

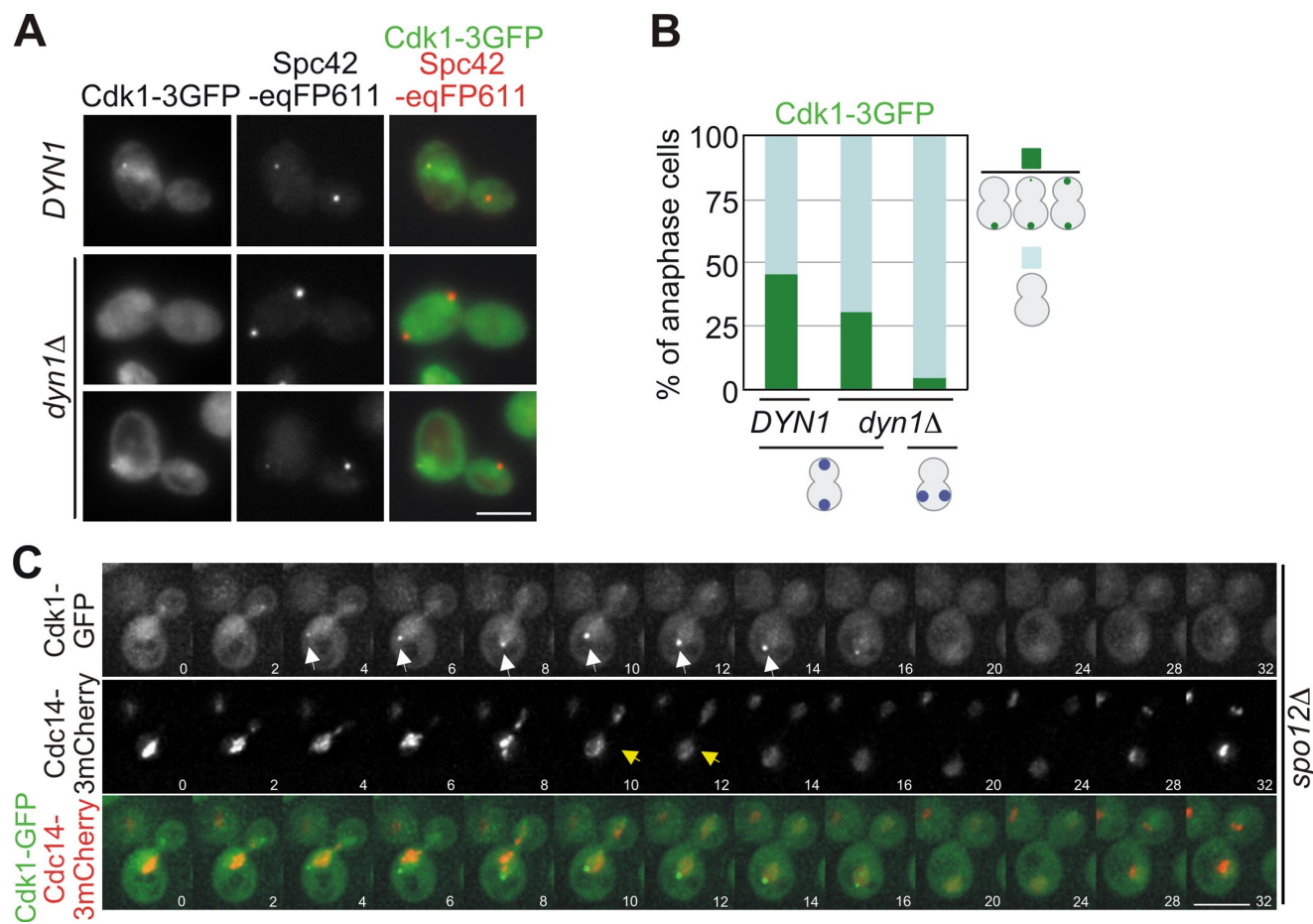
König et al., <http://www.jcb.org/cgi/content/full/jcb.200911128/DC1>

Figure S1. **SPB binding of Cdk1 in anaphase cells.** (A) Cdk1 binding to SPBs is reduced in *dyn1Δ* cells with a misaligned anaphase spindle. *DYN1 CDK1-3GFP SPC42-eqFP611* and *dyn1Δ CDK1-3GFP SPC42-eqFP611* cells were grown in YPAD medium at 14°C for 20 h. Cells with correctly aligned and misaligned anaphase spindles were analyzed for SPB binding of Cdk1. Bar, 5 μm. (B) Quantification of A. *n* > 50 anaphase cells per strain. (C) Time-lapse analysis of FEAR-defective *CDK1-GFP CDC14-3mCherry spo12Δ* cells. Cells were grown in SC medium at 30°C. Consecutive sections were taken every 2 min. Shown are deconvolved and projected images. Cdk1-GFP binds to the mSPB in early anaphase (*t* = 4 min) when Cdc14-3mCherry is still in the nucleolus. Cdc14-3mCherry becomes released from the nucleolus at *t* = 10–12 min. White arrows indicate Cdk1-GFP at mSPB and yellow arrows indicate release of Cdc14-3mCherry from the nucleolus. Bars, 5 μm.

