

1 **Supporting Information**

2 **Fig S1. Effects of fluorescent molecule-tagging of one copy of Sid4 on equational
3 segregation at meiosis I.**

4 Equational segregation frequencies of *cen2* in chiasma-forming *rec12⁺* (A) and chiasma-
5 lacking *rec12Δ* cells (B). No tagging: no Sid4-RFP; Sid4-RFP: cells containing one copy
6 of Sid4-RFP. Chromosome segregation was analyzed in >50 cells, and bars show
7 averages of more than three independent experiments. *P<0.05; **P<0.01 (Student's *t*-
8 test). ns: not significant. Error bars show standard error. Sid4-RFP was used instead of
9 Sid4-GFP for analyzing effects of Sid4 tagging, because segregation patterns of GFP-
10 visualized *cen2* cannot be assessed in cells containing GFP-visualized SPBs.

11

12 **Fig S2. Effects of chiasma formation and impairment of the error correction
13 mechanisms on sister centromere splitting and mobile activities of centromeres.**

14 (A) Sister centromere splitting frequencies in each cell. (B) Mean centromere velocities.
15 The centromere velocities were examined at each time point based on the datasets used
16 for Fig 2. (C) The mean of standard deviation (SD) of centromere positions relative to the
17 SPB. The mean SDs were obtained from SDs of centromere positions in each cell.
18 Numbers in parentheses indicate the number of centromeres examined. +: no other
19 mutations. Error bars show standard errors. Dotted lines in graphs distinguish results of
20 *rec12⁺* and *rec12Δ* cells. ns: not significant by Student's *t*-test (numbers in parenthesis
21 indicates a P value that is between 0.05 and 0.1). (D) Mean square displacement (MSD)
22 of centromeres relative to the SPB. Red dotted lines indicate 95% confidence intervals
23 obtained by the bootstrap method. The two rightmost graphs demonstrate superposition
24 of the average plots.

25

26 **Fig S3. Distribution of distances between splitting sister centromeres with respect to
27 angles of splitting sister centromeres**

28 (A) Distances between splitting sister centromeres and angles of splitting sister
29 centromeres relative to the spindle. Drawing demonstrates the angle of the splitting sister
30 centromeres to the spindle axis, “θ”, the distance between splitting centromeres, “D”, and
31 the projected distance along the spindle axis, “D·cosθ”. (B and C) 2D plots of the angles
32 and the distances (B) or the projected distances (C). WT w/o: not including a dataset of a

1 wild-type cell shown in Fig. 2.

2

3 **Fig S4. Distances of splitting sister centromeres in various cells.**

4 (A) Distribution of distances between splitting sister centromeres. (B) Mean distances
5 between splitting sister centromeres obtained by the bootstrap method. (C) Distribution
6 of the projected splitting distances. (D) The mean projected splitting distances obtained
7 by the bootstrap method. 60, 58, 132, 169, 22, and 109 splitting sister centromeres were
8 examined in wild-type, *mad2Δ*, *ark1-so*, *rec12Δ*, *mad2Δ rec12Δ*, and *ark1-so rec12Δ*
9 cells, respectively. *P<0.05; **P<0.01; ns: not significant (numbers show P values by
10 Studentized bootstrap analysis). Differences between chiasma-forming and chiasma-
11 lacking cells are not shown. In (B), numbers in parentheses at the top of graphs indicate
12 the numbers of cells examined. Dotted lines in graphs distinguish results of *rec12⁺* and
13 *rec12Δ* cells. WT w/o: not including a dataset of a wild-type cell shown in Fig. 2.

14

15

16 **Fig S5. Correlation analysis of centromere dynamics in *mad2Δ* and *ark1-so* cells at**

17 metaphase I.

18 (A and D) Auto-correlation and cross-correlation functions of centromere velocities in
19 *ark1-so* and *ark1-so rec12Δ* cells (A) and those in *mad2Δ* and *mad2Δ rec12Δ* cells (D).
20 Blue lines and red dotted lines respectively indicate mean correlation values and 95%
21 confidence intervals obtained by the bootstrap method. (B and E) P values of ACF and
22 CCF values at the indicated time points. (C and F) P values between *ark1-so* and *ark1-so*
23 *rec12Δ* CCF values (C) and between *mad2Δ* and *mad2Δ rec12Δ* CCF values (F) at the
24 indicated time points. The numbers of examined cells and total observation times (shown
25 in parentheses) are following. *ark1-so*: 11 cells (1432.2 s); *ark1-so rec12Δ*: 16 cells
26 (1392.0 s); *mad2Δ*: 10 cells (749.1 s); *mad2Δ rec12Δ*: 8 cells (479.4 s).

27

28 **Fig S6. Spore formation and viability and recombination in *dam1Δ* cells.**

29 (A) Number of spores in asci. The numbers in parentheses indicate numbers of examined
30 asci. (B) Spore viability. Spores in tetrads were dissected using a microneedle, and spore
31 viability was examined by colony formation. Numbers of examined spores are shown in
32 parentheses. (C) Meiotic recombination in *dam1Δ* cells. Left graph shows genetic

1 distance between the *leu1* and the *his2* loci examined by tetrad analysis. Numbers in
2 parentheses indicate numbers of examined tetrads. Middle and right graphs show gene
3 conversion rates of the *ade6-M26* (hot spot mutation) and *ade6-M375* mutations. Error
4 bars indicate standard errors ($n = 3$). ns: not significant by Student's *t*-test.

