Legends for Supplemental Figures

Supplemental Fig. 1
Overexpression of wild-type PINK1 using a plasmid vector protects PC3 cells from apoptosis induced by cisplatin (100 μM), arsenite (100 μM), H_2O_2 (1.2 mM), NaCl (300 mM), and tunicamycin (100 μM). The experiment was performed under conditions similar to those described in the legend to Fig. 1C. The vector was transfected 12 h prior to application of the agents. *Significantly different from the vehicle (-)-treated cells ($P<0.05$).

Supplemental Fig. 2
Regulation of Akt phosphorylation by PINK1. *A.* SH-SY5Y cells were transfected with plasmid vectors carrying GFP (p-GFP), wild type PINK1 (p-WT), and kinase dead mutant of PINK1 (p-KDD) 48 h before harvesting cells for Western blot analysis. *B.* Comparison of phosphorylation level of Akt between knockdown of PINK1 and overexpression of PINK1. SH-SY5Y cells were treated with indicated siRNAs for 72 h or with indicated adenovirus vectors for 48 h before harvesting cells for Western blot analysis.

Supplemental Fig. 3
Co-precipitation of overexpressed PINK1 with endogenous Akt protein. Forty-eight hours after infection with Ad-PINK1 (5 MOI), PINK1 (left panel) and Akt (right panel) were immunoprecipitated with anti-HA antibody and anti-Akt antibody, respectively, followed by Western blot analysis.

Supplemental Fig. 4
Presence of PINK1 in the cytoplasm. Forty-eight hours after infection with Ad-PINK1 (5 MOI), SH-SY5Y cells were immunostained either with anti-PINK1 antibody (left panel) or anti HA-tag antibody (right panel). The cells were co-stained with MitoTracker Orange for identifying mitochondria. Signals were only partly overlapped.

Figure S1

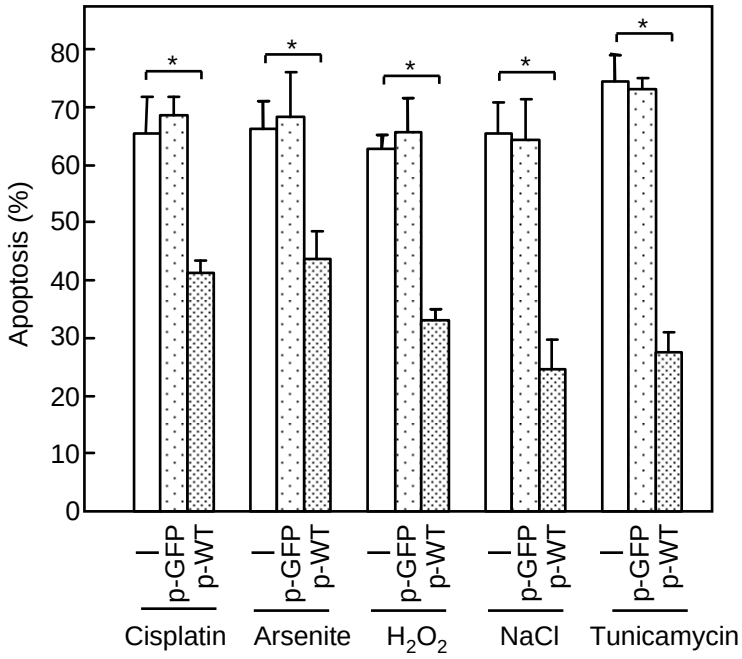

Apoptosis (%)
0
10
20
30
40
50
60
70
80
*
*
*
*
*
–
p-GFP
p-WT
Cisplatin
Arsenite
H_2O_2
NaCl
Tunicamycin

