APPENDIX

The Spo13/Meikin Pathway Confines the Onset of Gamete Differentiation to Meiosis II in Yeast

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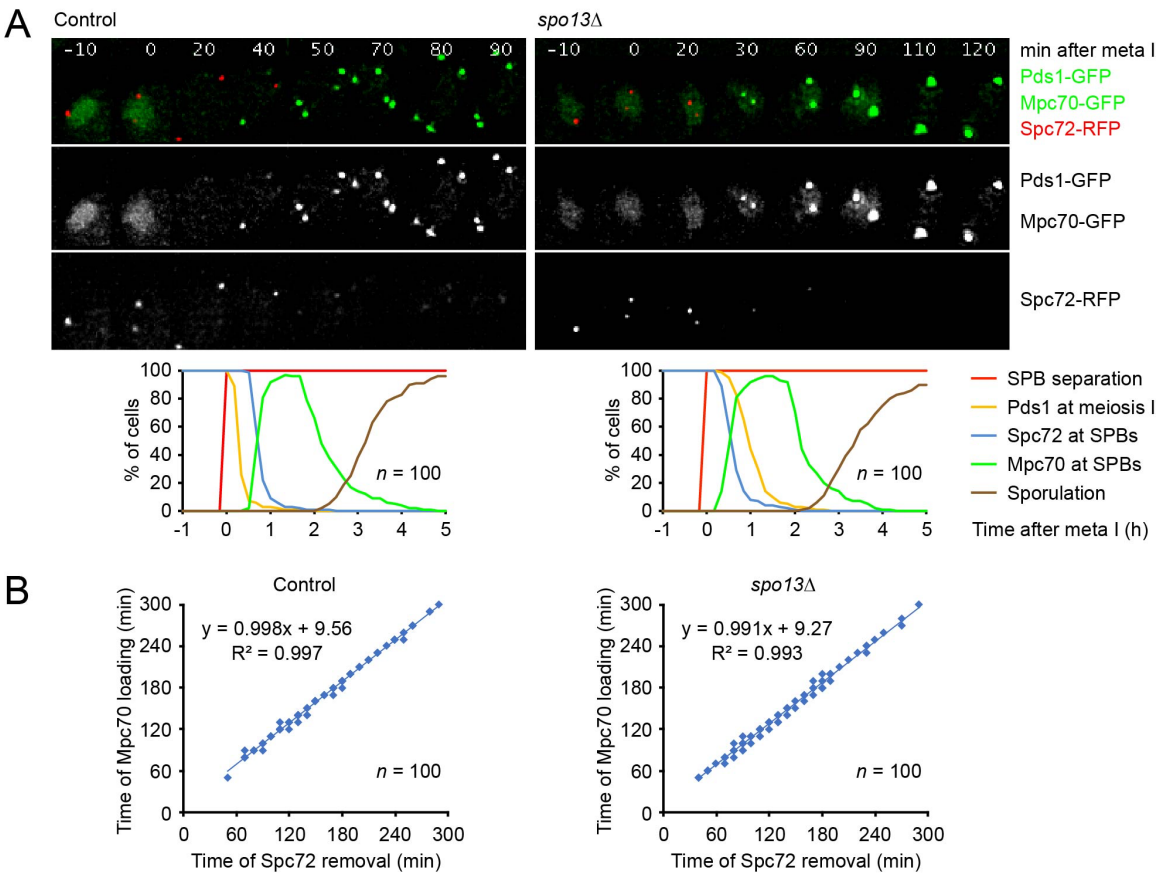
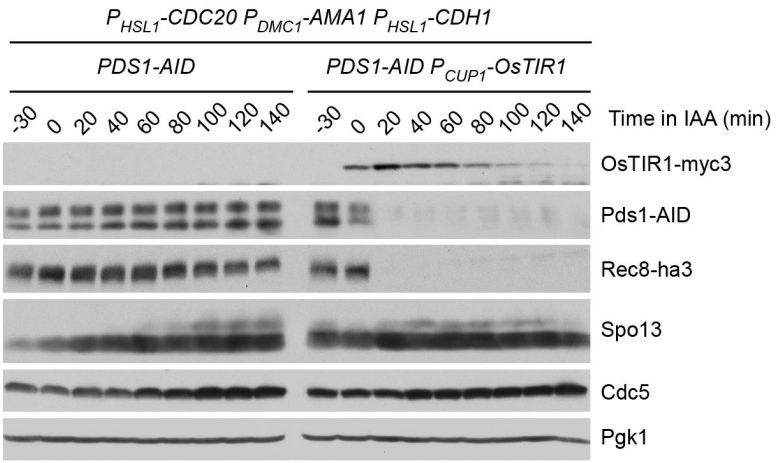


Appendix Figure S1. Exchange of Spc72 for Mpc70 at SPBs in control and *spo13Δ* cells

