

**Supplemental Table 1**

Yeast strains used in this study

Strain	Genotype
BY4741 <sup>1</sup>	<i>MATa his3Δ1 leu2Δ0 met15Δ0 ura3Δ0</i>
Y2454 <sup>1</sup>	<i>MATα mfa1Δ::MFA1pr-HIS3 can1Δ his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0</i>
YBL165b <sup>1</sup>	<i>MATα mfa1Δ::MFA1pr-HIS3 can1Δ his3Δ1 leu2Δ0 lys2Δ0 ura3Δ0 ipl1-321::natR</i>
SBY3	<i>Mata bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1</i>
SBY818	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1</i>
SBY1897	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1 mcm21ΔKAN</i>
SBY1983	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1 mcm22ΔKAN</i>
SBY5551	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1Δ:KAN mcm21ΔKAN</i>
SBY6133	<i>Mata PDS1-myc18:LEU2 LacOChrIII1437kb:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1 lys2Δ can1-100 ade2-1</i>
SBY6134	<i>Mata PDS1-myc18:LEU2 LacOChrIII1437kb:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1 lys2Δ can1-100 ade2-1 mcm21ΔKAN</i>
SBY6940	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1ΔKAN</i>
SBY6942	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1:pGAL-PDS1 mDB:URA3 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1</i>
SBY6943	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1:pGAL-PDS1 mDB:URA3 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1 mcm21ΔKAN</i>
SBY6944	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1:pGAL-PDS1 mDB:URA3 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1ΔKAN</i>
SBY6945	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1:pGAL-PDS1 mDB:URA3 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1Δ:KAN mcm21ΔKAN</i>
SBY6993	<i>Mata bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1ΔKAN</i>
SBY6998	<i>Mata PDS1-myc18:LEU2 bar1 ura3-1:pGAL-NC-MCD1:URA3 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1</i>

SBY6999 *Mata PDS1-myc18:LEU2 bar1 ura3-1:pGAL-NC-MCD1:URA3 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1 mcm21ΔKAN*

SBY7002 *Mata bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1 MCD1-6HA*

SBY7007 *Mata bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1 MCD1-6HA mcm21ΔKAN*

SBY7018 *Mata bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1 IPL1-Flag:KAN*

SBY7019 *Mata bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1 lys2Δ IPL1-Flag:KAN mcm21ΔKAN*

SBY7651 *Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1 lys2Δ SCC2-6HIS-3Flag:KAN*

SBY7652 *Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1 lys2Δ SCC2-6HIS-3Flag:KAN mcm21ΔKAN*

SBY7656 *Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1 mcm21ΔKAN mad3ΔKAN*

SBY7846 *Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:URA:TRP:mcd1-1 lys2Δ can1-100 ade2-1*

SBY7847 *Mata PDS1-myc18:LEU2 bar1 ura3-1:pGAL-NC-MCD1:URA3 leu2-3,112 his3-11:pCUP1-GFP12-LacI12:HIS3 trp1-1:256LacO:URA:TRP:mcd1-1 lys2Δ can1-100 ade2-1*

SBY7871 *Mata bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12<sub>4</sub>:HIS3 trp1-1:256LacO:TRP1 can1-100 ade2-1*

SBY7872 *Mata bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12<sub>4</sub>:HIS3 trp1-1:256LacO:TRP1 can1-100 ade2-1 lys2Δ mcm21ΔKAN*

SBY7873 *Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12<sub>4</sub>:HIS3 trp1-1:256LacO:TRP1 can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1ΔKAN*

SBY7874 *Mata PDS1-myc18:LEU2 bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-LacI12<sub>4</sub>:HIS3 trp1-1:256LacO:TRP1 lys2Δ can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1ΔKAN mcm21ΔKAN*

SBY7876 *Mata bar1 ura3-1 leu2-3,112:LEU2-TetR-GFP his3-11 trp1-1 ade2-1 CEN5(13 kb)-TetO-URA3*

