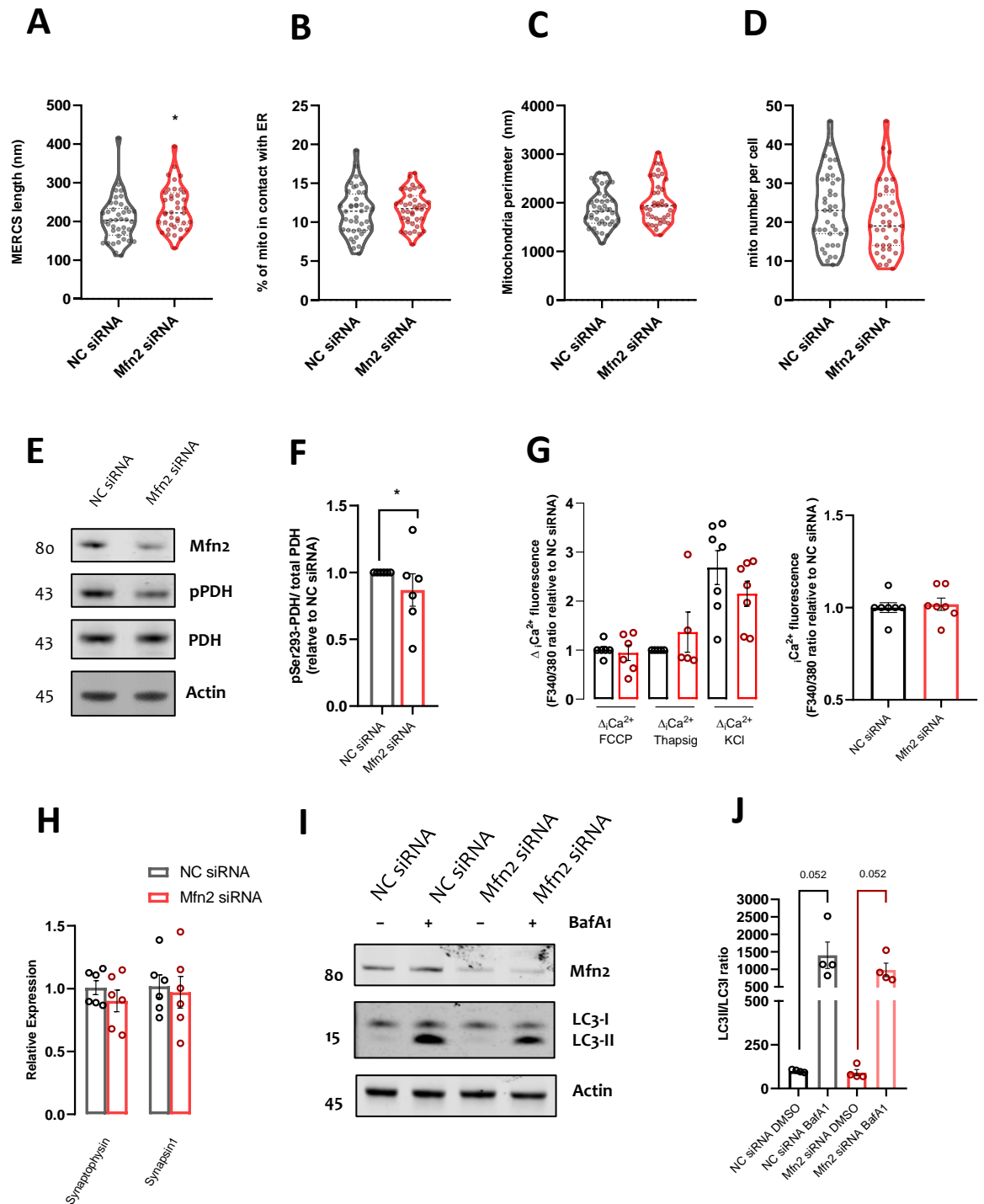