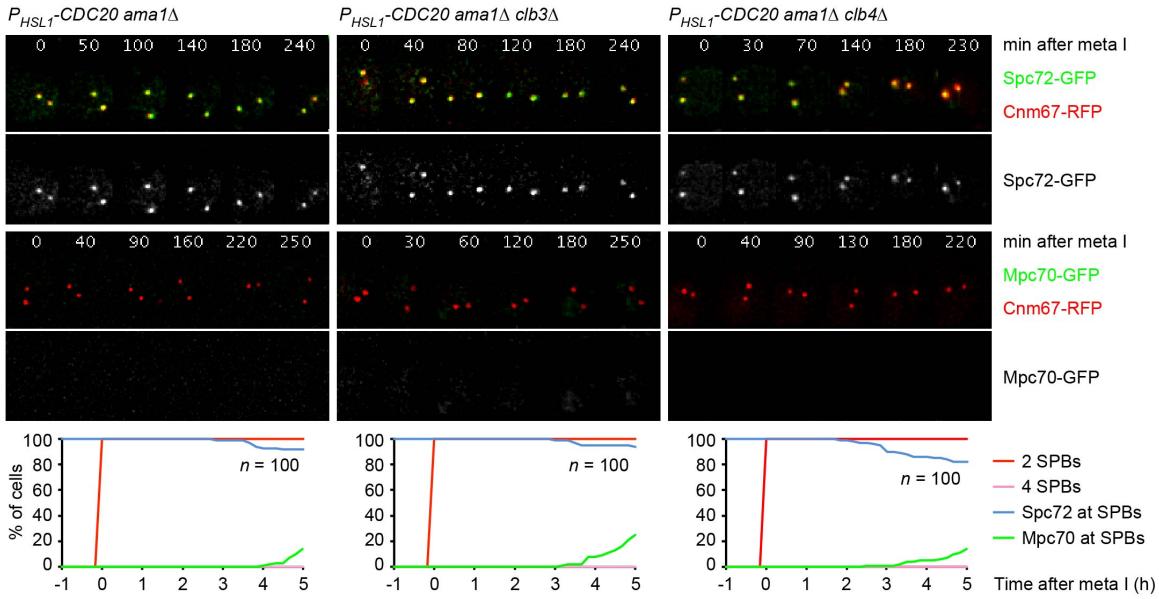
A. Imaging of Pds1-GFP, Mpc70-GFP, and Spc72-RFP in control and *spo13Δ* cells. Top, time-lapse series. Bottom, meiotic events were quantified in cells synchronized in silico to SPB separation at entry into metaphase I ($t = 0$). In *spo13Δ* cells, Pds1 degradation is delayed by 42 min (95% CI, 36-48; $P < 0.0001$), whereas Spc72 removal and Mpc70 loading are advanced by 8.7 min (95% CI, 3.6-13.8; $P = 0.0009$) and 9.8 min (95% CI, 4.7-14.9; $P = 0.0002$), respectively. Group means were compared using Welch's *t*-test.

B. Mpc70 loading correlates with Spc72 removal in control and *spo13Δ* cells from (A). Times were measured from the start of imaging at 3.5 h in SPM. The regression lines show similar slopes ($P = 0.53$) and comparable y-intercepts ($P = 0.045$; ANCOVA).



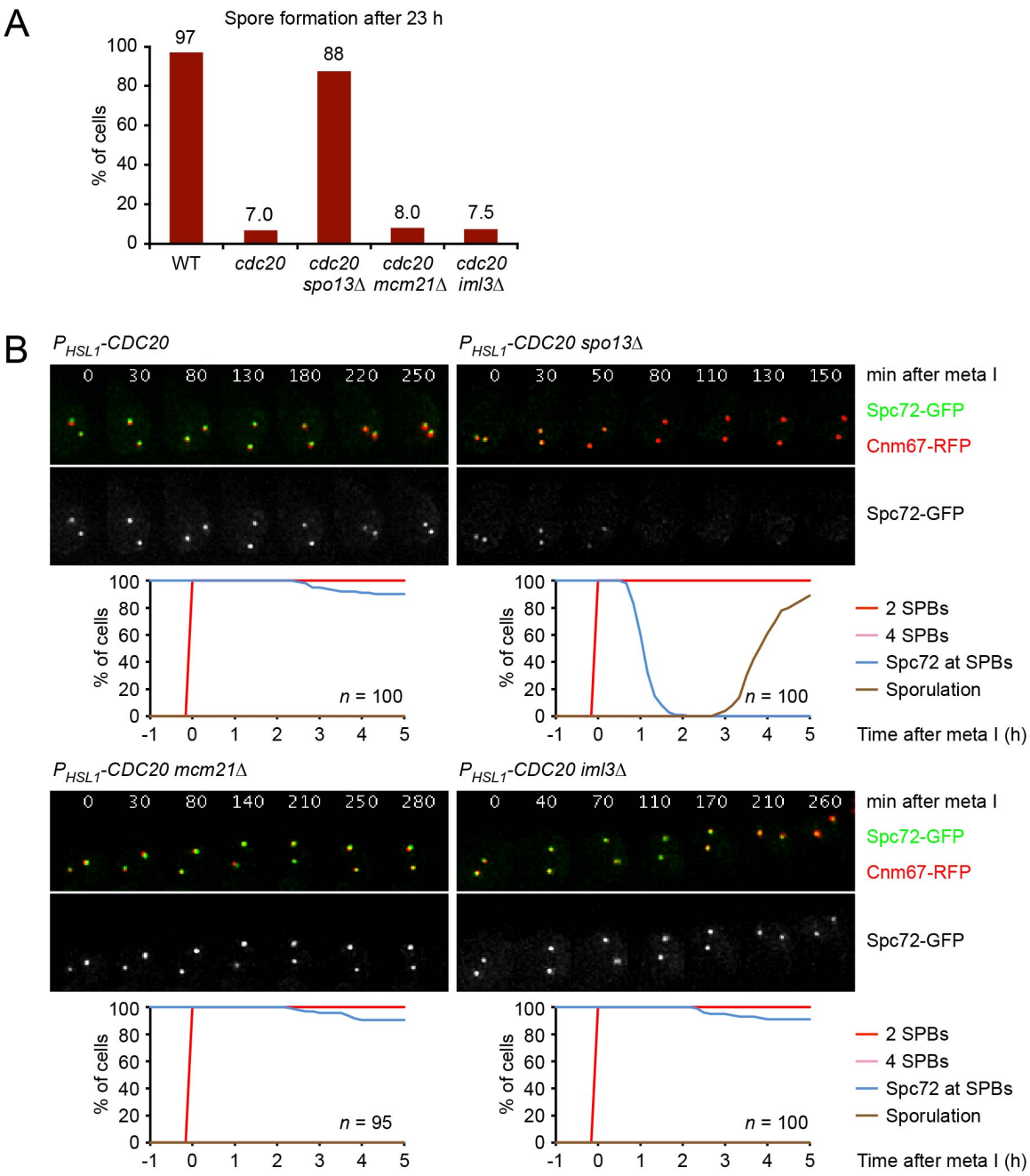
Appendix Figure S2. Spo13 is not cleaved upon activation of separase

