

# Supplementary material

**Supplementary Figure 1:** IC50 determination for SR-4835 in A2780 and A2780 cis (top panel) and Caov-3 and Caov-3 cis (bottom panel) by confluence measurements using the Incucyte® live cell imaging system. Shown is the normalized confluence non-linear fit curve with its 95% confidence interval relative to the 72h treatment with SR-4835 indicated as log[M] concentration. Vertical lines indicate the respective mean IC50 concentrations.

**Supplementary Figure 2:** (A) Cartoon depicting the consequences of CDK12 inhibition and IPA recognition on the bioinformatical analysis of gene expression by short-read RNA sequencing techniques. If analyzed on the gene level, all reads mapping to a gene are counted. When 3'-truncated variants occur, the number of reads/gene decreases due to missing exons. This can be resolved by analysis of the read data at the exon level and enumerating alternative exon usage per gene. Created with BioRender. (B) Impact of CDK12 inhibition by SR-4835 on the recognition of IPA sites on the generation of gene variants exemplified for the ATM gene. Shown are the results of the exon level analysis of the RNA sequencing data as logFC of the analyzed exon vs. all other exons. Possible interpretation of the results suggest the generation of two shorter variants in addition to the full length variant, albeit the 3' end of variant 1 is unclear.

**Supplementary Figure 3:** ORA reactome pathway analysis for A2780 cis, Caov-3 and Caov-3 cis after treatment with SR-4835. Top 20 pathways are shown. No significantly altered pathway were found for A2780 and SK-OV-3.

**Supplementary Figure 4:** (A) mRNA expression data for selected genes, as indicated, following treatment with 90 nM SR-4835, 0.3  $\mu$ M (Caov-3), 1  $\mu$ M (A2780), 3  $\mu$ M (SK-OV-3, COV362, A2780 cis, Caov-3 cis) cisplatin or 10  $\mu$ M (A2780, A2780 cis, Caov-3, Caov-3 cis), 30  $\mu$ M (SK-OV-3, COV362) olaparib. Bars indicate mean  $\pm$ SD (n $\geq$ 3). Statistical significance was calculated by Mann-Whitney U test; \*p<0.05. (B) Densitometrical analysis of immunoblots (n=3) corresponding

to Fig. 4B. Statistical analysis was done by 2way ANOVA with multiple comparisons by uncorrected Fisher's LSD. \*\*\*\*  $p < 0.0001$ ; \*\*\*  $p < 0.001$ ; \*\*  $p < 0.01$ ; ns, not significant.

**Supplementary Figure 5:** (A, B) IC50 determinations for cisplatin (A) and olaparib (B) in A2780, A2780 cis, SK-OV-3, Caov-3, Caov-3 cis and COV362. Graphs depict the mean  $\pm$  SEM pIC50 in  $-\log[M]$ , tables give the converted IC50 values in  $[\mu M]$  with their respective 95% confidence intervals. \*\*\*\*  $p < 0.0001$  by one-way ANOVA with uncorrected Fisher's LSD multiple comparison testing.

**Supplementary Figure 6:** Uncropped and unmodified Western Blot images used to generate Fig. 4B. Shown are the consecutive antibody incubations and multiple exposure times. Brightness and contrast were optimized on the respective whole image to generate Fig. 4B.

**Supplementary Figure 7:** IC50 determinations for cisplatin in A2780 cis after pre-treatment with SR-4835. A2780 cis cells were pre-treated with low doses of SR-4835 for 72h avoiding cell killing, as indicated. This was followed by a combination treatment with the same SR-4835 doses as indicated and increasing cisplatin concentrations (0-400  $\mu M$ ). IC50 concentrations were assessed by confluence measurements by live cell imaging. Vertical lines indicate mean IC50 concentrations for each SR-4835 pre-treatment concentration. No statistically significant differences were found.

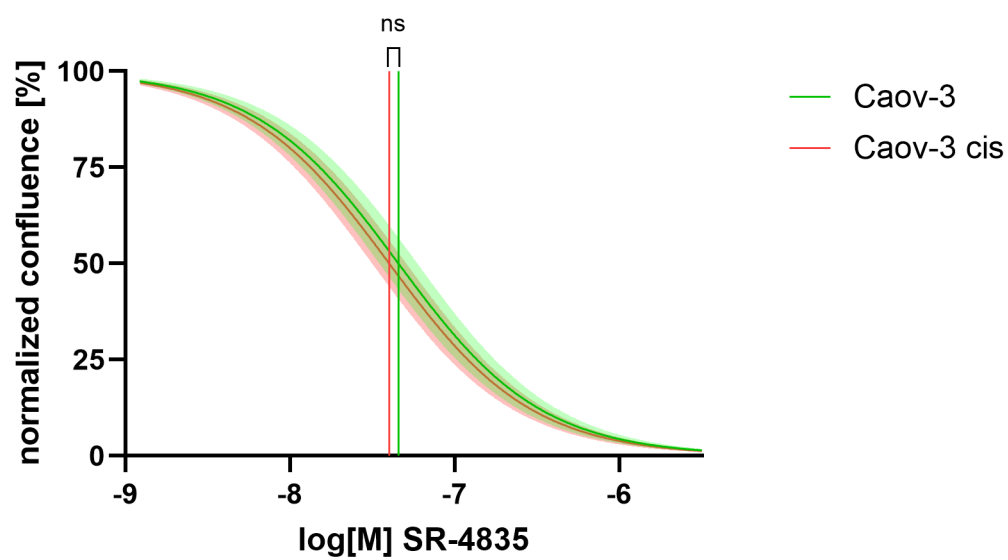
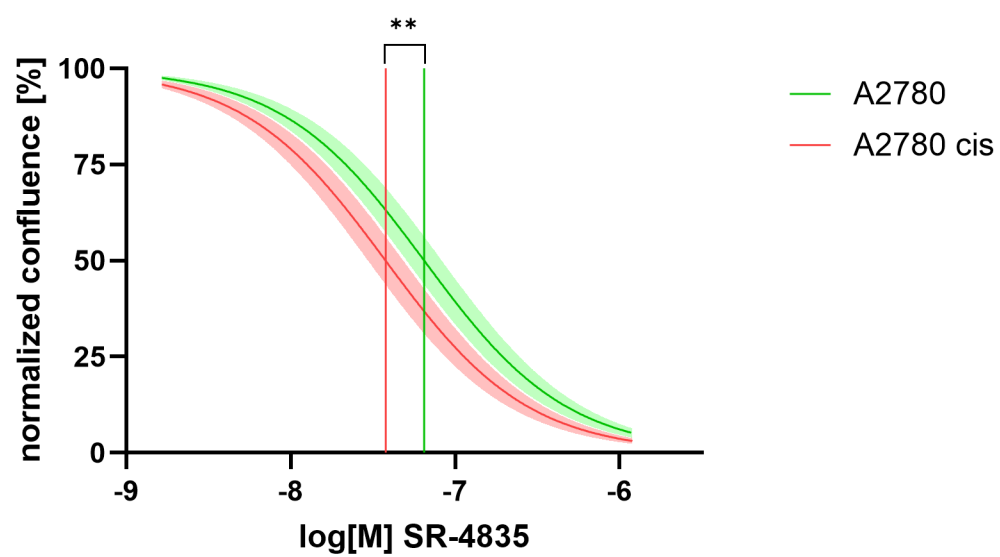
**Supplementary Table 1:** Mutations identified in the indicated OC cell lines by targeted exome sequencing. ClinVar database was interrogated to identify clinical significant mutations. Sequencing statistics and targeted genes are indicated in separate sheets.