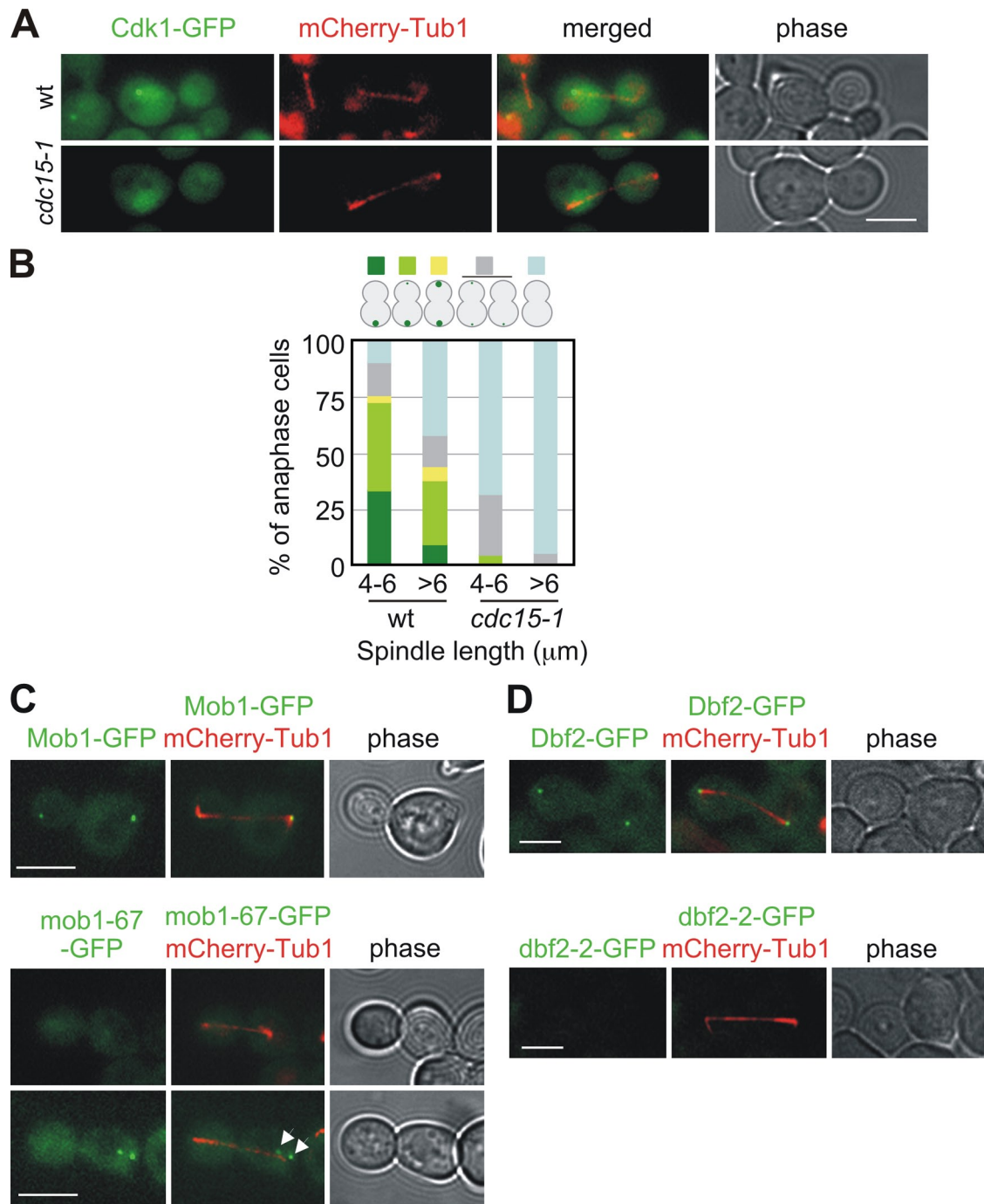


Figure S2. Role of MEN components for Cdk1 localization to the mSPB. (A) Cdc15 is required for anaphase SPB association of Cdk1. Synchronized wild-type cells and *cdc15-1* cells were grown in YPAD at 37°C. Anaphase cells were examined for SPB localization of Cdk1-GFP. (B) Quantification of A. $n > 50$ cells were analyzed per spindle length category. (C) *mob1-67*-GFP does not localize to SPBs at the restrictive temperature. *MOB1*-GFP and *mob1-67*-GFP cells with *mCherry-TUB1* were arrested in G1 by α -factor at 23°C. Synchronized cells were then released into a new