SBY7877 *Mata bar1 ura3-1 leu2-3,112:LEU2-TetR-GFP his3-11 trp1-1 ade2-1 CEN5(13 kb)-TetO-URA3 mcm21ΔKAN*

SBY7878 *Mata bar1 ura3-1 leu2-3,112:LEU2-TetR-GFP his3-11 trp1-1 ade2-1 CEN5(18 kb)-TetO-URA3*

SBY7879 *Mata bar1 ura3-1 leu2-3,112:LEU2-TetR-GFP his3-11 trp1-1 ade2-1 CEN5(18 kb)-TetO-URA3 mcm21ΔKAN*

SBY7880 *Mata ura3-1:URA3-TetO leu2-3,112:LEU2-TetR-GFP his3-11 trp1-1 ade2-1*

SBY7881 *Mata ura3-1:URA3-TetO leu2-3,112:LEU2-TetR-GFP his3-11 trp1-1 ade2-1 mcm21ΔKAN*

SBY8102 *Mata ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-Lacl12:HIS3 trp1-1 can1-100 ade2-1 MyxoGC-R-CEN3-PGK1-R:ChrIII*

SBY8103 *Mata ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-Lacl12:HIS3 trp1-1 can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1ΔKAN MyxoGC-R-CEN3-PGK1-R:ChrIII*

SBY8104 *Mata ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-Lacl12:HIS3 trp1-1 can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1ΔKAN MyxoGC-R-CEN3-PGK1-R:ChrIII*

SBY8105 *Mata ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-Lacl12:HIS3 trp1-1 can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1ΔKAN MyxoGC-R-CEN3-PGK1-R:ChrIII*

SBY8224 *Mata bar1 ura3-1 leu2-3,112 his3-11:pCUP1-GFP12-Lacl12<sub>4</sub>:HIS3 trp1-1 can1-100 ade2-1:7tetop-UB-R-IPL1:ADE2 ipl1Δ:KAN mcm21ΔKAN LacOChrIII1437kb:LEU2*

SBY8352 *Mata bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1 IPL1-GFP:KAN*

SBY8353 *Mata bar1 ura3-1 leu2-3,112 his3-11 trp1-1 can1-100 ade2-1 IPL1-GFP:KAN mcm21Δ:HIS3*

<sup>1</sup>All strains are isogenic with W303 except the denoted strains which are S288c.

### Supplemental Figure Legends

**Figure S1.** A representative image of unseparated and closely separated sister GFP foci in experiment outlined in Fig 2A.

**Figure S2.** Representative images of live, asynchronous *WT* and *mcm21Δ* cells containing *Ipl1-GFP* (SBY8352, SBY8353) are shown.

**Figure S3.** Loss of viability in *mcm21Δ deg-ipl1* mutants occurs during a single cell cycle. *WT*, *mcm21Δ*, *deg-ipl1*, and *mcm21Δ deg-ipl1* (SBY818, SBY1897, SBY6940, SBY5551) released from G1 into media containing doxycycline were plated for viability at the indicated time points. Pheromone was added back after bud emergence to prevent cells from entering a second cell cycle.

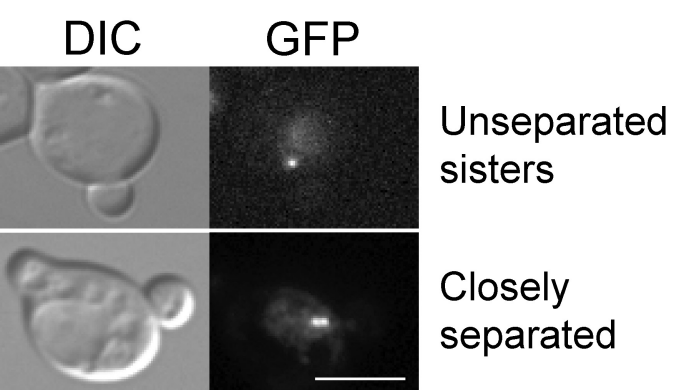
**Figure S4.** ChrIV segregation was analyzed in *mcm21 deg-ipl1* cells expressing *LacI<sub>4</sub>* that contained *LacO* sequences inserted at either the pericentromere or telomere (SBY7874, SBY8224) during a synchronous cell cycle in anaphase at a time when majority of the cells segregated their DAPI masses to opposite poles.

