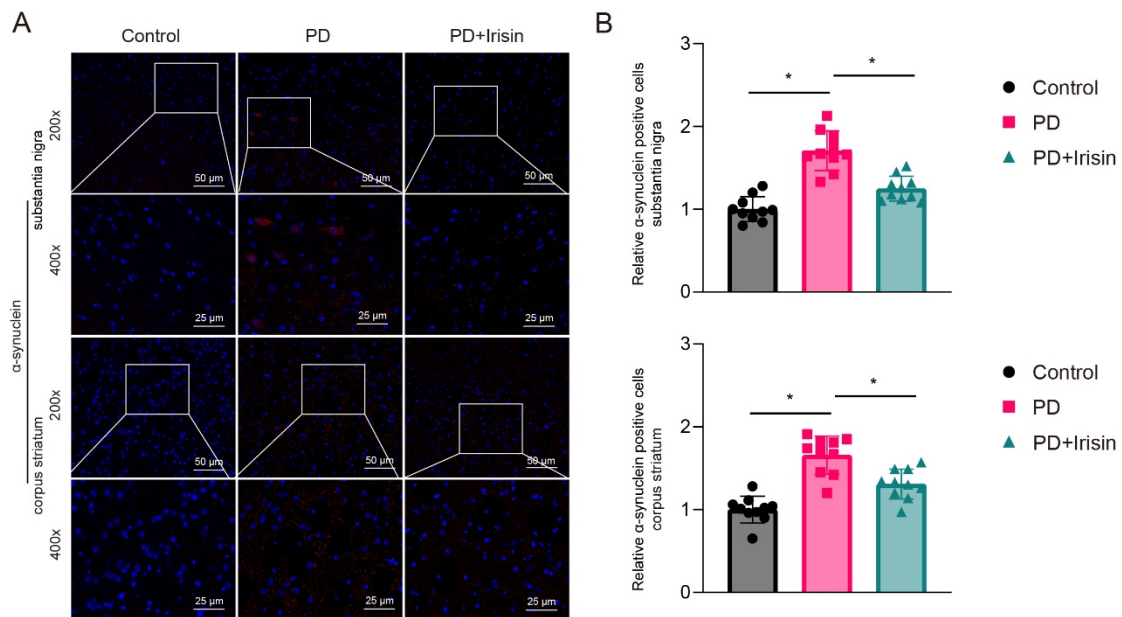
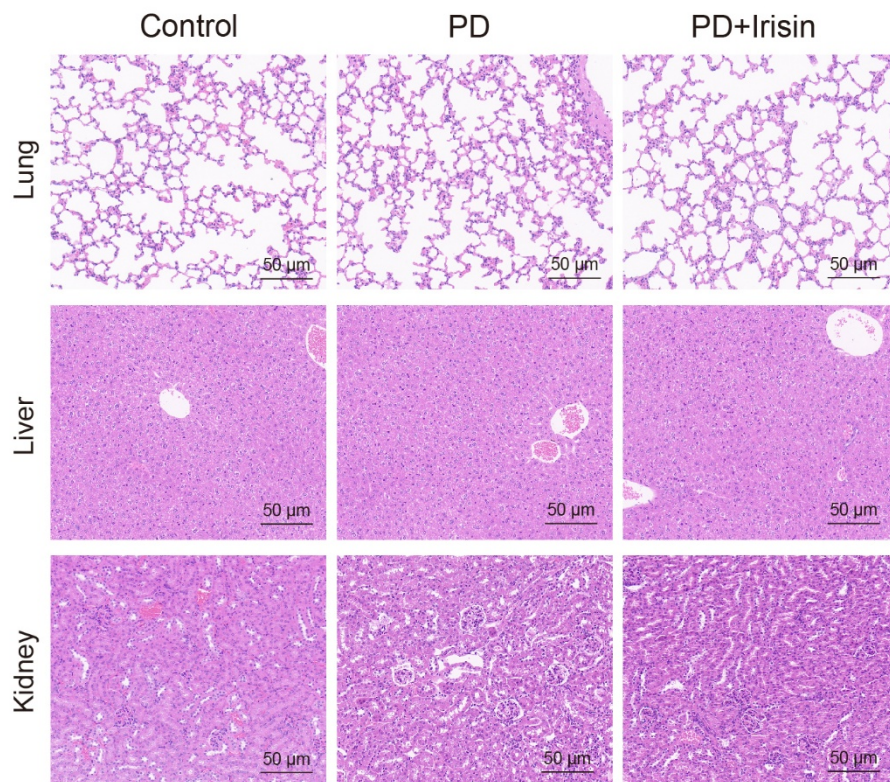