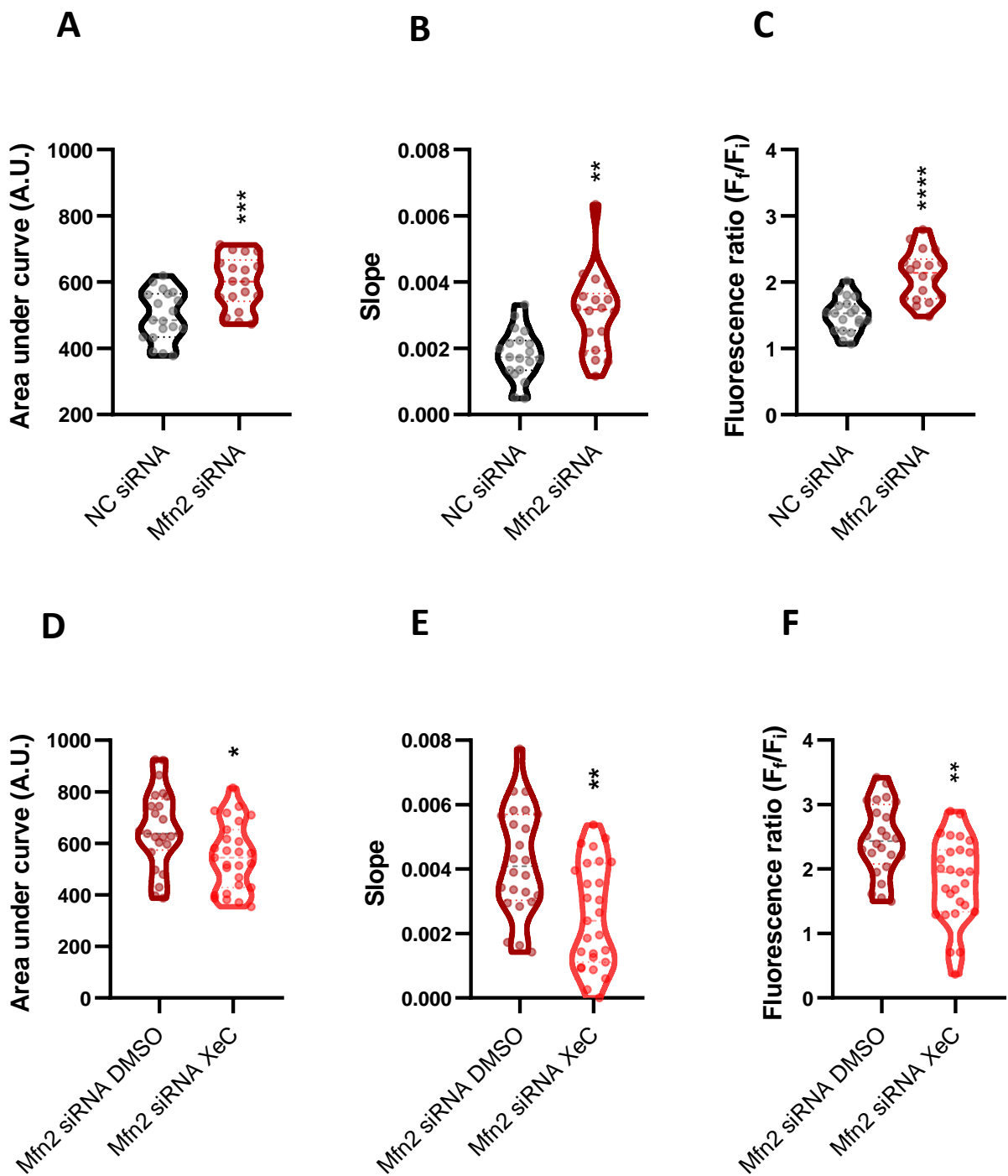


