**Description of Additional Supplementary Files**

**Data S1. Additional statistical analysis for this study.** The file contains results for statistical analysis in: **Figure 1c**, statistics for C71G$^{+/-}$ cells (iMGs vs PMPs and iPSCs) using two-way ANOVA and Šídák's multiple comparisons test. **Figure 1e**, additional comparisons not listed in the main text figure for WT iMGs and all comparisons for PFN1 C71G$^{+/-}$ iMGs, including LPS treated versus untreated conditions, using two-way ANOVA and Šídák's multiple comparisons test. **Sup. Figure 3,** additional comparisons not listed in Supplementary Figure 3 for WT iMGs and all comparisons for PFN1 M114T$^{+/-}$ and M114T$^{+/+}$ iMGs, including LPS treated versus untreated conditions, using two-way ANOVA and Šídák's multiple comparisons test. **Figure 4b and d**, the indicated comparison was assessed by two-way ANOVA followed by Tukey's multiple comparisons for WT vs C71G$^{+/-}$ iMGs (**b**) and WT vs M114T$^{+/-}$ vs M114T$^{+/-}$ iMGs (**d**). **Figure 4i**, statistics for WT vs C71G$^{+/-}$ iMGs within each treatment condition and additional comparisons not listed in main text figure across different treatment conditions, using two-way ANOVA and Šídák's multiple comparisons test. **Sup. Figure 7**, Additional comparisons not listed in Supplementary Figure 7 as in Figure 4i. **Figure 7e**, Additional comparisons not listed in the main text figure for WT iMGs untreated vs rapamycin and C71G$^{+/-}$ iMGs for both untreated vs rapamycin and untreated vs bafilomycin A.

**Data S2. Results of the differential expression analysis of C71G$^{+/-}$ vs WT iMGs using quantitative proteomics.** Data resulting from the mass spectrometry experiment using tandem mass tag (TMT) quantitative proteomics to identify differentially expressed proteins between PFN1 WT and C71G$^{+/-}$ iMGs. PFN1 WT iMGs was used as the reference condition. Data was analyzed using Scaffold Software as described in the methods. Statistics were determined using T-test and Benjamin-Hochberg correction for multiple testing. Differentially expressed proteins in herein have a *P*- value<0.00160 and are considered statistically significant with the Benjamin-Hochberg test.

**Data S3. Functional enrichment analysis of differentially expressed proteins identified from the quantitative proteomics study.** Enriched pathways identified using the Bioplanet 2019 library by Enrichr. Differentially expressed proteins used for this analysis are listed in **Data S2**.

**Data S4. Enrichr and Metascape analyses of differentially expressed proteins from the quantitative proteomics study.** Enrichr (KEEG pathway and Gene Ontology celullar component) and Metascape were used to determine enriched functional terms from the differentially expressed proteins listed in **Data S2**. The results of each analysis are presented in different tabs.

**Data S5. Results of differential gene expression analysis from the RNASeq dataset.** RNAseq data obtained from C71G$^{+/-}$ (CG) and M114T$^{+/-}$ (MThet) iMGs (ALS-PFN1 group) and WT controls (WT and WT2) were analyzed using DESeq2 package and Wald test.

**Data S6. Information for antibodies used herein.** The target, antibody species, clone information, supplier, catalog number (#), lot number, application and working dilution are provided.

**Supplementary Code 1.** ImageJ analysis script used to measure Iba1 signal in dead neuron injections tissue.