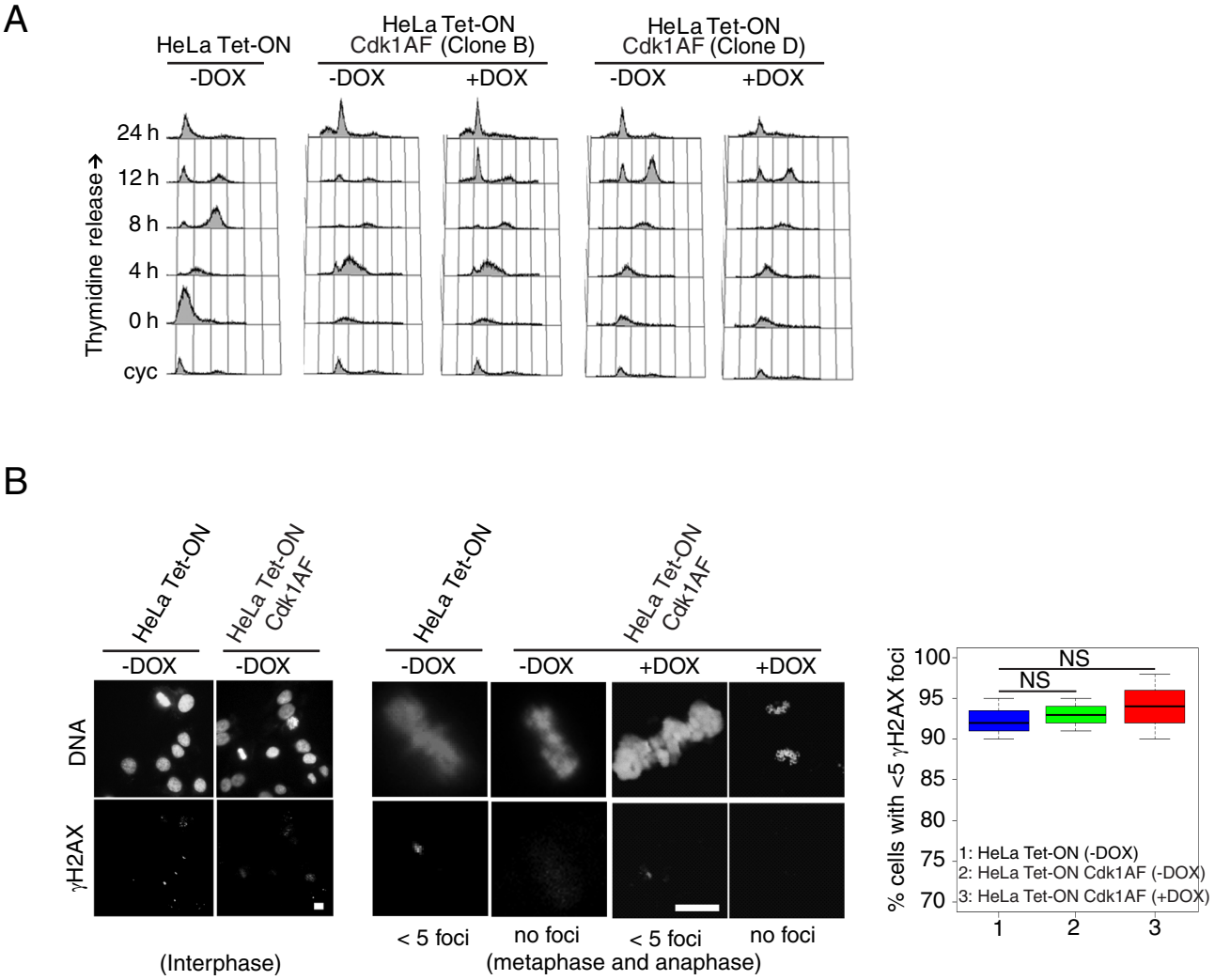
