Supplementary information to:

Arsenite impairs BRCA1-dependent DNA double-strand break repair, a mechanism potentially contributing to genomic instability

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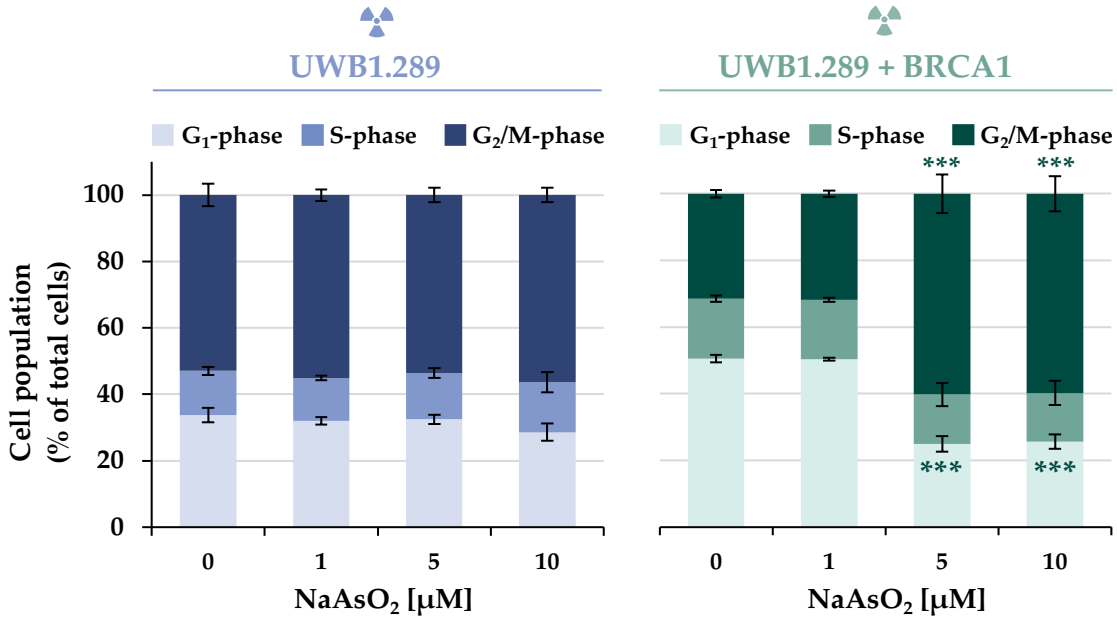

Figure S1: Cell cycle distribution of UWB1.289 and UWB1.289 + BRCA1 cells after treatment with $NaAsO_2$. Cells were pre-incubated with $NaAsO_2$ for 18 h, irradiated with 1 Gy, and post-incubated with $NaAsO_2$ for 8 h. Cell cycle distribution was analyzed by DAPI staining using flow cytometry. Shown are mean values ± standard deviations derived from three independent experiments. Statistically significant difference from control as determined by ANOVA followed by Dunnett`s T post hoc test: ***$p \leq 0.001$.

