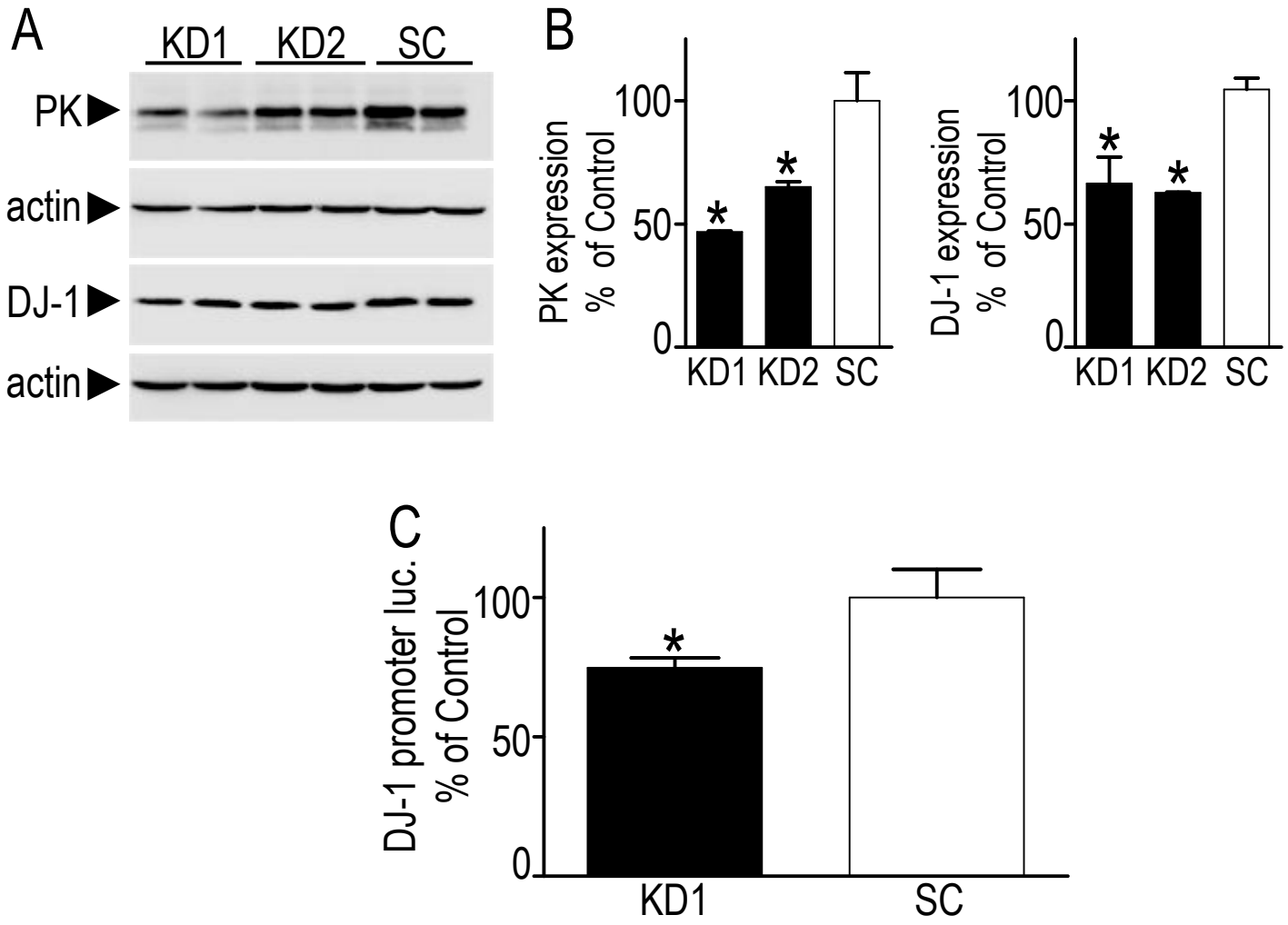

Fig. S1. The down-regulation of parkin levels by shRNA reduces DJ-1 expression and promoter transactivation. (**A**) Representative immunoblot showing the levels of parkin, DJ-1 and actin (loading control) in HEK293 cells transiently transfected with two shRNA (KD1 or KD2) targeting human parkin or with the control KD1 scramble shRNA (SC). (**B**) Densitometric analysis of parkin and DJ-1- like immunoreactivities normalized by actin signal and expressed as percent of control immunoreactivities recovered in SC-transfected cells. Bars represent the means ± SEM of 2-4 replicates. (**C**) Analysis of human DJ-1 promoter transactivation monitored as described in the "Methods". Bars correspond to activities expressed as percentage of the control SC-transfected cells and are means ± SEM of 4-6 replicates. **P*<0.05.

