

**Supplemental Data for Guertin et al's *Preclinical Evaluation of the WEE1 Inhibitor MK-1775 as Single Agent Anticancer Therapy***

**Supplemental Table 1. Proliferation assay EC<sub>50</sub> results for MK-1775 in diverse tumor cell lines.** As described in Materials and Methods, cell lines were plated and incubated with a 9-point titration of MK-1775 for a 96 hour treatment period. Cell proliferation was then determined with CellTiter-Glo and proliferation relative to DMSO-treated control cells was plotted against MK-1775 concentration to allow EC<sub>50</sub> calculation. Cells lines are grouped by tissue of origin.

**Supplemental Figure S1. MK-1775 treatment inhibits proliferation in diverse tumor cell lines.** Five hundred and twenty-two tumor cell lines were plated in triplicate and treated with a 9-point titration of MK-1775 ( $\mu$ M concentrations displayed on x-axis in log scale). After 96 hours proliferation was evaluated and cell line response data are represented on y-axis as fractional viability relative to DMSO-treated controls as a function of MK-1775 concentration. Cell lines are grouped by tumor type and each response curve denotes an individual line.

**Supplemental Figure S2. DNA damage induction by MK-1775 treatment requires mitogen stimulation.** ES-2 cells were serum starved for 36 hours at which point they were either left unstimulated or treated with 20% FBS. Cells cultured under both conditions received either DMSO or 500 nM MK-1775 for 24 hours before flow cytometry analysis of  $\gamma$ H2AX and DNA content. Left panel contains histograms of cell cycle distribution. The right panels contain scatter plots with gates indicating the  $\gamma$ H2AX population.

**Supplemental Figure S3. The MK-1775 response of three sensitive cell lines.** A2058, HT-29, and LoVo cells were treated with increasing concentrations of MK-1775 as indicated on x-axis. Proliferation is graphed as a percentage of DMSO-treated control cells at 72 hours following addition of drug. Dotted line indicates EC<sub>90</sub> effect of MK-1775.