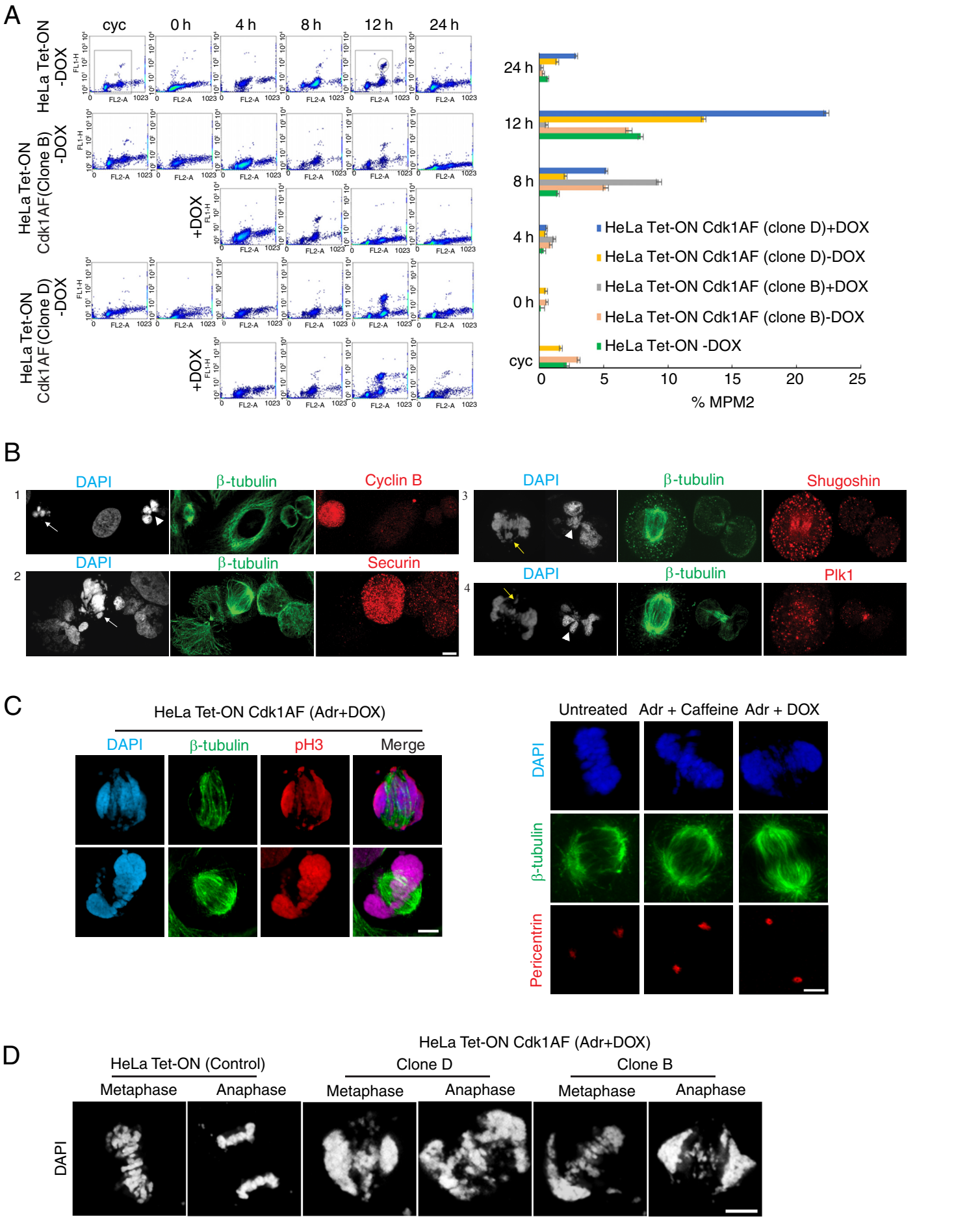