Activation of separase in metaphase I-arrested cells by auxin-inducible degradation of Pds1 causes cleavage of Rec8 but does not affect Spo13. *P_{HSL1}-CDC20 P_{DMC1}-AMA1 P_{HSL1}-CDH1* cells containing *PDS1-AID* or *PDS1-AID* plus *P_{CUP1}-OsTIR1-myc3* were arrested at metaphase I by depletion of APC/C activators. At 8 h in SPM, cells were treated with $CuSO_4$ to induce expression of the plant F-box protein OsTIR1. At 8.5 h in SPM (t = 0), the auxin IAA was added to activate separase through the degradation of Pds1-AID. Proteins were detected in whole-cell extracts by immunoblotting.



Appendix Figure S3. Analysis of *P*$_{HSL1}$*-CDC20 ama1Δ* cells lacking Clb3 or Clb4

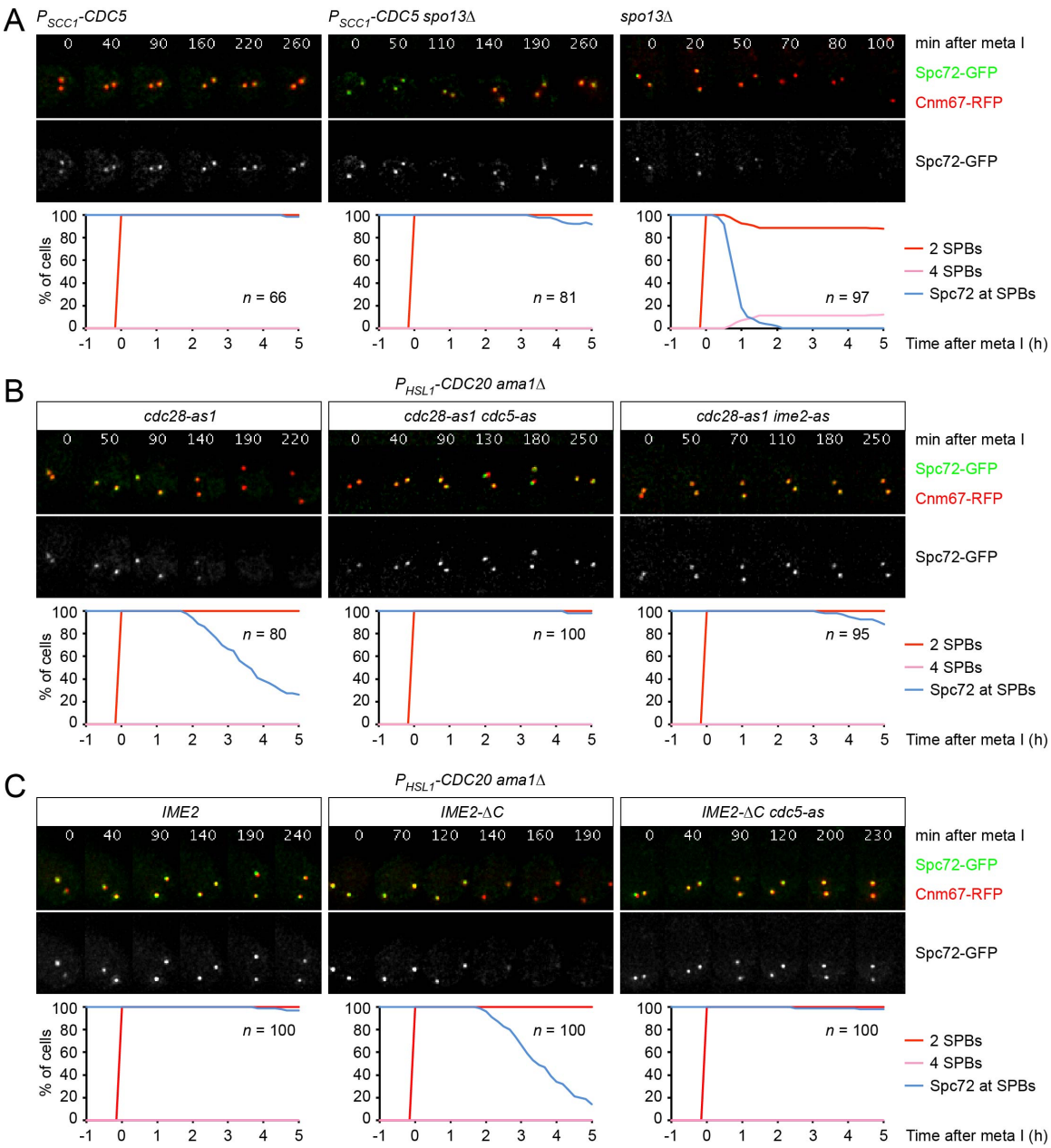
Imaging of SPBs (Cnm67-RFP) and Spc72-GFP or Mpc70-GFP in *P*$_{HSL1}$*-CDC20 ama1Δ* control cells and cells lacking Clb3 or Clb4. Top, time-lapse series. Bottom, meiotic events were quantified in cells synchronized in silico to SPB separation at entry into metaphase I (t = 0). Graphs show overlays of *SPC72-GFP* and *MPC70-GFP* strains.



