

## **Online supplement**

### **The deficiency of Maged1 attenuates Parkinson's disease progression by inhibiting apoptosis and enhancing autophagy in mice**

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## Supplemental Figures

**Figure S1**

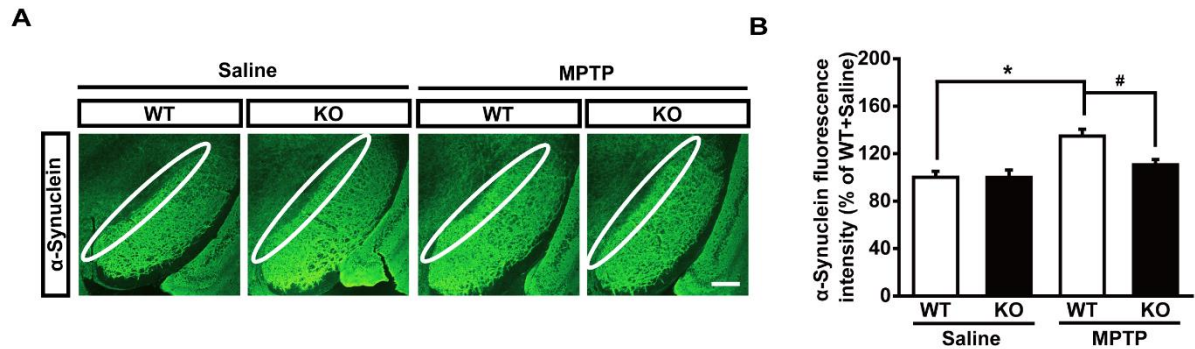


**Figure S1. (A)** Immunofluorescence staining for  $\alpha$ -Synuclein in the substantia nigra derived from WT or Maged1 KO mice with or without MPTP treatment. Scale bar: 200  $\mu$ m. **(B)** Quantification of  $\alpha$ -Synuclein fluorescence intensity in the substantia nigra (% of WT+saline), WT+saline: n=4, KO+saline: n=4, WT+MPTP: n=6, KO+MPTP: n=5. \*  $P < 0.05$ , #  $P < 0.05$ .

**Figure S2**

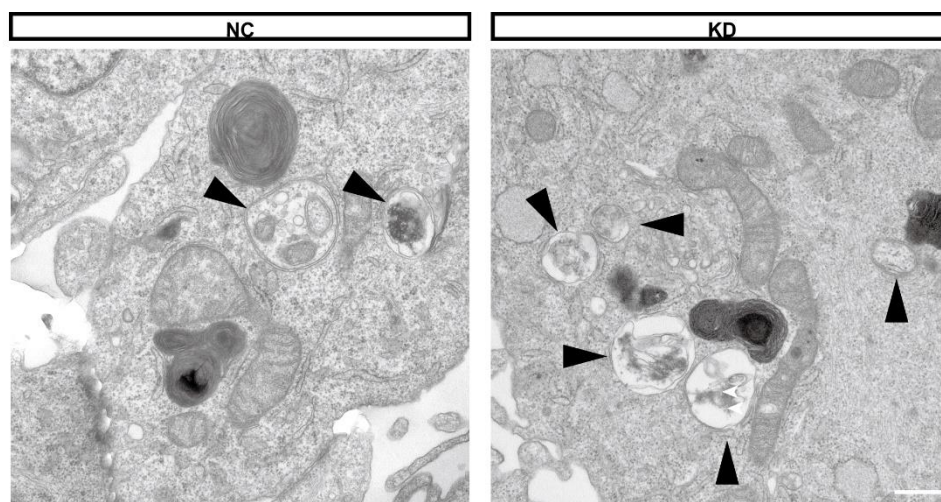


**Figure S2.** SH-SY5Y cells were transfected with NC or si-Maged1 and autophagosome was detected using transmission electron microscopy (TEM). Scale bar: 1  $\mu$ m. Black arrow

indicates autophagosome. NC: normal control, KD: knock down.

