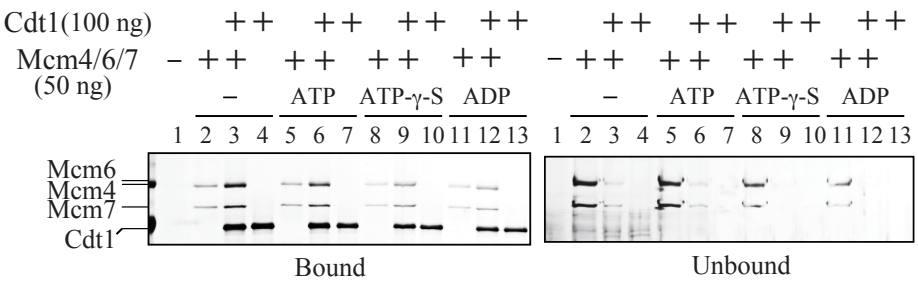
Supplemental figure legends

Supplemental figure 1. Cdt1 facilitates DNA binding of Mcm4/6/7. A biotin-labeled ssDNA oligonucleotide (37mer-dT_{40}) was incubated with indicated proteins in the gel-shift buffer as described in "EXPERIMENTAL PROCEDURES", and the oligonucleotide was pulled down using M-280 streptoavidin Dynabeads (Dynal). The bound (left panel) and unbound (right panel) proteins were then separated on a 4-20% polyacrylamide gel followed by silver staining. The amounts of protein added were; Mcm4/6/7, 50ng; Cdt1, 100ng.

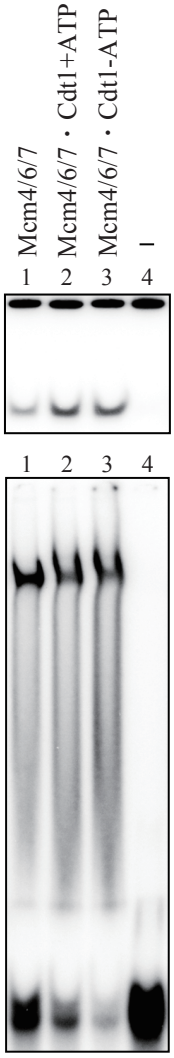
Supplemental figure 2. Mcm4/6/7-Cdt1 complex exhibits more efficient helicase activity than the Cdt1-free Mcm4/6/7. (A) DNA helicase (upper) and DNA-binding (lower) activities were examined on a partial heterodulpex substrates (10 fmole) or on ssDNA (20 fmole), respectively. The peak fraction of purified Mcm4/6/7 (lanes 1) and Mcm4/6/7-Cdt1 complex (lanes 2 and 3) complexes from glycerol gradient fractionation conducted as described in Fig. 6, were used for assays. ATP was either present (1 mM, lane 2) or absent (lane 3) in glycerol gradient centrifugation. In gel-shift assays, complexes were separated on a 5% native polyacrylamide gel. Reaction conditions were adjusted so that all the reactions are identical in terms of buffer conditions (50 mM Hepes-Na [pH 7.5], 87.5 mM Na-acetate, 11.25% glycerol, 0.375 mM EDTA, 0.75 mM DTT, 5 mM 2-mercaptoethanol, 0.0075% Triton X-100, 4 mM MgOAc, and 10 mM ATP) and the amount of Mcm4/6/7 present (75 ng). The half of the reactions was incubated at 30°C for gel-shift assays and the remainder was incubated at 37°C for DNA helicase assays. (B) The levels of DNA helicase activities, represented as per cent oligonucleotide displaced, are shown for each complex. The helicase activity of the isolated Mcm4/6/7-Cdt1 complex is higher than that of the Cdt1-free Mcm4/6/7 complex.

Supplemental figure 3. Bacterially produced Cdt1 stimulates the DNA binding and helicase activities of Mcm4/6/7. The wild-type or KKAA Cdt1 overproduced and purified from *E. coli* was titrated (6 ng, 12 ng, 24 ng, and 36 ng) in DNA binding (lower) and helicase (upper) assays for Mcm4/6/7 (25 ng).



