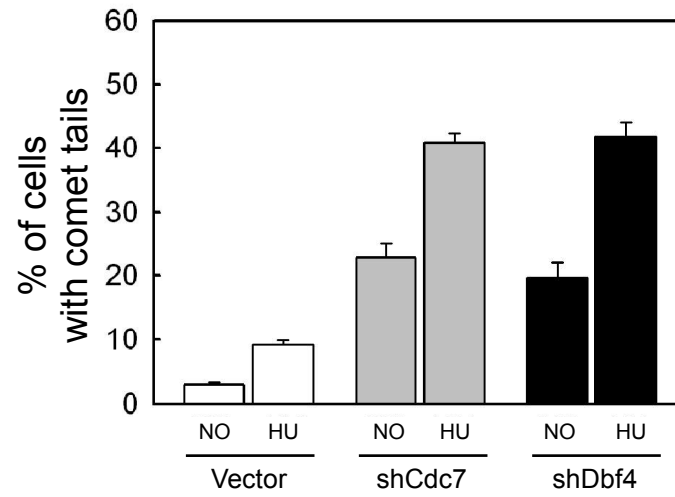
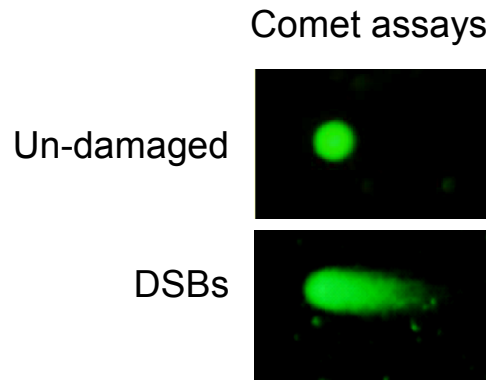
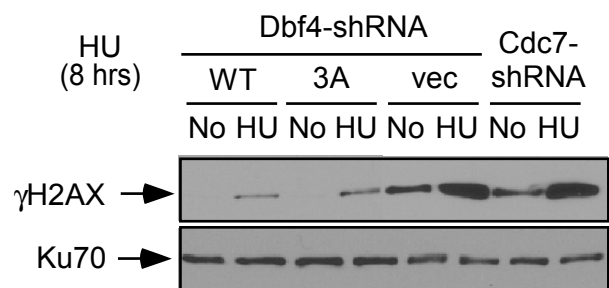


Supplementary Figure 2.





Supplementary Figure 4.

## Legends for Supplementary Figures

**Supplementary figure 1. The kinase activity of endogenous DDK to phosphorylate MCM2 is not changed after HU treatment.** (A). Endogenous Dbf4/Cdc7 was immunoprecipitated using anti-Cdc7 antibody with anti-flag antibody as a negative control. GST-MCM2 (1-169 aa) fragment was used in the *in vitro* kinase assay and phosphorylation was revealed by autoradiography ( $^{32}\text{P}$ ). (B). 293T cells were treated with or without HU (1 mM, 24 hr) and anti-Cdc7 immunoprecipitation was performed. GST-MCM2 (1-169 aa) fragment or GST was used as substrates in the *in vitro* kinase assay. Phosphorylation was revealed by autoradiography ( $^{32}\text{P}$ ) and the immunoprecipitation of endogenous Cdc7 is shown by Western blot.

**Supplementary figure 2. Suppression of Dbf4 or Cdc7 does not alter HU-induced cell cycle arrest.** FACS analysis was performed using U2OS cells infected with shRNAs for Dbf4 or Cdc7 before and after treatment of HU (1mM, 8hrs).

**Supplementary figure 3. Comet assays indicate DSB formation when replication forks are collapsed.** Neutral comet assays were performed using U2OS cells infected with shRNAs for Dbf4 or Cdc7 or pMKO vector control before or after treatment of HU (1 mM, 8 hrs). Nuclei with a comet tail larger than 2 nuclear diameters were counted as positive for cells containing substantial DSBs. Two typical cells with or without comet tails are shown on the left. Fifty cells were counted for each experiment and the percentage of nuclei with tails was the number of positive for DNA damage divided by the total number of nuclei counted. Results are presented as range of three independent experiments.

**Supplementary figure 4. Dbf4 and Cdc7 are important for fork protection.** U2OS cells expressing Flag-tagged wild-type Dbf4 (WT), the Dbf4-3A mutant (3A) or vector infected with Dbf4-shRNAs, or U2OS cells infected with Cdc7shRNAs were mock-treated (No) or treated with HU (1 mM, 8 hrs). Chromatin fractions were prepared and immunoblotted with anti- $\gamma\text{H2AX}$  and Ku70 antibodies.