

Supplementary Information

RECQL5 cooperates with Topoisomerase II alpha in DNA decatenation and cell cycle progression.

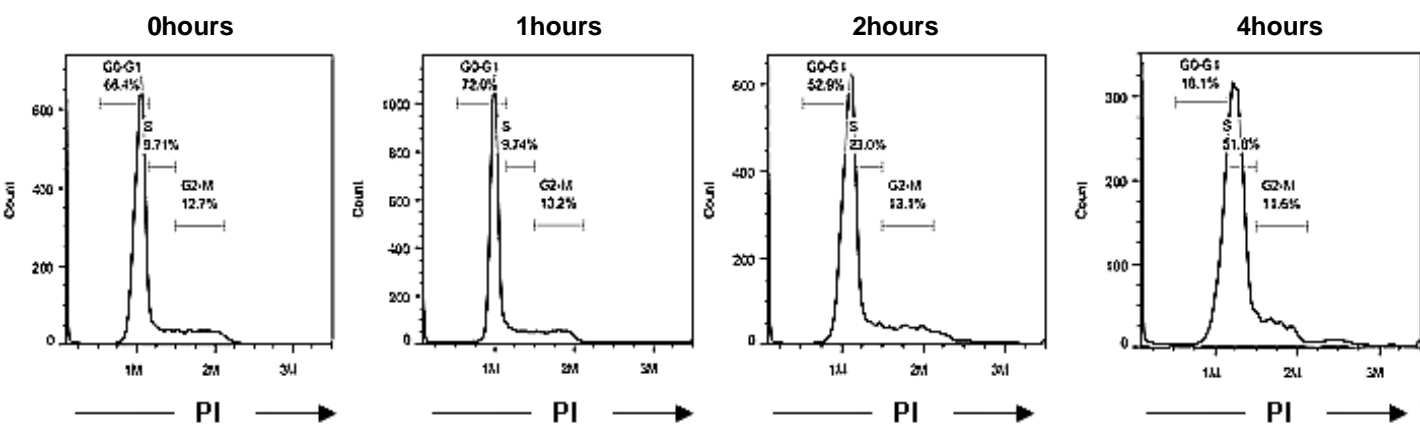
Mahesh Ramamoorthy, Takashi Tadokoro, Ivana Rybanska, Avik K. Ghosh, Robert Wersto, Alfred May, Tomasz Kulikowicz, Peter Sykora, Deborah L. Croteau and Vilhelm A. Bohr.

Supplementary Figure S1-S4
Supplementary Table S1

	Number of events	
	WI38 shScrambled	WI38 shRECQL5-2
Entangled	15.217	30.434
Under condensed	10.0562	25.878

Table S1. Quantification of metaphase spreads from shScrambled and shRECQL5-2 depleted WI38 cells. At least 50 metaphase spreads per cell line were analyzed.