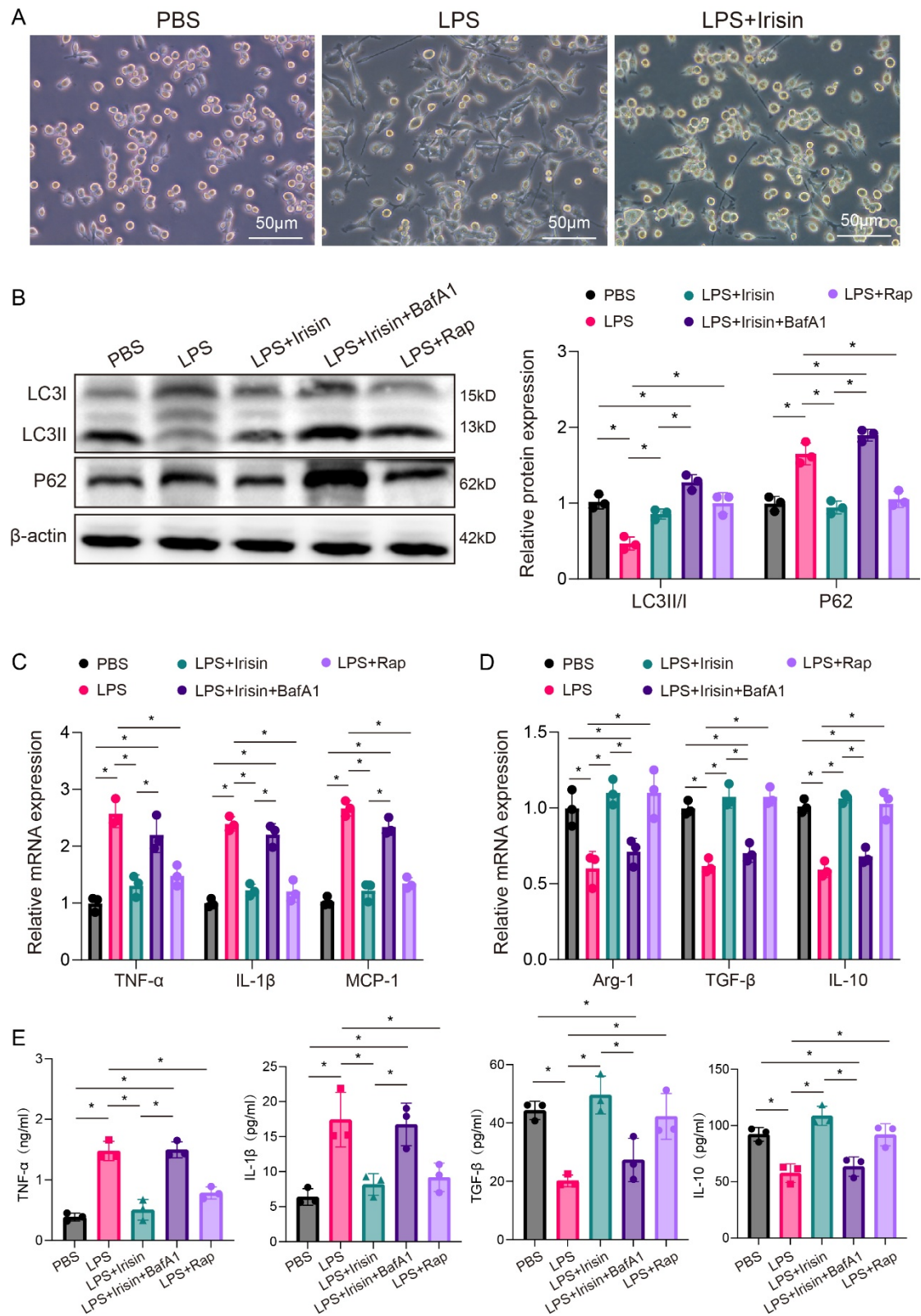


Supplementary Figure 1. Irisin suppresses α -synuclein expression in the substantia nigra and striatum of mice

A-B: Representative immunohistochemical images showing α -synuclein expression in the substantia nigra of the control, PD, and PD + Irisin groups. Scale bars: 50 μ m. Control vs PD, $*p < 0.05$; PD + Irisin vs PD, $*p < 0.05$.



Supplementary Figure 2. Safety assessment of irisin treatment *in vivo*



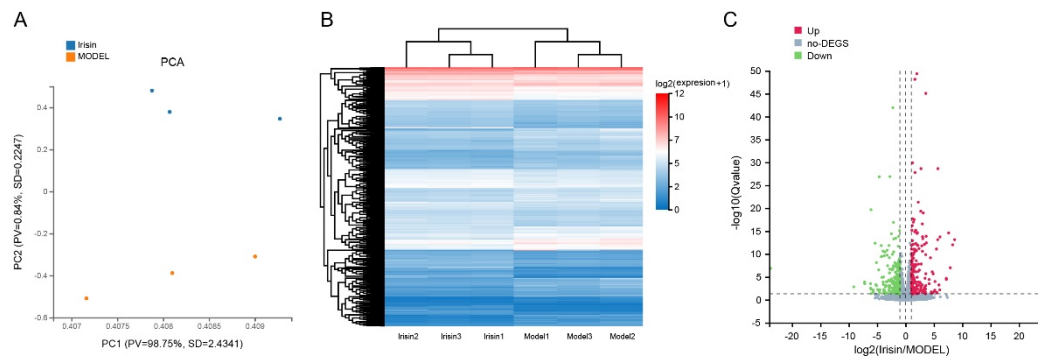
Supplementary Figure 3. Effects of LPS, irisin, Rapamycin, and Bafilomycin A1 on BV2 microglial cells

A: Morphological changes of BV2 treated with LPS and irisin.

B: WB analysis of LC3 and P62 protein levels in BV2 cells treated with Rapamycin (Rap, an autophagy activator) or Bafilomycin A1 (BafA1, an autophagic flux inhibitor). The effects of these treatments on autophagy were confirmed by changes in LC3 and P62 levels. PBS vs LPS, $*p < 0.05$; LPS vs LPS + Irisin, $*p < 0.05$; LPS + Irisin vs LPS + Irisin + BafA1, $*p < 0.05$; PBS vs LPS + Irisin + BafA1, $*p < 0.05$; LPS vs LPS + Rap, $*p < 0.05$.

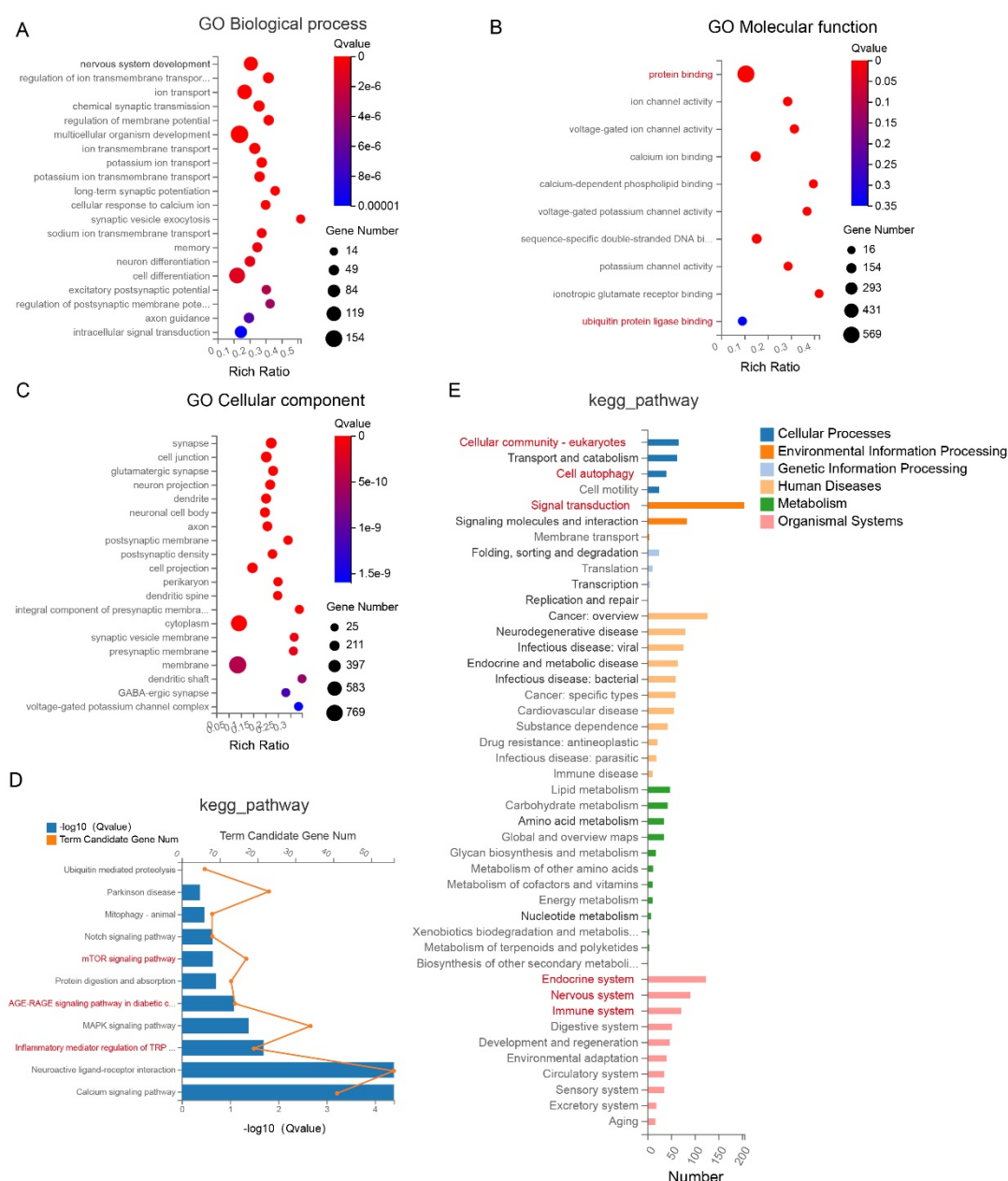
C-D: QRT-PCR was used to detect the mRNA expression levels of pro-inflammatory markers (C) and anti-inflammatory markers (D) in each treatment group. PBS vs LPS, $*p < 0.05$; LPS vs LPS + Irisin, $*p < 0.05$; LPS + Irisin vs LPS + Irisin + BafA1, $*p < 0.05$; PBS vs LPS + Irisin + BafA1, $*p < 0.05$; LPS vs LPS + Rap, $*p < 0.05$.