cell cycle at 37°C. Cells in anaphase were examined for Mob1-GFP and *mob1-67*-GFP localization. Most of cells did not show discrete GFP signal (top *mob1-67* panel). Some cells showed *mob1-67*-GFP dots in the cytoplasm that did not colocalize with SPBs (white arrows in the bottom *mob1-67* panel). (D) *dbf2-2*-GFP does not localize to SPBs at the restrictive temperature. *DBF2*-GFP and *dbf2-2*-GFP cells with *mCherry-TUB1* were arrested in G1 by α -factor at 23°C. Cells were then released into a new cell cycle at 37°C. Cells in anaphase were examined for Dbf2-GFP and *dbf2-2*-GFP localization. *dbf2-2* cells did not show discrete GFP signals. Bars, 5 μ m.

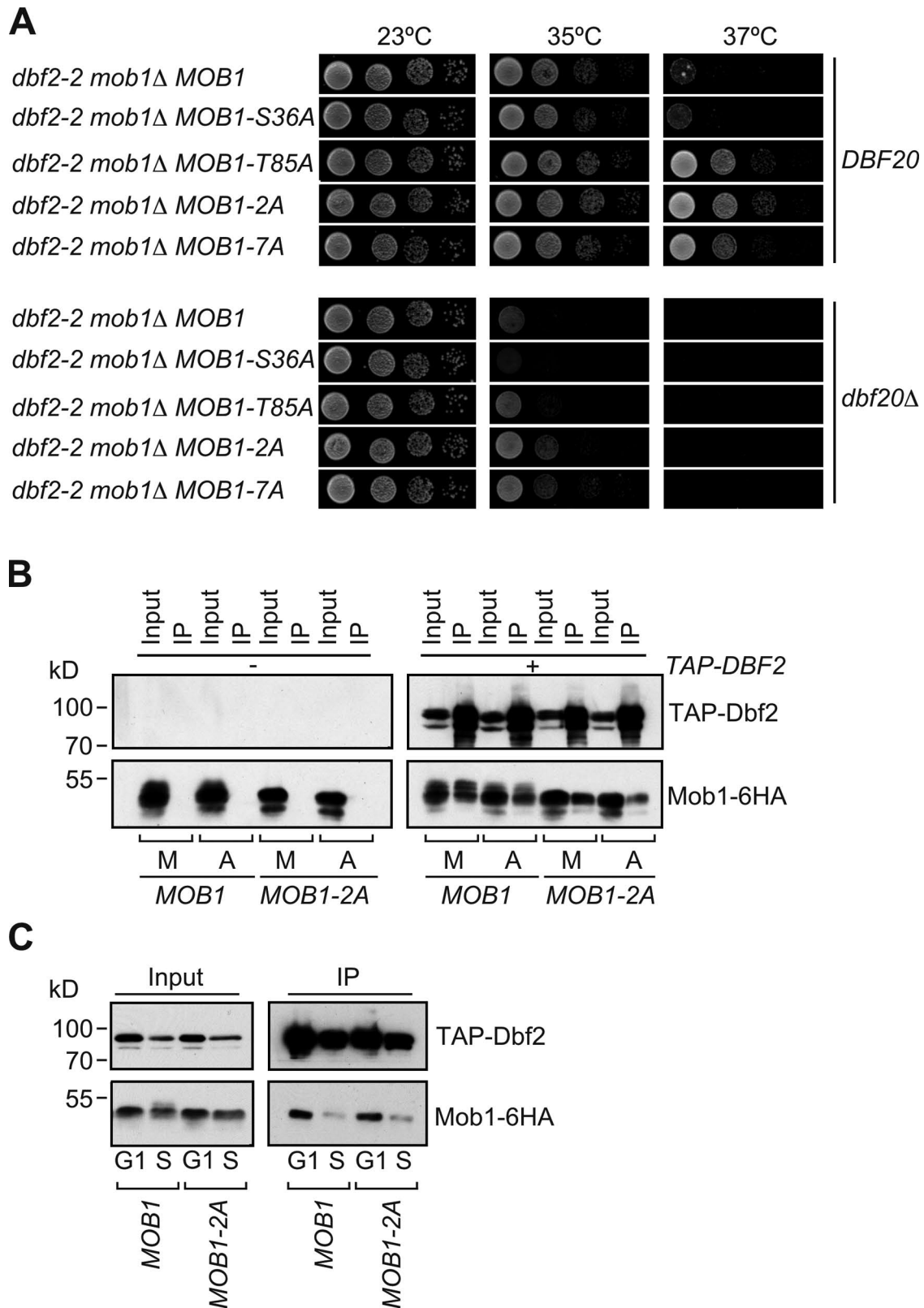


Figure S3. **Role of Mob1 phosphorylation by Cdk1.** (A) Log-phase cells with indicated genotypes were serially diluted 10-fold in PBS and spotted onto YPD plates. Plates were incubated for 2 d at 23, 35, or 37°C. (B and C) Mob1 phosphorylation does not affect complex formation of Mob1 and Dbf2. (B) TAP-Dbf2 was immunoprecipitated (IP) from metaphase (M) and anaphase (A) cells. (C) Cells arrested in G1 or S phase were used for the TAP-Dbf2 immunoprecipitation experiment. (B and C) Mob1 was detected with anti-HA antibodies.