**Supplementary Table 2:** List of differentially expressed genes after SR-4835 treatment of A2780, A2780 cis, SK-OV-3, Caov-3, Caov-3 cis and on the global level (different sheets) with their log fold change (logFC) ranked by the adjusted p-value (adj. P. Val). Summary sheet indicates the number of down-, not significantly, and up-regulated genes.

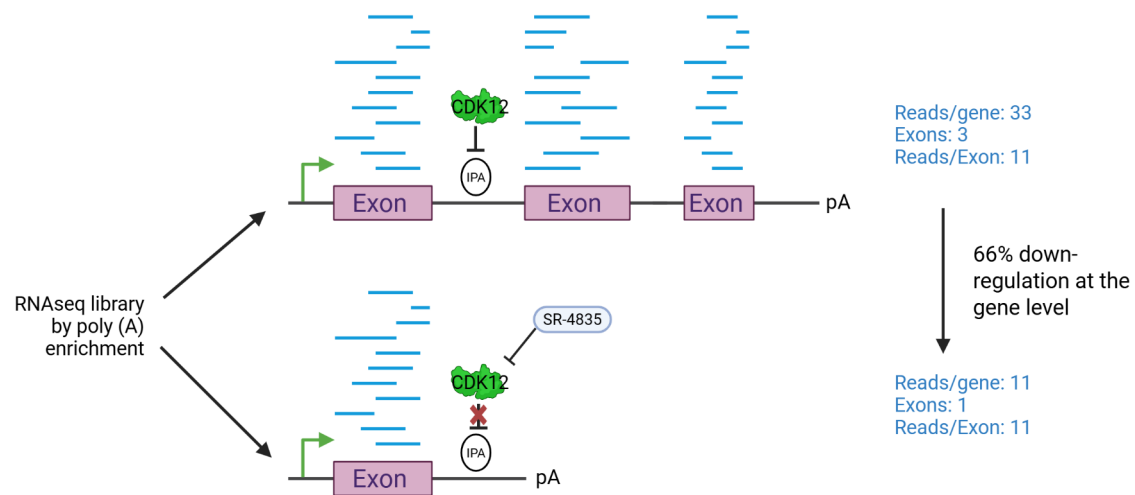
**Supplementary Table 3:** List of genes with alternative exon usage after SR-4835 treatment of A2780, A2780 cis, SK-OV-3, Caov-3, Caov-3 cis and on the global level (different sheets) ranked by the adjusted p-value (adj. P. Val). Number of intronic and terminal polyadenylation sites, as well as number of exons are indicated. Summary sheet indicates the number of genes with significant and not significant alternative exon usage.

**Supplementary Table 4:** List of significantly enriched pathways (ORA reactome) in the transcriptome analysis on the gene level after SR-4835 treatment of A2780, A2780 cis, SK-OV-3, Caov-3, Caov-3 cis and on the global level (different sheets).

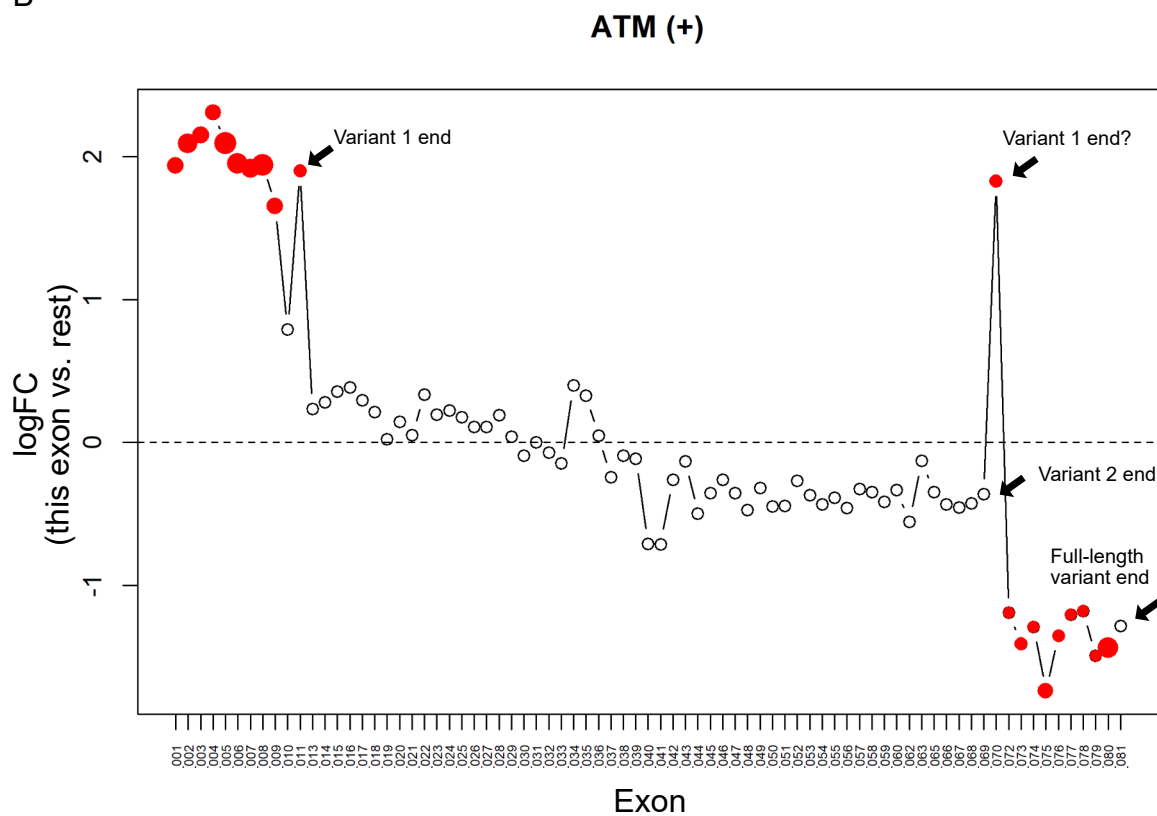
**Supplementary Table 5:** Detailed and average HSA scores for each OC cell line for SR-4835/cisplatin and SR-4835/olaparib combinations (different sheets). Statistical significance of the mean deviation from zero was evaluated by one-sided Student's t-tests.