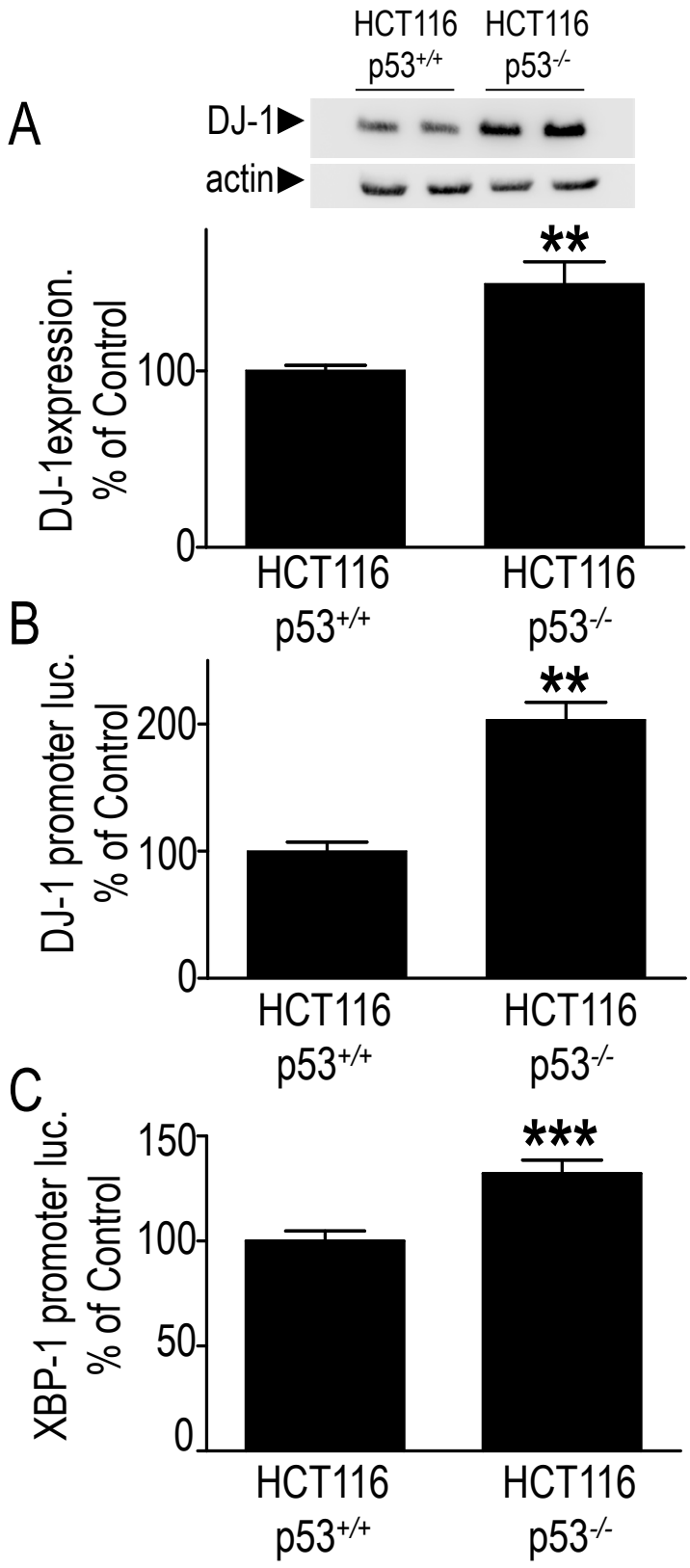

Fig. S2. Depletion of endogenous p53 increases DJ-1 expression and DJ-1 and and XBP-1 promoter transactivation in human colorectal carcinoma cells (HCT116). (A) Analysis of DJ-1 expression in wild-type HCT116 cells or *p53 null* HCT116 cells as described in Methods and represent the means ± SEM of 3-6 replicates. Bars are expressed as percent of control promoter activities measured in TPS-treated HCT116 $p53^{+/+}$ and represent the means ± SEM of 3-6 replicates. **(B-C)** Wild-type HCT116 cells or *p53 null* HCT116 cells were transiently transfected with human DJ-1- (A) or mouse XBP-1- (B) luciferase promoter constructs then treated with thapsigargin (TPS, 1μM, 16 hours). Twenty-four hours after transfection, promoter transactivation analyses were performed as described in the "Methods". Bars are expressed as percent of control promoter activities measured in TPS-treated HCT116 $p53^{+/+}$ and represent the means ± SEM of 3-6 replicates. $^{**}P<0.01$, $^{**}P<0.001$.

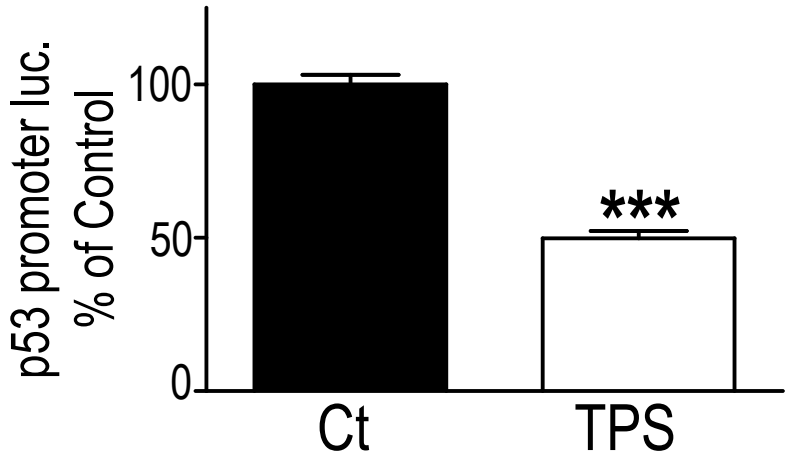

