Figure S2  BrdU and EdU cause prolonged DNA synthesis, cell cycle slowing and DNA damage (related to Figure 3).  
A. Sytox Green stained cells (from Figures 3A, 3B) were analyzed by flow cytometry to highlight progression to 4C DNA (second S-phase; using modified cytometer settings). Non-incorporating (non-inc) or hsv-tk\(^\ast\) hENT\(^\ast\) cells (Inc) at indicated doses of BrdU or EdU. Note that 4C peak accumulation is consistent with septation index peaks (Figure 3A, 3B), indicating the second S-phase after release. B. Forward scatter (FSC) dynamics of cells in A, indicating cell size during experiment. Left-shift toward smaller cell size (M-phase) occurs slightly later than septation (S-phase; Figure 3A, 3B). C. Cells were stained with DAPI and aniline blue to detect nuclei and septa, respectively, before or after 6h of BrdU or EdU treatment. Wild-type (wt) incorporating cells elongate during prolonged exposure. Both chk1\(^\Delta\) and rad3\(^\Delta\) hsv-tk\(^\ast\) hENT\(^\ast\) cells continue to septate and divide, and many cells mis-segregate DNA (indicated by arrows). mrc1\(^\Delta\) and cdc1\(^\Delta\) hsv-tk\(^\ast\) hENT\(^\ast\) cells show an intermediate phenotype in EdU. Scale bar 10 µm. D. Abnormal DNA segregation events were scored as the percentage of cut or anucleate cells in the total population during BrdU treatment. Shown are combined data from 2 independent experiments, displayed as proportion of abnormal segregants ±95% CI. E. Abnormally segregated nuclei during EdU exposure. Shown are combined data from 2 independent experiments, displayed as proportion of abnormal segregants ±95% CI.