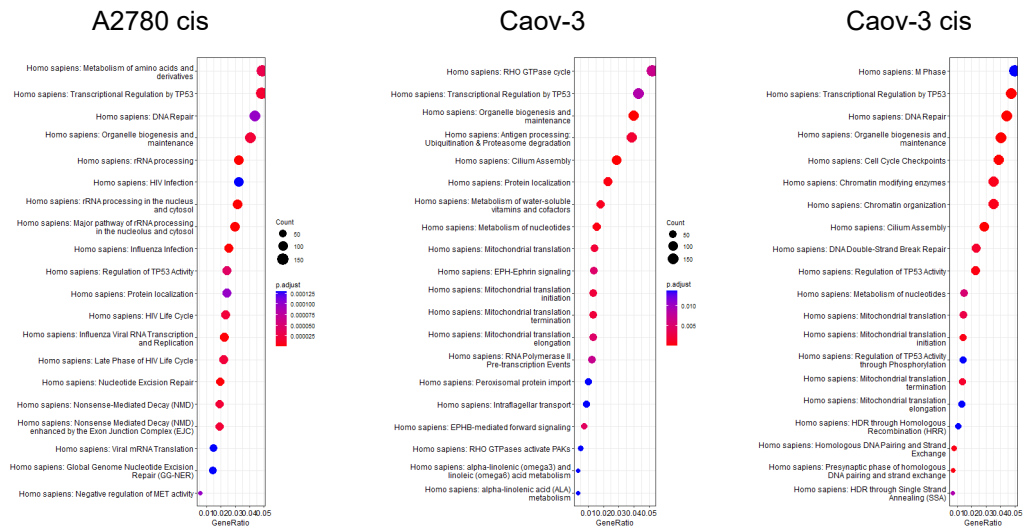


A

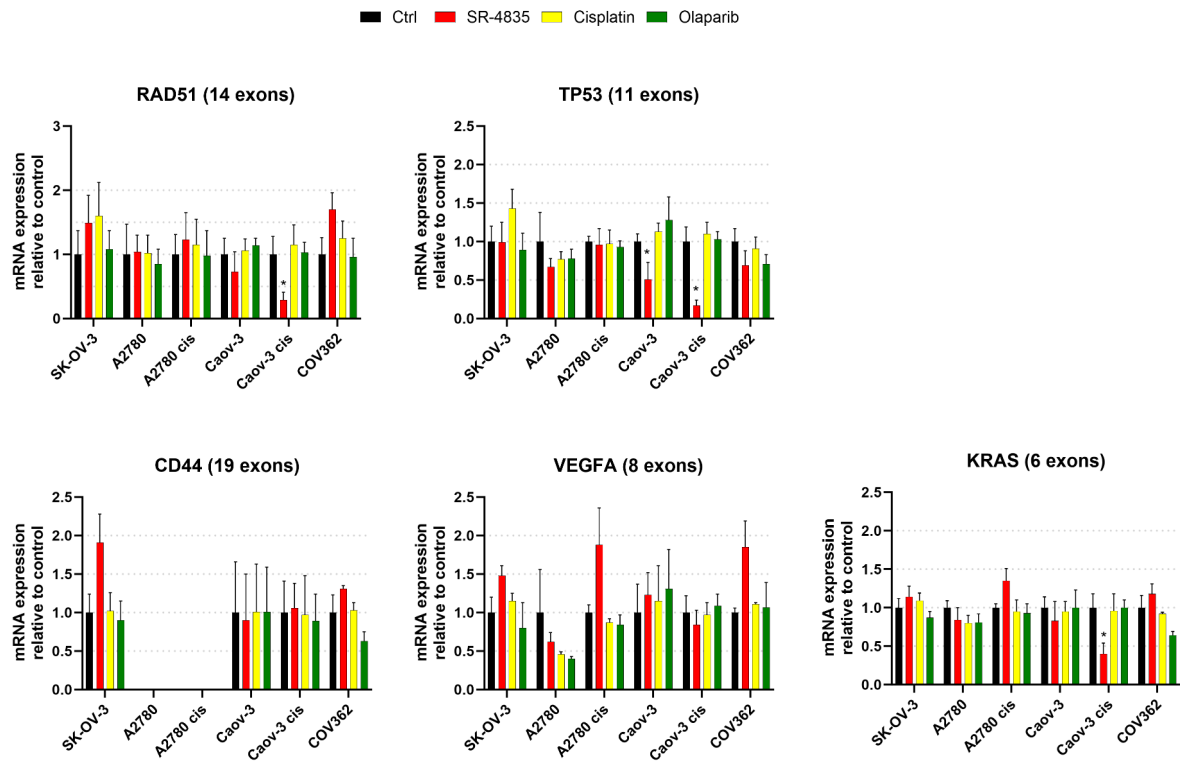


B

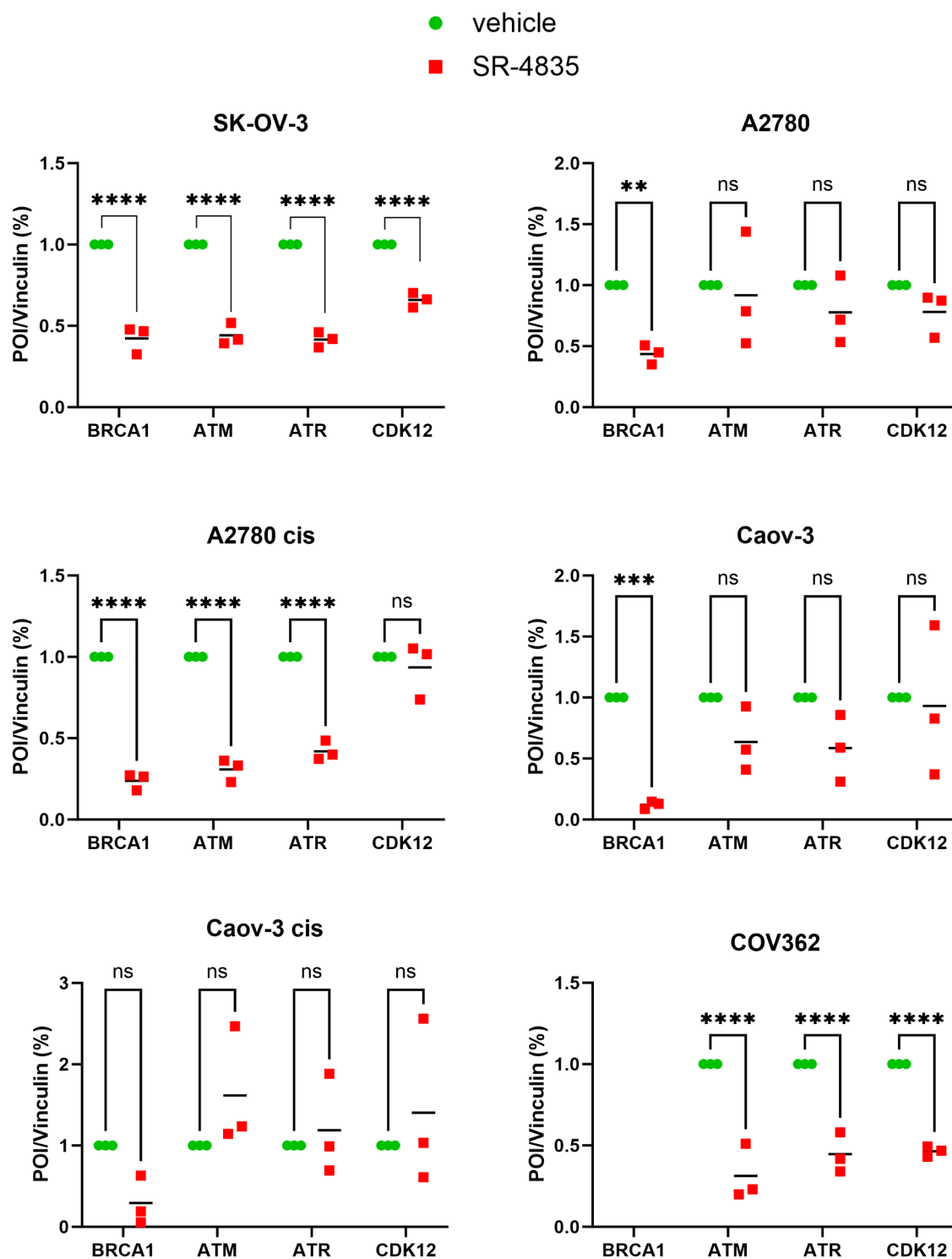




Supplementary Figure 3, Santer et al., 2025

