

Supplementary Material

Chromatin-bound oxidized α -Synuclein causes strand breaks in neuronal genomes in *in vitro* models of Parkinson's disease

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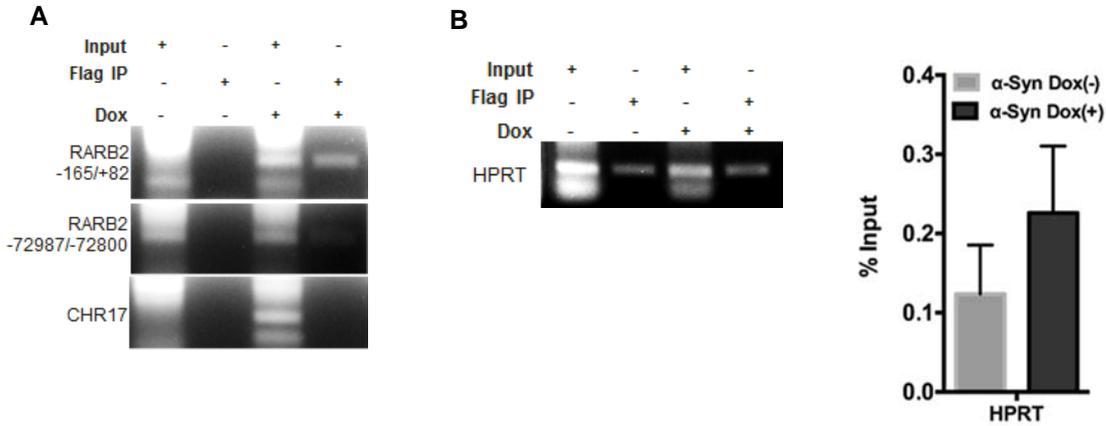
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Supplementary Material includes one table and three figures.

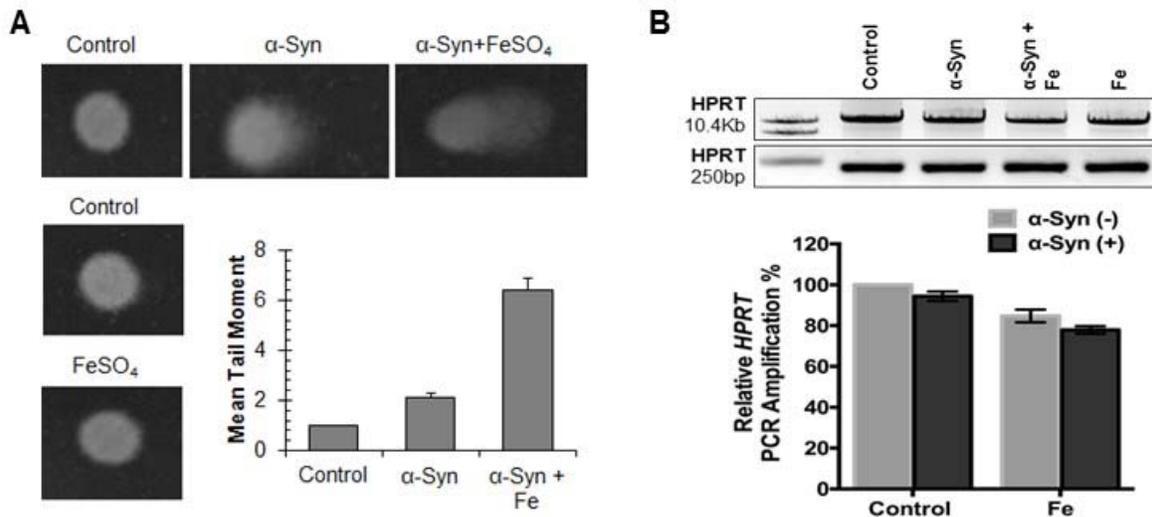
Supplementary Table 1. Primer oligonucleotide sequences used for quantitative real time PCR amplification of ChIP products in Fig. 1D and Fig. S1.

Primer sequences used in ChIP analysis	
RAR β 2 promoter (-165/+82) F	5'- CTCTGGCTGTCTGCTTTTGC-3'
RAR β 2 promoter (-165/+82) R	5'-CATGGGGGAATTCTGGTCCC-3'
RAR β 2 distal (-72987/-72800) F	5'-TGGTTAGGTTTGGGCGTGTT -3'
RAR β 2 distal (-72987/-72800) R	5'-GGGGCTATGACTGAAGTGCT-3'
Chromosome 17 F	5'-TACTATCCCCGTGCTTCCCA-3'
Chromosome 17 R	5'-CATTGAGGAGGGGGCAACAT-3'
HPRT F	5'-TGACCTTGATTTATTTTGCATACC-3'
HPRT R	5'-CGAGCAAGACGTTTCAGTCCT-3'

Supplementary Fig. S1. (Linked to Fig. 1D). Chromatin Immunoprecipitation (ChIP) using FLAG antibody from pCW-iFLAG- α -Syn SHSY-5Y cells after Dox induction (72 h) and PCR amplification of indicated genomic segments using three randomly selected primer pairs. The PCR products were separated in 1% agarose gel electrophoresis. (A) Representative agarose gel separation of ChIP-PCR products for proximal and distal regions of RARB2 and a randomly selected region in chromosome (CHR) 17 and HPRT (B). Representative agarose gel image (left) and histogram of RT-PCR analysis for HPRT gene (right) are shown. Other details are in Material and Methods.



Supplementary Fig. S2. (Linked to Fig. 2). (A) Alkaline Comet assay from SHSY-5Y cells treated with 300 nM recombinant α -Syn with or without 200 μ M FeSO₄. A synergistic increase in mean comet tail moment was observed by α -Syn + Fe. FeSO₄ alone at the same concentration causes negligible genome damage. (B) Amplification of *HPRT* gene by LA-PCR assay from neural progenitor cells treated with 300 nM recombinant α -Syn with or without 200 μ M FeSO₄. Cells treated with both α -Syn and Fe showed ~22% decrease in amplified product, indicating increased DNA strand breaks. Whereas, α -Syn or Fe alone caused negligible DNA damage.



Supplementary Fig. S3. (Linked to Fig. 10). Protein DNA docking models demonstrating binding of α -Syn (PDB: 1XQ8) N-terminal amino acid residues to the crystal structure of B-DNA duplex (PDB:3IXN). **(A)** Model 2 shows α -Syn amino acid residues Lys-45, Val-55, Lys-32, Glu-28 interaction with BDNA. **(B)** Model 3 shows α -Syn amino acid residues Lys-43, Thr-54, Val-55, Lys-58. Model 2 and Model 3 represents the second and third best structures from the bigger cluster after refinement.

