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Supplemental Data

Intercalating TOP2 poisons attenuate topoisomerase action at higher concentrations

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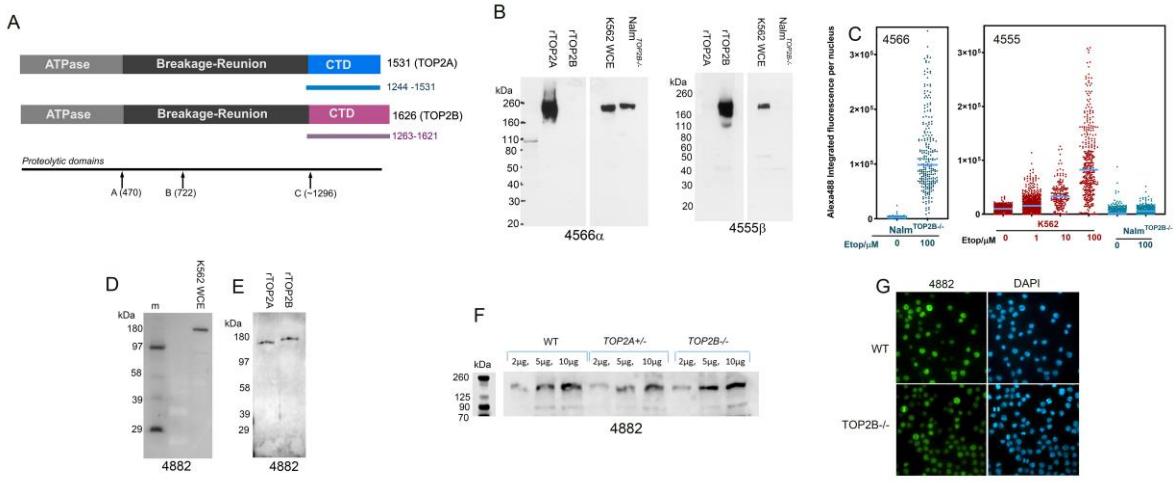
Supplemental Figure Legends

Supplemental Figure 1. Characteristics of inhouse antibodies used in this study. (A) TOP2 domain structure and location of immunogen polypeptide regions. The divergent C-terminal domains of TOP2A and TOP2B are coloured. Polyclonal antibodies 4566 & 4555 were raised in rabbits immunized with GST-fusion proteins containing amino acids 1244-1531 of TOP2A or 1263-1621 of TOP2B respectively. Polyclonal 4882 was produced in a rabbit immunized with a calf thymus TOP2 prep containing predominantly C-terminally truncated TOP2. **(B)** Western blot validation of antisera 4566 and 4555. rTOP2A & rTOP2B, recombinant TOP2A and B respectively, expressed in and purified from yeast; K562 WCE, whole cell extract derived from K562 cells; Nalm$^{TOP2B-/-}$, whole cell extract derived from TOP2B null Nalm6 cells (human pre-B leukemia cell line). **(C)** TARDIS analysis in K562 and Nalm$^{TOP2B-/-}$ cells, confirming lack of etoposide induced signal in TOP2B null Nalm$^{TOP2B-/-}$ cells (but efficient induction of signal in K562 cells) with antibody 4555, but efficient induction of signal in these cells with antibody 4566. **(D-F)** Western blot validation of antibody 4882 (note TOP2A and TOP2B frequently fail to resolve from each other in WCEs run on mini-gels). **(G)** Nuclear immunofluorescent staining pattern with antibody 4882 in WT and TOP2B null Nalm6 cells.



Supplemental Figure 1