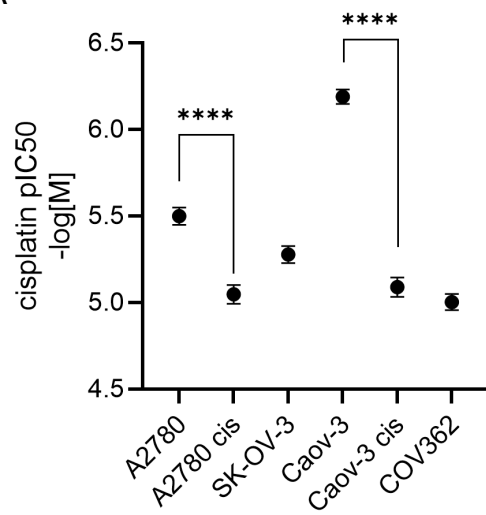


Supplementary Figure 4A, Santer et al., 2025



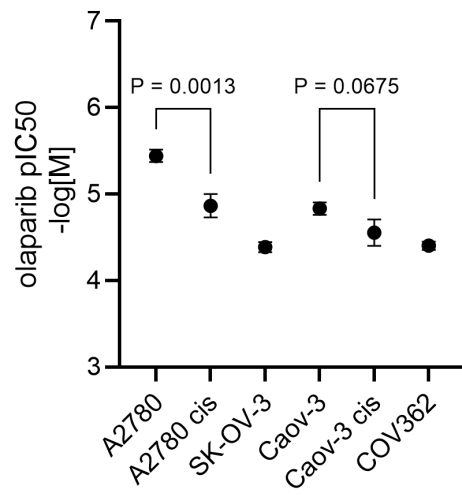
Supplementary Figure 4B, Santer et al., 2025

