementary Fig. 2 | Decreased expression of *IncMtDloop* selectively occurs in neuronal cells of 3xTg. (a) Representative images of RNAscope ISH and immunostaining showing loss of *IncMtDloop* expression (green) in 3xTg hippocampal neurons at DIV14. (b) Relative intensities of *IncMtDloop* fluorescent signals illustrated in a). Bars indicate mean $\pm$ SEM. n (cells) = 35-40 per group. **** $P < 0.001$, by unpaired t -test. (c) ssRT-qPCR analysis of *IncMtDloop* expression in wild type and 3xTg hippocampal neurons at DIV14. Bars indicate mean $\pm$ SEM. $n = 6$ per group. * $P < 0.05$, by unpaired t -test. (d) Representative images of RNAscope ISH and immunostaining showing few changes in levels of *IncMtDloop* expression (green) in 3xTg hippocampal microglia cultures at DIV14. (e) Relative intensities of *IncMtDloop*

fluorescent signals illustrated in d). Bars indicate mean $\pm$ SEM. n (cells) = 28-35 group. By unpaired *t*-test. (f) ssRT-qPCR analysis of *lncMtDloop* expression in wild type and 3xTg hippocampal microglia cultures at DIV14. Bars indicate mean $\pm$ SEM. n = 6 per group. By unpaired *t*-test. (g) Representative images of RNAscope ISH and immunostaining showing few changes in levels of *lncMtDloop* expression (green) in 3xTg hippocampal astrocyte cultures at DIV14. (f) Relative intensities of *lncMtDloop* fluorescent signals illustrated in g). Bars indicate mean $\pm$ SEM. n (cells) = 30-37 per group. By unpaired *t*-test. (i) ssRT-qPCR analysis of *lncMtDloop* expression in wild type and 3xTg hippocampal astrocyte cultures at DIV14. Bars indicate mean $\pm$ SEM. n = 6 per group. By unpaired *t*-test.



