**Supplementary Figures**

**Supplementary Figure S1. A.** HCT116 cells were treated with increasing doses of BOS172722 for 24 and 48 h. Immunoblotting was performed using antibodies against p-histone H3, total histone H3 and cleaved PARP. Tubulin was used as loading control. **B.** Table showing the fold change difference of GI50 at indicated incubation time of BOS172722 compared with GI50 of BOS172722 incubated for 96h. HCT116 cells were treated with BOS172722 for different incubation time and cells were washed to remove BOS172722 and replenished with media without BOS172722. Cells were grown further till 96h post-treatment and were fixed with MTT to determine GI50. Results were average of 2 independent experiments. **C**. Immunofluorescence images showing the localization of the indicated kinetochore proteins in HeLa cells, in the absence or presence of 200nM BOS172722. The white boxes are enlarged to highlight kinetochores.

**Supplementary Figure S2. A.** Box and whiskers plot of GI50s categorised by tissue of origin. Top: average doubling time of cell lines above (upper) and below (lower) the median, averages are shown in bold. **B**. Scatter dot plot of Emax of 5 days of 5M BOS172722 treatment in 10 TNBC and 9 non-TNBC cell lines **C-E.** relative fold change of MPS1 mRNA level normalised to housekeeping gene PPIA (C) and 18S (D) in 11 TNBC and 10 non-TNBC cell lines; and doubling time in 10 TNBC and 9 non-TNBC cell lines (E). Triangles in non-TNBC group indicate HER2 positive and overexpressed cell lines. Lines indicate median in eachgroup, *p*-value representsa two-tailed Mann Whitney test.

**Supplementary Figure S3.** BOS172722 was given at 50mg/kg orally twice a week (Day 0 and 4) for 47 days to MDA-MB-231 tumour bearing mice. Graphs showing tumour volume. 16 mice per group were treated.

**Supplementary Figure S4. A.** Distribution of chromosome numbers in MDA-MB-231 after incubation with 1nM Paclitaxel or vehicle (DMSO) for 36h. Bars depict the average of percentage values from three independent experiments with >20 metaphases scored each. **B-C.** Quantification of chromosome alignment errors in HeLa cells. HeLa cells were incubated for 90 minutes with MG132, Paclitaxel, BOS172722 or the combination of both, as indicated. Defects were categorized as normal: with all chromosomes aligned, minor; 1-3 chromosomes unaligned, major: >3 chromosomes unaligned. >100 cells were analysed for each replicate condition. Representative images (B) and graph with mean values and standard deviation from three biological replicates (C) were shown. **D.** Bar graph representing the extent of synergy observed after incubation with Paclitaxel and BOS172722 for distinct periods of time. Combinations of BOS172722 with 1 and 2nM Paclitaxel were added to MDA-MB-231 cells and removed by washing once after the indicated time. All assays were read after 5 days (120h). Values are means of at least three independent experiments, error bars represent standard deviation. **E**. Quantification of synergism in relation to the time of addition of BOS172722 or Paclitaxel. Left graph: MDA-MB-231 cells were incubated with a dilution series of Paclitaxel and the MPS1 inhibitor (37.5, 75, 150nM) added either simultaneously (0h), 12 or 24h after addition of Paclitaxel. Right graph: MDA-MB-231 cells were incubated with a dilution series of BOS172722 and Paclitaxel (1 or 2nM) added either simultaneously (0), 12 or 24h after addition of the MPS1 inhibitor. The difference in synergy at simultaneous addition is a consequence of the different experimental setup. Values are means of at least three independent experiments, error bars represent standard deviation.