Appendix Figure S4. Control of spore formation by Spo13 does not require intact kinetochores

A. Quantification of spore formation in the wild-type (WT) and the indicated *P_{HSL1}-CDC20* strains at 23 h in SPM. Data are mean values of two cultures (n = 200 cells per culture).

B. Imaging of SPBs (Cnm67-RFP) and Spc72-GFP in *P_{HSL1}-CDC20* control cells and cells lacking Spo13, Mcm21, or Iml3. Top, time-lapse series. Bottom, Spc72's presence at SPBs was quantified in cells synchronized in silico to SPB separation at entry into metaphase I (t = 0). While *P_{HSL1}-CDC20 mcm21Δ* and *P_{HSL1}-CDC20 iml3Δ* cells are defective in kinetochore assembly, they retain Spc72 at SPBs. Data are representative of two independent experiments.



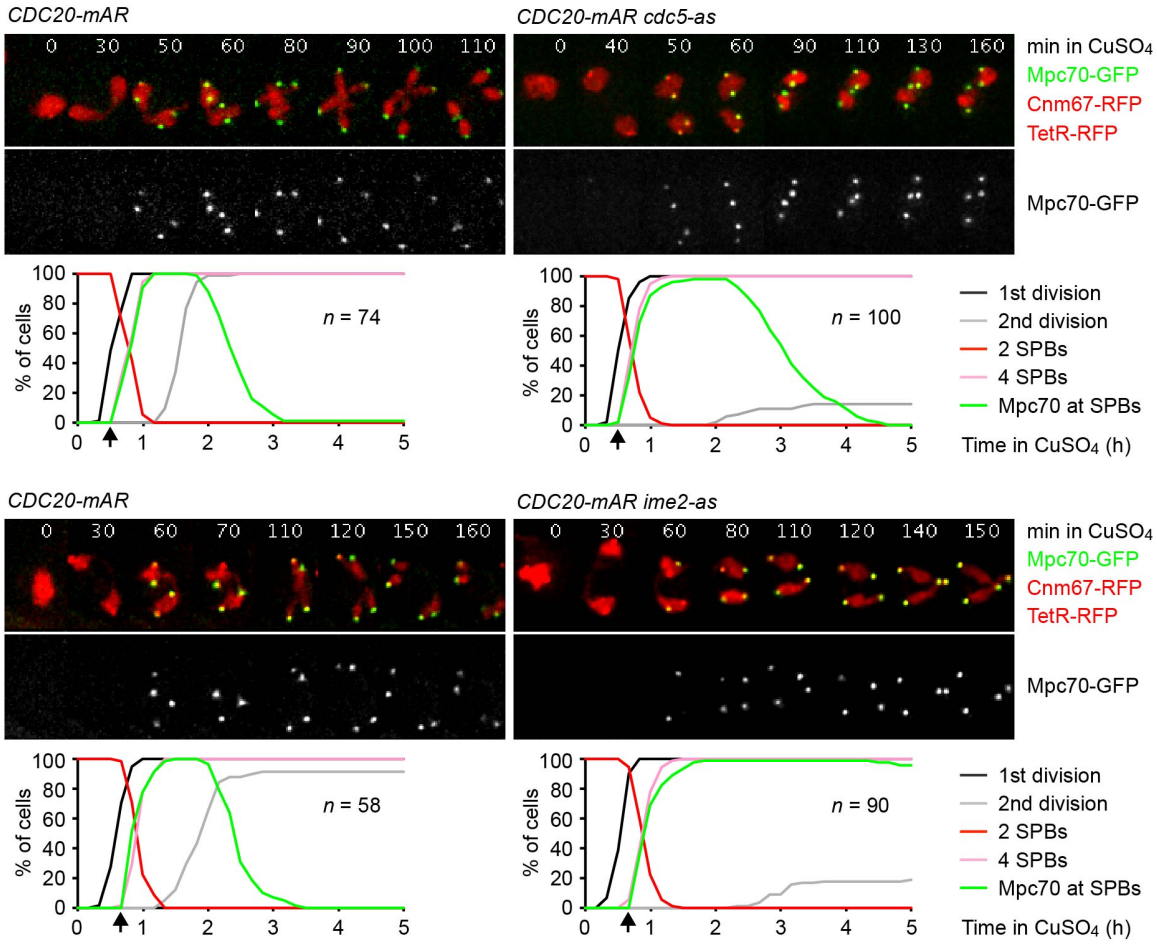
Appendix Figure S5. Spc72 removal requires the activities of Cdc5 and Ime2

A. Imaging of SPBs (Cnm67-RFP) and Spc72-GFP in cells lacking Cdc5 (P_{SCC1}-*CDC5*) and/or Spo13. Top, time-lapse series. Bottom, Spc72's presence at SPBs was quantified in cells synchronized in silico to SPB separation at entry into metaphase I ($t = 0$).