A



B

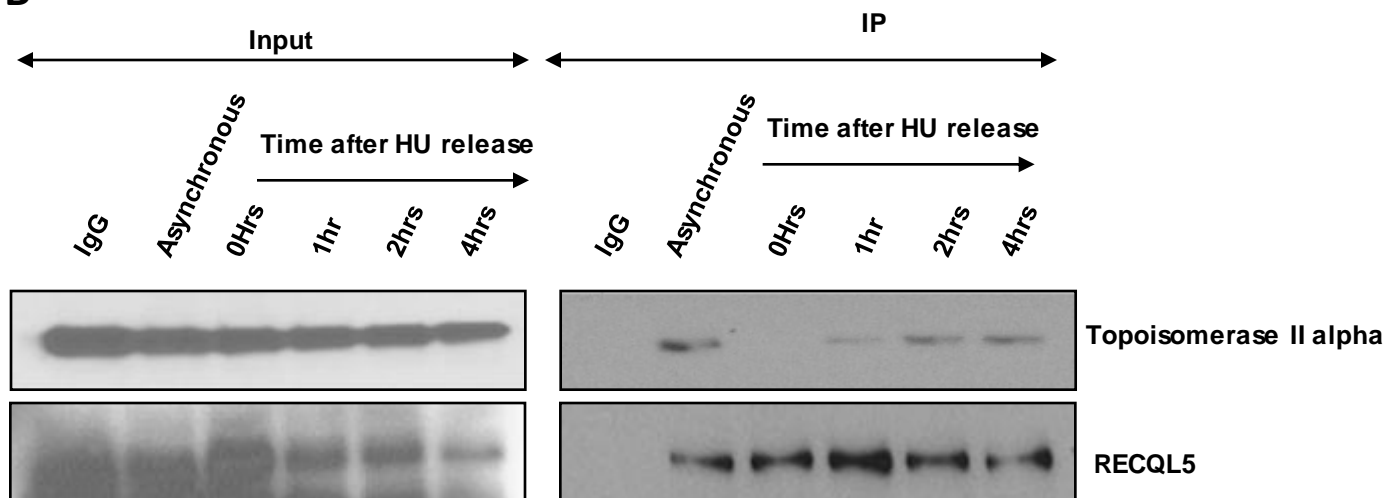


Figure S1 RECQL5 physically interacts with Topoisomerase II α during S phase. . (A) Representative flow cytometry histograms of HeLa cells analyzed after 2 mM hydroxy urea treatment and released for the time indicated. DNA content stained with PI was plotted against cell number. . (B) Immunoprecipitation of RECQL5 probed with indicated antibodies.

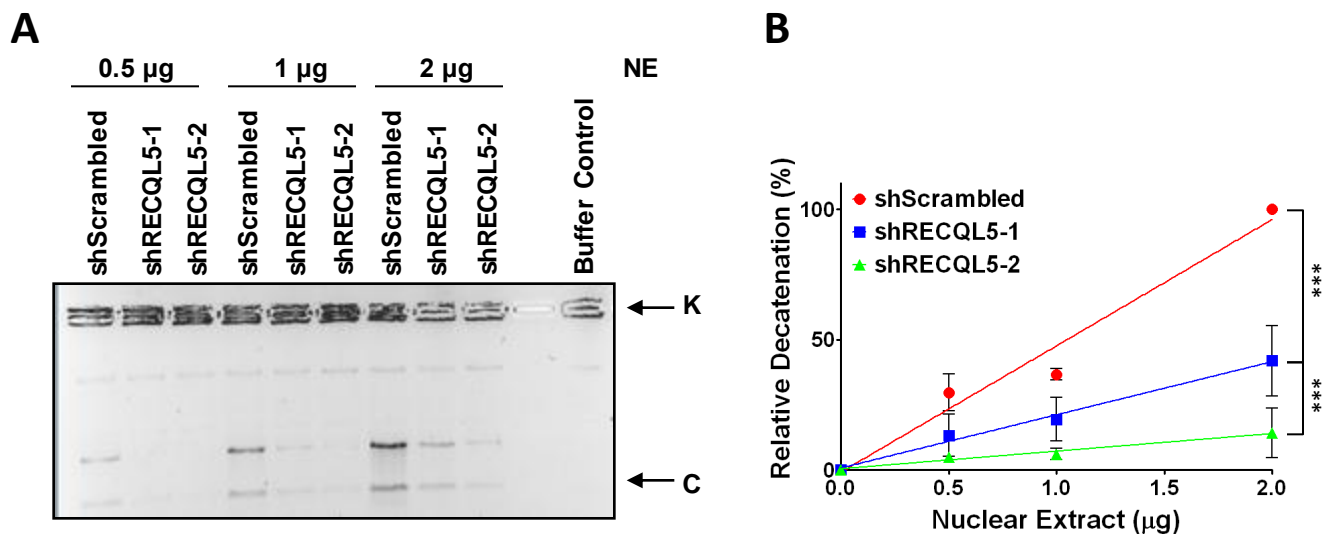


Figure S2 RECQL5 stimulates Topoisomerase II α catalyzed DNA decatenation. (A) Decatenation assay carried out with the nuclear extracts of WI38 shScrambled, shRECQL5-1 and shRECQL5-2 at indicated concentrations. (B) Quantification of the decatenation was performed by measuring the intensity of the decatenated bands. Error bars represent \pm SD, n=3. ***p< 0.0001, f test.