(E) ELISA results showing the levels of inflammatory factors in the supernatants of BV2 cells treated with LPS, irisin, Rap, or BafA1. PBS vs LPS, $*p < 0.05$; LPS vs LPS + Irisin, $*p < 0.05$; LPS + Irisin vs LPS + Irisin + BafA1, $*p < 0.05$; PBS vs LPS + Irisin + BafA1, $*p < 0.05$; LPS vs LPS + Rap, $*p < 0.05$.



Supplementary Figure 4. Differentially expressed genes (DEGs) between the PD and irisin-treated groups

A-C: Volcano plot showing the DEGs in the substantia nigra samples between the PD and Irisin-treated groups. The screening criteria for DEGs were set at a p -value < 0.05 and fold change > 1 .



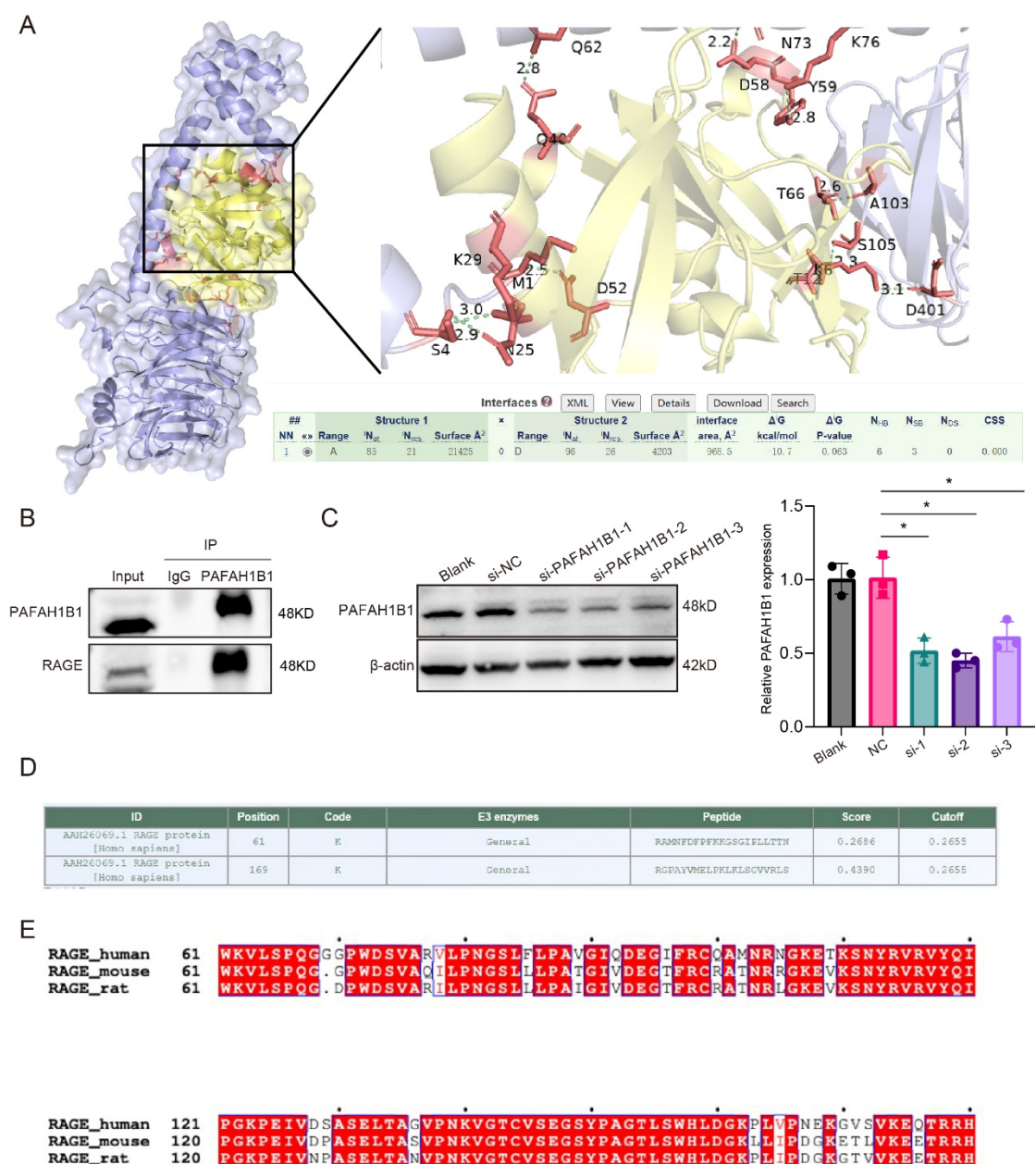
Supplementary Figure 5. GO and KEGG pathway enrichment analyses of DEGs

A: Gene Ontology (GO) analysis of Biological Process (BP) terms for the DEGs.