B. Imaging of SPBs (Cnm67-RFP) and Spc72-GFP in P_{HSL1}-*CDC20 ama1Δ cdc28-as1* control cells and cells containing *cdc5-as* or *ime2-as*. At metaphase I (8 h in SPM), cells were treated with 1NM-PP1 (to inhibit Cdc28-as1) plus either CMK (to inhibit Cdc5-as) or 1Na-PP1 (to inhibit Ime2-as). Spc72's presence at SPBs was analyzed as in (A).

C. Imaging of SPBs (Cnm67-RFP) and Spc72-GFP in P_{HSL1}-*CDC20 ama1Δ* cells containing wild-type *IME2*, *IME2-ΔC*, or *IME2-ΔC* plus *cdc5-as*. Cdc5-as was inhibited with CMK at 6.5 h in SPM. Spc72's presence at SPBs was analyzed as in (A).

Data information: data in (B) and (C) are representative of two independent experiments.



Appendix Figure S6. MP assembly does not require Cdc5 or Ime2 activity at metaphase II

CDC20-mAR control cells and cells containing *cdc5-as* (top) or *ime2-as* (bottom) were released from the metaphase I-arrest with $CuSO_4$ at 7 h in SPM (t = 0). At anaphase I (arrows), cells were treated with CMK (to inhibit Cdc5-as) or 1Na-PP1 (to inhibit Ime2-as). Panels, time-lapse series from the imaging of Mpc70-GFP, SPBs (Cnm67-RFP), and nuclei (TetR-RFP). Graphs, quantification of meiotic events.

Appendix Table S1. *Saccharomyces cerevisiae* SK1 Strains Used in this Study

Figure	Strain[1]	Genotype[2]
1A	Z17009	*PDS1myc18::KlTRP1*
1A	Z32767	*PDS1myc18::KlTRP1 cdc20::P_{HSL1}-CDC20-HphMX4*
1A	Z32771	*PDS1myc18::KlTRP1 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::HIS3MX6*
1A	Z38457	*PDS1myc18::KlTRP1 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::HIS3MX6::spo13-m2::HphMX4*
1A	Z32768	*PDS1myc18::KlTRP1 cdc20::P_{HSL1}-CDC20-HphMX4 clb1Δ::BleMX4*
1A	Z35062	*cdc20::P_{HSL1}-CDC20-HphMX4 clb3Δ::TRP1*
1A	Z32769	*PDS1myc18::KlTRP1 cdc20::P_{SCC1}-CDC20-KanMX4 clb4Δ::KanMX4*
1B	Z35411	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2*
1B	Z35412	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2 cdc20::P_{HSL1}-CDC20-HphMX4*
1B	Z35414	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::HIS3MX6*
1B	Z35415	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2 cdc20::P_{HSL1}-CDC20-HphMX4 clb1Δ::KanMX4*
1C	Z33199	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4*
1C	Z32054	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4*
1C	Z21998	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4*
1C	Z35190	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4*
1D	Z35268	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::BleMX4*
1D	Z35266	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::HIS3MX6::spo13-m2::HphMX4*
1D	Z35269	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 clb1Δ::BleMX4*
1D	Z35455	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::BleMX4*
1D	Z35189	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::HIS3MX6::spo13-m2::HphMX4*
1D	Z35456	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 clb1Δ::BleMX4*
2A	Z32389	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3*
2A	Z35509	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spo13Δ::HIS3MX6*
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2A	Z35230	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spo13Δ::BleMX4*
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2B	Z36062	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::NatMX4 cdh1::P_{HSL1}-CDH1-BleMX4*
2B	Z36065	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 cdh1::P_{HSL1}-CDH1-HIS3MX6 spo13Δ::BleMX4*