Fig. S3. Thapsigargin lowers p53 promoter transactivation in human embryonic kidney cells (HEK293). HEK293 cells were transiently transfected with human p53 promoter and treated with thapsigargin (TPS, 1μM, 16 hours) or with the equivalent volume of DMSO (Ct, control vehicle). Twenty-four hours after transfection, promoter transactivation analyses were performed as described in the "Methods". Bars are expressed as percent of control and are means ± SEM of 12 replicates. ***P<0.001.

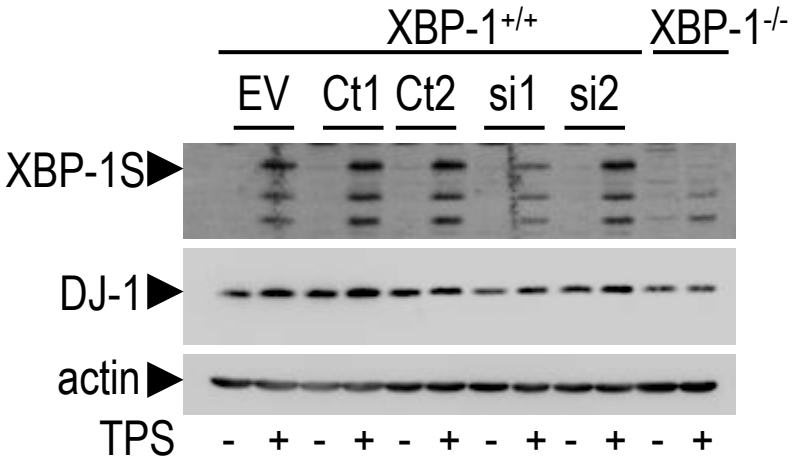

Fig. S4. shRNA-induced reduction of XBP-1S levels lowers DJ-1 expression. The XBP-$1^{+/+}$ fibroblasts were transfected with an empty vector (EV) or with the indicated controls (Ct1 and Ct2) or two previously described (see supplementary material Table S3) XBP-1S-directed (si1 and si2) shRNA. Twenty-four hours after transfection, cells were treated for 8 hours with thapsigargin (TPS, 300nM) then harvested and analyzed for XBP-1, DJ-1 and actin expressions. The gel represents a typical immunoblot showing the levels of XBP-1S, DJ-1 and actin in XBP-$1^{+/+}$ or XBP-$1^{-/-}$ (used as control of XBP-1S depletion).

