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Supplemental Material to:

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Saccharomyces cerevisiae Dma proteins participate in cytokinesis by controlling two different pathways

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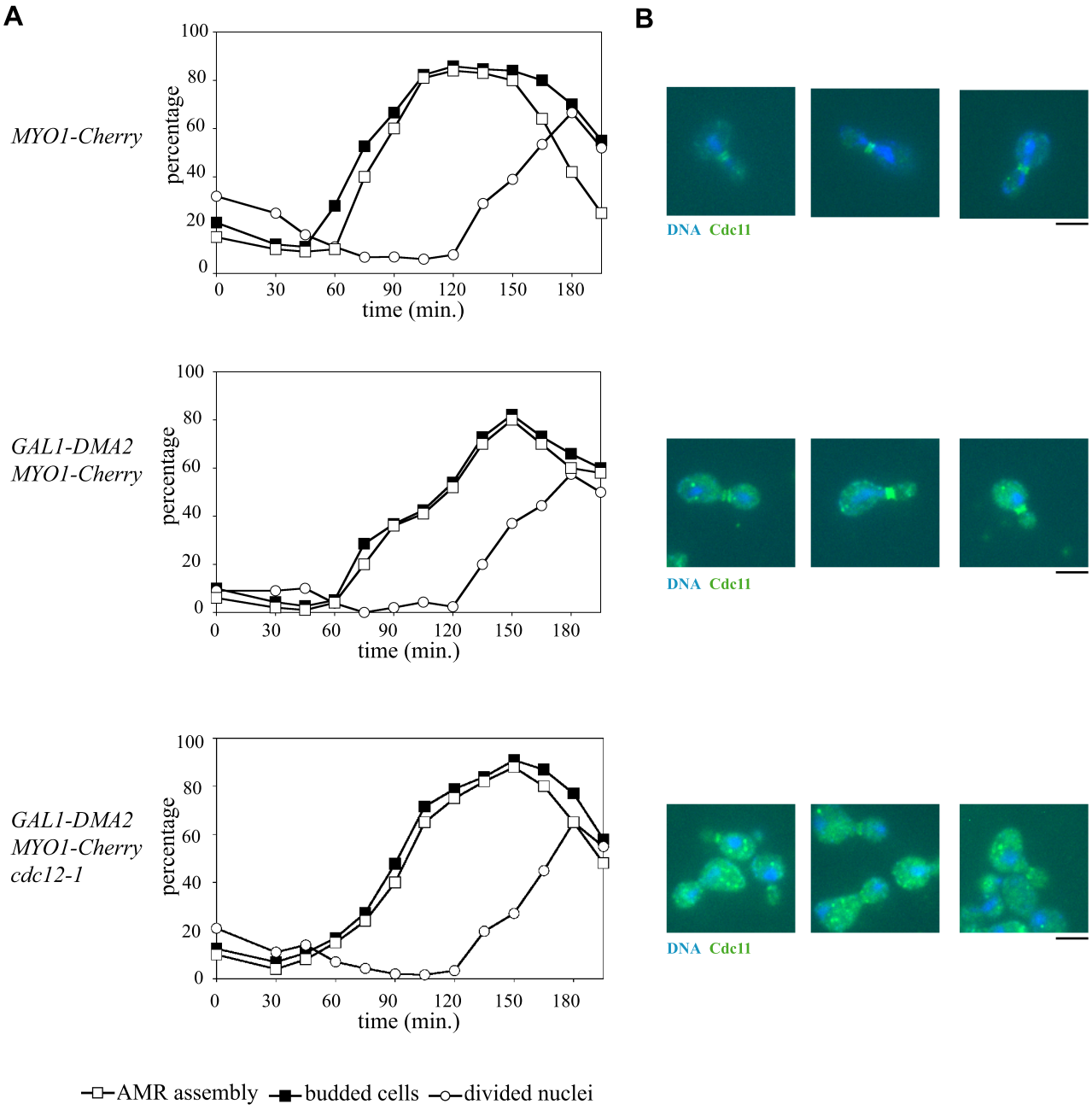
A
B
MYO1-Cherry
GAL1-DMA2
MYO1-Cherry
GAL1-DMA2
MYO1-Cherry
cdc12-1
percentage
time (min.)
0
20
40
60
80
100
0
30
60
90
120
150
180
DNA Cdc11
AMR assembly
budded cells
divided nuclei