A

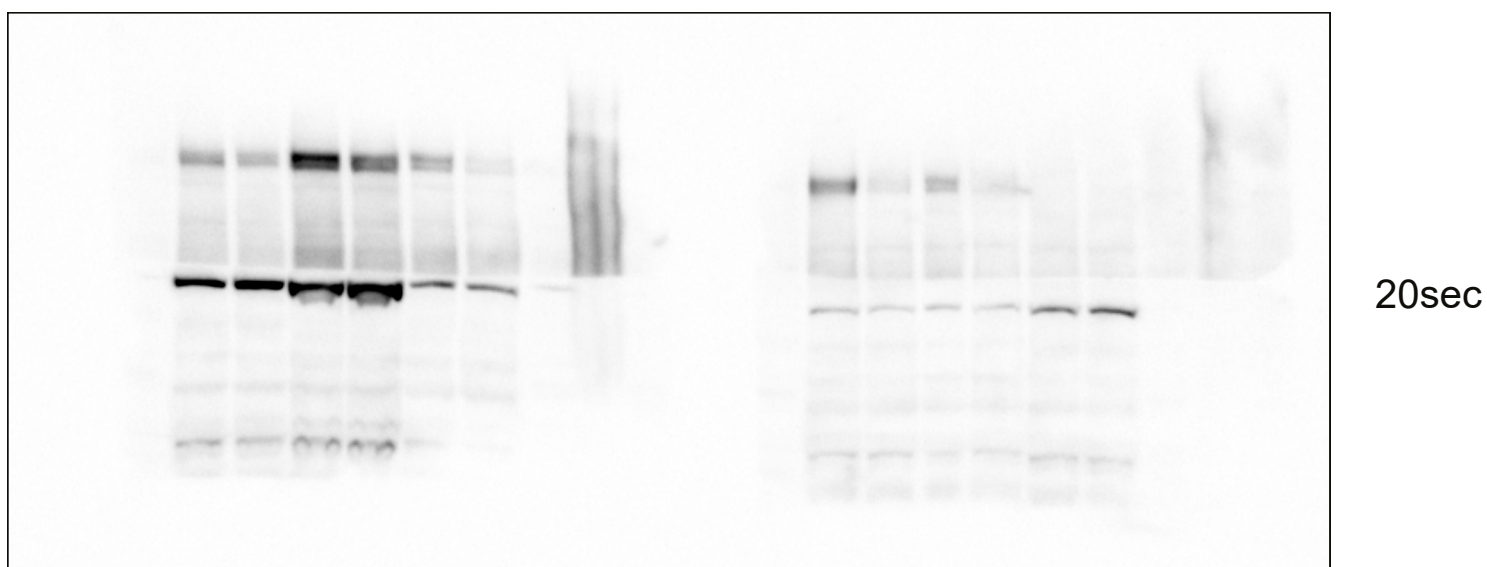
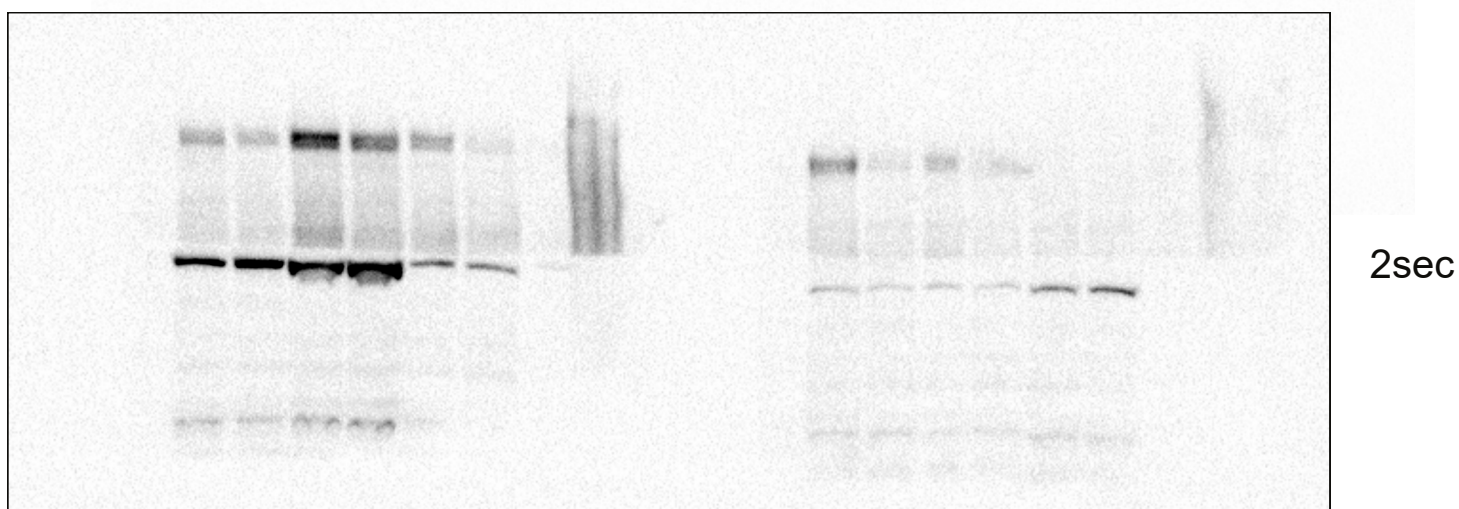
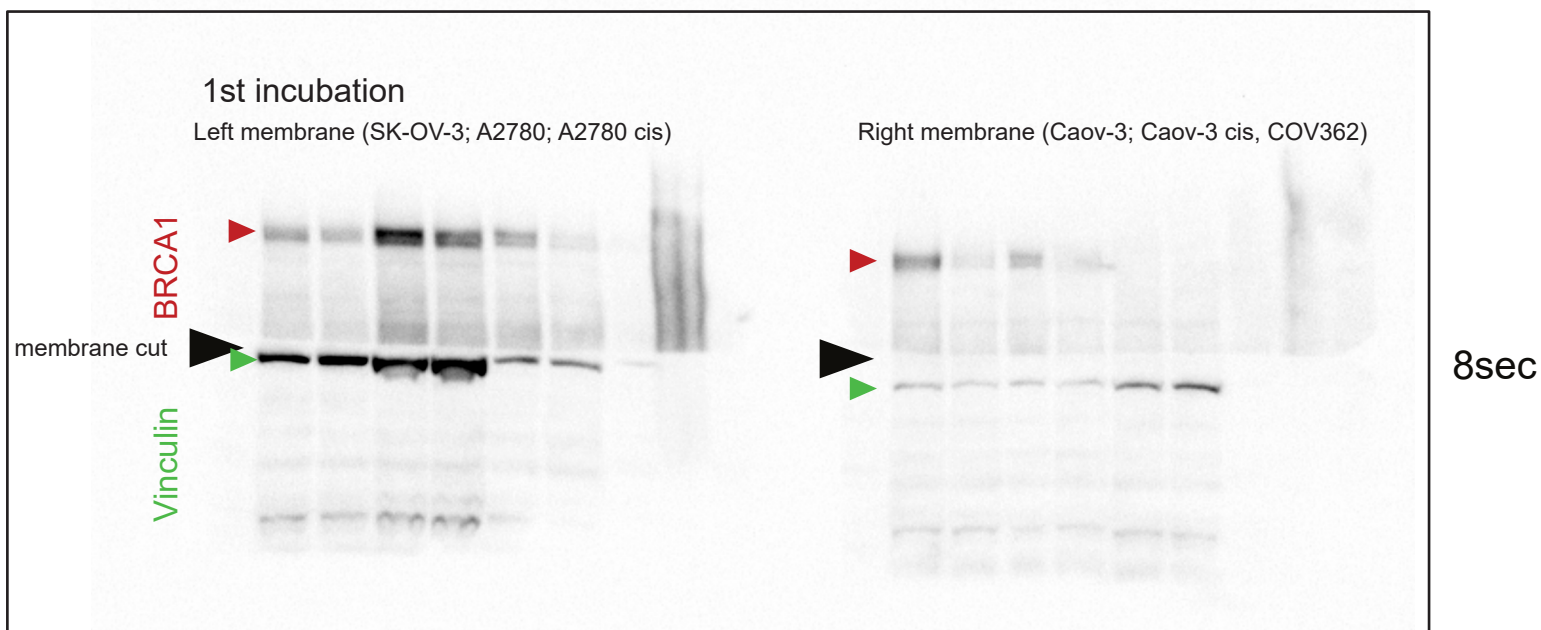
cisplatin [ $\mu$ M]