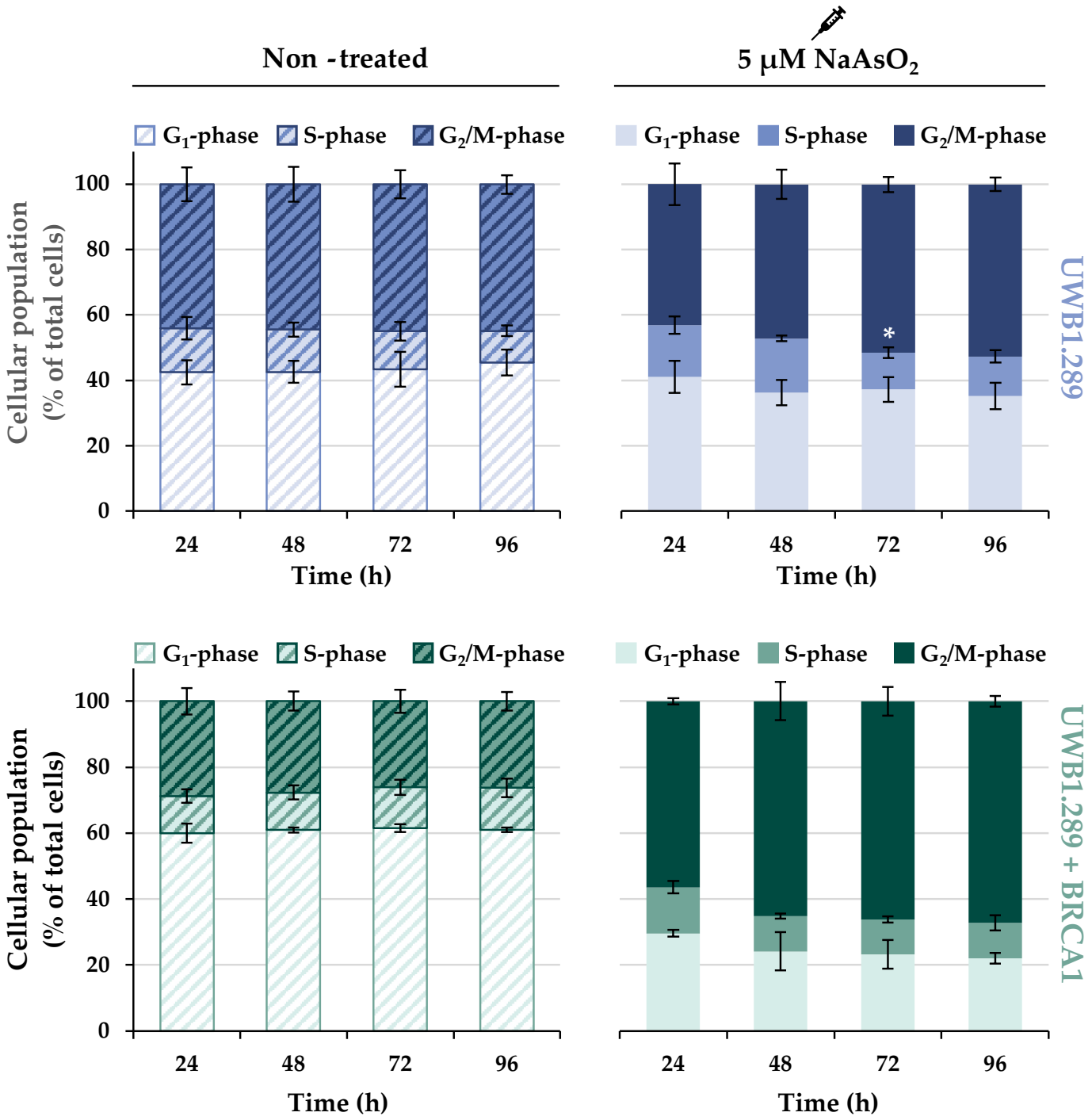

Figure S2: Time-dependent analysis of the cell cycle distribution of UWB1.289 and UWB1.289 + BRCA1 cells after treatment with $NaAsO_2$. Cells were treated with 5 µM $NaAsO_2$ and post-incubated for 24 h to 96 h. Cell cycle distribution was analyzed by DAPI staining using flow cytometry. Shown are mean values ± standard deviations derived from three independent experiments. Statistically significant difference from control as determined by ANOVA followed by Dunnett`s T post hoc test: *$p \leq 0.05$.