2B	Z23218	*cdc20::P_{CLB2}-CDC20-HphMX4 ura3::P_{CUP1}-CDC20-URA3 hrr25Δ::KanMX4::HRR25-HIS3*
2B	Z29809	*cdc20::P_{CLB2}-CDC20-HphMX4 ura3::P_{CUP1}-CDC20-URA3 mpc54Δ::AurCMX4 mpc70Δ::BleMX4*
2C	Z32389	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3*
2C	Z32513	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 clb1Δ::BleMX4*
2C	Z32388	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 cdc28-as1*
2C	Z32908	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3*
2C	Z32907	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 clb1Δ::BleMX4*
2C	Z32545	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 cdc28-as1*
3A	Z36338	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spc72Δ::KanMX4 leu2::SPC72-LEU2*
3A	Z36513	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spc72Δ::KanMX4 leu2::spc72-7-LEU2*
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3B	Z36546	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spc72Δ::KanMX4 leu2::SPC72-LEU2*
3B	Z36545	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spc72Δ::KanMX4 leu2::spc72-7-LEU2*
3C	Z35213	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spo13Δ::BleMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3*
3C	Z35397	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spo13Δ::BleMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2::P_{GAL1}-SPC72-LEU2*
4A	Z33847	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 spo13Δ::HIS3MX6::spo13-mD::HphMX4*
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5B	Z38261	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 CDC5-ha3::URA3 spo13::Ha3-SPO13-NatMX4*
5C	Z12185	Wild-type
5C	Z17454	*CDC5-ha3::URA3*
5C	Z6619	*CDC5-ha3::URA3 spo13Δ::HIS3MX6*
5D	Z37002	*cdc20::P_{HSL1}-CDC20-HphMX4 cdc5::P_{SCC1}-CDC5-KanMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 ura3::P_{GAL1}-PBD-ha3-URA3*
5D	Z37001	*cdc20::P_{HSL1}-CDC20-HphMX4 cdc5::P_{SCC1}-CDC5-KanMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 ura3::P_{GAL1}-PBD-FAA-ha3-URA3*
5E	Z35197	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3*
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6A,C	Z35833	*ndt80Δ::NatMX4 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3*
6A,C	Z38067	*ndt80Δ::NatMX4 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2*
6A,C	Z38066	*ndt80Δ::NatMX4 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 ura3::P_{GAL1}-CDC5-ha3-URA3*
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6B,D	Z38441	*ndt80Δ::NatMX4 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2 ura3::P_{GAL1}-CDC5-ha3-URA3 spo13Δ::HIS3MX6::spo13-10A::HphMX4*
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7A	Z36179	*cdc20-3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3*
7A	Z36076	*cdc20-3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2*
7A	Z36358	*cdc20-3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2 ura3::P_{GAL1}-SPO13-URA3*
7B	Z36140	*cdc20-3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 ura3-P_{GAL1}-CDC5-ha3-URA3*
7B	Z36418	*cdc20-3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 trp1-P_{GAL1}-CDC5-TRP1 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2*
7B	Z36419	*cdc20-3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 trp1-P_{GAL1}-CDC5-TRP1 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2 ura3::P_{GAL1}-SPO13-URA3*
7C	Z38279	*cdc20-3 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2 ura3::P_{GAL1}-CDC5-ha3-URA3*
7C	Z38417	*cdc20-3 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 trp1::P_{GAL1}-NDT80-TRP1*
7C	Z38280	*cdc20-3 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 trp1::P_{GAL1}-NDT80-TRP1 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2 ura3::P_{GAL1}-CDC5-ha3-URA3*
8A	Z32534	*cdc28-as2 MPC70/MPC70-eGFP::KanMX4 NDT80/NDT80-sfGFP::KanMX4 ura3::P_{HIS3}-mScarlet-I-TUB1-URA3 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::NatMX4*
8B	Z35798	*cdc28-as2 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::NatMX4*
8C	Z32534	*cdc28-as2 MPC70/MPC70-eGFP::KanMX4 NDT80/NDT80-sfGFP::KanMX4 ura3::P_{HIS3}-mScarlet-I-TUB1-URA3 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::NatMX4*
8C	Z38193	*cdc28-as2 MPC70/MPC70-eGFP::KanMX4 NDT80/NDT80-sfGFP::KanMX4 ura3::P_{HIS3}-mScarlet-I-TUB1-URA3 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::NatMX4 spo13Δ::BleMX4*
8C	Z35946	*cdc28-as2 MPC70/MPC70-eGFP::KanMX4 NDT80/NDT80-sfGFP::KanMX4 ura3::P_{HIS3}-mScarlet-I-TUB1-URA3 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::NatMX4 cdc5-as::HphMX4*
8D	Z36338	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spc72Δ::KanMX4 leu2::SPC72-LEU2*
8D	Z36209	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spc72Δ::KanMX4 leu2::spc72-7-LEU2*
8D	Z38848	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 spc72Δ::KanMX4 leu2::spc72-7-LEU2 cdc5-as::HphMX4*
9A	Z31260	*cdc20::P_{CLB2}-CDC20-HphMX4 ura3::P_{CUP1}-CDC20-URA3 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 leu2/leu2::P_{URA3}-tetR-tdTomato-LEU2*