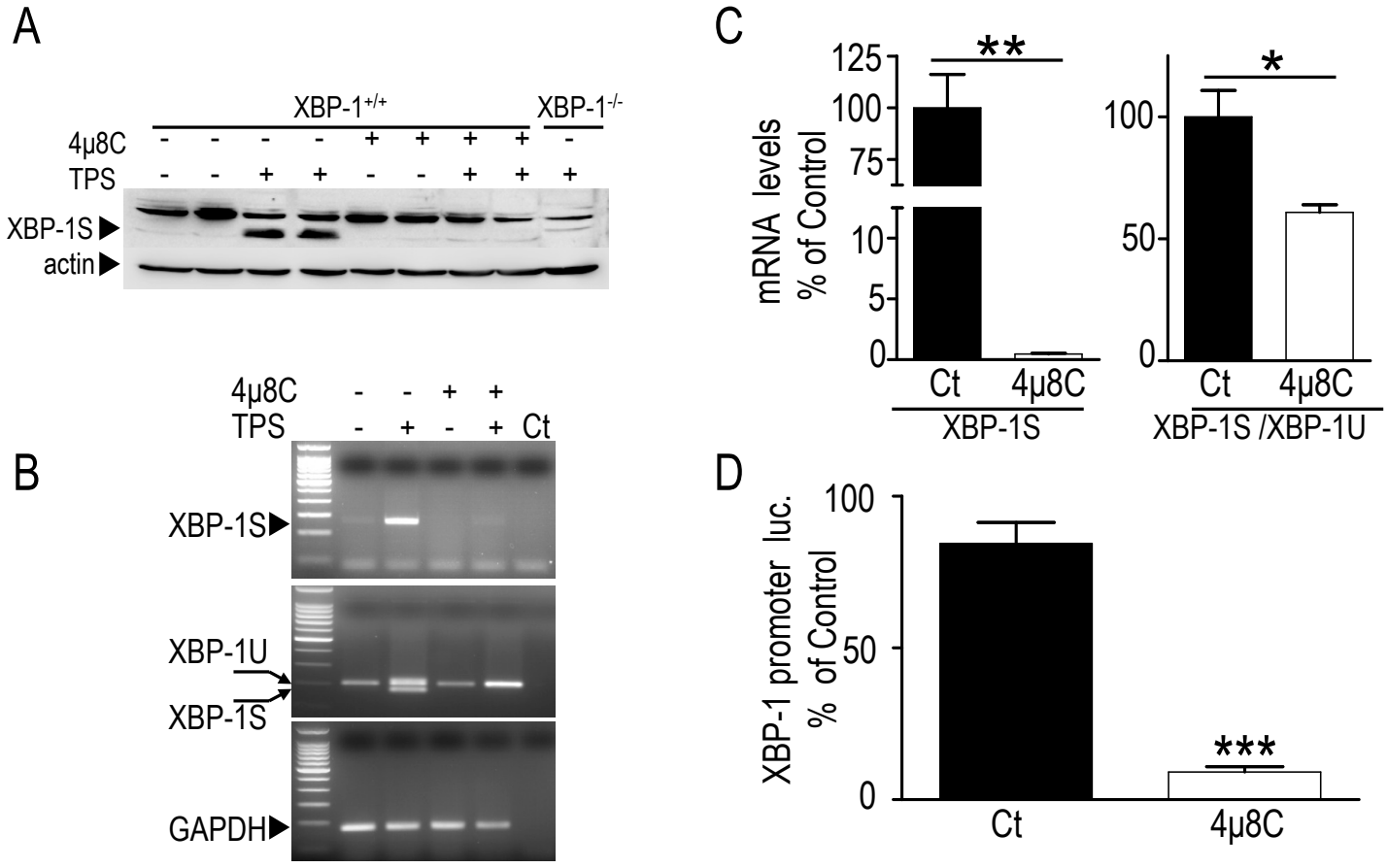

Fig. S5. The IRE-1α inhibitor 4μ8C reduces XBP-1S expression, mRNA levels and promoter transactivation. (A) A representative immunoblot showing the expression of the spliced form of XBP-1 (XBP-1S) in wild-type (XBP-$1^{+/+}$) or XBP-1-deficient fibroblasts (XBP-$1^{-/-}$) treated overnight with (DMSO: control vehicle), TPS (1μM), 4μ8C (50μM) or with both TPS and 4μ8C. Three independent experiments were performed in 2 replicates. (**B**) RT-PCR analysis of XBP-1S (upper panel), total XBP-1 mRNA expression (middle panel) and GAPDH (lower panel) in SH-SY5Y cells treated as above. Two independent experiments were performed in 3 replicates. (**C**) XBP-1S (left panel) and total XBP-1 (right panel) mRNA levels obtained as described in the "Methods" in SH-SY5Y cells treated as above. Bars are expressed as percent of control