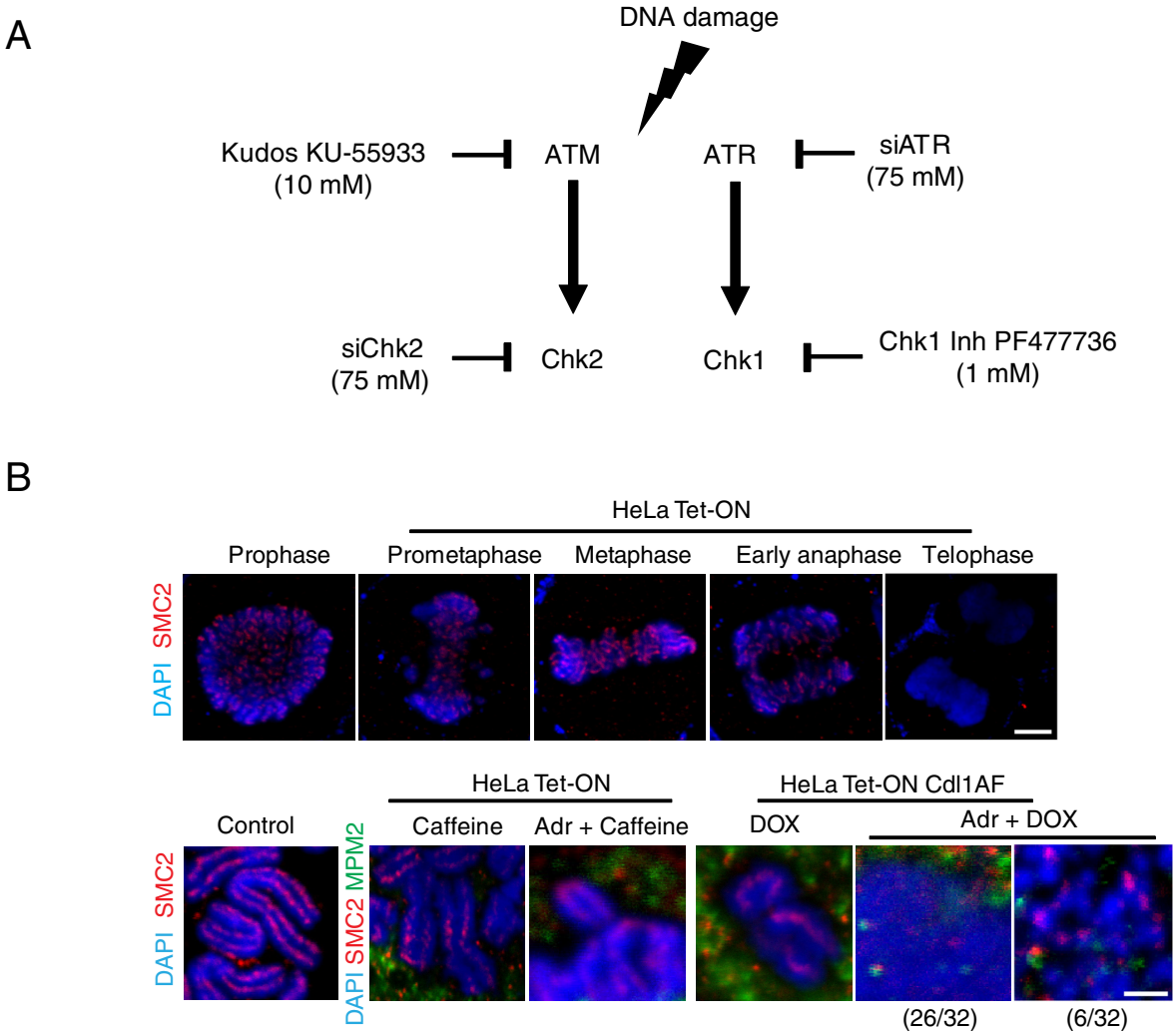
Supplementary Figure S1