9A	Z31259	*cdc20::P_{CLB2}-CDC20-HphMX4 ura3::P_{CUP1}-CDC20-URA3 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 leu2/leu2::P_{URA3}-tetR-tdTomato-LEU2 cdc28-as1*
9B	Z38565	*cdc20::P_{HSL1}-CDC20-HphMX4 trp1::P_{CUP1}-cdc20-3-TRP1 ama1Δ::NatMX4 leu2::P_{DMC1}-cAMA1-LEU2 cdh1::P_{HSL1}-CDH1-BleMX4 MPC70/MPC70-eGFP::KanMX4 [pRS426-P_{TEF2}-mRFP-spo20$^{51-91}$]*
9C	Z38604	*cdc20::P_{HSL1}-CDC20-HphMX4 trp1::P_{CUP1}-cdc20-3-TRP1 ama1Δ::NatMX4 leu2::P_{DMC1}-cAMA1-LEU2 cdh1::P_{HSL1}-CDH1-BleMX4 MPC70/MPC70-eGFP::KanMX4 his3/his3::P_{HIS3}-eGFP-TUB1-HIS3 [pRS426-P_{TEF2}-mRFP-spo20$^{51-91}$]*
9C	Z38606	*cdc20::P_{HSL1}-CDC20-HphMX4 trp1::P_{CUP1}-cdc20-3-TRP1 ama1Δ::NatMX4 leu2::P_{DMC1}-cAMA1-LEU2 cdh1::P_{HSL1}-CDH1-BleMX4 MPC70/MPC70-eGFP::KanMX4 his3/his3::P_{HIS3}-eGFP-TUB1-HIS3 cdc28-as2 [pRS426-P_{TEF2}-mRFP-spo20$^{51-91}$]*
EV1A	Z35411	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2*
EV1A	Z35414	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::HIS3MX6*
EV1A	Z35415	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2 cdc20::P_{HSL1}-CDC20-HphMX4 clb1Δ::KanMX4*
EV1A	Z39534	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2 ama1Δ::NatMX4*
EV1A	Z39537	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2 ama1Δ::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::HIS3MX6*
EV1A	Z39536	*his3::P_{HIS3}-eGFP-TUB1::HIS3 leu2::P_{URA3}-tetR-tdTomato::LEU2 ama1Δ::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 clb1Δ::KanMX4*
EV1B	Z31931	*cdc20::P_{SCC1}-CDC20-HphMX4 ama1Δ::CaURA3 cdh1::P_{HSL1}-CDH1-HphMX4 SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4*
EV1B	Z31930	*cdc20::P_{SCC1}-CDC20-HphMX4 ama1Δ::CaURA3 cdh1::P_{HSL1}-CDH1-HphMX4 SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 spo13Δ::BleMX4*
EV1B	Z32205	*cdc20::P_{SCC1}-CDC20-HphMX4 ama1Δ::CaURA3 cdh1::P_{HSL1}-CDH1-BleMX4 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4*
EV1B	Z32206	*cdc20::P_{SCC1}-CDC20-HphMX4 ama1Δ::CaURA3 cdh1::P_{HSL1}-CDH1-BleMX4 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 spo13Δ::HIS3MX6*
EV1C	Z26184	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4*
EV1C	Z31931	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{SCC1}-CDC20-HphMX4 ama1Δ::CaURA3 cdh1::P_{HSL1}-CDH1-HphMX4*
EV1C	Z31930	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{SCC1}-CDC20-HphMX4 ama1Δ::CaURA3 cdh1::P_{HSL1}-CDH1-HphMX4 spo13Δ::BleMX4*
EV2	Z33848	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3*
EV2	Z33847	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 spo13Δ::HIS3MX6::spo13-mD::HphMX4*
EV2	Z33845	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2/leu2::P_{GAL1}-clb1-mDK-LEU2*
EV2	Z33846	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2/leu2::P_{GAL1}-clb1-mDK-LEU2 spo13Δ::HIS3MX6::spo13-mD::HphMX4*
EV3A	Z35835	*ndt80Δ::NatMX4 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2 ura3::P_{GAL1}-CDC5-ha3-URA3*
EV3A	Z35491	*ndt80Δ::NatMX4 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2 ura3::P_{GAL1}-CDC5-ha3-URA3 spo13Δ::BleMX4*