|            | IC50 | 95% CI     | n |
|------------|------|------------|---|
| A2780      | 3.17 | 2.50-4.01  | 5 |
| A2780 cis  | 8.98 | 7.05-11.41 | 5 |
| SK-OV-3    | 5.28 | 4.26-6.55  | 4 |
| Caov-3     | 0.65 | 0.54-0.77  | 5 |
| Caov-3 cis | 8.15 | 6.41-10.38 | 5 |
| COV362     | 9.94 | 8.11-12.17 | 6 |

B

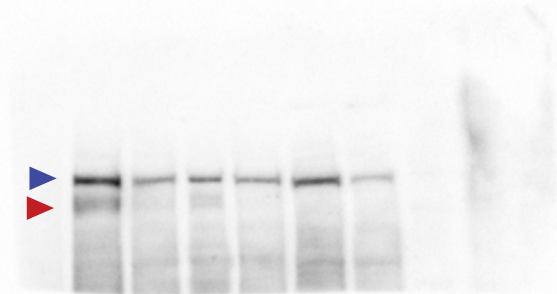
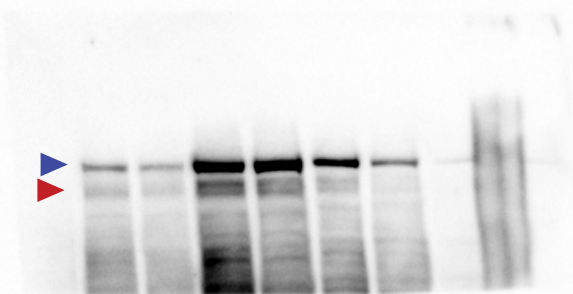
olaparib [ $\mu$ M]

|            | IC50  | 95% CI      | n |
|------------|-------|-------------|---|
| A2780      | 3.61  | 2.50-5.21   | 3 |
| A2780 cis  | 13.60 | 6.33-28.53  | 3 |
| SK-OV-3    | 40.98 | 31.48-53.31 | 3 |
| Caov-3     | 14.66 | 10.82-19.87 | 3 |
| Caov-3 cis | 27.85 | 11.29-64.57 | 3 |
| COV362     | 39.49 | 31.46-49.59 | 3 |

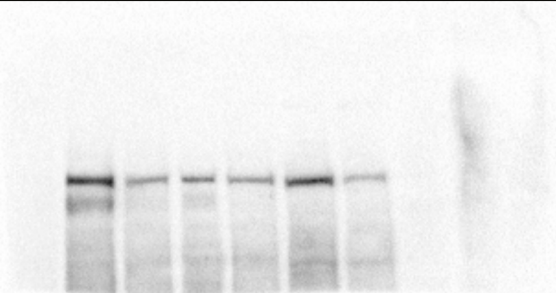


2nd incubation (upper membrane)

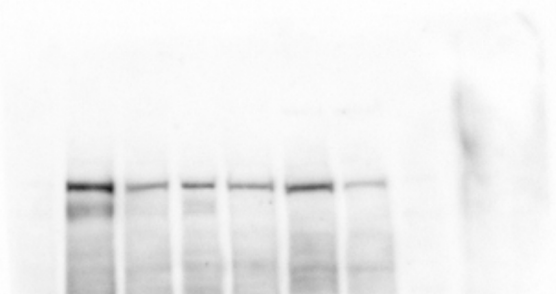
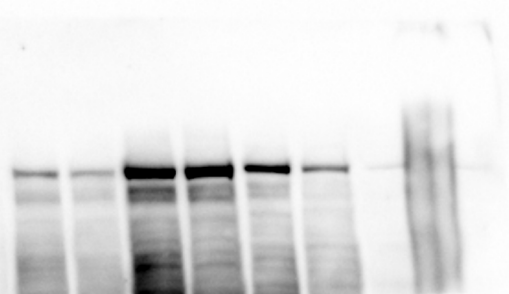
ATM



10sec



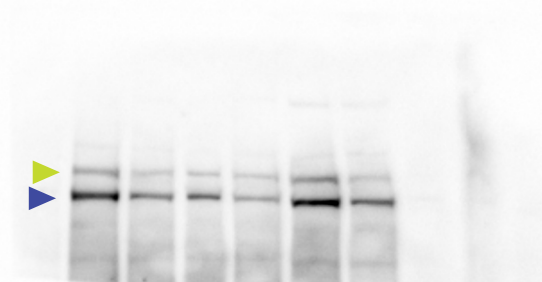
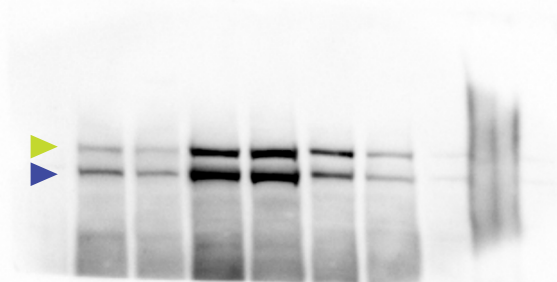
3sec