# Supplementary Figure 1



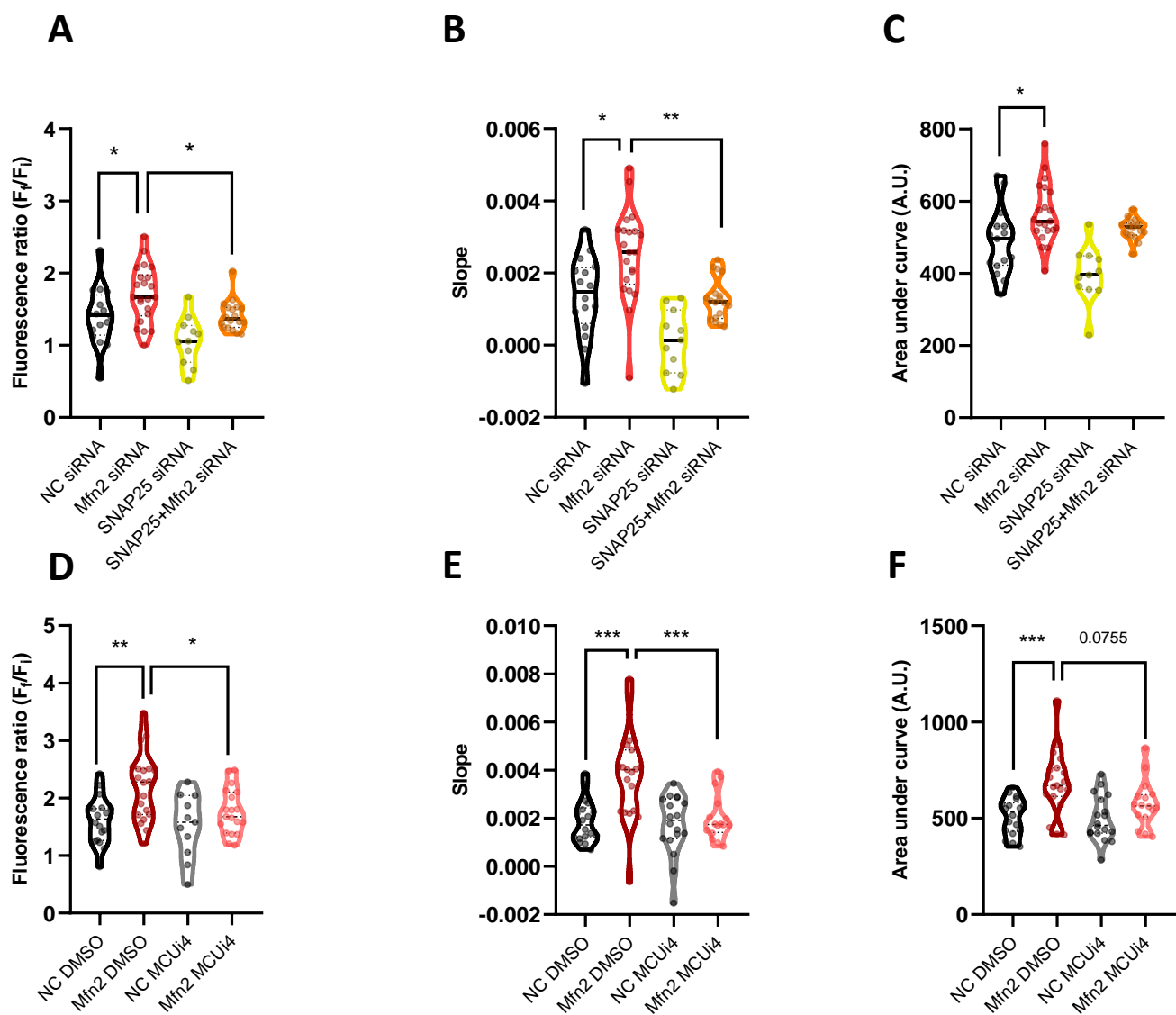
**Supplementary Figure 1 Analysis of MERCs, Ca<sup>2+</sup> imaging and dynamics, mRNA synaptic protein levels and autophagy in NC and Mfn2 siRNA conditions** **A)** Further analysis of TEM figures shown in Fig.1A of NC siRNA and Mfn2 siRNA SH-SY5Y cells. Violin graphs show quantification of MERCs length **B)** % of mitochondria in contact with ER **C)** average mitochondrial perimeter per cell and **D)** number of mitochondria per cell. (*n* = 4 independent cultures, 9-10 cells analyzed per culture). **E)** Representative immunoblots of NC siRNA or Mfn2 siRNA SH-SY5Y cells. Blots were probed with antibodies against Mfn2, pSer293-PDH, total PDH and actin which was used as a loading control. **F)** Bar graph shows ratio pSer293-PDH and total PDH protein levels (*n* = 5 independent cultures). **G)** Quantifications of Ca<sup>2+</sup> traces in Figure 1H **H)** Graph shows the quantification of gene expression analysis of vesicle-related proteins synaptophysin and synapsin-1 by qPCR (*n* = 6 independent cultures). **I)** Representative immunoblots of NC siRNA or Mfn2 siRNA SH-SY5Y cell treated with 50 nM Bafilomycin A1 (BafA1) or non-treated for 5h to block autophagy. **J)** Bar graph shows ratio LC3II to LC3I protein levels (*n* = 4 independent cultures). Data shown as mean  $\pm$  SEM. \* *p*  $\leq$  0.05