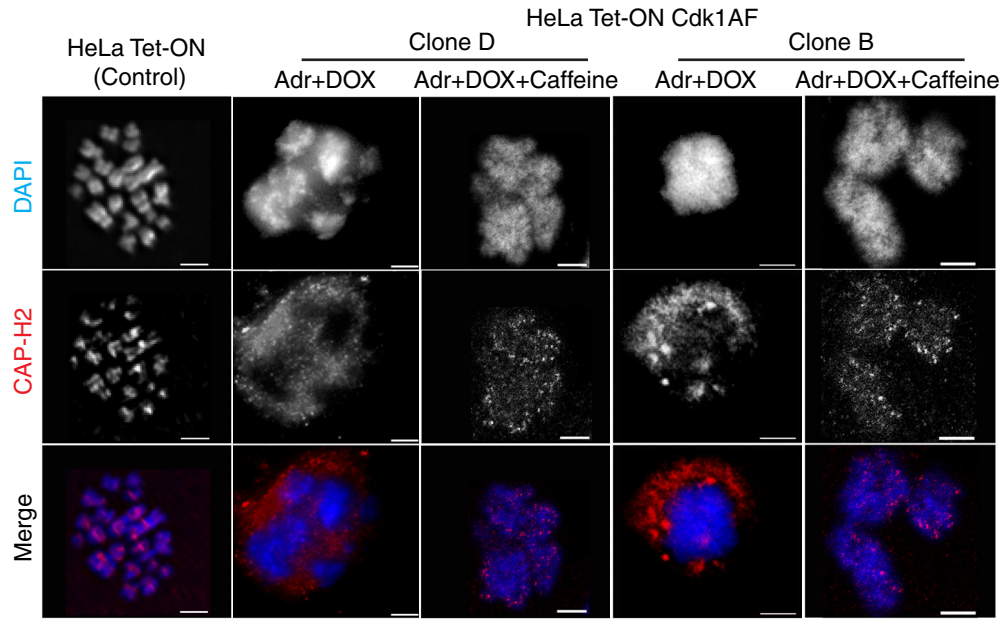
Supplementary Figure S2



Supplementary Figure S3



Supplementary Figure S4



Supplementary Figure S5

Antigen	Catalog number	Species	Usage and dilution					Source
			Usage and dilution	IF	WB	IP	FACS	
ACA (anti-crest antWBody)		Human	IF	1:1000				Kindly donated by Dr. Brian Gabrielli, University of Queensland Diamantina Institute.
ATR (N-19)	sc-1887	Goat	WB		1:200			Santa Cruz Biotechnology
b-actin	ab3280-500	Mouse	WB		1:800			Abcam
b-tubulin	T 0198	Mouse	IF	1:1000				Sigma-Aldrich
b-tubulin	RB-9249-P	Rabbit	IF	1:500				NeoMarkers
BubR1 [8G1]	ab4637	Mouse	IF	1:500				Abcam
CAP-D2	sc-101012	Mouse	IF/WB	1:500	1:500			Santa Cruz Biotechnology
CAP-D3	SC-101016	Mouse	WB		1:500			Santa Cruz Biotechnology
CAP-G	sc-101014	Mouse	IF/WB	1:500	1:500			Santa Cruz Biotechnology
CAP-G2	HPA 026631	Rabbit	IF/IP	1:500		1:10		Sigma-Aldrich
CAP-G2		Rabbit	WB		1:500			Kindly donated by Dr Keiji Kimura, University of Tsukuba
CAP-H	HPA002647	Rabbit	WB		1:500			Sigma-Aldrich
CAP-H2		Rabbit	WB		1:500			Kindly donated by Dr J.M Peters, The Research Institute of Molecular Pathology (IMP)
Cdk1/Cdc2 p34	610037	Mouse	WB/IP		1:500	1:100		Becton Dickinson
Chk1 (G-4)	sc-8408	Mouse	WB		1:500			Santa Cruz Biotechnology
Chk2 (A-11)	sc-17747	Mouse	WB		1:500			Santa Cruz Biotechnology
Cyclin B1 (H-433)	sc-752	Rabbit	IF/WB	1:500	1:500			Santa Cruz Biotechnology
Flag M2	F 3165	Mouse	WB/IP		1:500	1:50		Sigma-Aldrich
GFP	ab13970	Chicken	IF/WB	1:500	1:500			Abcam
Giantin (N-18)	sc-46993	Goat	IF	1:500				Santa Cruz Biotechnology
Histone H3	ab1791	Rabbit	WB		1:1000			Abcam
Lamin A/C	2032	Rabbit	IF	1:500				Cell Signaling Technology
MPM2	05-368	Mouse	IF/FACS	1:1000			1:200	Upstate Biotechnology
ORC2 (3G6)	sc-32734	Rat	WB		1:500			Santa Cruz Biotechnology
p53	sc-126 (DO-1)	Mouse	WB		1:500			Santa Cruz Biotechnology
Pericentrin	ab28144	Mouse	IF	1:500				Abcam
Phospho - Chk1 (Ser 345)	2341	Rabbit	WB		1:500			Cell Signaling Technology
Phospho-Chk1 (S317)	IHC-00068	Rabbit	WB		1:500			Bethyl Laboratories
Phospho-Chk1 (Ser317)	2344	Rabbit	WB		1:500			Cell Signaling Technology