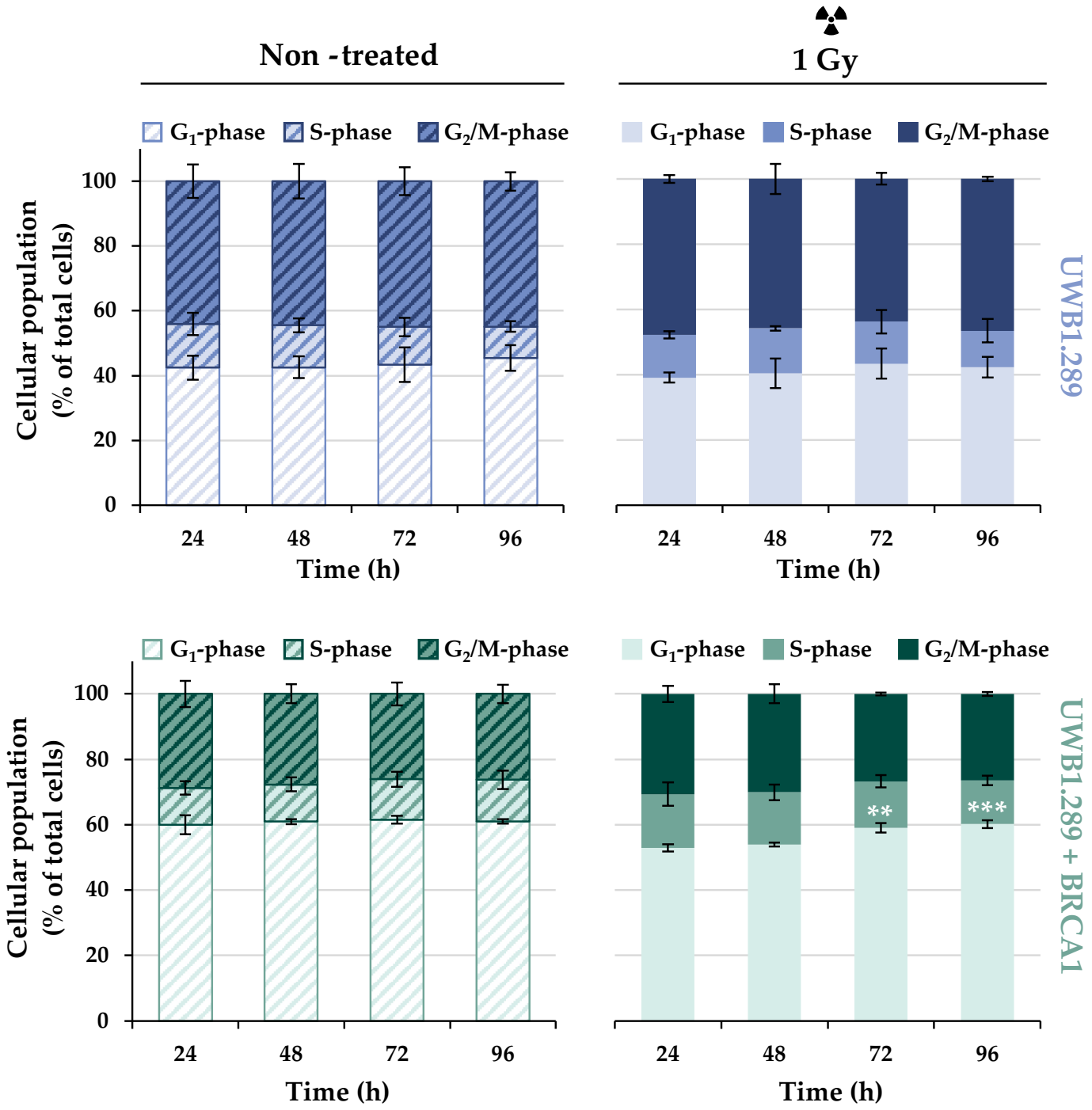

Figure S3: Time-dependent analysis of the cell cycle distribution of UWB1.289 and UWB1.289 + BRCA1 cells after exposure to IR. Cells were irradiated with 1 Gy and post-incubated for 24 h to 96 h. Cell cycle distribution was analyzed by DAPI staining using flow cytometry. Shown are mean values ± standard deviations derived from three independent experiments. Statistically significant difference from control as determined by ANOVA followed by Dunnett`s T post hoc test: **$p \leq 0.01$, ***$p \leq 0.001$.