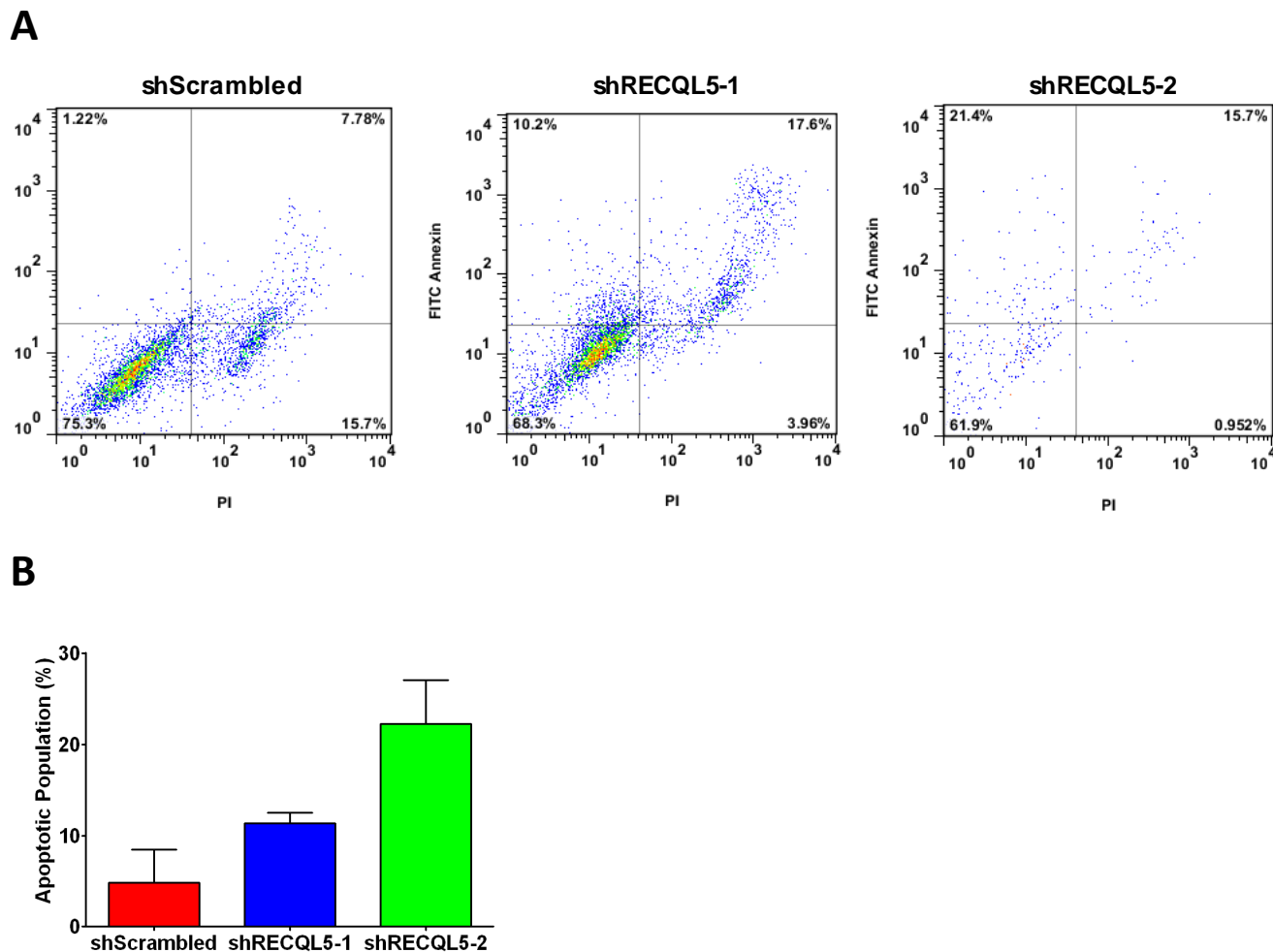
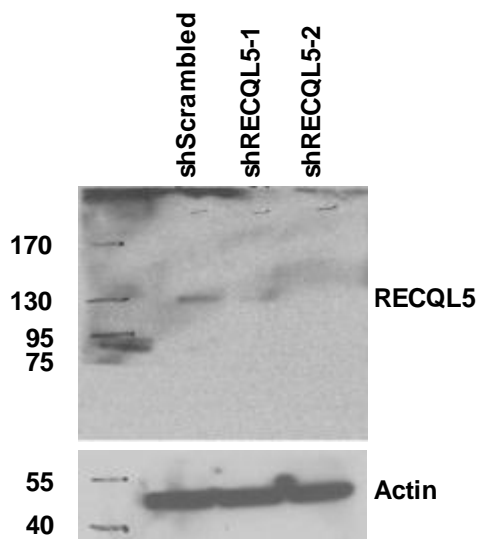


Figure S3 Depletion of RECQL5 increases apoptosis and activates a G2/M checkpoint. (A) Dot plots of FITC-Annexin versus Propidium Iodide (PI) two parameter flow cytometry of WI38 shScrambled, shRECQL5-1 and shRECQL5-2 cells. **(B)** Quantification of Annexin positive and PI negative cells in WI38.