Ct and are the means ± S.E.M of 6 replicates. (**D**) Analysis of mouse XBP-1 promoter transactivation in the above conditions was performed as described in Methods. Data are expressed as percentage of control (Ct) and are the means ± S.E.M of 6 replicates. **P*<0.05, ***P*<0.01, ****P*<0.001.

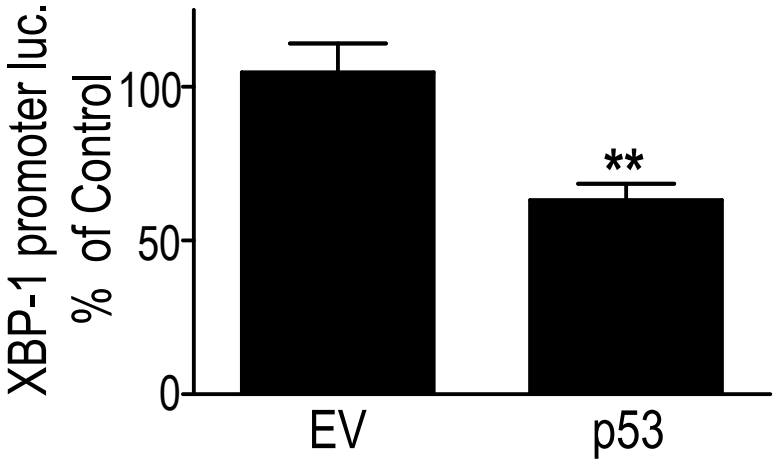

Fig. S6. p53 overexpression represses XBP-1 promoter transactivation in human embryonic kidney cells (HEK293). HEK293 cells were transiently transfected with the mouse XBP-1 promoter and either an empty vector (EV) or the cDNA of human p53 protein (p53). Twenty-four hours after transfection, XBP-1 promoter transactivation analysis was performed as described in the "Methods". Bars are expressed as percent of EV-transfected cells and correspond to means ± S.E.M of 3 independent experiments performed in 3 replicates. ***P*<0.01.