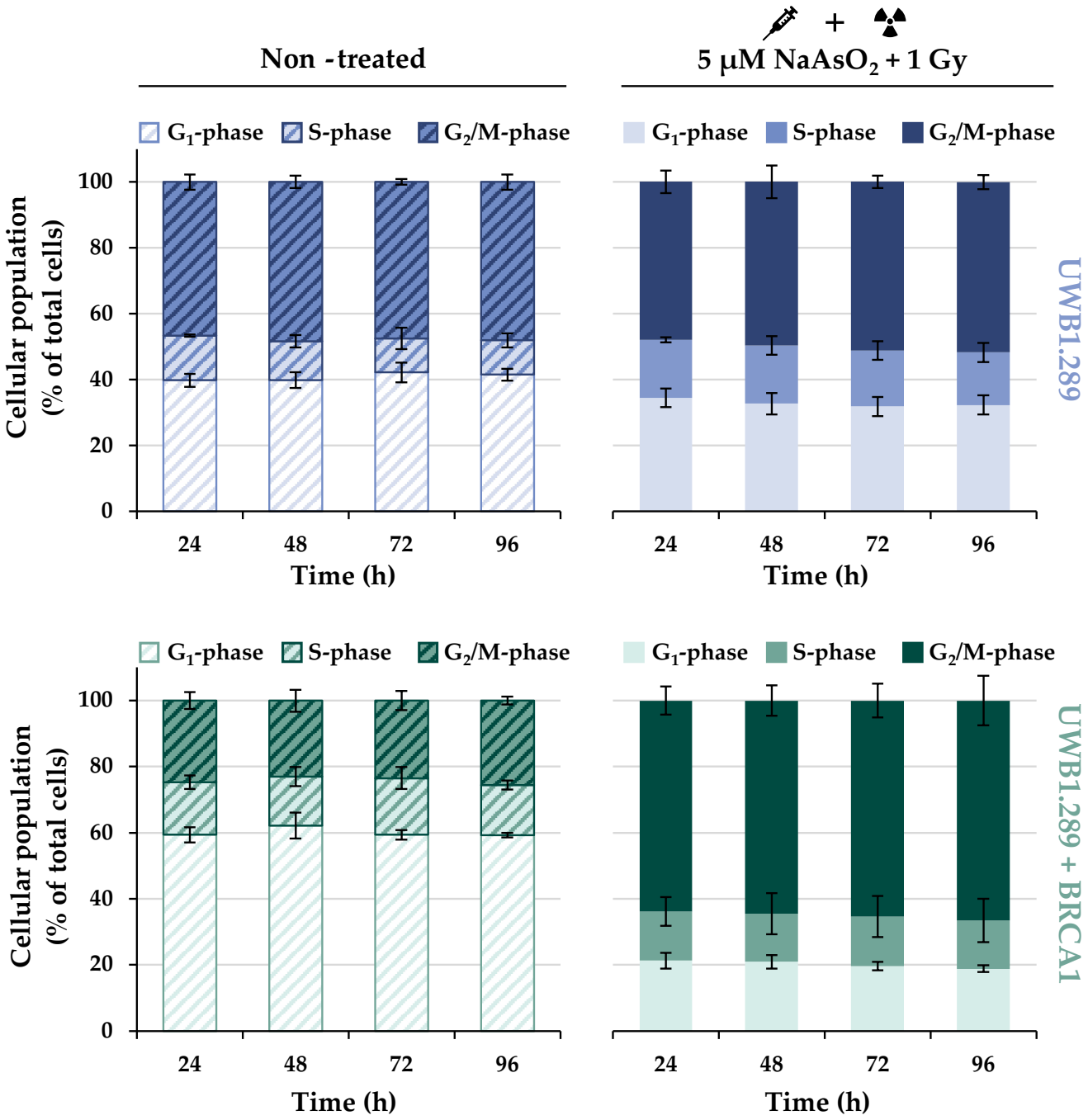

Figure S4: Time-dependent analysis of the cell cycle distribution of UWB1.289 and UWB1.289 + BRCA1 cells after co-exposure to $NaAsO_2$ and IR. Cells were pre-incubated with $NaAsO_2$ for 18 h, irradiated with 1 Gy, and post-incubated with $NaAsO_2$ for 24 h to 96 h. Cell cycle distribution was analyzed by DAPI staining using flow cytometry. Shown are mean values ± standard deviations derived from three independent experiments. Statistically significant difference from control as determined by ANOVA followed by Dunnett`s T post hoc test.

