

Supplementary Figures and Figure Legends

Fig. S1 The colocalization of KDM6A with RPA1 in HeLa cells treated with CPT.

Representative images with scale bars (5 μm) are shown.

Fig. S2 KDM6A could recruit Ku70 protein on nascent DNA through TPR domain.

(A) CPT treatment could enhance the interaction of KDM6A with Ku70. Co-IP was performed in cells treated with CPT followed by western blot with indicated antibodies.

(B) The association of KDM6A with Ku70 in HeLa cells treated with CPT. Representative images with scale bars (5 μm) are shown. (C) Detection of the

interaction between SND1 either with Ku70 or PCNA. SND1 was immunoprecipitated using specific antibodies against SND1, followed by western blot with indicated

antibodies. (D) Co-IP confirmed that TPR domain mediated the interaction of

KDMA6A and Ku70. (E) SND1 knockdown attenuated the enrichment of KDM6A and Ku70 on nascent DNA which could be restored by wild-type SND1 and TSNA Δ truncation.

Fig. S3. KDM6A regulated the stability of replication forks through SUMOylation.

(A) Predicted SUMOylation sites in KDM6A protein. (B) BrdU and EdU were incorporated into nascent DNA for observing the formation of replication forks foci in HeLa cells. The histogram showed the number of replication foci formation in indicated cells. * $p < 0.05$.

Fig. S4. The characteristic of KDM6A expression in various tumors.

(A) A pancancer analysis of KDM6A expression using TCGA data base. (B) KDM6A expression and mutations in ESCC was analyzed using data from public database Cbioportal. * $p < 0.05$, ** $p < 0.01$. *** $p < 0.001$.

Fig. S5. KDM6A mutation involved in the genomic stability of ESCC.

(A) Cellular viability in the presence of genotoxin CPT for ESCC Kyse150 cells knocked down

endogenous KDM6A and followed by transfection with the plasmid of KDM6Awt or KDM6A mutants respectively. (B) Western blot for phospho-RPA32^{S4/S8} and γ H2AX detection in ESCC cells lacking KDM6A with KDM6Awt or KDM6A mutant plasmid transfection respectively.