B: GO analysis of Molecular Function (MF) terms for the DEGs.

C: GO analysis of Cellular Component (CC) terms for the DEGs. The DEGs were predominantly enriched in synapse formation, cell junctions, and glutamatergic synapses.

D-E: KEGG pathway enrichment analysis of the DEGs.



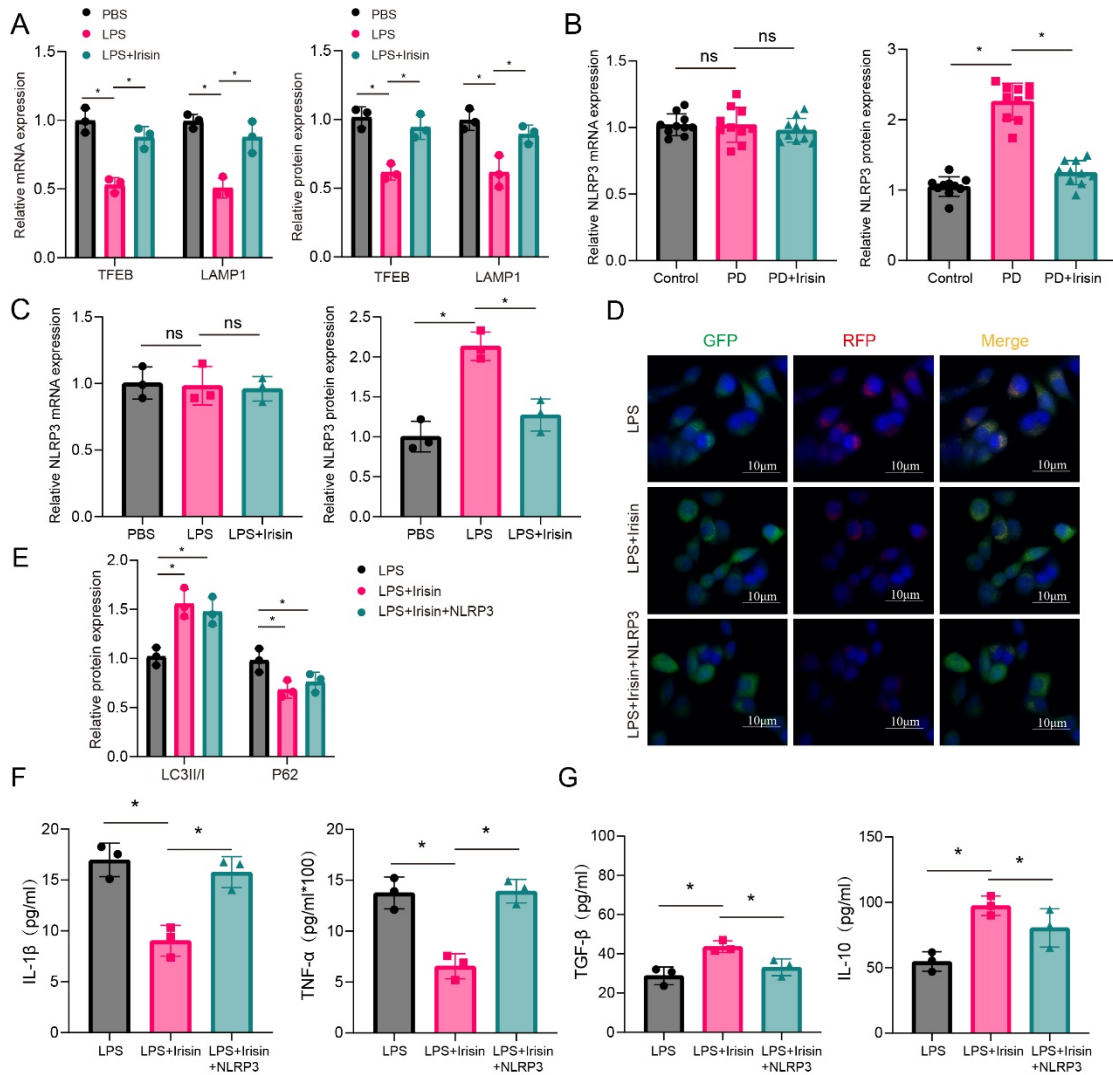
Supplementary Figure 6. Analysis of RAGE ubiquitination and related molecules

A: Molecular docking of PAFAH1B1 (PDB: P43034) and UB from the EMBL-EBI database, showing a protein-binding free energy of -10.7 kcal/mol, suggesting a possible interaction between PAFAH1B1 and ubiquitin.

B: Co-IP results confirming the binding interaction between PAFAH1B1 and RAGE in mice.

C: Screening of si-PAFAH1B1 sequences. Si-PAFAH1B1-2 was selected for subsequent studies based on its inhibitory effect on PAFAH1B1 expression in BV2 cells. NC vs si-1, * $p < 0.05$; NC vs si-2, * $p < 0.05$; NC vs si-3, * $p < 0.05$.

D-E: Prediction of ubiquitination modification sites of RAGE using the database GPS-uber. K61 and K169 sites were predicted to have ubiquitination modification, and they were found to be highly conserved among the three species.



Supplementary Figure 7. Related factors in the TFEB-NLRP3 pathway

A: QRT-PCR and WB analysis of the expression of lysosomal markers TFEB and LAMP1 in BV2 models. PBS vs LPS, $*p < 0.05$; LPS + Irisin vs LPS, $*p < 0.05$.

B: QRT-PCR and WB analysis of NLRP3 mRNA levels in the striatum and substantia nigra of PD models. mRNA: Control vs PD, not significant; PD + Irisin vs PD, not significant. Protein: Control vs PD, $*p < 0.05$; PD + Irisin vs PD, $*p < 0.05$.

C: QRT-PCR and WB analysis of NLRP3 mRNA and protein levels in BV2 cells. mRNA: PBS vs LPS,

not significant; LPS + Irisin vs LPS, not significant. Protein: PBS vs LPS, $*p < 0.05$; LPS + Irisin vs LPS, $*p < 0.05$.

D: Autophagy flux analysis in BV2 cells with NLRP3 overexpression.

E: QRT-PCR and WB analysis of LC3 and P62 protein levels in BV2 cells. LPS vs LPS + Irisin, $*p < 0.05$; LPS + Irisin vs LPS + Irisin + NLRP3, $*p < 0.05$.

F-G: Analysis of inflammatory markers in BV2 cells with NLRP3 overexpression. LPS vs LPS + Irisin, $*p < 0.05$; LPS + Irisin vs LPS + Irisin + NLRP3, $*p < 0.05$.