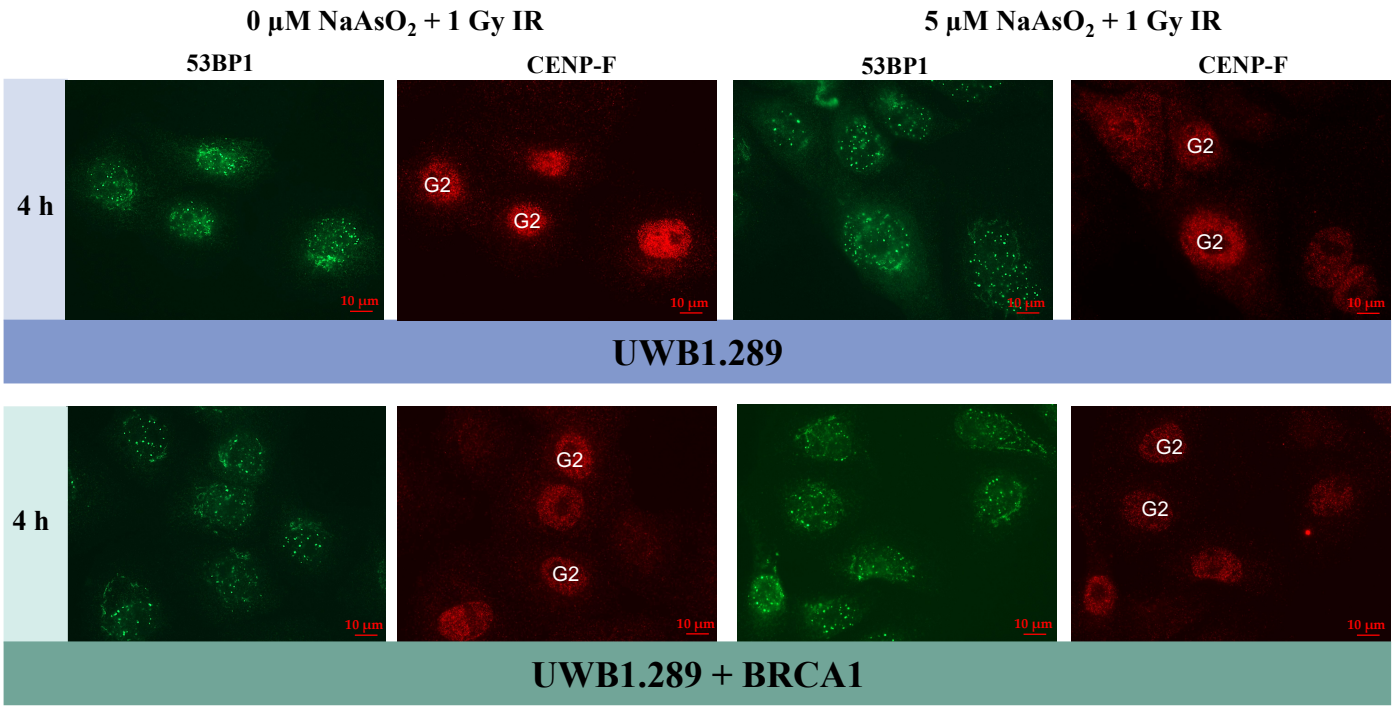

Figure S5: Impact on 53BP1 foci formation in UWB1.289 and UWB1.289 + BRCA1 cells after treatment with $NaAsO_2$. Cells were pre-incubated with $NaAsO_2$ for 18 h, irradiated with 1 Gy, and post-incubated with $NaAsO_2$ 4 h. The cells were stained using specific primary and secondary antibodies against 53BP1, as well as CENP-F against the centromere protein F.