Phospho-Chk2 (Thr68)	2661	Rabbit	WB		1:500			Cell Signaling Technology
Phospho-histone H2AX (ser139)	2577S	Rabbit	IF/WB	1:500	1:500			Cell Signaling Technology
Phospho-Histone H3 (Ser10)	sc-8656	Rabbit	IF/WB	1:500	1:500			Santa Cruz Biotechnology
Phospho-Histone H3 (Ser10)	9701	Rabbit	IF/WB	1:1000	1:500			Cell Signaling Technology
Phospho-p53 (Ser15)	9284	Rabbit	WB		1:500			Cell Signaling Technology
Phospho-Serine/Threonine	PP2551	Rabbit	IP			1:10		ECM Biosciences
Phospho-Tyrosine	PP2221	Rabbit	IP			1:10		ECM Biosciences
PLK1 (polo-like kinase 1)	ab17057	Mouse	IF	1:500				Abcam
Rad51(H-92)	sc-8349	Rabbit	IF	1:500				Santa Cruz Biotechnology
Securin	ab3305	Mouse	IF	1:500				Abcam
Shugoshin (Sgo1)	H00151648-M01	Mouse	IF	1:500				Abnova
SMC2	A300-058A	Rabbit	IF/WB/IP	1:500	1:1000	1:50		Bethyl Laboratories
SMC2	AM05324PU-N	Mouse	WB		1:1000			Acris
SMC4	A300-064A	Rabbit	IF	1:500				Bethyl Laboratories

Supplementary Figure Legends

Figure S1. Comparable cell cycle progression profiles of HeLa Tet-ON and HeLa Tet-ON Cdk1AF cells.

A HeLa Tet-ON, HeLa Tet-ON Cdk1AF clone B and clone D cells were synchronised by a combined serum starvation and thymidine protocol (Materials and Methods), then washed and released into medium with doxycycline (DOX, 1 μg/ml) or without DOX as described in Fig. 1B. Cells at 0, 4, 8, 12 and 24h post-release were collected for FACS.

B Synchronised HeLa Tet-ON and HeLa Tet-ON Cdk1AF cells were released into medium with doxycycline (DOX, 1 μg/ml) or without DOX. Cells were collected at 24h post-release and stained with γH2AX (red, bottom panel) and DAPI (blue, top panel). Images taken using 10 x objective are shown in the left panel; images taken by 60 x objectives are shown in the middle panel. Representative images for interphase cells, metaphase and anaphase cells with <5 γH2AX foci and no γH2AX are shown. Scale bar represents 5 μm. Box plots for percentage of cells with <5 γH2AX foci are shown in the right panel. There is no significant difference (NS) between HeLa Tet-ON (-DOX) and HeLa Tet-ON Cdk1AF (-DOX); between HeLa Tet-ON (-DOX) and HeLa Tet-ON Cdk1AF (+DOX).

