

Figure S1. A representation of the gating used for flow cytometry analysis. The top row (a) shows the dot plots of the untreated condition and the bottom row (b) shows the dot plots of B4S5E4 1.1:1 90°C condition. On both rows the leftmost dot plot gates the population of 'Cells' from debris, the middle one is a subset of 'Cells' and gates the 'Live Cells' from 'Dead Cells' with high propidium iodide content, and the rightmost plot is a subset of 'Live Cells' that gates for live cells positive for GFP expression. FL1 is the green channel indicating GFP expression (x-axis) and FL-2 is the yellow channel indicating autofluorescence (y-axis). 2-D gating is used to ensure that autofluorescent cells are not gated as GFP positive.

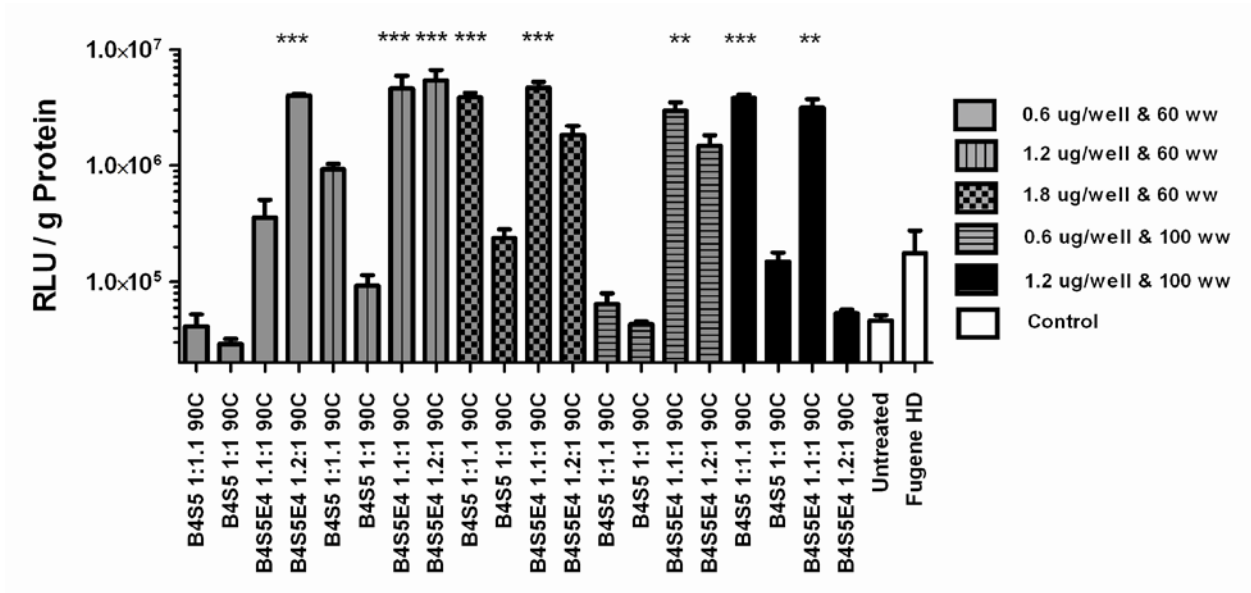


Figure S2. Preliminary screening of EPH4 cells at different polymer to DNA weight ratios and DNA dose per well to optimize transfection conditions. The cells were plated at a density of 150,000 cell/mL and screened with the 96-well plate protocol described in Materials and Methods. The transfection efficacy is plotted in terms of relative light units (RLU) per gram of protein in each well. The results are presented as mean \pm SEM (n=4).

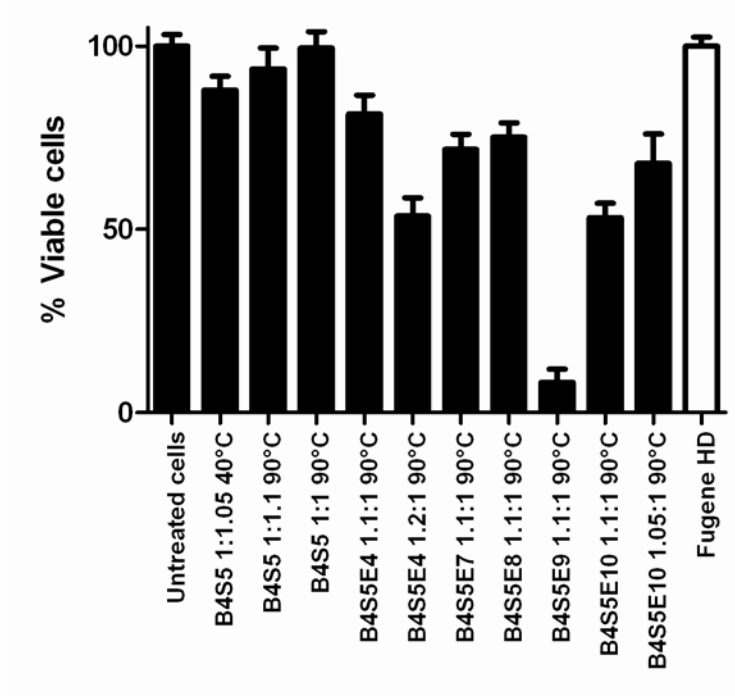


Figure S3. EPH4 cytotoxicity data presented as percent viable cells normalized to untreated control. The results are presented as mean \pm SEM (n=8). A 96-well plate cell titer assay kit (Promega CellTiter 96 AQueous One) was used to determine viability. The absorbance at 490 nm was measured at 4 hrs.