Supplementary Figure 8 (Uncropped and unedited blot/gel images):

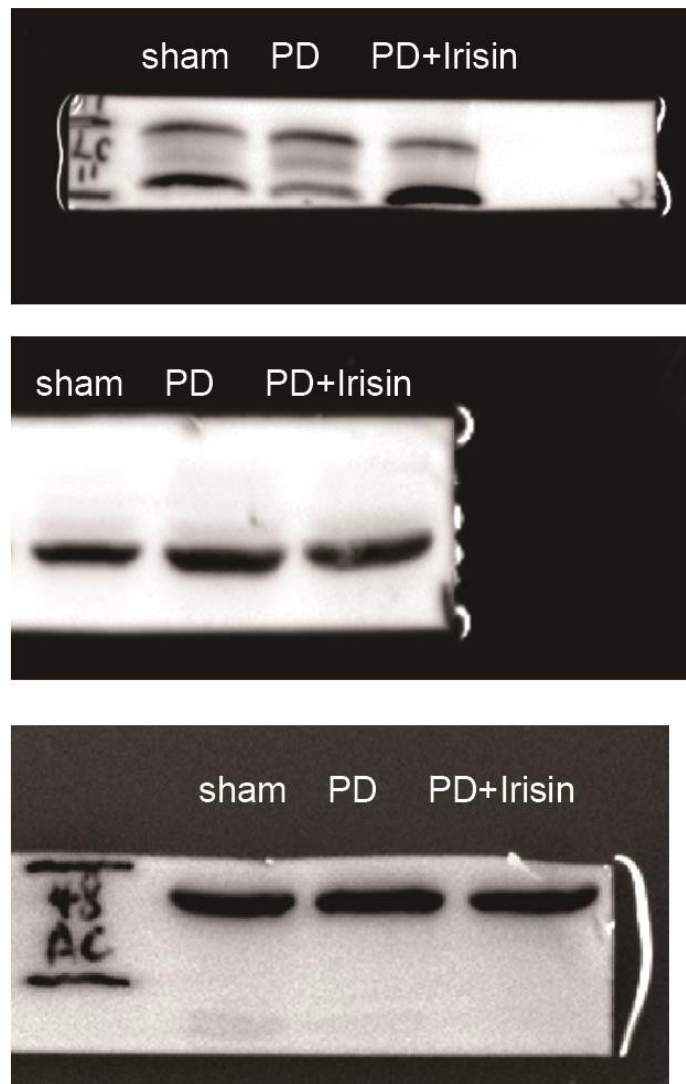
1. Figure 5E:

Figure 5E



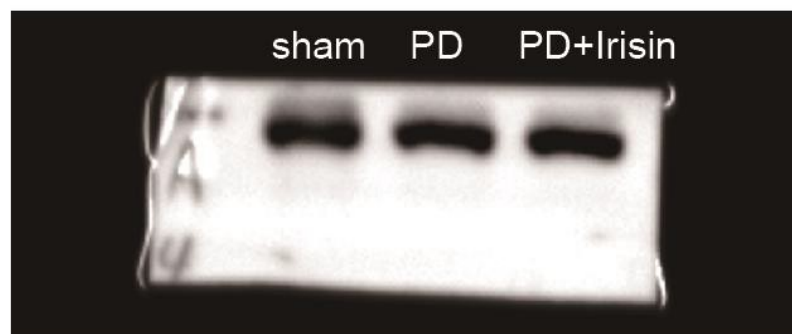
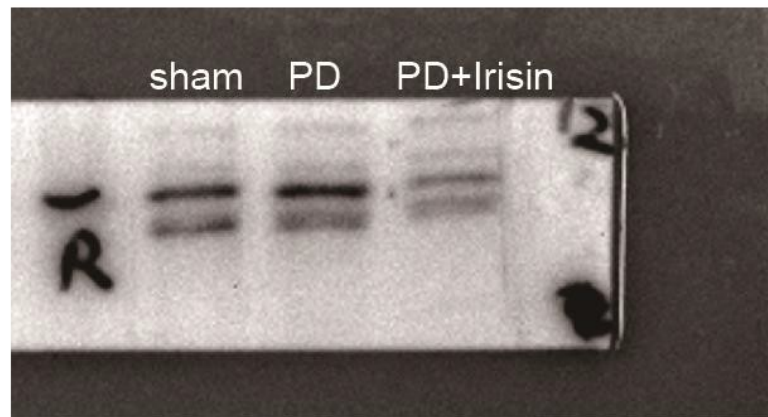
2. Figure 5F:

Figure 5F



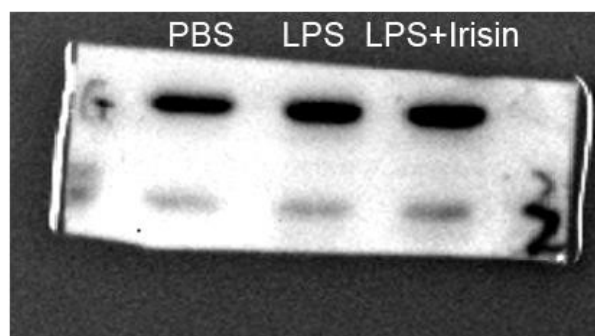
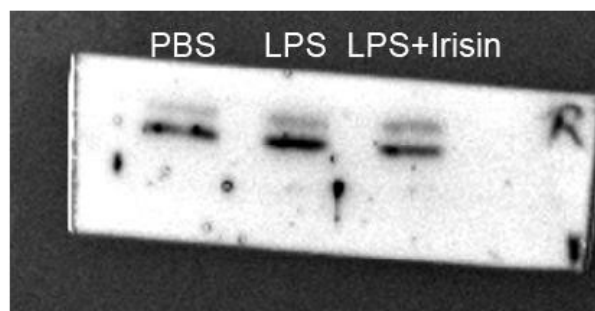
3. Figure 6B:

Figure 6B



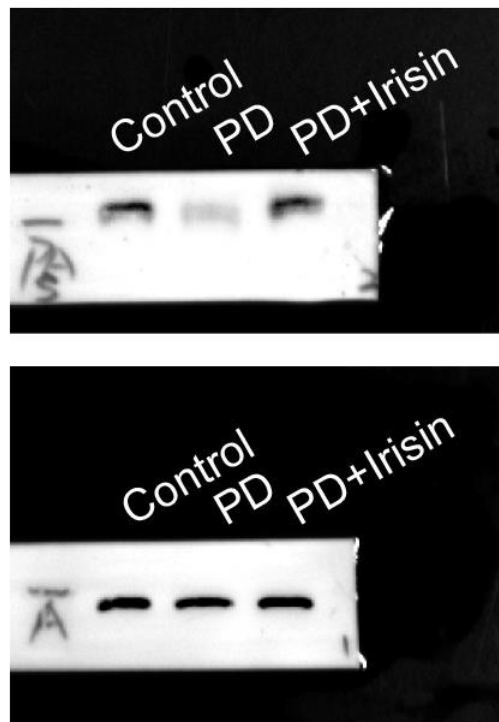
4 Figure 6D:

Figure 6D



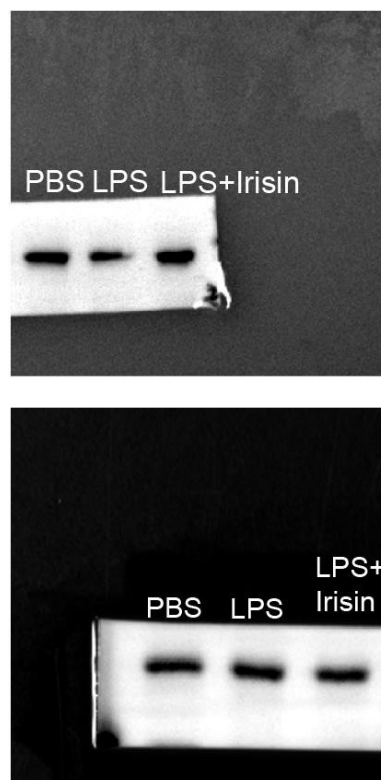
5. Figure 6G:

Figure 6G



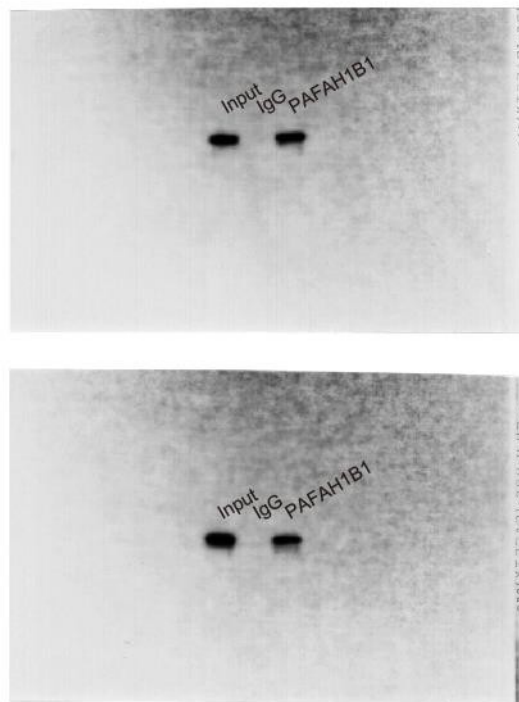
6. Figure 6H:

Figure 6H



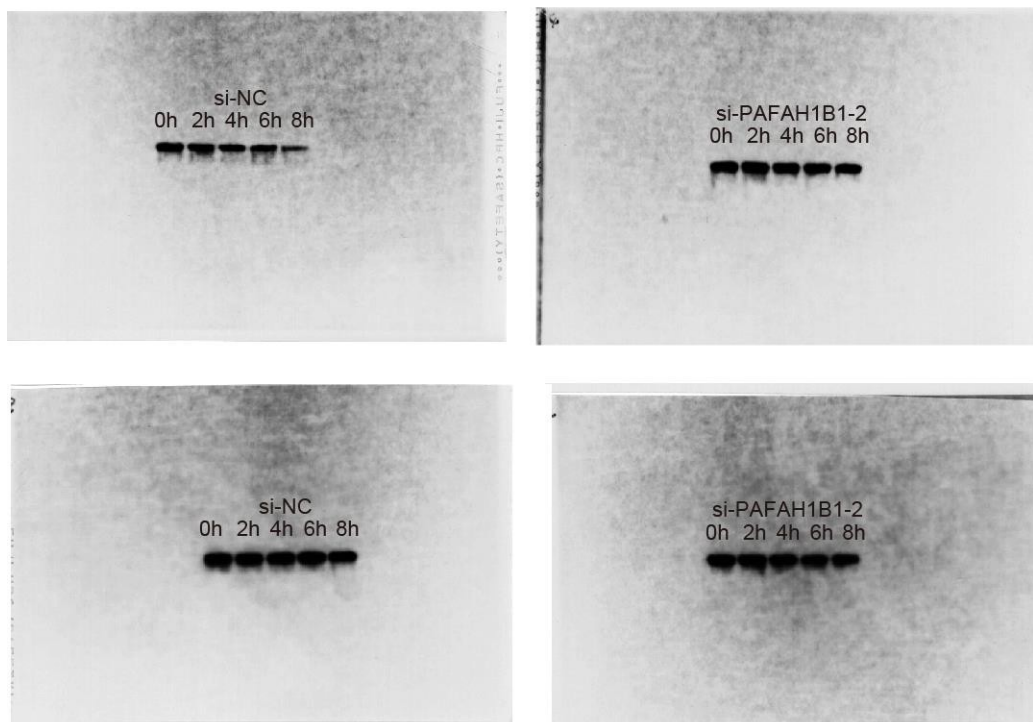
7. Figure 7C:

Figure 7C



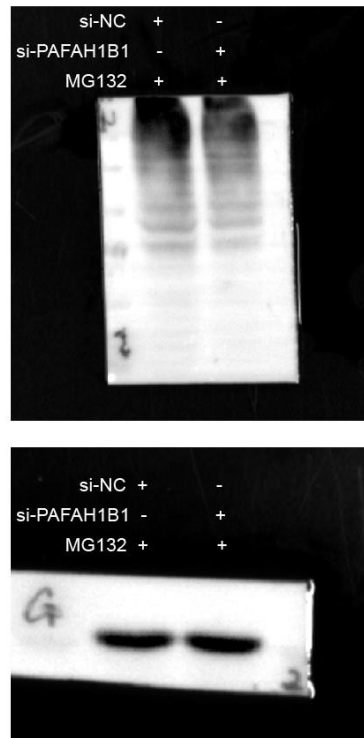
8. Figure 7E

Figure 7E



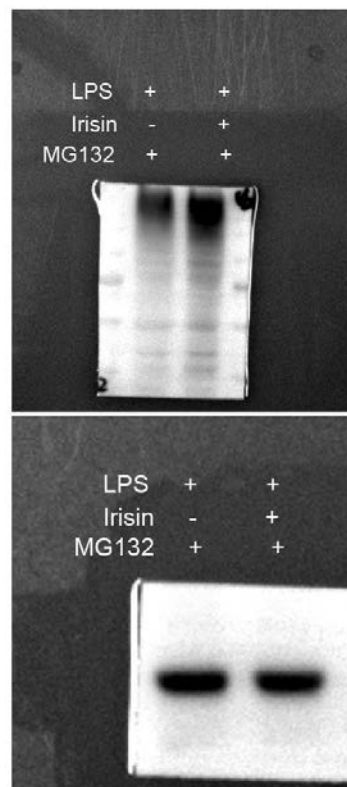
9. Figure 7F:

Figure 7F



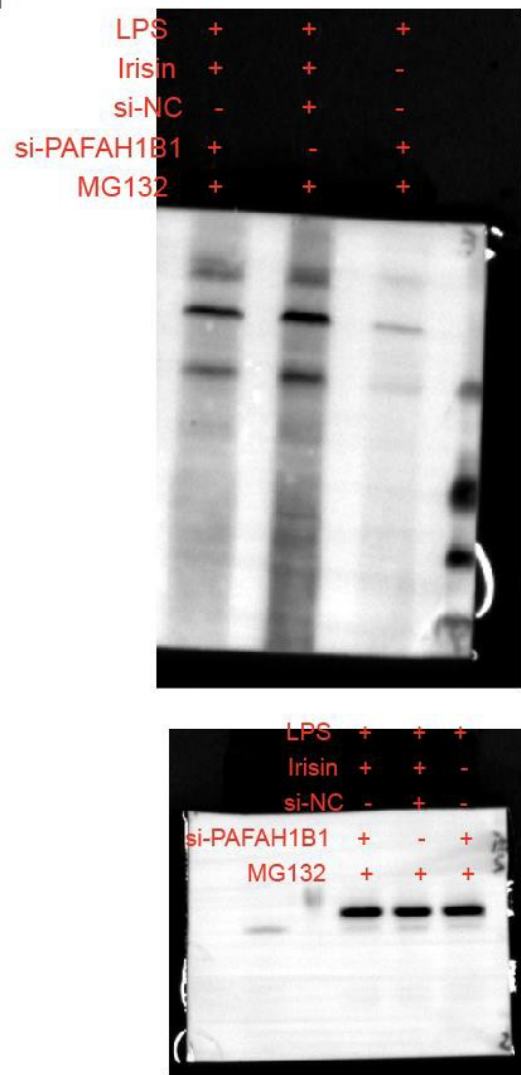
10. Figure 7G:

Figure 7G



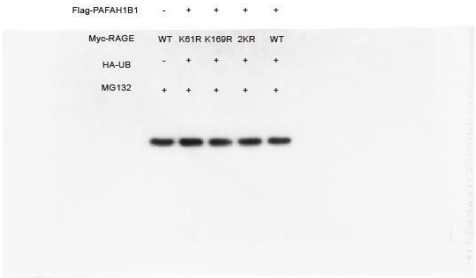
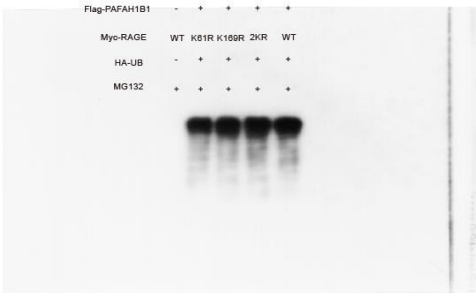
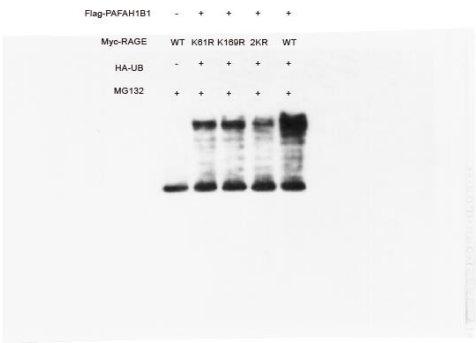
11. Figure 7G:

Figure 7H



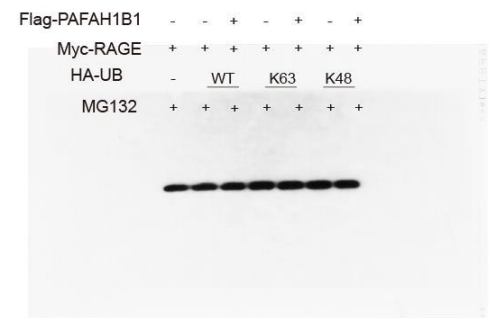
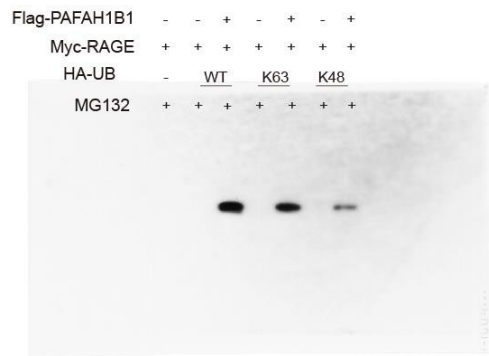
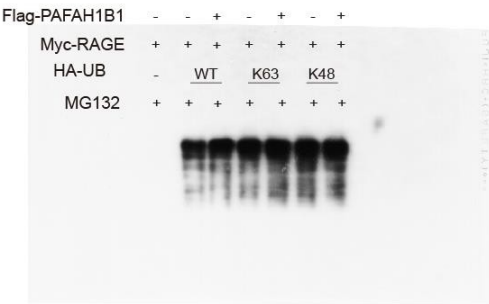
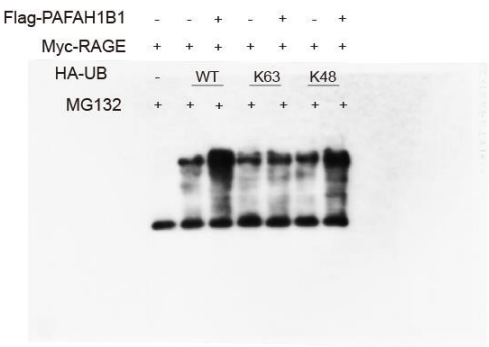
12. Figure 8C:

Figure 8C



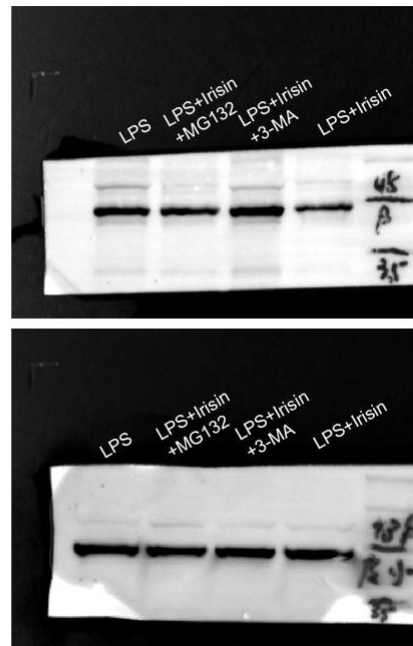
13. Figure 8D:

Figure 8D



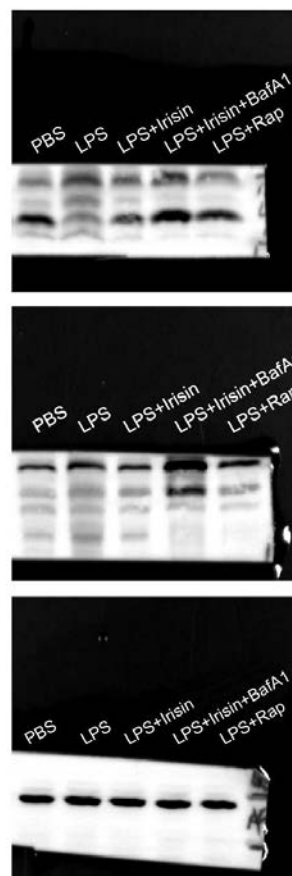
14. Figure 9H:

Figure 9H



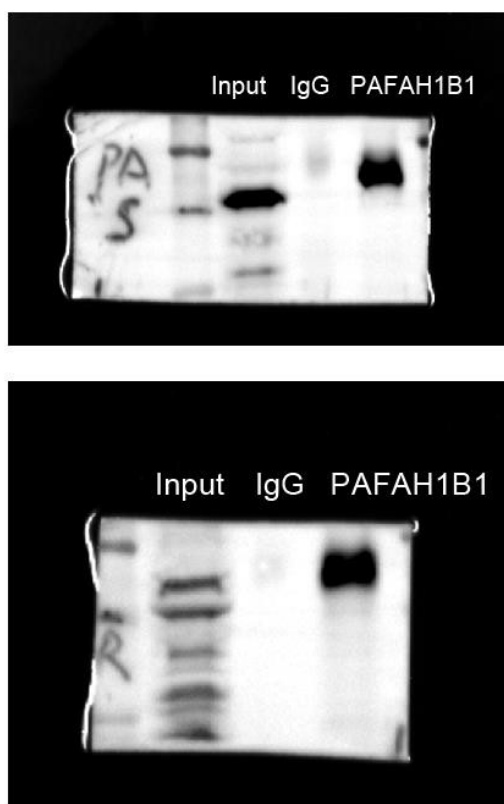
15. Figure S3B:

Figure S3B



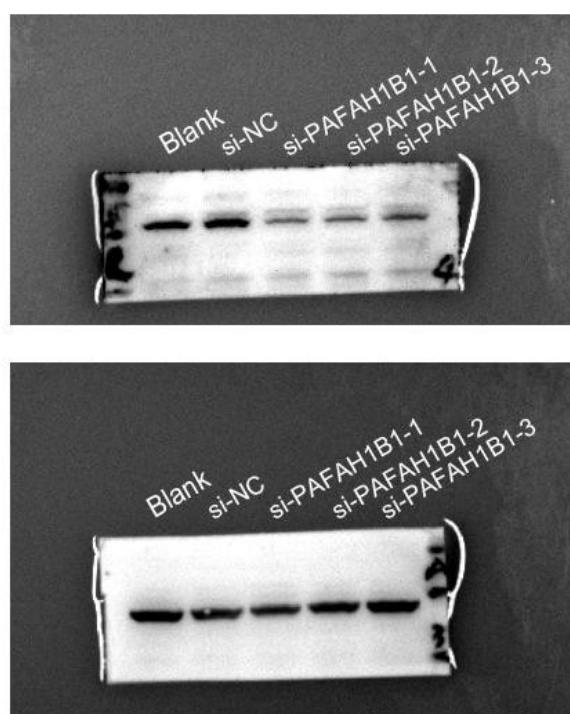
16. Figure S6B:

Figure S6B



17. Figure S6C:

Figure S6C



Supplementary Table 1 Basic information of the clinical study population

Category	Control (N=120)	PD (N=120)	t/x ² value	p value
Age (years)	66.45±9.04	64.62±10.84	1.42	0.16
Gender (male/female)	68/52	69/51	0.02	0.90
Course of the disease	--	6.87±4.48	--	--
BMI (kg/m ²)	22.65±3.16	22.92±2.84	0.70	0.49
Smoking (%)	26.67	25.00	0.07	0.79

Supplementary Table 2 si-RNAs for the PAFAH1B1 gene

si-RNAs	Primer sequences (5'-3')	
si-PAFAH1B1-1	Sense	GGCUCCAGAAAGUUCAUAUTT
	Antisense	AUAUGAACUUUCUGGAGCCTT
si-PAFAH1B1-2	Sense	GCAGAU AUGACGAUUA AAUUTT
	Antisense	AUUUAAUCGUCAUAUCUGCTT
si-PAFAH1B1-3	Sense	GGUCAUAGGAGUCCAGUUATT
	Antisense	UACUGGACUCCUAUGACCTT

Supplementary Table 3 Primer sequences for qRT-PCR

Gene	Species	Primer sequences (5'-3')	
β -actin	Mouse	Forward	AGTGTGACGTTGACATCCGT
		Reverse	GCAGCTCAGTAACAGTCCGC
TNF- α	Mouse	Forward	CCCTCACACTCAGATCATCTTCT
		Reverse	GCTACGACGTGGGCTACAG
IL-1 β	Mouse	Forward	GGAGAGGTTTCGTAGCTTAACG
		Reverse	CCACACAGCACTATATCTGGGT
MCP-1	Mouse	Forward	TAGACTTTCGGGAAAAACGTGTG
		Reverse	AGAAGCTCTGTTGACTGGTGA
Arg-1	Mouse	Forward	GTCCCAGACATCAGGGAGTAA
		Reverse	TCGGATACTTCAGCGTCAGGA
TGF- β	Mouse	Forward	CCCCTGGAAAGAATGGAGATG
		Reverse	TCCAAACCACTGAAACCTCTG
IL-10	Mouse	Forward	CCACCACGCTCTTCTGTCTAC
		Reverse	AGGGTCTGGGCCATAGAACT
RAGE	Mouse	Forward	AGTTGACGGACCCCAAAAG
		Reverse	AGCTGGATGCTCTCATCAGG