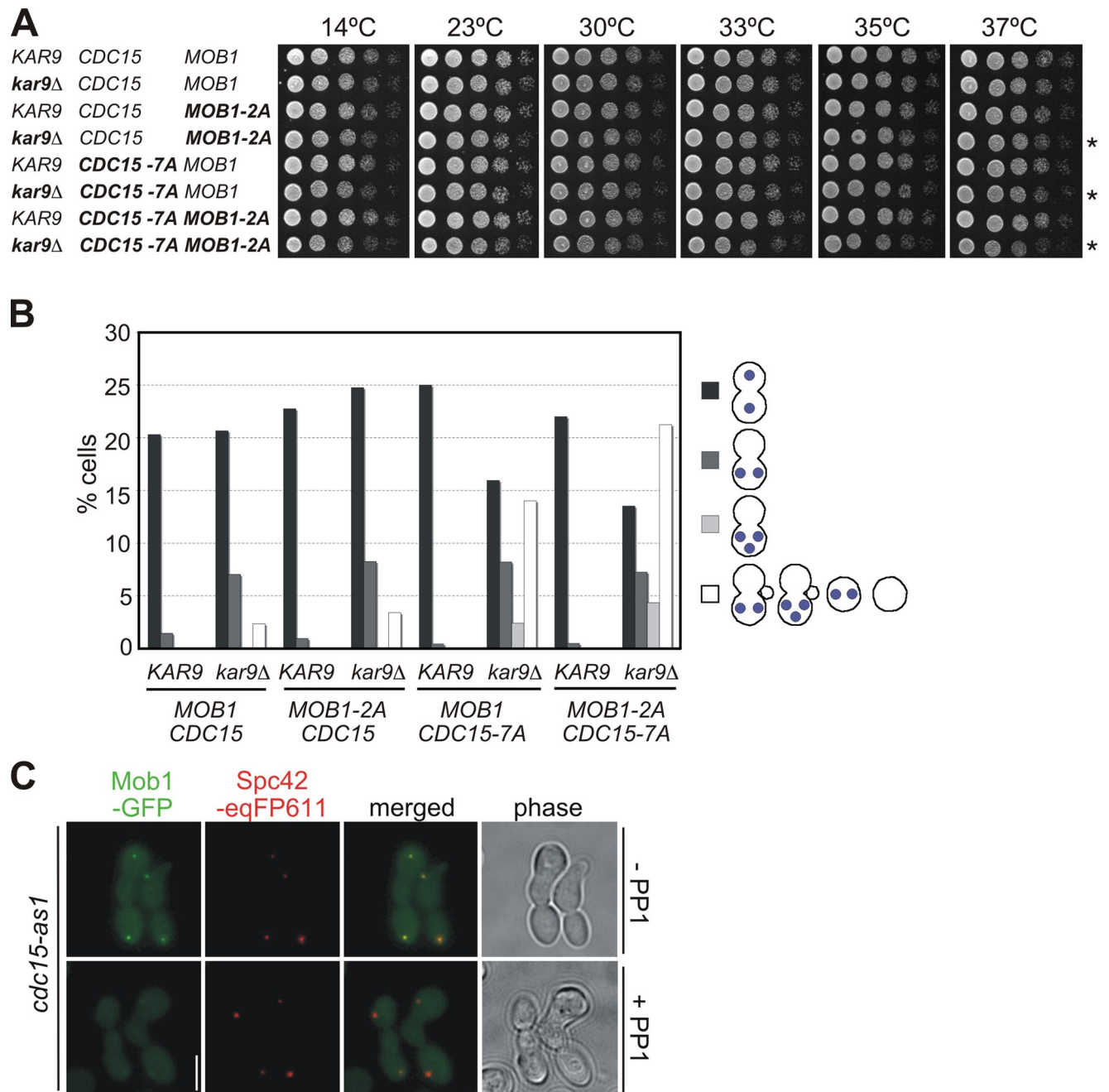


Figure S4. ***CDC15-7A* and *MOB1-2A* mutants show genetic interactions with *kar9Δ*; role of Cdc15 activity for Mob1 SPB binding.** (A) Log-phase cells with indicated genotypes were serially diluted fivefold in PBS and spotted onto YPD plates. Plates were incubated for 2–3 d at indicated temperatures. Strains marked with asterisks show slightly reduced growth at 37°C. (B) Strains with indicated genotypes were pregrown at 23°C and then incubated for 3 h at 30°C. Cells were fixed, and DNA was stained with DAPI. More than 200 cells were analyzed and categorized per strain as illustrated in the figure. (C) Binding of Mob1-GFP to the mSPB requires kinase activity of Cdc15. *CDC15* (not depicted) and *cdc15-as1* anaphase cells with *MOB1-GFP SPC42-eqFP611* were grown in YPAD in the presence (+PP1) or absence (–PP1) of the inhibitor PP1 and analyzed by fluorescence microscopy 70 min after release from α -factor arrest. Bar, 5 μ m.