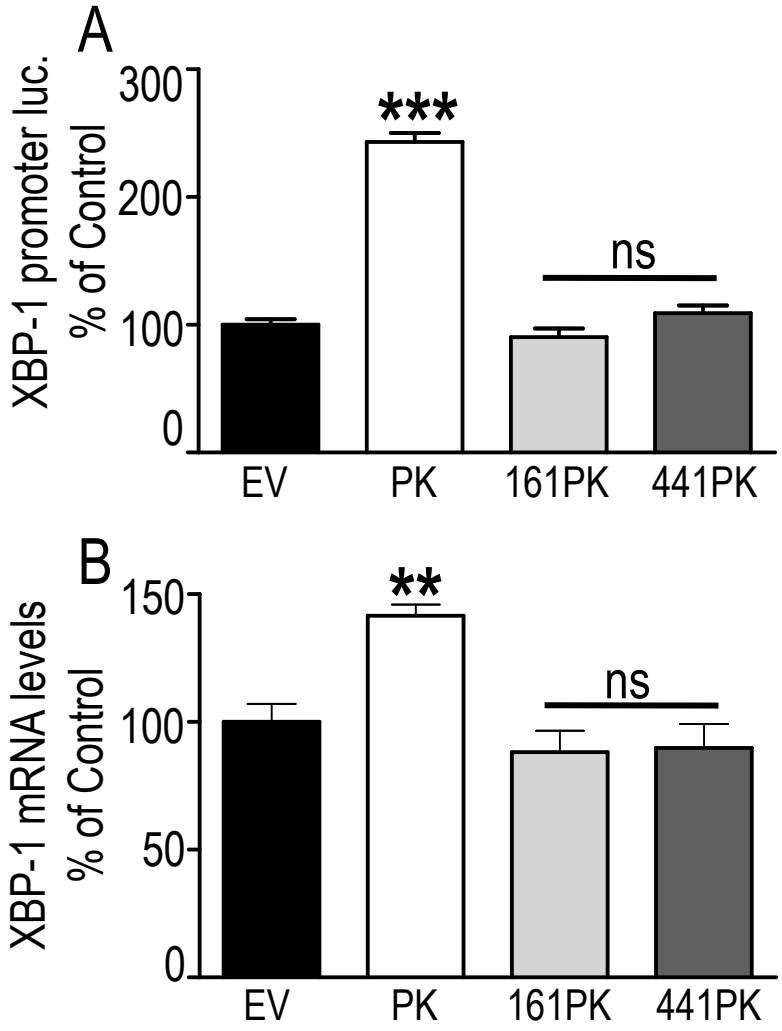

Fig. S7. Familial Parkinson's disease associated mutations abolish parkin ability to up-regulate XBP-1 promoter transactivation and mRNA levels in HEK293 cells. (A,B) HEK293 human embryonic kidney cells were stably transfected with the indicated cDNAs then mouse XBP-1 promoter transactivation analysis (A) and mRNA levels (B) were performed as described in the "Methods". Bars are expressed as percent of EV-transfected cells and are the means ± S.E.M of 3-6 replicates. ***P*<0.01, ****P*<0.001, ns: not statistically significant.

Table S1. Vectors and sequences used for shRNA, 3′UTR-luciferase constructs and microRNA strategy.

We based our primer design on previous studies (Veeriah et al.,2010; and Lee et al; 2003). Note: primer position is relative to the transcription start site.

Name	Sequence	Position/Reference	Species
H1-XBP1i#1 (control)	5′-TCTTAAAGGTGGTAGTATA-3′	nt.1370-1388	mouse
H1-XBP1i#2 (control)	5′-GGATTCATGAATGGCCCTT-3′	nt.1558-1576	mouse
U6-XBP-1i#4	5′-GGATTCATGAATGGCCCTTA-3′	nt.1558-1577 at 3′UTR (Lee et al., 2003)	mouse
U6-XBP-1i#5	5′-GGTCTGCTGAGTCCGCAGCA-3′	nt.463-483, against human and mouse	mouse/human
pSUPER.puro	empty vector	OligoEngine	
PARK2 KD-1	5′-AGTGCCGTATTTGAAGCCTCAGGAACAAC-3′	(Veeriah et al., 2010)	human
PARK2 KD-2	5′-AAATGGAGGCTGCATGCACATGAAGTGTC-3′	(Veeriah et al., 2010)	human
PARK2 KD-1-scramble	5′-GCGGATGATCTGAACAGCATTCGAACTCA-3′		human
pSUPER.neo+GFP	empty vector	OligoEngine	

1.

Table S2. Primers used for mutagenesis. The mutations have been described in previous studies (Abbas et al., 1999; Bertoli-Avella et al., 2004; West et al., 2002). All the constructs have been made in the pGL2 basic vector (Promega) and verified by sequencing. The mutated nucleotides are underlined and in bold. The position of deleted nucleotides is indicated in brackets in the indicated sequences.