**Figure S3**

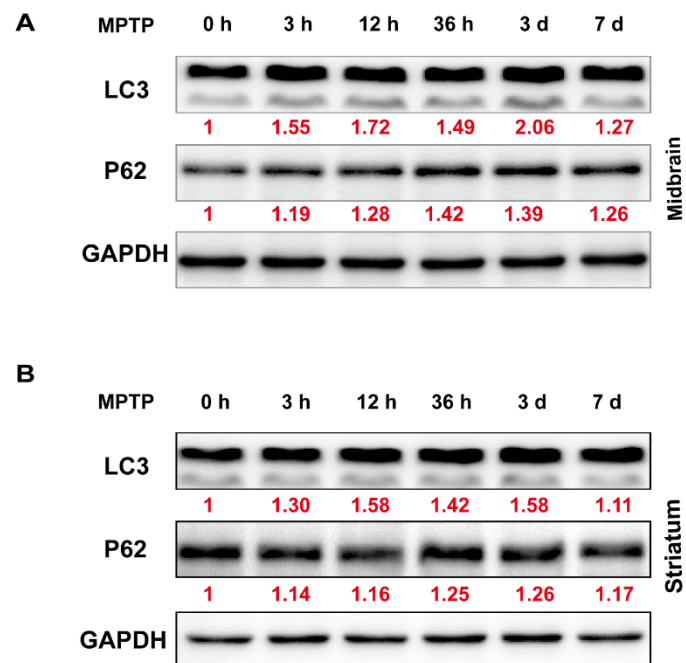


**Figure S3. (A and B)** Western blot analyses illustrating the expression of LC3 and P62 in midbrain (A) and striatum (B) at different time points after MPTP treatment.

**Figure S4**

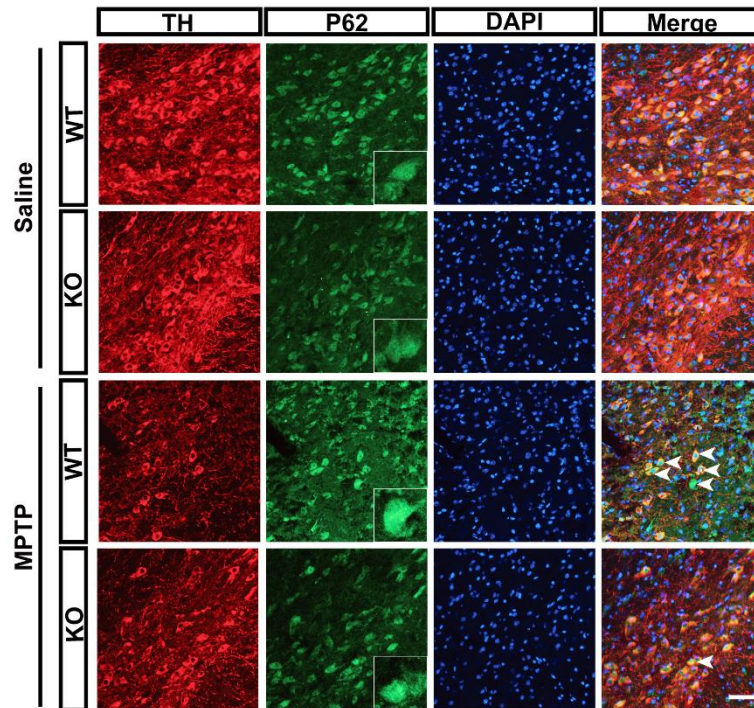


**Figure S4.** Immunofluorescence staining for TH (red) and P62 (green) in the substantia nigra derived from WT or Maged1 KO mice (induced or not induced with MPTP), nuclei were counterstained with DAPI (blue). Scale bar: 50  $\mu$ m. White arrow indicates P62-aggregated TH-positive neuron.

