



**Figure S1 Cartoon Illustration of Primers Used for PCR Validation in of HCT116<sup>ΔNQO2</sup> Cells.**

PCR amplification of wild-type NQO2 gene using primers Exon4\_F1 and Exon4\_R1 will generate a PCR product of 425bp, while the PCR product of the knockout gene is estimated to be 363bp if exonuclease excise single stranded nucleotides up to the targeted cut sites. PCR amplification of the 5' flank and 3' flank regions in genomes where full-length PCR products are absent will show that the two ends of exon4 still exist in the genomic DNA. This suggests that a chromosomal translocation event has occurred.

**Table S1 Oligonucleotides Used for CRISPR Cassette Construct.**

Guide RNA oligos	Sequence
Exon4_46_top	CACCGACCTTGCTTGTAGGCTTCG
Exon4_46_bottom	AAACCGAACCTACAAGCAAAGGT
Exon4_108_top	CACCGTGAGCAGAAAAAGGTTGGG
Exon4_108_bottom	AAACCCGAACCTTTCTGCTCA

**Table S2 Oligonucleotides Used for PCR Validation of HCT116<sup>ΔNQO2</sup> cells**

Primers	Sequence
Exon4_F1	TGCTAGGTAGCAAGTGCTCAATC
Exon4_R1	CTTCCAGAACGCAGCACAAAACTC
Exon4_F2	CTGACCTAGTGTATTTCAGGTTGTTTCTC
Exon4_R2	GTTTCCACTCCATAATTGAAAACCTCAGG
46oF1	ATGGAAGCAGGAACCTAAACTCA
46oR1	GTACATGCTGGCTAGAAAACAC
108oF1	AGTAGGACATCACAGAAGTTGGG
108oR1	TTCTGCTCAAAGGTTCGCC