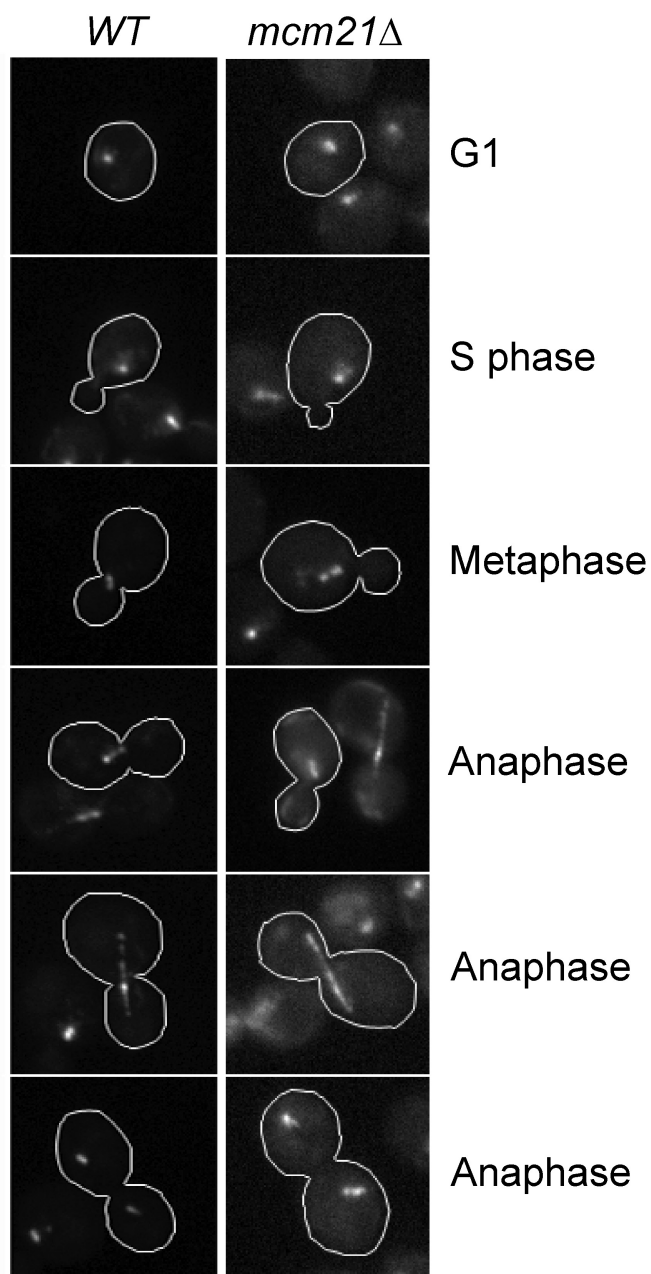
**Figure S5.** *IPL1* and *MCD1* genetic interaction profiles. Synthetic lethal and sick genetic interactions between the hypomorphic alleles *ipl1-321*, *mcd1-1*, and indicated yeast deletion strains.