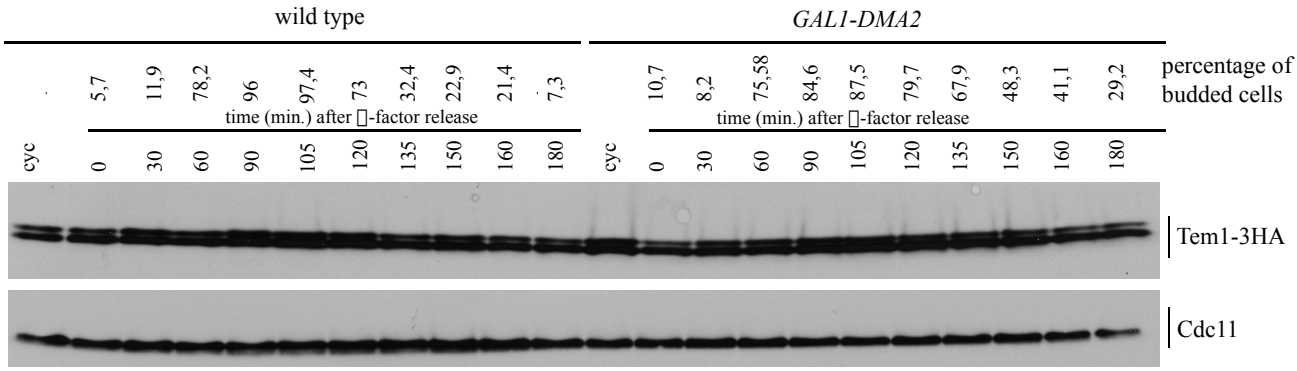
wild type
GAL1-DMA2
percentage of budded cells
5,7
11,9
78,2
96
97,4
73
32,4
22,9
21,4
7,3
10,7
8,2
75,58
84,6
87,5
79,7
67,9
48,3
41,1
29,2
time (min.) after α-factor release
time (min.) after α-factor release
cyc
0
30
60
90
105
120
135
150
160
180
cyc
0
30
60
90
105
120
135
150
160
180
Tem1-3HA
Cdc11