EV3B	Z38331	*cdc20::P_{HSL1}-CDC20-URA3 ama1Δ::NatMX4*
EV3B	Z38332	*cdc20::P_{HSL1}-CDC20-URA3 ama1Δ::NatMX4 cdc28-as2*
EV3B	Z38333	*cdc20::P_{HSL1}-CDC20-URA3 ama1Δ::NatMX4 spo13Δ::HIS3MX6::spo13-10A::HphMX4*
EV3B	Z38335	*cdc20::P_{HSL1}-CDC20-URA3 ama1Δ::NatMX4 spo13Δ::HIS3MX6::spo13-10A::HphMX4 cdc28-as2*
EV3B	Z38334	*cdc20::P_{HSL1}-CDC20-URA3 ama1Δ::NatMX4 spo13Δ::HIS3MX6::spo13-10D::HphMX4*
EV3B	Z38336	*cdc20::P_{HSL1}-CDC20-URA3 ama1Δ::NatMX4 spo13Δ::HIS3MX6::spo13-10D::HphMX4 cdc28-as2*
EV3C	Z33437	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4*
EV3C	Z38465	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 spo13Δ::HIS3MX6::spo13-10A::HphMX4*
EV3C	Z38763	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 spo13Δ::HIS3MX6::spo13-10D::HphMX4*
EV4A	Z36471	*cdc20-3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 cdc5-as::HphMX4*
EV4A	Z36472	*cdc20-3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 cdc5-as::HphMX4 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2*
EV4A	Z36076	*cdc20-3 SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 his3::P_{GPD}-GAL4^{484}-ER-HIS3 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2*
EV4B	Z38637	*cdc20-3 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3/his3::P_{GPD}-GAL4^{484}-ER-HIS3 trp1::P_{GAL1}-NDT80-TRP1 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2 ura3::P_{GAL1}-CDC5-ha3-URA3*
EV4B	Z38638	*cdc20-3 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 his3/his3::P_{GPD}-GAL4^{484}-ER-HIS3 trp1::P_{GAL1}-NDT80-TRP1 leu2::P_{GAL1}-IME2-ΔC-ha3-LEU2 ura3::P_{GAL1}-CDC5-ha3-URA3 mpc54Δ::AurCMX4*
EV5A	Z31670	*SPC72-eGFP::KanMX4 ura3::P_{HIS3}-mScarlet-I-TUB1-URA3*
EV5A	Z31671	*SPC72-eGFP::KanMX4 ura3::P_{HIS3}-mScarlet-I-TUB1-URA3 cdc28-as1*
EV5A	Z31400	*MPC70/MPC70-eGFP::KanMX4 ura3::P_{HIS3}-mScarlet-I-TUB1-URA3*
EV5A	Z31401	*MPC70/MPC70-eGFP::KanMX4 ura3::P_{HIS3}-mScarlet-I-TUB1-URA3 cdc28-as1*
EV5B	Z32038	*MPC70/MPC70-eGFP::KanMX4*
EV5B	Z32450	*MPC70/MPC70-eGFP::KanMX4 cdc28-as1*
EV5B	Z32037	*MPC54/MPC54-eGFP::KanMX4*
EV5B	Z32036	*MPC54/MPC54-eGFP::KanMX4 cdc28-as1*
EV5B	Z32038	*MPC70/MPC70-eGFP::KanMX4*
EV5B	Z32450	*MPC70/MPC70-eGFP::KanMX4 cdc28-as1*
EV5B	Z32035	*SPO74/SPO74-sfGFP::KlTRP1*
EV5B	Z32040	*SPO74/SPO74-sfGFP::KlTRP1 cdc28-as1*
S1	Z32536	*MPC70/MPC70-eGFP::KanMX4 PDS1-mNeonGreen::KlTRP1 SPC72-mScarlet::KlTRP1*
S1	Z32535	*MPC70/MPC70-eGFP::KanMX4 PDS1-mNeonGreen::KlTRP1 SPC72-mScarlet::KlTRP1 spo13Δ::BleMX4*
S2	Z37615	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::NatMX4 leu2::P_{DMC1}-cAMA-LEU2 cdh1::P_{HSL1}-CDH1-BleMX4 REC8-ha3::URA3 PDS1-AID*::KanMX4*
S2	Z37616	*cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::NatMX4 leu2::P_{DMC1}-cAMA-LEU2 cdh1::P_{HSL1}-CDH1-BleMX4 REC8-ha3::URA3 PDS1-AID*::KanMX4 ura3::P_{CUP1}-OsTIR1-myc3::URA3*

S3	Z33437	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3*
S3	Z35320	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 clb3Δ::TRP1*
S3	Z33291	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 clb4Δ::KanMX4*
S3	Z32908	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3*
S3	Z38575	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 clb3Δ::TRP1*
S3	Z32988	*MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 clb4Δ::HphMX4*
S4A	Z12185	Wild-type
S4A,B	Z35267	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4*
S4A,B	Z35268	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 spo13Δ::BleMX4*
S4A,B	Z39487	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 mcm21Δ::BleMX4*
S4A,B	Z39485	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 iml3Δ::CaURA3MX4*
S5A	Z32337	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc5:P_{SCC1}-CDC5-KanMX4*
S5A	Z32336	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc5:P_{SCC1}-CDC5-KanMX4 spo13Δ::HIS3MX6*
S5A	Z32335	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 spo13Δ::HIS3MX6*
S5B	Z35198	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 cdc28-as1*
S5B	Z35370	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 cdc28-as1 cdc5-as::HphMX4*
S5B	Z35200	*SPC72-eGFP::KanMX4 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 cdc28-as1 ime2::KanMX4::ime2-as-LEU2*
S5C	Z33437	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3*
S5C	Z38204	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 IME2-ΔC-ha3-HIS3MX6*
S5C	Z39253	*SPC72-eGFP::KlTRP1 CNM67-tdTomato::NatMX4 cdc20::P_{HSL1}-CDC20-HphMX4 ama1Δ::CaURA3 IME2-ΔC-ha3-HIS3MX6 cdc5-as::HphMX4*
S6	Z24053	*cdc20::P_{CLB2}-CDC20-KanMX6 trp1::P_{CUP1}-CDC20-TRP1 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 leu2/leu2::P_{URA3}-tetR-tdTomato-LEU2*
S6	Z31053	*cdc20::P_{CLB2}-CDC20-KanMX6 trp1::P_{CUP1}-CDC20-TRP1 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 leu2/leu2::P_{URA3}-tetR-tdTomato-LEU2 cdc5-as::HphMX4*
S6	Z31260	*cdc20::P_{CLB2}-CDC20-HphMX4 ura3::P_{CUP1}-CDC20-URA3 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 leu2/leu2::P_{URA3}-tetR-tdTomato-LEU2*
S6	Z31398	*cdc20::P_{CLB2}-CDC20-HphMX4 ura3::P_{CUP1}-CDC20-URA3 MPC70/MPC70-eGFP::KanMX4 CNM67-tdTomato::NatMX4 leu2/leu2::P_{URA3}-tetR-tdTomato-LEU2 ime2::KanMX4::ime2-as-LEU2*

[1]Strains are listed for each figure from left to right and/or top to bottom. [2]All SK1 strains are diploid with the background *MATa/MATα ho::LYS2 lys2 ade2Δ::hisG trp1Δ::hisG leu2Δ::hisG his3Δ::hisG ura3*. Mutations are homozygous unless stated otherwise.