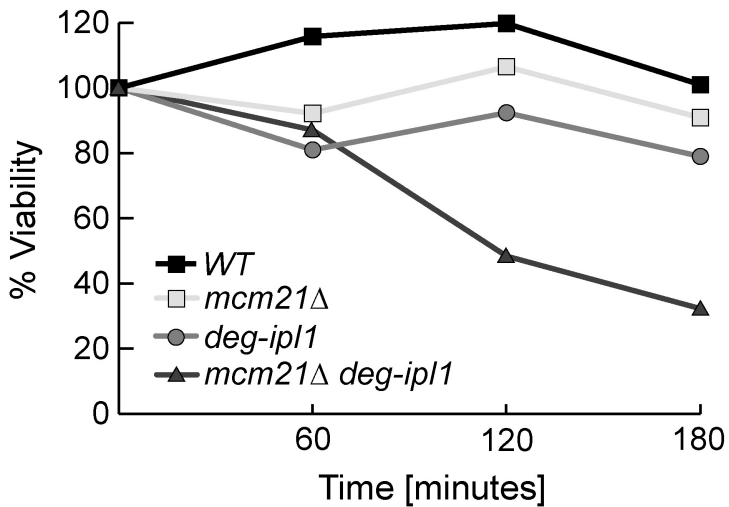
**Figure S6.** ChIP was performed on nocodazole-arrested *WT* and *mcm21Δ* cells containing *Mcm21-Flag*. Three-fold dilutions of both crude extract and immunoprecipitate samples were subjected to PCR reactions. The schematic illustrates

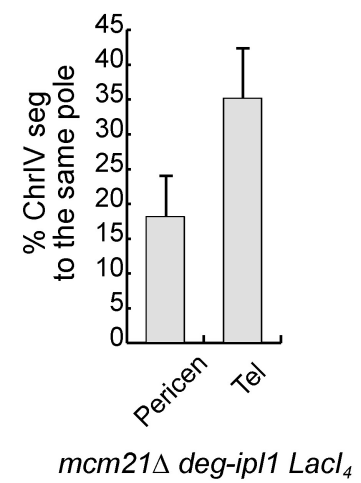
distance from the centromere on ChrIII as well as CAR and non-CAR sites as indicated by the orange rings.

**Figure S7.** Percentage of cells with buds was scored in the experiment described in Figure 6B and 6C.

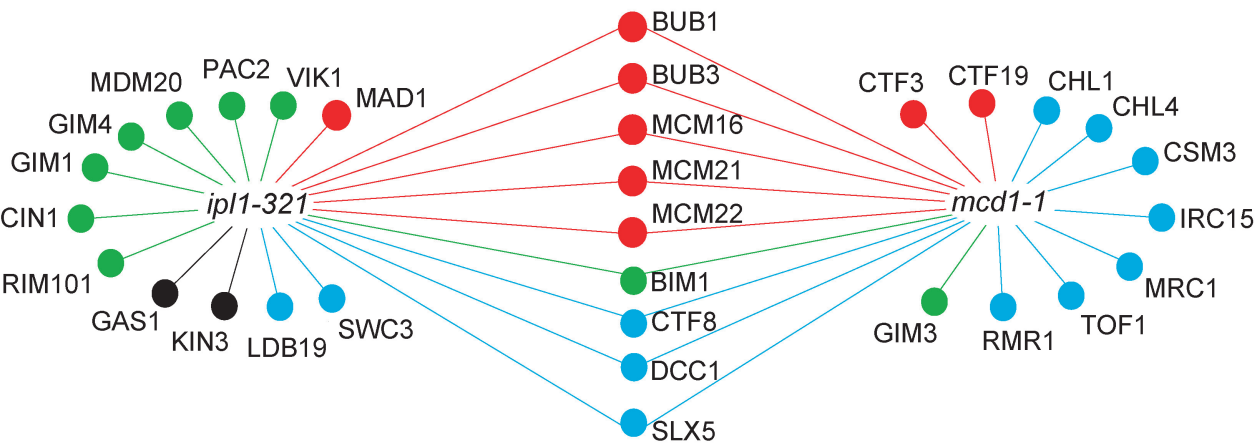












● spindle checkpoint/  
kinetochore

● genomic integrity

● cytoskeleton

● other