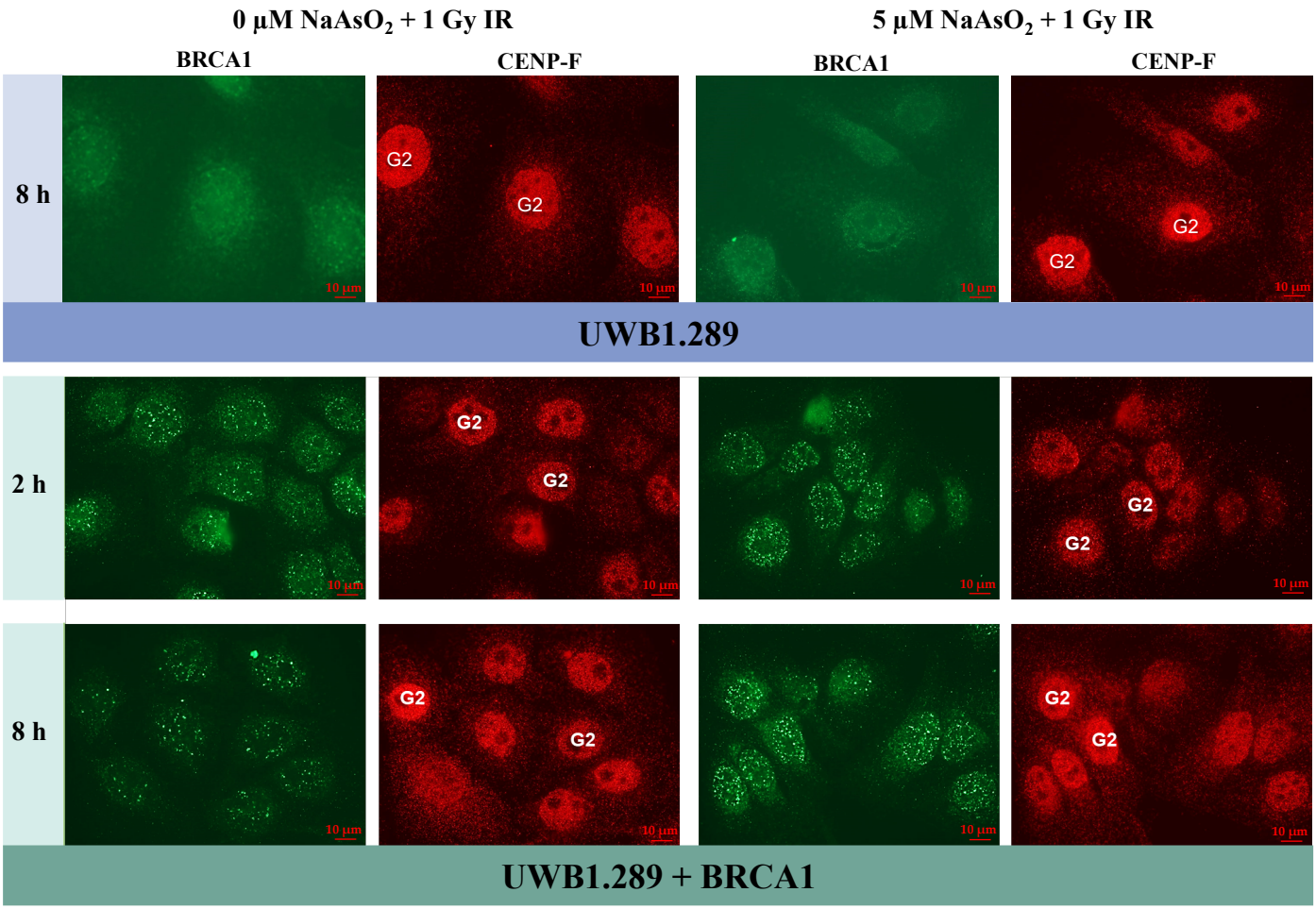

Figure S6: Impact on BRCA1 foci formation in UWB1.289 and UWB1.289 + BRCA1 cells after treatment with $NaAsO_2$. Cells were pre-incubated with $NaAsO_2$ for 18 h, irradiated with 1 Gy, and post-incubated with $NaAsO_2$ 2 h and 8 h. The cells were stained using specific primary and secondary antibodies against 53BP1, as well as CENP-F against the centromere protein F.