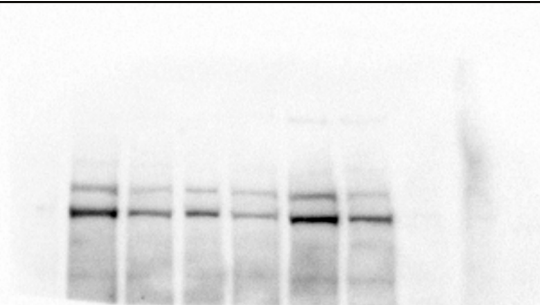
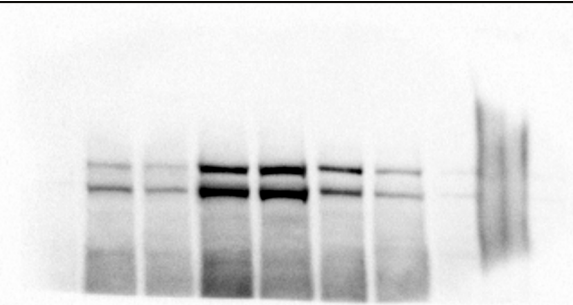
15sec

3rd incubation (upper membrane)

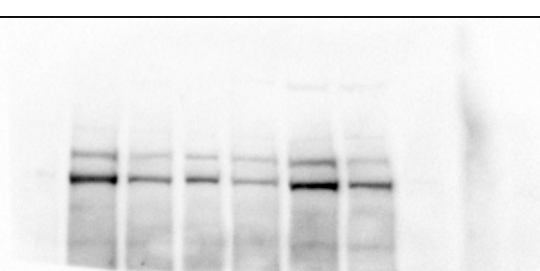
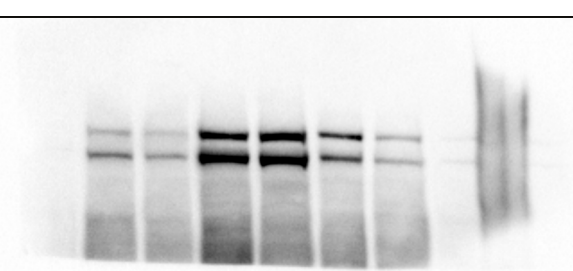
ATR



10sec



3sec

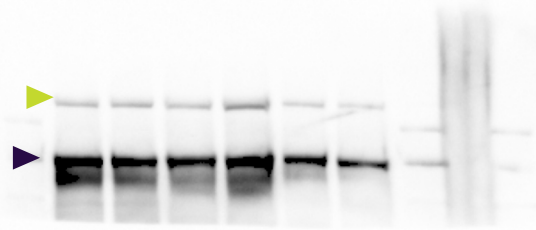


5sec

#### 4th incubation (upper membrane)

Left membrane (SK-OV-3; A2780; A2780 cis)

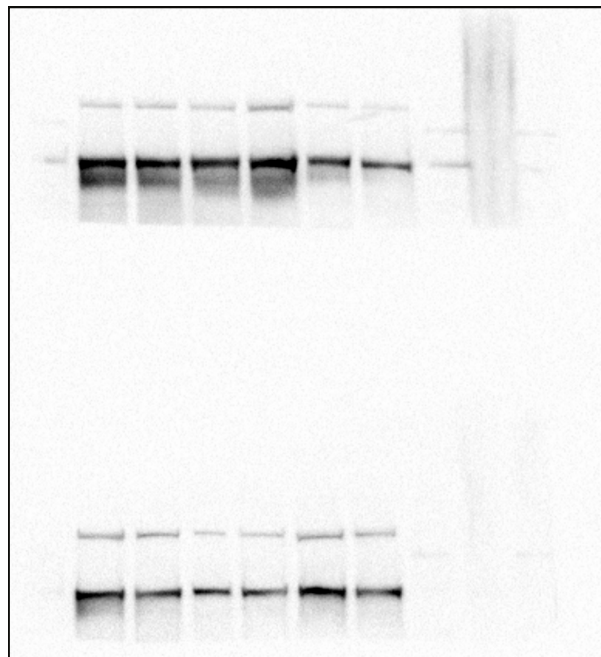
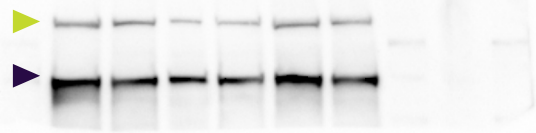
CDK12



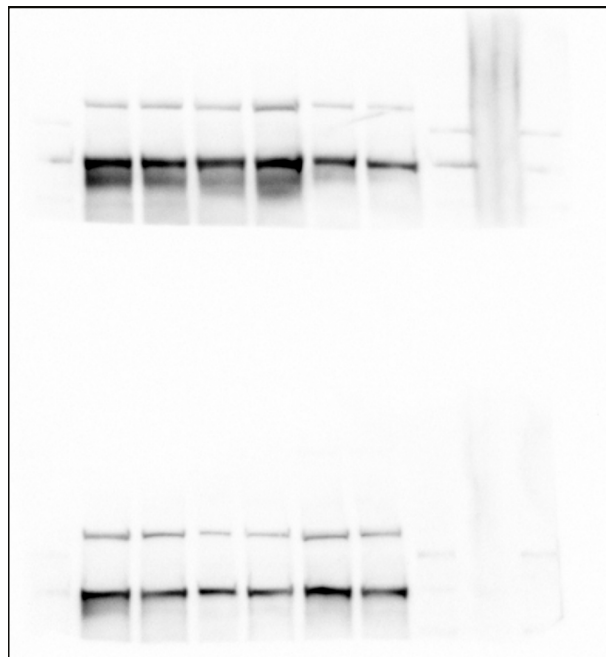
10sec

Right membrane (Caov-3; Caov-3 cis, COV362)

