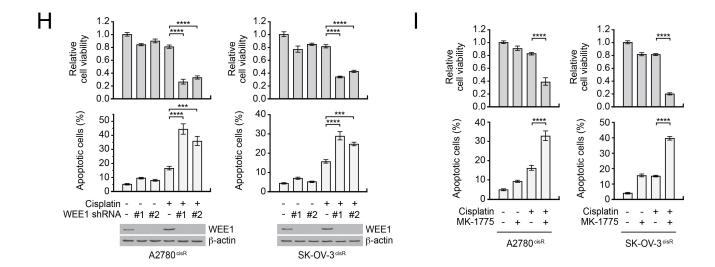


Supplementary Fig. S6 continued



Supplementary Fig. S6. A and B, Human cell stress array proteome profiling of A2780<sup>cisR</sup> cells with DGKA knockdown and cisplatin treatment. Cells were treated with a sublethal dose (5 ug/ml) of cisplatin for 48 hr. Density analysis was performed using ImageJ software. C, Phospho-JNK and JNK levels in A2780cisR and SK-OV-3cisR with DGKA knockdown and cisplatin treatment. Cells were treated with sublethal doses of cisplatin (5 µg/ml A2780cisR and 2 µg/ml SK-OV-3cisR) for 48 hr. Phospho-JNK Thr183/Tyr185 and JNK levels were assessed by immunoblotting. **D**, Effect of targeting c-JUN on cisplatin-dependent cell viability and apoptotic cell death E. In vitro DGKA kinase assay using c-JUN as a substrate. Recombinant c-JUN was incubated with GST fused DGKA variants enriched from 293T cells. Antibodies against phospho-cJUN S63 and pan-phospho-serine/threonine were used to detect phosphorylation in c-JUN. F and G, PA relocates c-JUN to the nucleus in DGKA knockdown cells with cisplatin treatment. Immunofluorescence staining of c-JUN in A2780cisR cells with DGKA knockdown and cisplatin (5 µg/ml) and PA (10 µM) treatment. Representative images from three independent experiments are shown in (F). Scale bars represent 10 µm. c-JUN cytosol/nucleus ratio of (F) is shown in (G). H and I, Genetic or pharmacological inhibition of WEE1 sensitizes ovarian cancer cells to cisplatin treatment. Effect of WEE1 knockdown (H) or WEE1 inhibitor MK-1775 treatment (I) on cisplatin-dependent cell viability (upper panels) and apoptotic cell death (lower panels). Stable WEE1 knockdown cells were treated with sublethal doses of cisplatin (A2780cisR: 5 uM, SK-OV-3cisR: 2 uM) for 48 hr for (H). Cells were treated with 100 nM of MK-1775 and sublethal doses of cisplatin for 48 hr for (I). Cell viability and apoptosis were assessed by CellTiter-Glo luminescent cell viability assay and Annexin V staining, respectively. Results of one representative experiment from three (D and F-I) and two (A-C and E) independent experiments are shown. Error bars represent SD. P values were determined by one-way ANOVA (ns: not significant; \*P < 0.05; \*\*\*P < 0.001; \*\*\*\*P < 0.0001).