## Supplemental Tables

**Table S1. Antibodies**

| Target antigen       | Vendor or Source          | Catalog    | Working        |
|----------------------|---------------------------|------------|----------------|
| Maged1               | Proteintech               | 22053-1-AP | 1:2000 for WB; |
| P62/SQSTM1           | Proteintech               | 18420-1-AP | 1:1000 for WB; |
| Bcl2                 | Proteintech               | 12789-1-AP | 1:1000 for WB  |
| LC3A/B               | Cell Signaling Technology | 12741#     | 1:1000 for WB  |
| $\alpha$ -synuclein  | Cell Signaling Technology | 4179#      | 1:200 for IF   |
| Cleaved PARP         | Cell Signaling Technology | 5625#      | 1:1000 for WB  |
| Caspase-3            | Cell Signaling Technology | 9662#      | 1:1000 for WB  |
| $\beta$ 3-Tubulin    | Cell Signaling Technology | 5568#      | 1:1000 for IF  |
| Akt                  | Cell Signaling Technology | 9272S#     | 1:1000 for WB  |
| p-Akt (T308)         | Cell Signaling Technology | 9275S#     | 1:1000 for WB  |
| p-Akt (S473)         | Cell Signaling Technology | 9271T#     | 1:1000 for WB  |
| S6                   | Cell Signaling Technology | 2317S#     | 1:1000 for WB  |
| p-S6 (S235/236)      | Cell Signaling Technology | 2211S#     | 1:1000 for WB  |
| p70S6K               | Cell Signaling Technology | 2708#      | 1:1000 for WB  |
| p-p70S6K             | Cell Signaling Technology | 95796#     | 1:1000 for WB  |
| mTOR                 | Cell Signaling Technology | 2972#      | 1:1000 for WB  |
| p-mTOR (S2448)       | Cell Signaling Technology | 2971#      | 1:1000 for WB  |
| Tyrosine Hydroxylase | Sigma                     | T2928      | 1:1000 for WB; |
| Iba1                 | Wako                      | 019-19741  | 1:1000 for IF  |
| GFP                  | abcam                     | ab13970    | 1:200 for IF   |
| Bax                  | Bioworld Technology       | BS2538     | 1:1000 for WB  |
| Actin                | Bioworld Technology       | BS6007MH   | 1:1000 for WB  |
| GAPDH                | Bioworld Technology       | AP0063     | 1:1000 for WB  |

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|                         |                     |           |         |
|-------------------------|---------------------|-----------|---------|
| Goat Anti-Mouse IgG     | Bioworld Technology | BS10006   | 1:400   |
| Goat anti-Mouse IgG     | Bioworld Technology | BS50350   | 1:10000 |
| Goat anti-Rabbit IgG    | Bioworld Technology | BS13278   | 1:10000 |
| Donkey anti-Rabbit IgG  | Thermo Fisher       | SA5-10038 | 1:1000  |
| (H+L) Cross-Adsorbed    |                     |           |         |
| Alexa Fluor 488         | Jackson             | 703-545-  | 1:400   |
| AffiniPure Donkey Anti- |                     | 155       |         |

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**Table S2. Reagents**

| Description                        | Source / Repository |
|------------------------------------|---------------------|
| RIPA                               | Sigma-Aldrich       |
| DAPI                               | Sigma-Aldrich       |
| CCK-8                              | Dojindo             |
| DMEM/F12                           | KeyGEN BioTECH      |
| MPP <sup>+</sup>                   | Sigma               |
| MPTP                               | Sigma               |
| Laminin                            | Sigma               |
| pentobarbital                      | Sigma               |
| BCA protein quantitative kit       | ThermoFisher        |
| Lipofectamine-iMAX                 | ThermoFisher        |
| Lipofectamine 2000                 | ThermoFisher        |
| Protease Inhibitor Cocktail        | Thermo Fisher       |
| phosphatase inhibitors             | Thermo Fisher       |
| Accutase                           | ThermoFisher        |
| B27                                | ThermoFisher        |
| Glutamax                           | ThermoFisher        |
| Penicillin and Streptomycin        | ThermoFisher        |
| bFGF-2                             | Peprotech           |
| EGF                                | Peprotech           |
| cAMP                               | Peprotech           |
| GDNF                               | Peprotech           |
| BCA Protein Assay kit              | Beyotime            |
| polyvinylidene difluoride membrane | Millipore           |
| Neurobasal medium                  | ThermoFisher        |