Table S1. Yeast strains used in this study

Name	Relevant genotype
yRF41	*MATa, dma2::LEU2, dma1::TRP1*
yRF99	*MATa, ura3::URA3::GAL1-DMA2 (single integration)*
yRF214	*MATa, TEM1::HA3::KlURA3*
yRF490	*MATa, hof1::KanMX*
yRF663	*MATa, hof1::KanMX, ura3::URA3::GAL1-DMA2 (single integration)*
yRF700	*MATa, [YEp13]*
yRF701	*MATa, hof1::KanMX, ura3::URA3::GAL1-DMA2 (single integration), [YEp13]*
yRF729	*MATa, ade2-1, trp1-1, can1-100, leu2-3,112, his3-11,15, ura3, GAL, psi+, hof1::KanMX, dma2::LEU2, dma1::TRP1*
yRF850	*MATa, iqg1::IQG1-GFP::LEU2*
yRF851	*MATa, CYK3-GFP::URA3*
yRF903	*MATa, CYK3-3HA::SpHIS5*
yRF938	*MATa, dma2::LEU2, dma1::TRP1, CYK3-3HA::SpHIS5*
yRF942	*MATa, hof1::KanMX, ura3::URA3::GAL1-DMA2 (single integration), [YEp-CHS2]*
yRF966	*MATa, hof1::KanMX, ura3::URA3::GAL1-DMA2 (single integration), [YEp-BNI5]*
yRF979	*MATa, hof1::KanMX, ura3::URA3::GAL1-DMA2 (single integration), [YEp-CYK3]*
yRF997	*MATa, hof1::KanMX, ura3::URA3::GAL1-DMA2 (single integration), [YEplac181]*
yRF999	*MATa, hof1::KanMX, ura3::URA3::GAL1-DMA2 (single integration), [YEplac112]*
yRF1000	*MATa, [YEplac112]*
yRF1002	*MATa, [YEplac181]*
yRF1083	*MATa, ura3::URA3::GAL1-DMA2 (single integration), iqg1::IQG1-GFP:LEU2*
yRF1085	*MATa, ura3::URA3::GAL1-DMA2 (single integration), CYK3-3HA::SpHIS5*
yRF1103	*MATa, ura3::URA3::GAL1-DMA2 (single integration), CYK3-GFP::URA3*
yRF1138	*MATa, [YEp96 CUP1-6HIS-UBI4]*
yRF1246	*MATa, dma2::HPHMx, dma1::LEU2kl, CYK3-8x GFP::URA3*
yRF1156	*MATa, iqg1-1, ura3::URA3::GAL-DMA2 (single integration)*
yRF1157	*MATa, hof1::KanMX, ura3::URA3::GAL1-DMA2 (single integration), bub2::HIS3*
yRF1234	*MATa, hof1::KanMX, ura3::URA3::GAL1-DMA2 (single integration), [pRS315 Dbf2-1c]*
yRF1268	*MATa, INN1-GFP-klTRP1*
yRF1271	*MATa, MYO1-CHERRY-hphNT1*
yRF1285	*MATa, ura3::URA3::GAL1-DMA2 (single integration), MYO1-CHERRY-hphNT1*
yRF1286	*MATa, ura3::URA3::GAL1-DMA2 (single integration), INN1-GFP-klTRP1*

yRF1308	*MATa, TEM1::HA3::KlURA3, [YEp96 CUP1 6HIS-UBI4]*
yRF1310	*MATa, hof1::KAN, ura3::URA3::GAL1-DMA2 (single integration), INN1-GFP-klTRP1*
yRF1355	*MATa, [YEp96 CUP1-UBI4]*
yRF1359	*MATa, TEM1::HA3::KlURA3, [YEp96 CUP1-UBI4]*
yRF1400	*MATa, ura3::URA3::GAL1-DMA2 (single integration), TEM1::HA3::KlURA3*
yRF1412	*MATa, IQG1-3HA::klTRP1, cdh1::LEU2*
yRF1414	*MATa, ura3::URA3::GAL1-DMA2 (single integration), TEM1::HA3::KlURA3,[YEp96 CUP1-6HIS-UBI4]*
yRF1415	*MATa, ura3::URA3::GAL1-DMA2 (single integration), TEM1::HA3::KlURA3, [YEp96 CUP1-UBI4]*
yRF1457	*MATa, his3, ura3, trp1, 6lexAOP-LEU2, [pSH18-34], [pEG202-p53], [pJG4-5-SV40]*
yRF1465	*MATa, TEM1::HA3::KlURA3, MYO1-CHERRY-hphNT1, [YEp96 CUP1-6HIS-UBI4]*
yRF1541	*MATa, ura3::URA3::GAL1-DMA2 (single integration), MYO1-CHERRY-hphNT1, cdc12-1*
yRF1563	*MATa, his3::TUB1-GFP::HIS3, MYO1-CHERRY-hphNT1*
yRF1564	*MATa, ura3::URA3::GAL1-DMA2 (single integration), MYO1-CHERRY-hphNT1, his3::TUB1-GFP::HIS3*
yRF1584	*MATa , dma1::TRP1, dma2::HPHMx, MYO1-CHERRY-hphNT1*
yRF1585	*MATa, dma1::TRP1, dma2::HPHMx, MYO1-CHERRY-hphNT1, his3::TUB1-GFP::HIS3*
yRF1588	*MATa, cyk3::natNT2*
yRF1589	*MATa, dma2::LEU2, dma1::TRP1, cyk3::natNT2*
yRF1595	*MATa, ade2-1, trp1-1, can1-100, leu2-3,112, his3-11,15, ura3, GAL, psi+ , dma2::LEU2, dma1::TRP1, chs2::natNT2*
yRF1650	*MATa, his3, ura3, trp1, 6lexAOP-LEU2, [pSH18-34], [pEG202-TEM1], [pJG4-5-IQG1]*
yRF1651	*MATa, his3, ura3, trp1, 6lexAOP-LEU2, [pSH18-34], [pEG202], [pJG4-5-IQG1*
yRF1652	*MATa, his3, ura3, trp1, 6lexAOP-LEU2, [pSH18-34], [pEG202-TEM1], [pJG4-5]*
yRF1663	*MATa, hof1::KanMX, [p425GAL1- DMA1-2HA]*
yRF1667	*MATa, his3, ura3, trp1, 6lexAOP-LEU2, [pSH18-34], [pEG202-TEM1], [pJG4-5-IQG1], [p425GAL1- DMA2-2HA]*
yRF1677	*MATa, dma2::HPHMx, dma1::LEU2kl, iqg1-1*
yRF1749	*MATa, cyk3:: natNT2, ura3::URA3::GAL1-DMA2 (single integration)*
yRF1750	*MATa, chs2:: natNT2, ura3::URA3::GAL1-DMA2 (single integration)*
yRF1795	*MATa, ura3::URA3::GAL1-DMA2 (single integration), MYO1-CHERRY-hphNT1, [pRS315 CHS2-GFP]*
yRF1796	*MATa, MYO1-CHERRY-hphNT1, [pRS315 CHS2-GFP]*