Figure S1

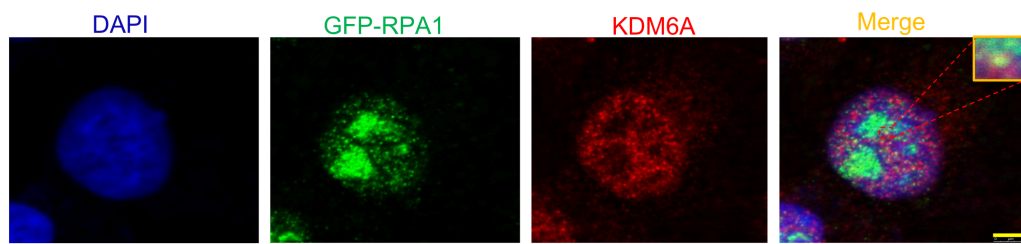


Figure S2

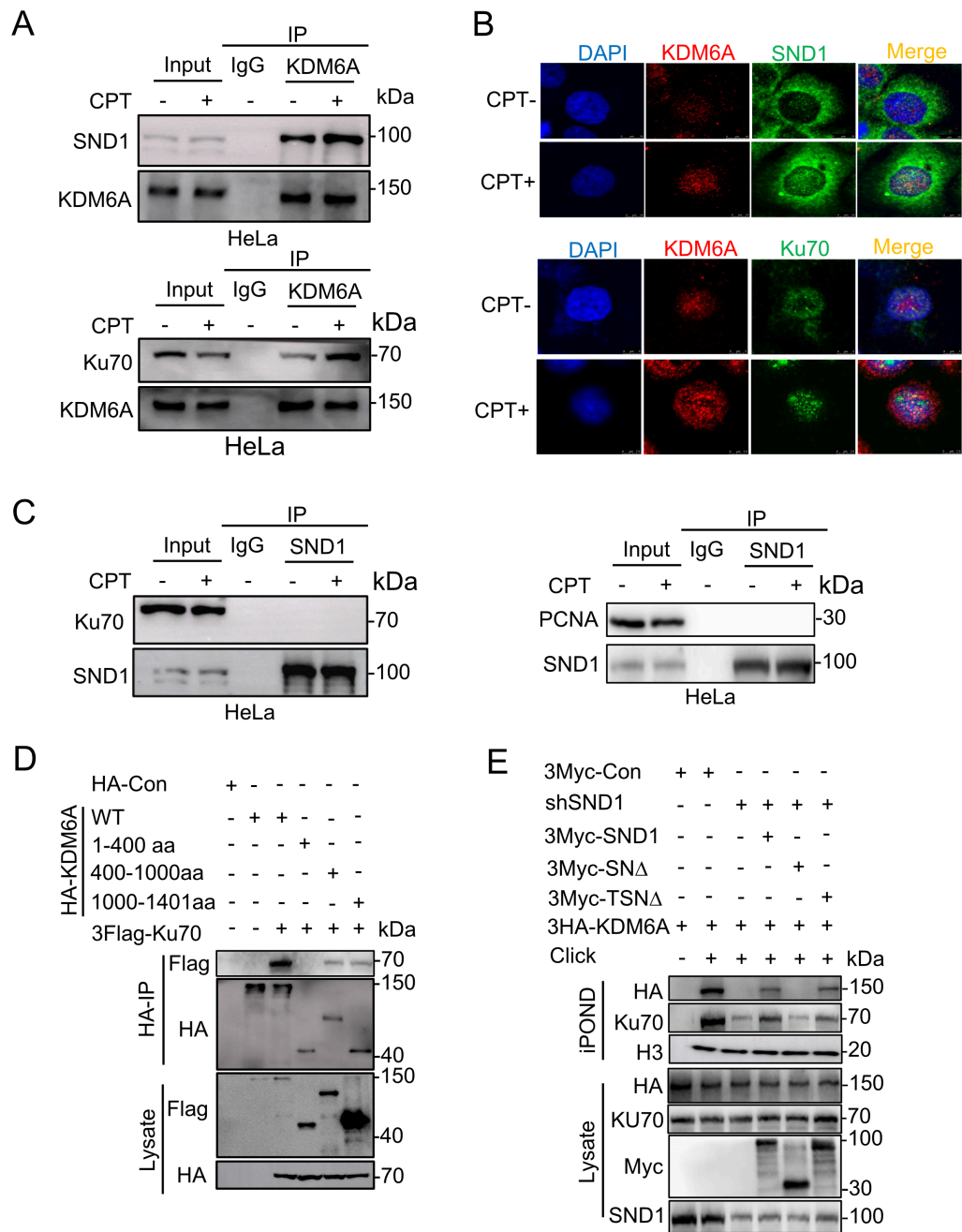


Figure S3

A

Results for putatifs SUMO site									
Position K	Sequence	Best PS	Consensus direct			Consensus Inverted			
			Type	PSd	DB Hit	Type	PSi	DB Hit	
K2	-----MKSCGVSLATAA	None	None	None	None	None	None	None	1
K23	AAAAAFGDEEKGGMAAGKASGE	Low	None	None	None	None	Low	None	1
K24	AAAAFGDEEKGMPLAGKASGES	Low	None	None	None	Consensus inv	Low	None	1
K86	AVRCYESLIKAEKVESDEF	Low	NDSM	Low	8	None	None	None	1
K90	YESLILKAEKVESDFFCQLG	High	Weak Consensus	None	2	Strong consensus inv	High	None	1
K111	HFNLLLEDYFKALSAYQRYYS	None	None	None	None	None	None	None	1
K151	FHYNAFQWAIKAEFOEVLVDF	None	None	None	None	None	None	None	1
K242	LQTNLSAQVKAATVLQQLGM	None	None	None	None	None	None	None	2
K265	TVDLLGDKATKESVAIQYLQK	None	None	None	None	None	None	None	1
K275	KESVAIQYLQKSLADPNSGQ	None	None	None	None	None	None	None	1
K299	FLGRCYSSIGKVDAFISYRQ	None	None	None	None	None	None	None	2
K313	AFISYRQSIDKSEASADTWCS	None	None	None	1	None	None	None	1
K716	SGGQQGITLTKESKFSGNILT	None	None	None	None	None	None	None	1
K719	QQGITLIKESKSGNILLTPE	High	None	None	None	Consensus inv	High	None	1
K867	EGMEESQSPKTDLLLVNHKF	Low	Consensus	Low	None	None	None	None	1
K905	EVLKACRNLGKNGLSNSSL	None	None	None	None	None	None	None	1
K978	TVIRGLAGAKLGLGFSTKT	Low	Strong Consensus	Low	None	None	None	None	1
K1076	TSSDNSGRRRKGFFKTIKFGI	None	None	None	None	None	None	None	1
K1095	GTNIDLSDKRWLQLHELTK	None	None	None	None	Consensus inv	None	None	1
K1265	RYENKIQSVKSLVFMVHLSW	None	None	None	None	None	None	None	1
K1287	MARNIKVSDPKLFEMIKYCLL	Low	None	None	None	Strong consensus inv	Low	None	1
K1301	MIKYCLRLTKQCOTLREALI	None	None	None	None	None	None	None	3
K1324	GKEIIWHGRTEKPEAHYCSIC	None	None	None	1	None	None	None	1
K1379	LENFVVLEQYKMEDLMQVYDQ	None	Weak Consensus	None	None	None	None	None	1