C Cells were treated as described in Fig. S1B, cells were collected and stained Rad51 (red, bottom panel) and DAPI (blue, top panel). Representative images for HeLa Tet-ON (-DOX) and HeLa Tet-ON Cdk1AF (-DOX) are shown.

Figure S2. Sustained pH3 staining on decondensed chromosomes in DNA-damaged Cdk1AF cells.

A HeLa Tet-ON, HeLa Tet-ON Cdk1AF clone B and clone D cells were synchronised and released as described in Fig. S1A. Asynchronous (cyc) cells, and cells at 0, 4, 8, 12 and 24h post-release were collected for MPM2-FACS analysis. Scatter plots corresponding to

different time points are displayed in the left panel. Histogram shows percentage of MPM2 positive cells in different time points in the right panel.

B Representative immunofluorescence micrographs indicating mitotic hallmarks exhibited by HeLa Tet-ON Cdk1AF cells with DNA damage (synchronised and treated as described in Fig. 1B) are shown with DAPI-stained DNA, cyclin B (red, row 1), securin (red, row 2), shugoshin (red, row 3) and Plk1 (red, row 4). White arrows indicate prematurely decondensed chromosomes in metaphase, white arrowheads indicate chromosomes trapped in the cleavage furrow in late anaphase and telophase, and yellow arrows indicate fragmented chromatin in metaphase.

C Synchronised HeLa Tet-ON Cdk1AF cells were treated as described in Fig. 1B. Cells were collected and stained with anti-pH3 (red), β-tubulin antibodies (green) with DNA counterstained with DAPI (left hand side). Cells co-stained with anti- β-tubulin (green) and anti- pericentrin (red) antibodies to visualise centrosomal foci are shown (right).

D Synchronised HeLa Tet-ON, HeLa Tet-ON Cdk1AF clone B and clone D cells were treated as described in Fig. 1B. Cells were collected for DAPI staining. Representative images for abnormal metaphase and anaphase in Hela Tet-ON Cdk1AF clone B and clone D cells are shown, with HeLa Tet-ON as controls. Scale bar represents 5 μm.

Figure S3. Loss of axial condensin localisation in damaged U2OS Cdk1AF cells.

A Schematic diagram for siRNA and inhibitors used in Fig. 2D.

B Top panel shows immunofluorescence staining of SMC2 (red) during different phases of mitosis in cells with no drug treatment. Metaphase spread (bottom right) of damaged cells treated with caffeine (Adr+Caffeine) show axial localisation of SMC2 within each chromatid of the chromosomes. In damaged Cdk1AF cells (Adr+DOX), SMC2 is displaced from the chromatins or associates only loosely on fragmented chromosomes.

Chromosomes are stained with anti- SMC2 (red) and anti-MPM2 (green) and counterstained with DAPI. Normal mitotic cells with no drug treatment were used as control (bottom left). Scale bar represents 5 μm.

C Synchronised HeLa Tet-ON and HeLa Tet-ON Cdk1AF cells were treated as described in Fig. 1B, trapped with nocodazole, harvested, hypotonically-swollen, fixed and stained for SMC4 (red) and MPM2 (green), with chromosomes counterstained with DAPI. Representative images are shown. Scale bar represents 5 μm.

Figure S4. Caffeine treatment restores condensins localisation in mitotic chromosomes. Synchronised HeLa Tet-ON, HeLa Tet-ON Cdk1AF clone B and clone D cells were treated as described in Fig. 1B. HeLa Tet-ON Cdk1AF clone B and clone D cells were collected at T=16 and stained for CAP-H2 (red) and DAPI (blue), with HeLa Tet-ON untreated cells as a control. Representative images are shown. Scale bar represents 5μm.

Figure S5. List of primary antibodies used in this study