Suppl. figure 1: Septin ring destabilization does not rescue the AMR dynamic defects due to Dma2 excess. A-B: Exponentially growing cultures of *MYO1-Cherry*, *GAL1-DMA2 MYO1-Cherry* and *GAL1-DMA2 cdc12-1 MYO1-Cherry* cells were arrested in G1 by α-factor and released from G1 arrest in YEPRG at 23°C (time 0). At the indicated times after release, cell samples were taken for scoring budding, nuclear division and AMR disassembly (A). Pictures were taken 120 minutes after release to show in situ immunofluorescence analysis of ring deposition (Cdc11) and nuclei (DNA). bar: 5μm.

Suppl. figure 2: Tem1 total levels are not affected by Dma2 overproduction. Exponentially growing YEPR cultures of *TEM1-3HA* and *TEM1-3HA GAL1-DMA2* cells were arrested in G1 by α-factor and released from G1 arrest in fresh YEPRG medium at 25°C (time = 0). At the indicated times, cell samples were taken for scoring budding and for determining Tem1 levels by western blot analysis with anti-HA and anti-Cdc11 (loading control) antibodies.

Video 1 Time lapse analysis of Myo1-Cherry dynamics with respect to mitotic spindle in wild type cells (related to Figure 3A). 5 min interval.
Video 2 Time lapse analysis of Myo1-Cherry dynamics with respect to mitotic spindle in *GAL1-DMA2* cells (related to Figure 3A). 5 min interval.
Video 3 Time lapse analysis of Chs2 localization at the bud neck with respect to actomyosin ring constriction in wild type cells (related to Figure 4B). 2 min interval.
Video 4 Time lapse analysis of Chs2 localization at the bud neck with respect to actomyosin ring constriction in *GAL1-DMA2* cells (related to Figure 4B). 2 min interval.
Video 5 Time lapse analysis of Myo1-Cherry dynamics with respect to mitotic spindle in wild type cells (related to Figure 6A). 3 min interval.
Video 6 Time lapse analysis of Myo1-Cherry dynamics with respect to mitotic spindle in *dma1Δ dma2Δ* cells (related to Figure 6A). 3 min interval.