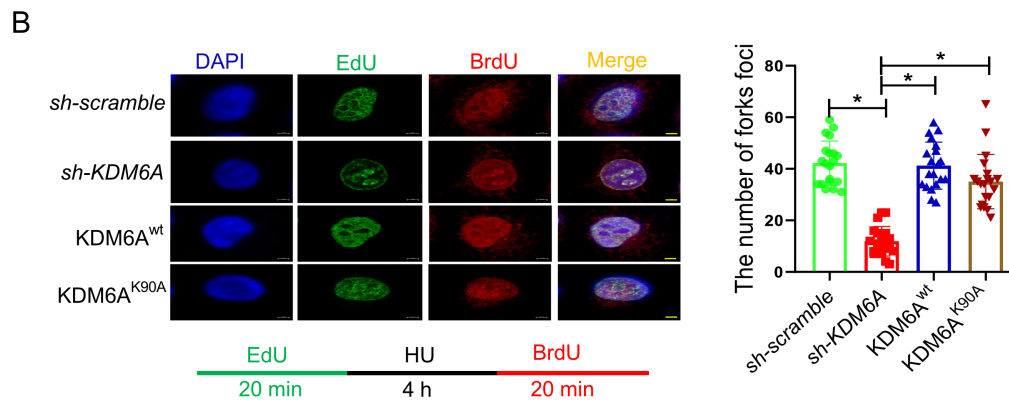


Figure S4

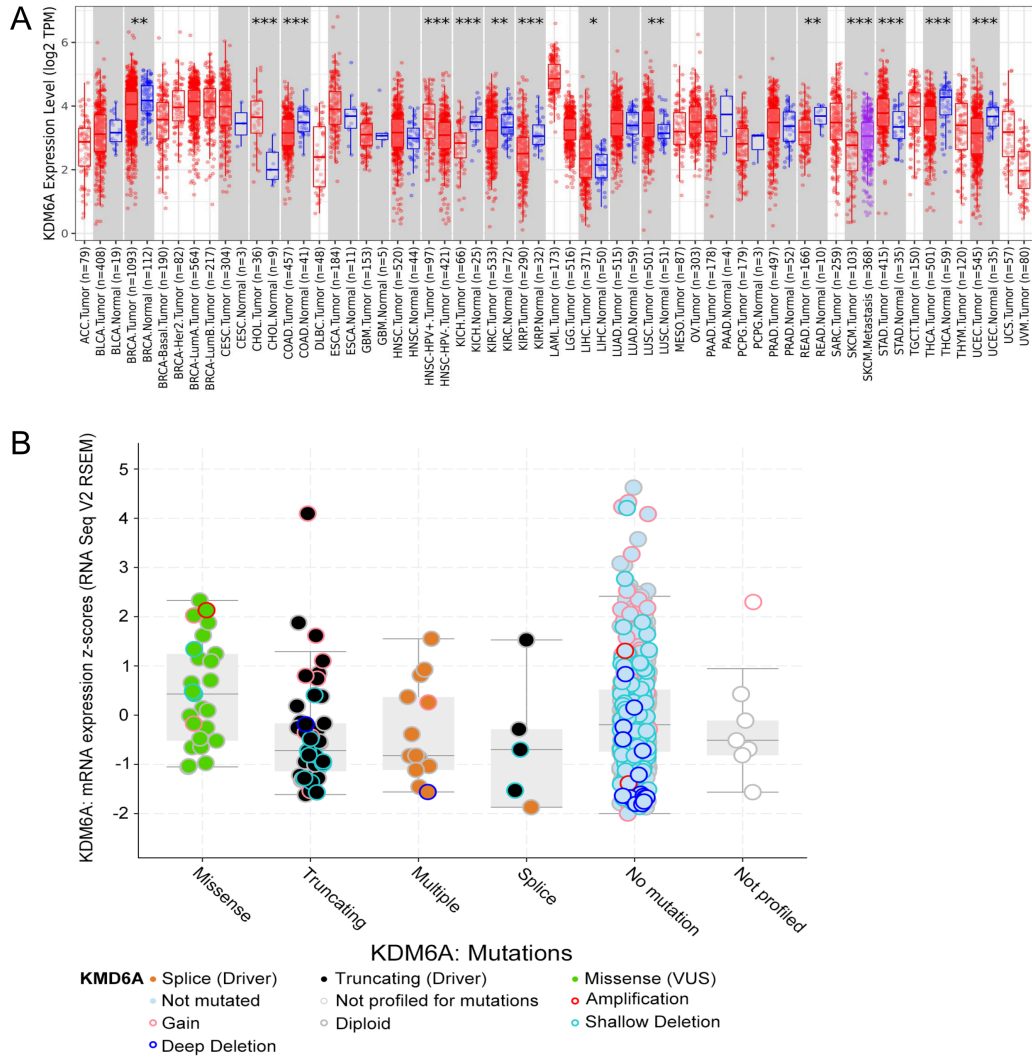


Figure S5