CDK12

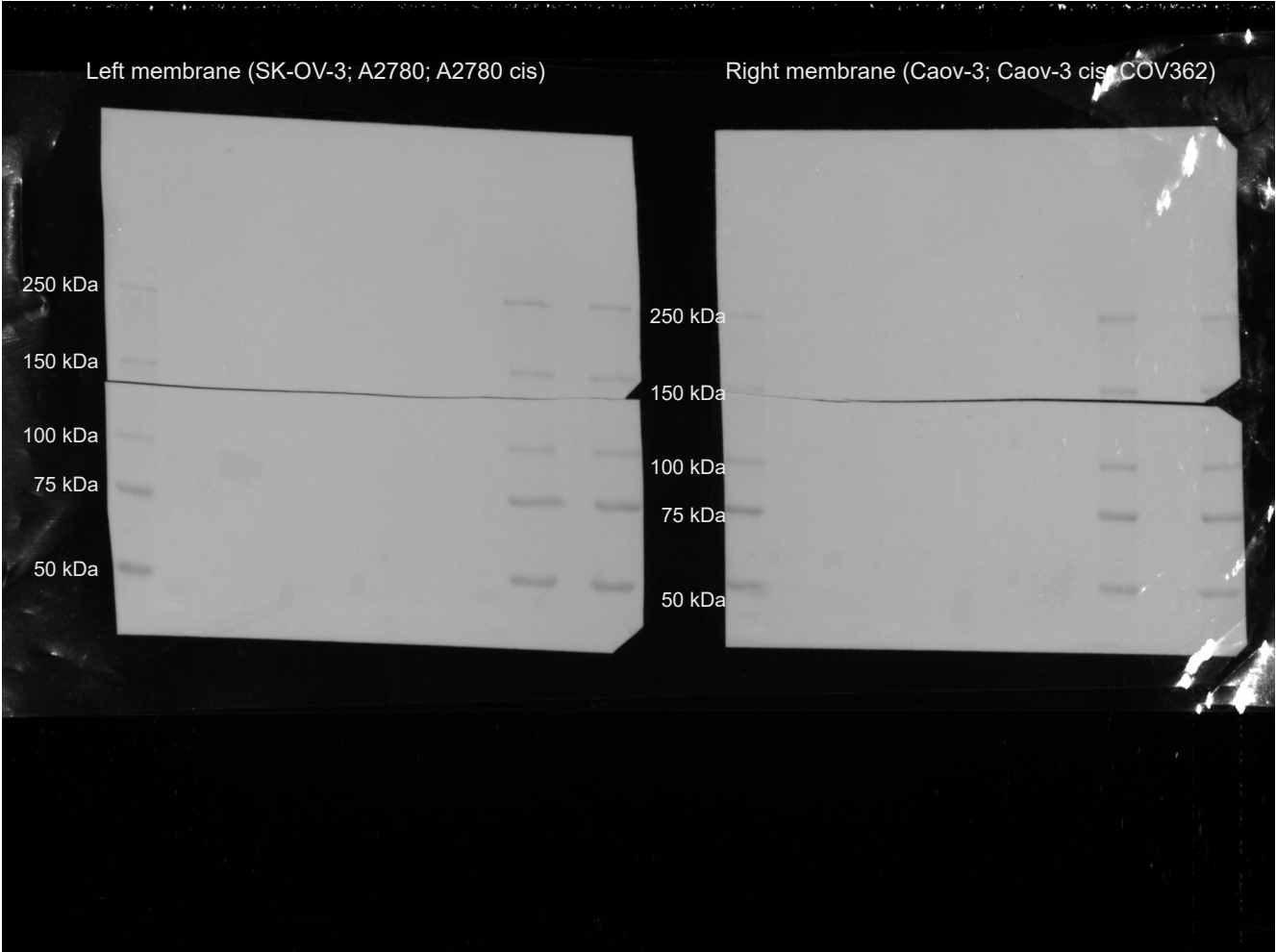


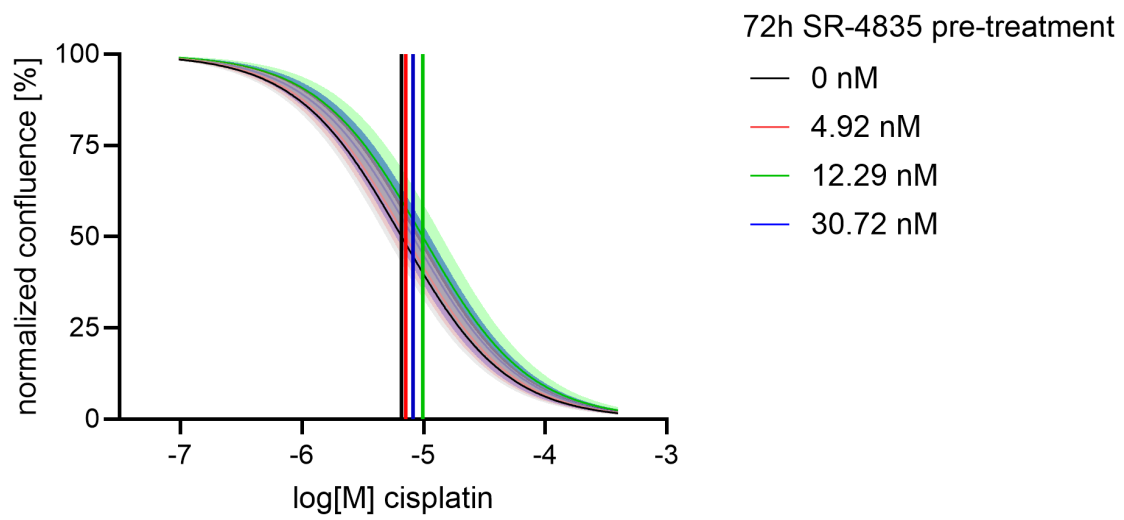
1sec



5sec

Brightfield





| Pre-treatment | cisplatin       |            |   |
|---------------|-----------------|------------|---|
| SR-4835 [nM]  | IC50 [ $\mu$ M] | 95% CI     | n |
| 0             | 6.60            | 4.99-8.72  | 3 |
| 4.92          | 7.12            | 5.45-9.30  | 3 |
| 12.29         | 9.87            | 6.85-14.18 | 3 |
| 30.72         | 8.18            | 5.96-11.20 | 3 |