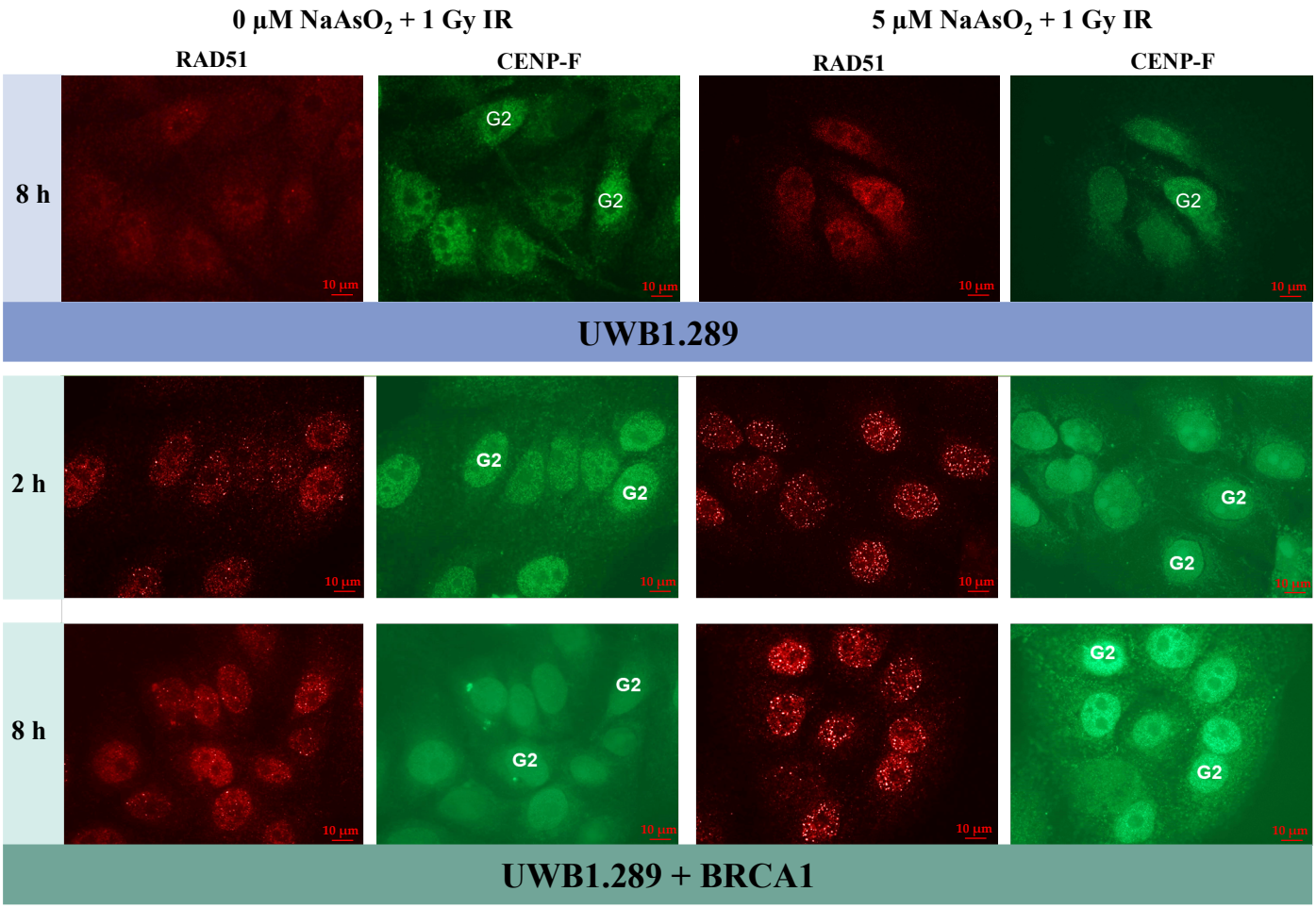

Figure S7: Impact on RAD51 foci formation in UWB1.289 and UWB1.289 + BRCA1 cells after treatment with $NaAsO_2$. Cells were pre-incubated with $NaAsO_2$ for 18 h, irradiated with 1 Gy, and post-incubated with $NaAsO_2$ 2 h and 8 h. The cells were stained using specific primary and secondary antibodies against 53BP1, as well as CENP-F against the centromere protein F.

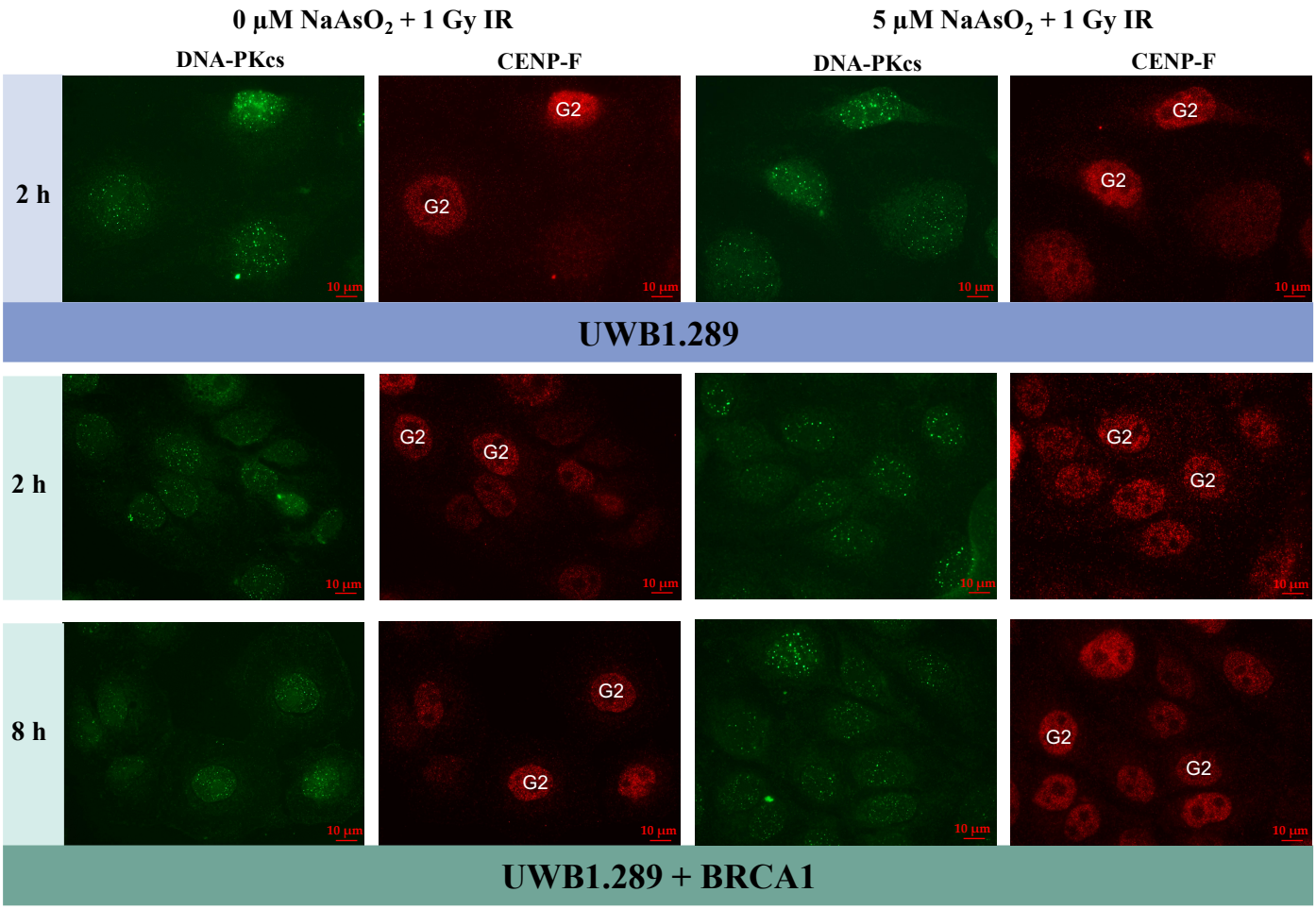

Figure S8: Impact on DNA-PKcs foci formation in UWB1.289 and UWB1.289 + BRCA1 cells after treatment with $NaAsO_2$. Cells were pre-incubated with $NaAsO_2$ for 18 h, irradiated with 1 Gy, and post-incubated with $NaAsO_2$ 2 h and 8 h. The cells were stained using specific primary and secondary antibodies against DNA-PKcs, as well as CENP-F against the centromere protein F.