Figure S2

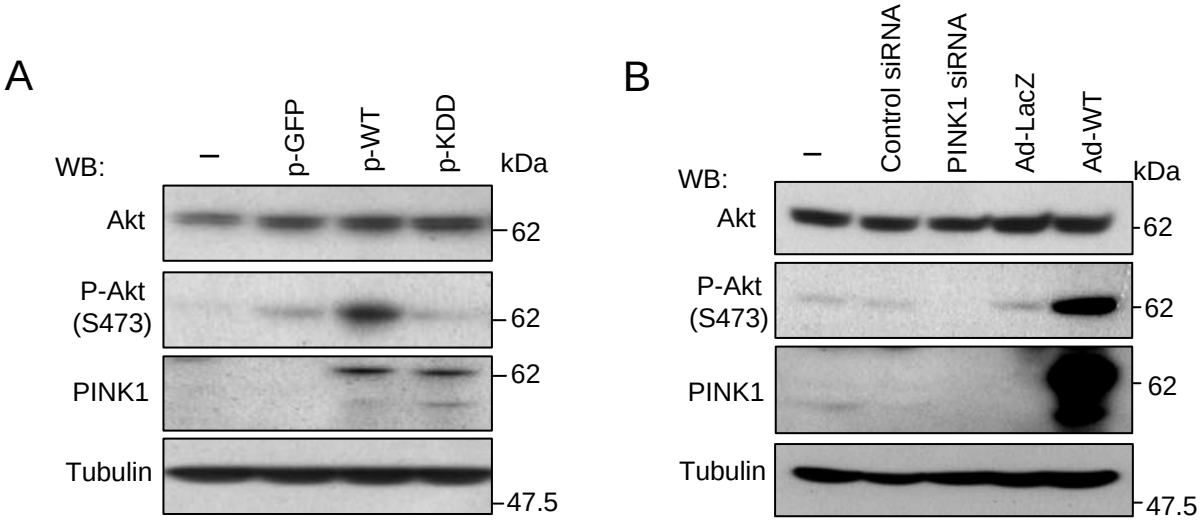

A
WB:
–
p-GFP
p-WT
p-KDD
kDa
Akt
62
P-Akt
(S473)
62
PINK1
62
Tubulin
47.5
B
WB:
–
Control siRNA
PINK1 siRNA
Ad-LacZ
Ad-WT
kDa
Akt
62
P-Akt
(S473)
62
PINK1
62
Tubulin
47.5

Figure S3

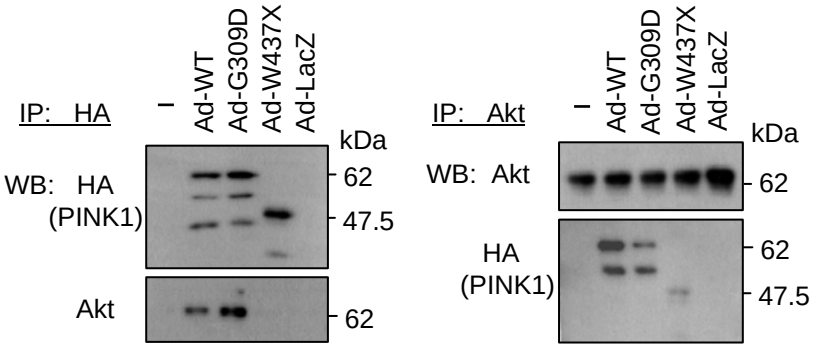
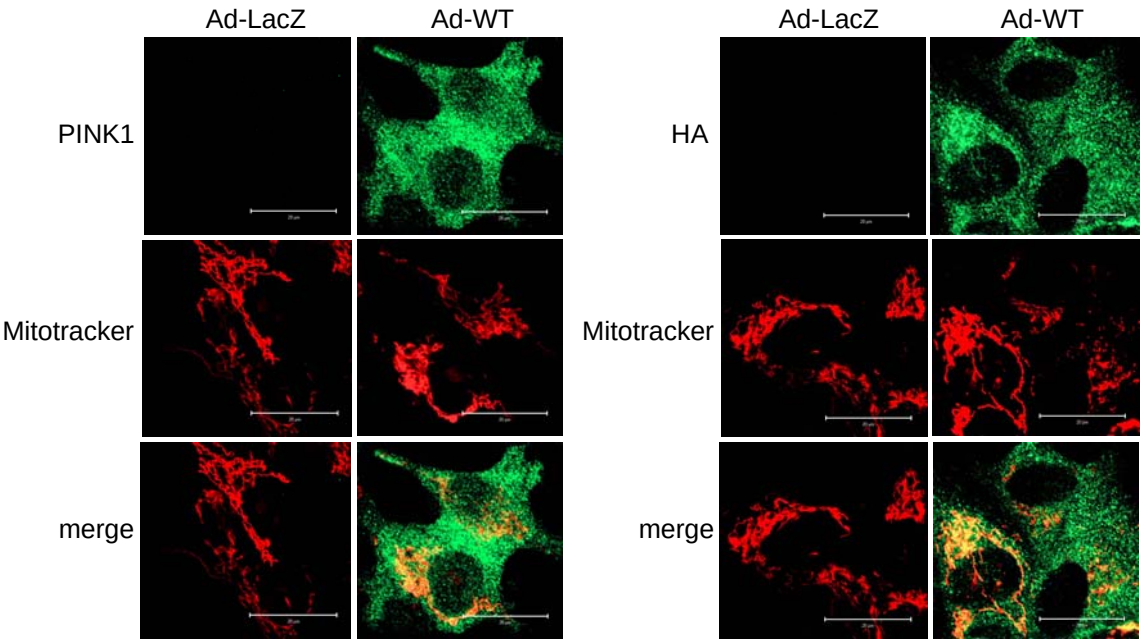

IP: HA
–
Ad-WT
Ad-G309D
Ad-W437X
Ad-LacZ
kDa
WB: HA (PINK1)
62
47.5
Akt
62
IP: Akt
–
Ad-WT
Ad-G309D
Ad-W437X
Ad-LacZ
kDa
WB: Akt
62
HA (PINK1)
62
47.5

Figure S4


Ad-LacZ
Ad-WT
PINK1
Mitotracker
merge
Ad-LacZ
Ad-WT
HA
Mitotracker
merge