Supplementary Material

# Supplementary Figures

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**Supplementary Figure 1.** Modulation of cell viability bymiR-335. (**A**) qRT-PCR analysis of miR-335 in BV2 cells transfected with a pre-miR-335 or pre-NC for 24 h. (**B**) qRT-PCR analysis of miR-335 in SH-SY5Y cells transfected with a pre-miR-335 or pre-NC for 24 h. (**C**) BV2 cells were transfected with a pre-miR-335 or pre-NC for 24 h and then stimulated with LPS for additional 24 h. (**D**) SH-SY5Y cells with or without overexpression of α-syn were transfected with a pre-miR-335 or pre-NC for 24 h. Cell metabolic activity was determined by MTS metabolism assay and cell membrane integrity by LDH activity in the supernatant. Results are presented as mean ± SEM of three independent experiments performed in duplicates and normalized to control cells transfected with pre-NC.



**Supplementary Figure 2** Modulation of RIP1, RIP3 and MLKL by miR-335. (**A**) BV2 cells were co-transfected with a LRRK2-Wt or empty vector plasmid and with a pre-miR-335 or pre-NC for 24 h. (**B**) SH-SY5Y neuroblastoma cells with or without overexpression of α-syn were transfected with pre-miR-335 or pre-NC for 24 h. Total protein extracts were prepared for Western blot analysis of RIP1, RIP3 and MLKL. Representative immunoblots are presented. β-actin was used as loading control. Values are expressed as mean ± SEM of three independent experiments.



**Supplementary Figure 3** Modulation of proinflammatory mRNA levels by miR-335. BV2 cells were co-transfected with a plasmid encoding a constitutively active form of MEK1 (MEK1\*), or empty vector and pre-miR-335/pre-NC for 24 h, and then stimulated with LPS for additional 24 h. TNF-α and IL-1β mRNA levels were measured by qRT-PCR. Results are expressed as mean ± SEM from two independent experiments.

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**Supplementary Figure 4** *In silico* analysis of has-miR-335-3p targeting of α-syn (SNCA) 3’-UTR region using TargetScan and PicTar databases.

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**Supplementary Figure 5** Modulation of cell viability by LRRK2-Wt. (**A**) BV2 cells were co-transfected with a LRRK2-Wt or empty vector plasmid and with a pre-miR-335 or pre-NC for 24 h, and then stimulated with LPS for additional 24 h. (**B**) SH-SY5Y cells overexpressing α-syn were co-transfected with a LRRK2-Wt or empty vector plasmid and with a pre-miR-335 or pre-NC for 24 h. Cell metabolic activity was determined by MTS metabolism assay and cell membrane integrity by LDH activity in the supernatant. Results are presented as mean ± SEM of three independent experiments performed in duplicates and normalized to control cells transfected with pre-NC.

# Supplementary Material

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**Figure 4**. Original and uncropped blot p-p38.

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**Figure 4**. Original and uncropped blot p38.



**Figure 4**. Original and uncropped blot p-JNK.



**Figure 4**. Original and uncropped blot JNK.



**Figure 4**. Original and uncropped blot p-ERK1/2.



**Figure 4**. Original and uncropped blot ERK1/2.

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**Figure 4**. Original and uncropped blot p-IкB.

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**Figure 4**. Original and uncropped blot IкB.