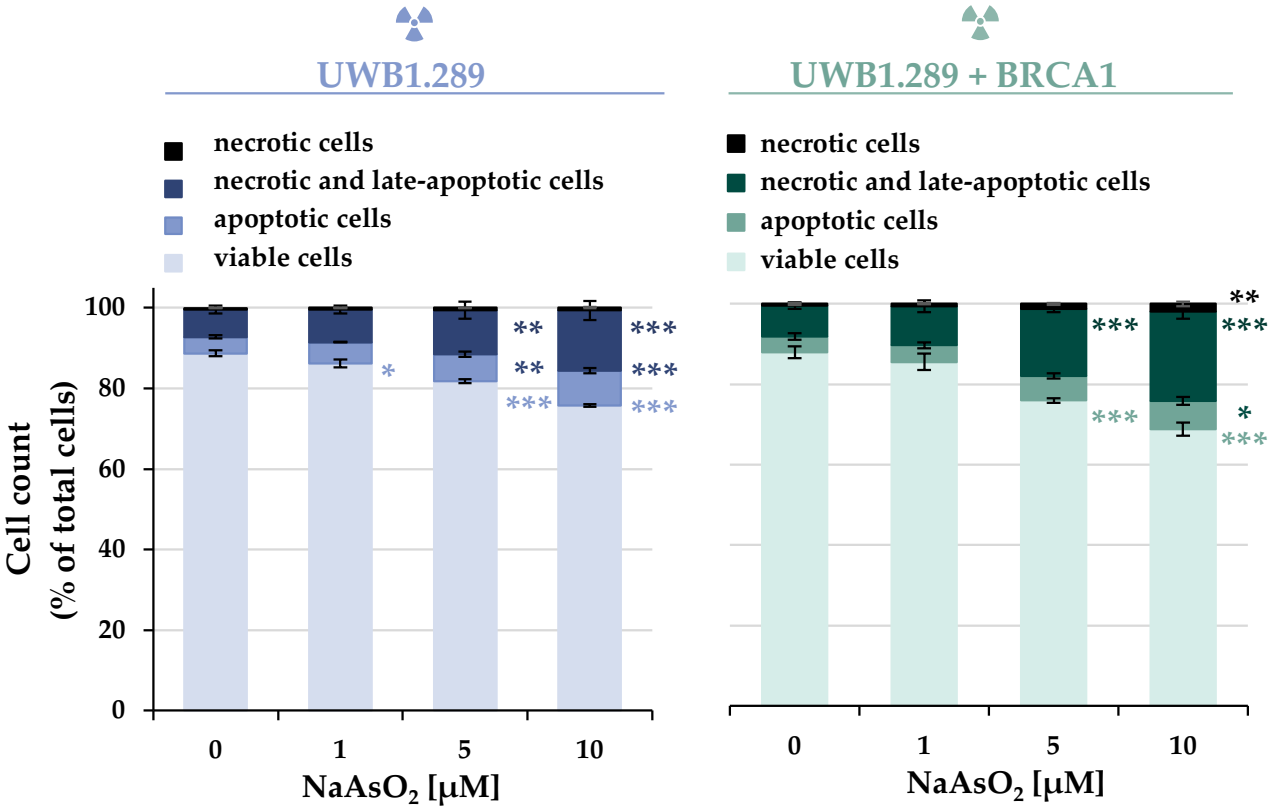

Figure S9: Analysis of apoptotic and necrotic cells after treatment with $NaAsO_2$. Cells were pre-incubated with $NaAsO_2$ for 18 h, irradiated with 1 Gy, and post-incubated with $NaAsO_2$ for 8 h To distinguish between necrotic, necrotic, and late-apoptotic, apoptotic, and viable cells, cells were stained with Annexin V-FITC and propidium iodide (PI). Cell count was then analyzed using flow cytometry. Shown are mean values ± standard deviations derived from three independent experiments. Statistically significant difference from control as determined by ANOVA followed by Dunnett`s T post hoc test: *$p \leq 0.05$, **$p \leq 0.01$, ***$p \leq 0.001$.