Supplement Figure 1, You and Masai

A



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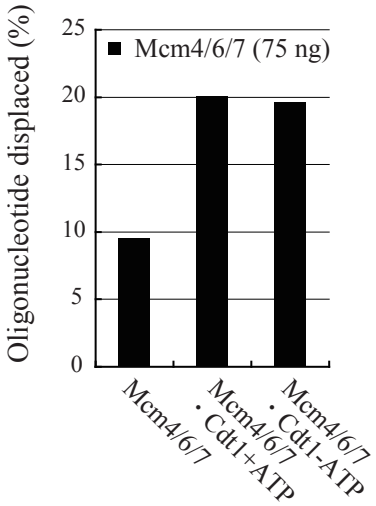
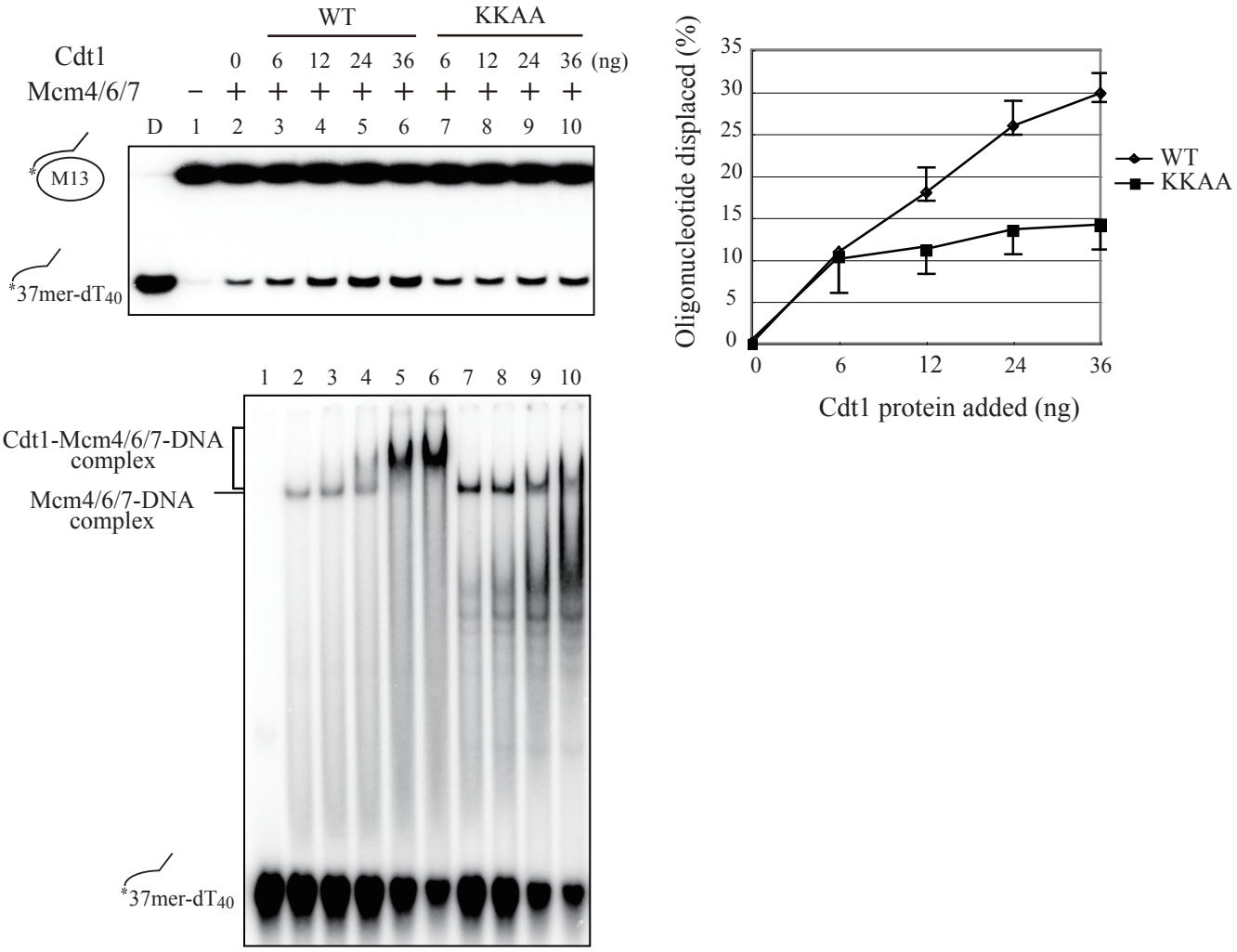


Supplement Figure 2, You and Masai



Supplement Figure 3, You and Masai