

Figure S1. Combinational effects of AZD4547 and BEZ235 in HPV⁺ UPCI-SCC-154 and HPV⁻ UT-SCC-60A cells. Cell viability after single and combination treatments with AZD4547 and BEZ235 for 48 h, assessed with WST-1 analysis. CI was calculated according to the Highest Single Agent approach. A CI <1 indicates a positive combination effect and a CI >1 a negative effect. The dotted line represents the effect of the best single drug, thus a combinational effect below that line shows a positive combination effect (equivalent with CI <1). Significance was tested for HPV⁺ UPCI-SCC-154 cells between (A) the combination of AZD4547 (5 μ M) and BEZ235 (0.5 μ M), and the best single drug effect of AZD4547 (5 μ M) or BEZ235 (0.5 μ M); (B) the combination of AZD4547 (5 μ M) and BEZ235 (1 μ M), and the best single drug effect of AZD4547 (5 μ M) or BEZ235 (1 μ M); (C) the combination of AZD4547 (10 μ M) and BEZ235 (0.25 μ M) and the best single drug effect of AZD4547 (10 μ M) or BEZ235 (0.25 μ M); and (D) the combination of AZD4547 (10 μ M) and BEZ235 (1 μ M), and the best single drug effect of AZD4547 (10 μ M) or BEZ235 (1 μ M). Significance was tested for HPV⁻ UT-SCC-60A cells between (E) the combination of AZD4547 (5 μ M) and BEZ235 (0.5 μ M), and the best single drug effect of AZD4547 (5 μ M) or BEZ235 (0.5 μ M); (F) the combination of AZD4547 (5 μ M) and BEZ235 (1 μ M), and the best single drug effect of AZD4547 (5 μ M) or BEZ235 (1 μ M); (G) the combination of AZD4547 (10 μ M) and BEZ235 (0.25 μ M) and the best single drug effect of AZD4547 (10 μ M) or BEZ235 (0.25 μ M); and (H) the combination of AZD4547 (10 μ M) and BEZ235 (1 μ M), and the best single drug effect of AZD4547 (10 μ M) or BEZ235 (1 μ M). Significance was assessed using Student's t-test between the combination and the best single drug effect. *P<0.05 and ***P<0.001 vs. best single drug effect. The mean \pm SD of three independent experiments is presented. CI, combination index; HPV, human papillomavirus.

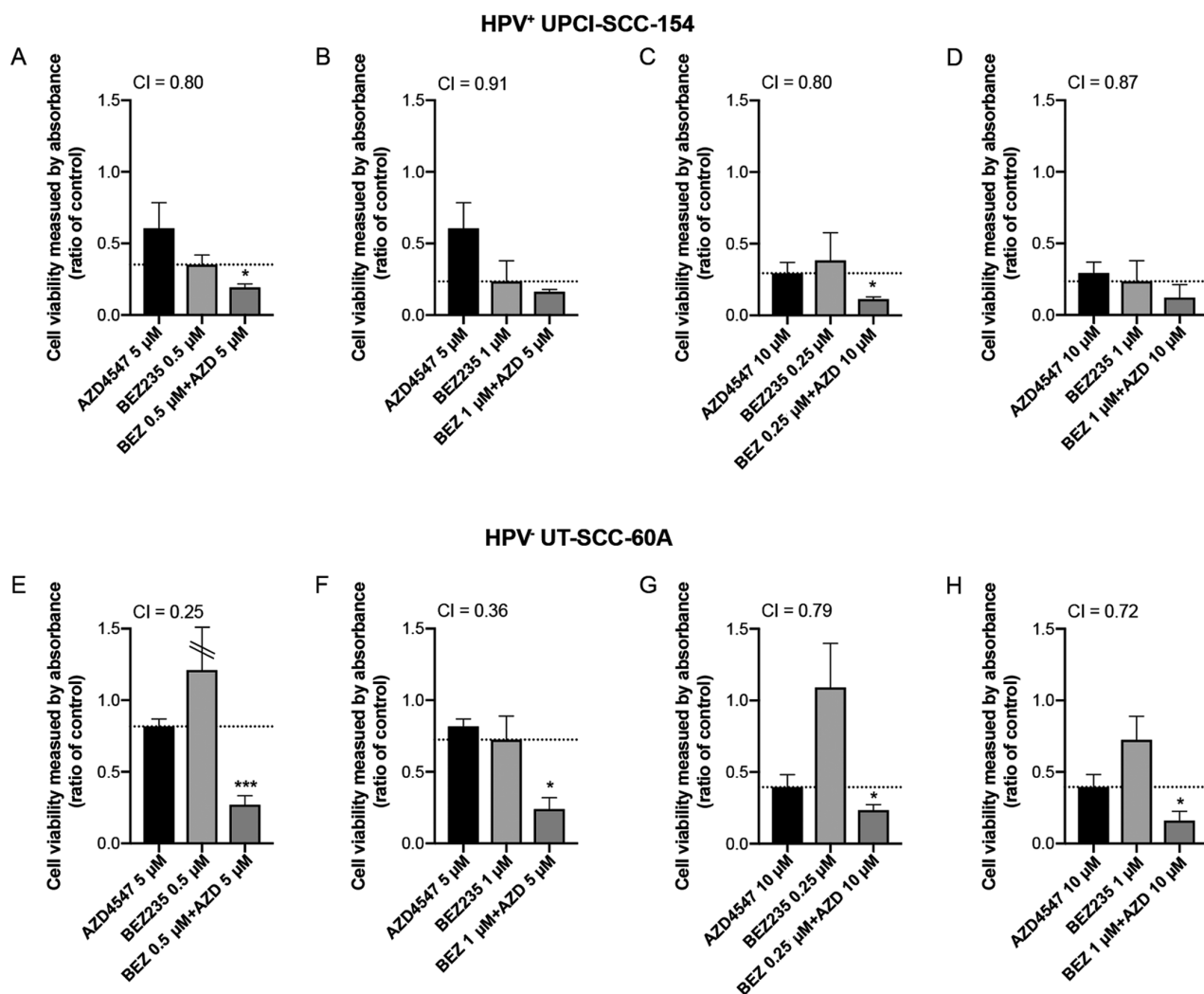


Figure S2. Heatmap of CI from combination treatment with AZD4547 and BEZ235 in HPV⁺ UPCI-SCC-154 and HPV⁻ UT-SCC-60A cells. CI from effects on viability were calculated according to the Highest Single Agent approach. A CI <1 denotes a positive combination effect, represented by green color, and a CI >1 a negative effect, represented by red color. Viability was evaluated using WST-1 assays at 24, 48 and 72 h, and each experiment was repeated three times. Significant differences between the best single drug effect and the combination effect are shown. *P<0.05, **P<0.01, ***P<0.001 vs. best single drug effect. CI, combination index; HPV, human papillomavirus; N.d., not defined; ns, not significant.