A



B

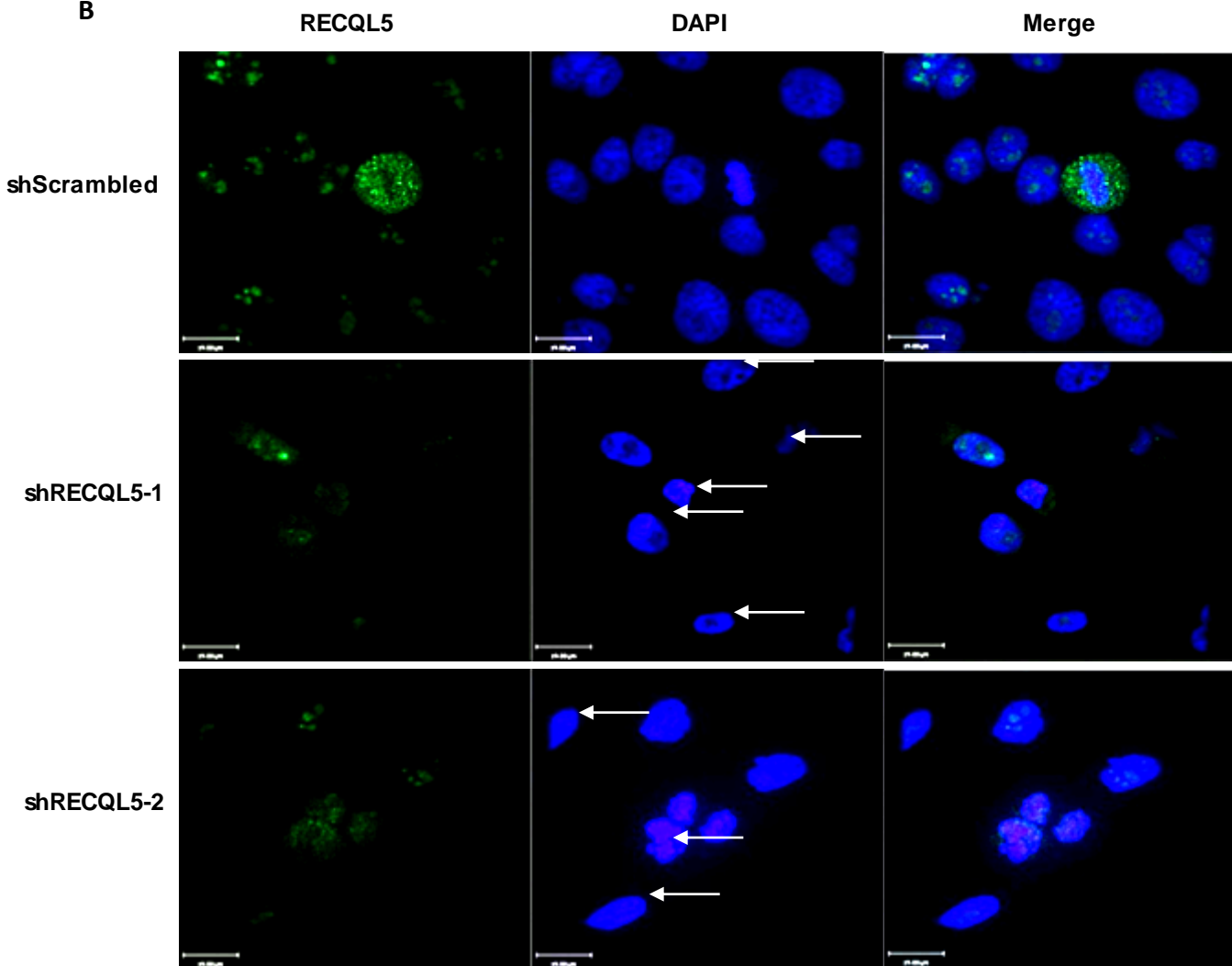


Figure S4 Validation of RECQL5 Antibody generated. Validation of rabbit polyclonal antibody raised against recombinant RECQL5 in HeLa shScrambled, shRECQL5-1 and shRecQL5-2 using (A) western blot and (B) immunofluorescence.