# Supplementary Figure 2



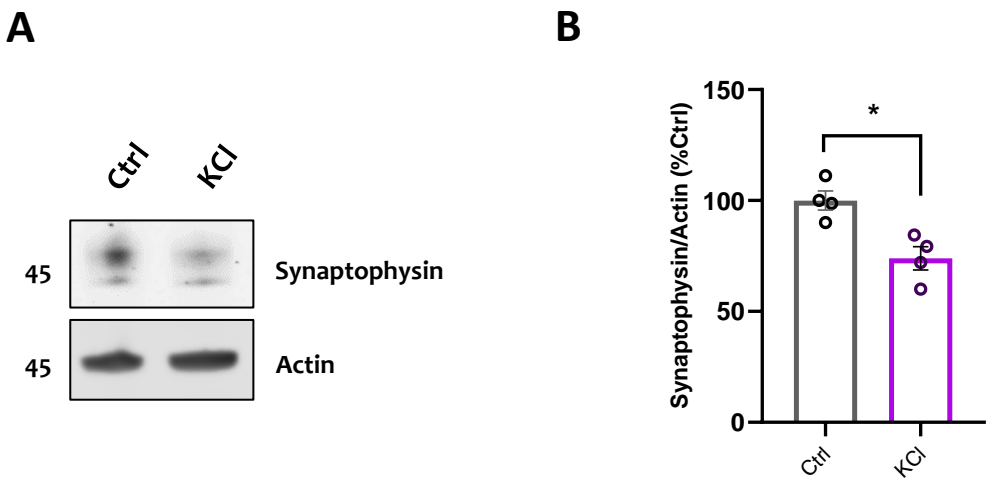
**Supplementary Figure 2** Further quantification of SypHy curves in Figure 2H and Figure 4E A) Violin plots show quantification of curve and data in Figure 2 G-H, area under the curve B) slope of curve and C) ratiometric analysis of  $F_t/F_i$ . D) Violin plots show quantification of curve and data in Figure 4D-E area under the curve E) slope of curve and F) ratiometric analysis of  $F_t/F_i$ . \*  $p \leq 0.05$ , \*\*  $p \leq 0.01$

# Supplementary Figure 3



**Supplementary Figure 3** Further quantification of SypHy curves in Figure 3D and Figure 4I. Violin plots show quantification of curve and data in Fig.3D **A**) ratiometric analysis of  $F_t/F_0$  **B**) slope of curve and **C**) area under the curve. Violin plots show quantification of curve and data in Fig.4I **D**) ratiometric analysis of  $F_t/F_0$  **E**) slope of curve and **F**) area under the curve. \*  $p \leq 0.05$ , \*\*  $p \leq 0.01$

# Supplementary Figure 4



**Supplementary Figure 4 Synaptophysin immunoblots of control SH-SY5Y cells or cells treated with KCl 50 mM stimulation** **A)** Representative immunoblots of control SH-SY5Y cells or cells treated with brief KCl 50 mM 15 mins stimulation and lysed 24h later. Blots were probed with antibodies against Synaptophysin and actin which was used as a loading control. **B)** Bar graph shows the amounts of synaptophysin analyzed once standardized to actin content in each sample ( $n = 4$  independent cultures) Data shown as mean  $\pm$  SEM. \*  $p \leq 0.05$ .