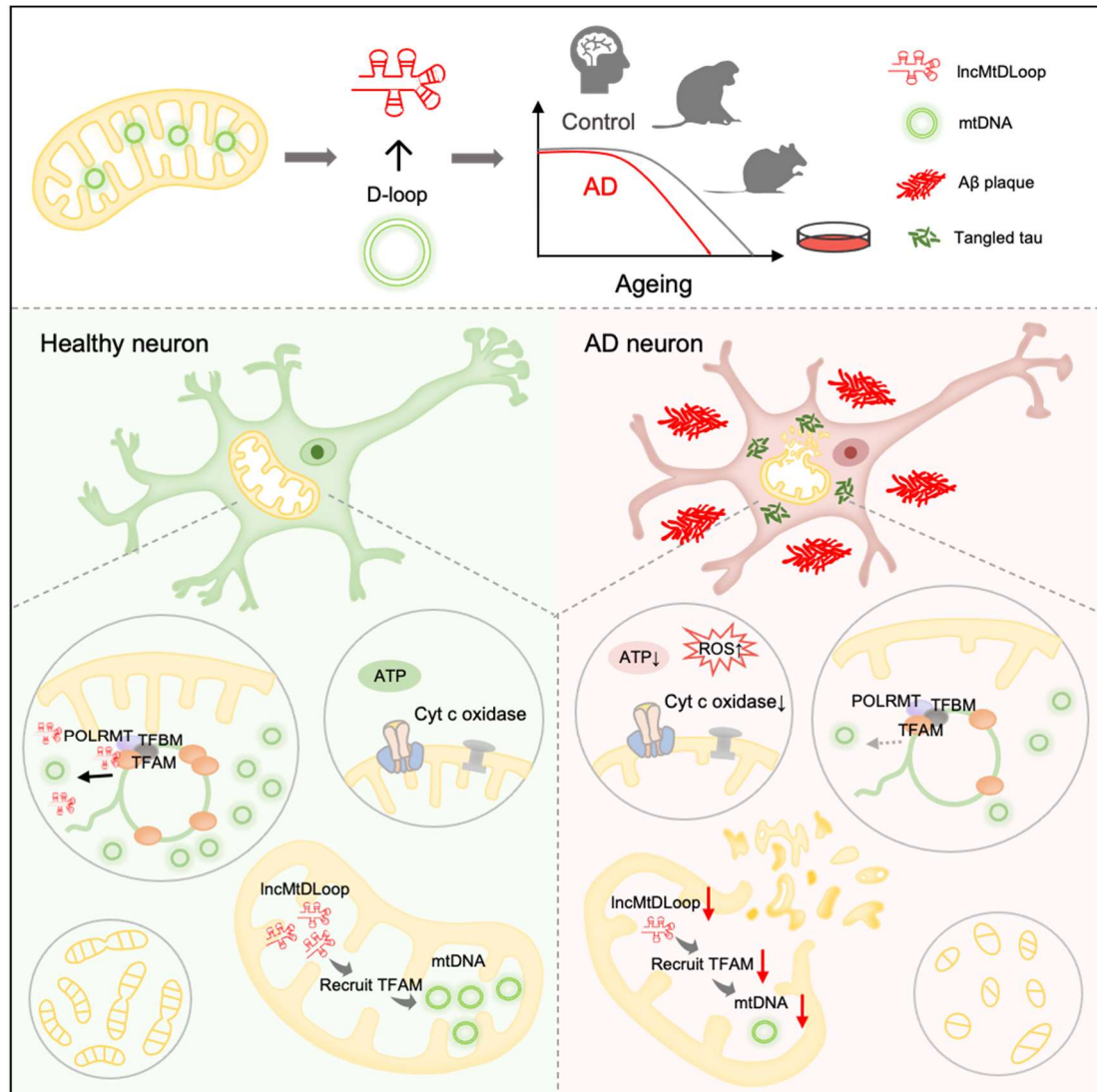
Supplementary Fig. 3 | Identification of key binding partners of *lncMtDloop*. (a) Silver staining gel showing *lncMtDloop* RNA pull-down with mitochondrial and cytosolic lysate preparations. Specific bands were cut and for the analysis of mass spectrum. +, *lncMtDloop*; —, anti-*lncMtDloop*. (b and c) The fold of enrichment of specific proteins compared with control group in the mitochondrial and cytosolic preparations in the products of *lncMtDloop* RNA pull-down. (d and e) Interaction propensity and discriminative power for *lncMtDloop* binding to p32, indicating C terminus of p32 tends to bind to the 3' or 5' terminus of *lncMtDloop*. (f) The score of

interaction between different motifs of *lncMtDloop* and p32.



Supplementary Fig. 4 | Restoration of *lncMtDloop* expression alleviates AD-related biochemical and pathological changes. (a) Representative IF images showing HT7, TAU-5 and AT8 in wild type and 3xTg primary hippocampal neurons. Wild type and 3xTg primary hippocampal neurons were infected with Lv-*lncMtDloop* at DIV5, cells were collected and prepared for tests at DIV14. (b-d) Relative intensities of HT7, TAU-5 and AT8 fluorescent signals illustrated in a). Bars indicate mean $\pm$ SEM. * P <0.05, ** P <0.01, by one-way ANOVA with Tukey's multiple comparisons test. (e) Representative IF images showing AT8 in hippocampus of wild type and 3xTg mice w/o overexpressing *lncMtDloop*. (f) Relative intensities of AT8 fluorescent signals

illustrated in (E). Bars indicate mean $\pm$ SEM. $n = 6$ mice per group. * $P < 0.05$, ** $P < 0.01$, **** $P < 0.001$, by one-way ANOVA with Tukey's multiple comparisons test.



Supplementary Fig. 5. Schematic diagram of the mechanism of the *lncMtDloop* in the regulation of mitochondrial function in healthy and AD neurons.