Name	Sequence	Domain targeted by mutation/reference
hParkin K161N forward	5′-GAGTGCAGCCGGGAAA**T**CTCAGGGTACAGTGC-3′	Domain SH2-like (Abbas et al., 1999)
hParkin K161N reverse	5′-GCACTGTACCCTGAG**A**TTTCCCGGCTGCACTC-3′	
hParkin C418R forward	5′-CCACCAAGCCCC**C**GTCCCCGCTGCCATG-3′	Domain R2 (Bertoli-Avella et al., 2004)
hParkinC418R reverse	5′-CATGGCAGCGGGGAC**G**GGGCTTGGTGG-3′	
hParkin C441R forward	5′-CCGCAGCCCCAG**C**GCAGGCTCGAGTGG-3′	Domain R2 (West et al., 2002)
hParkin C441R reverse	5′-CCACTCGAGCCTGC**G**CTGGGGCTGCGG-3′	
hp.DJ-1ΔXBP-1 forward	5′-TCGGCCG(-----)GCAGCGTGAGGCCAAGGC-3′	Deletion of XBP-1 binding site (−79/−74) in the human DJ-1 promoter. 5′-GACGTGAC-3′/3′-CTGCACTG-5′
hp.DJ-1ΔXBP-1 reverse	5′-ACGCTGC(-----)CGGCCGAGTCTACCTCGG-3′	
GL primer 1 forward	5′-TGTATCTTATGGTACTGTAACTG-3′	Forward primer for pGL2 basic vector
GL primer 2 reverse	5′-CTTTATGTTTTTGGCGTCTTCCA-3′	Reverse primer for pGL2 and pGL3 basic vectors
RV primer 3 forward	5′-CTAGCAAAATAGGCTGTCCC-3′	Forward primer for pGL3 basic vector
mp.XBP-1Δp53 forward	5′-GTGGGGGGGGGGG(-----)AGCGCCGGACATTAAAATGGGG-3′	Deletion of p53 putative binding site (−373/−354) in the mouse XBP-1 promoter. 5′-GGGGACGGGACGGGGGACGT-3′ 3′-CCCCTGCCCTGCCCCCTGCA-5′
mp.XBP-1Δp53 reverse	5′-CCCCATTTTAATGTCCGGCGCT(-----)CCCCCCCCCCCAC-3′	
mp.XBP-1 fgt(1) 581 forward	5′-CGAGCTCGTGCTGAGATTACAAGTGAGTACTACC-3′	(581 nucleotides) contain a SacI site
mp.XBP-1 fgt(2) 438 forward	5′-CGAGCTCCTGCTGTACTGGGAAGTGGG-3′	(438 nucleotides) contain a SacI site
mp.XBP-1 fgt(3) 285 forward	5′-CGAGCTCGTCTGCCGAAGATCCTTGGAC-3′	(285 nucleotides) contain a SacI site
mp.XBP-1 fgt(4) 139 forward	5′-CGAGCTCCCAATAAGTGATGAATATACCCGCGCG-3′	(139 nucleotides) contain a SacI site
mp.XBP-1fgts reverse	5′-CCCAAGCTTCACCACCATAGCCAG-3′	contain a HinDIII site

Table S3. Primers used for quantitative and/or semi quantitative PCR. We based our primer design on previous studies (Acosta-Alvear et al., 2007; Iwawaki et al., 2004)

Name	Sequence of forward primer	Sequence of reverse primer	Species/Reference
mDJ-1	5′-GGAGATGCAAAAACGCAGGG-3′	5′-TCCTCCTGGAAGAACCACCA-3′	mouse
mActin	5′-CACCATCGGTTGTTAGTTGCC-3′	5′-CAGGTGTCGATGCAAACGTT-3′	mouse
hGAPDH	5′-TGGGCTACACTGAGCACCAG-3′	5′-CAGCGTCAAAGGTGGAGGAG-3′	human
mTop1	5′-TGCCTCCATCACACTACAGC-3′	5′-CGCTGGTACATTCTCATCAGG-3′	mouse
hXBP-1	5′-GGAGTTAAGACAGCGCTTGG3′	5′-CACTGGCCTCACTTCATT-CC-3′	human
hTOP1	5′-CCCTGTACTTCATCGACAAGC-3′	5′-CCACAGTGTCCGCTGTTTC-3′	human
hDJ-1	5′-GCCTGATTCTTACAAGCCGG-3′	5′-CAAGCGCAAACTCGAAGCT-3′	human
mXBP-1	5′-TGACGAGGTTCCAGAGGTG-3′	5′-TGCAGAGGTGCACATAGTCTG-3′	mouse
hXBP-1 spliced	5′-AGCTTTTACGAGAGAAAACTCA-3′		human (based on the strategy described previously (Acosta-Alvear et al., 2007))
m/hXBP-1 spliced		5′-GCCTGCACCTGCTGCG-3′	mouse/human (based on the strategy described previously (Acosta-Alvear et al., 2007))
hXBP-1 com	5′-GAACCAGGAGTTAAGACAGCG-3′	5′-ATCTGAAGAGTCAATACCGCC-3′	human (based on the strategy described previously (Iwawaki et al., 2004))
mDJ-1	5′-GGATGCTGGGAACCGAACCTGGGTC-3′	5′-CTGGGCTACAGCCTGAACCCCACAC-3′	mouse