5

6 **Fig S7. Ark1 localization in wild-type and *dam1Δ* cells at meiosis I.**

7 (A) Changes in Ark1 localization during meiosis I. Analysis of live cells producing
8 mCherry-tagged α-tubulin and GFP-tagged Ark1 at meiosis I. Numbers indicate time in
9 minutes. Bar: 4 μm. (B) Juxtaposition of Ark1 punctate signals with Mis12 kinetochore
10 signals at meiosis I. Bar: 2 μm.

11

12 **Fig S8. Dynamics of centromeres and the spindle and correlation analysis of
13 centromere movements in *dam1Δ* cells.**

14 (A) Maximum spindle length at metaphase I. Numbers in parentheses indicate the number
15 of spindles examined. (B) The mean standard deviations of centromere positions relative
16 to the SPB. Numbers in parentheses indicate the number of examined centromeres. (C)
17 The MSD of centromeres relative to the SPB in *dam1Δ* and *dam1Δ rec12Δ* cells. Dotted
18 lines indicate 95% confidence intervals obtained by the bootstrap method. Grey lines
19 indicate MSD plots of wild-type or *rec12Δ* cells. (D) Auto-correlation and cross-
20 correlation functions of centromere velocities in *dam1Δ* and *dam1Δ rec12Δ* cells. (E and
21 F) P values of ACF and CCF values (E) and those between *dam1Δ* and *dam1Δ rec12Δ*
22 CCF values (F) at the indicated time points. (G) Changes in centromere–SPB and SPB–
23 SPB distances during anaphase I in *dam1Δ rec12Δ* cells. (H) Loss of anaphase A
24 centromere movements in *dam1Δ rec12Δ* cells. Photos show changes in centromere
25 positions during anaphase I, and numbers show time in minutes from the start of anaphase.
26 Dotted lines and arrowheads show positions of the SPB and centromeres, respectively.
27 Bar: 2 μm. Graph shows mean centromere–SPB distances during metaphase I (Meta) and
28 anaphase I (Ana). Numbers in parentheses indicate the number of examined centromeres.
29 Data of *rec12Δ* cells adopted from our previous published results [22]. In (B) and (C),
30 data of wild-type and *rec12Δ* cells were adopted from Fig S2. Error bars in graphs show
31 standard errors. In (A), (B), and (H), dotted lines in graphs distinguish results of *rec12⁺*
32 and *rec12Δ* cells (A and B) or those of *rec12Δ* and *dam1Δ rec12Δ* cells (H). *P<0.0005;

1 ns: not significant (Student's *t*-test). In (B) to (F), the numbers of examined cells and total
2 observation times (shown in parentheses) are following. *dam1Δ*: 8 cells (970.2 s); *dam1Δ*
3 *rec12Δ*: 8 cells (735.9 s).

4

5 **Fig S9. Possible assembly/disassembly dynamics of kMTs during centromere
6 oscillation and elimination of erroneous attachments in mitosis and meiosis I.**

7 In this model, we assume that each MT bundle interacting with a single kinetochore or
8 associated sister kinetochores undergoes assembly/disassembly switching cooperatively,
9 whereas MT bundles interacting with different kinetochores or associated sister
10 kinetochores undergo switching independently. In addition, MT disassembly drives
11 centromere movements, whereas MT assembly does not. (A) Possible
12 assembly/disassembly dynamics of properly attached kMTs. Centromere movement and
13 kMT dynamics: direction of centromere movement and kMT dynamics. Transition state:
14 two possible situations of kMT dynamics at the reversal of the movements. At the
15 transition state, simultaneous disassembly of kMTs extending from opposite poles causes
16 centromere stretch (left), whereas their simultaneous assembly causes centromere
17 relaxation (right). These tensile changes would cause coordinated switching of kMT
18 dynamics and reversal of homologous centromere movements. (B) A problem with the
19 model of Aurora B-dependent elimination of improper attachments in fission yeast.
20 Because sister centromeres separate from each other after their bipolar attachment, the
21 orientation of kinetochores and centromeres easily changes. Consequently, when bi-
22 oriented attachment of a single centromere or sister centromeres takes place in mitosis or
23 meiosis I, respectively, improper attachment sites cannot approach the Aurora B-enriched
24 region, and the kinetochore components cannot undergo Aurora B-dependent
25 phosphorylation. Red dotted boxes indicate kinetochore-MT interactions, and predicted
26 positions of kinetochores and the Aurora B-enriched region. (C) Possible
27 assembly/disassembly dynamics of improperly attached kMTs in mitosis or meiosis I. In
28 this case, one of sister centromeres (Mitosis) or one sister centromere pair (Meiosis I) is
29 improperly attached to opposite poles, and the MT bundles interacting with different
30 kinetochores or associated sister kinetochores extend forward or rearward of the moving
31 centromeres during centromere oscillation. During poleward centromere movements, the
32 leading kMTs undergo disassembly, while the trailing kMTs undergo assembly as in (A),

1 and in this case, attachment elimination should not occur (Centromere movement & kMT
2 dynamics). However, there are occasions, in which one of the leading kMTs that are
3 improperly attached undergoes assembly (b) and is selectively eliminated due to sister
4 chromatid cohesion-dependent load (Mitosis) or a chiasma-dependent minus end-directed
5 load (Meiosis I) (kMT detachment). Such a situation would never arise at proper
6 attachment sites. L-kMT: leading kMT; T-kMT: tailing kMT. Assembly/disassembly
7 dynamics of kMTs interacting with the designated kinetochores are shown in boxes at the
8 bottom (kMT dynamics). For simplicity, only one kMT attached to each single
9 kinetochore is shown.

10

11 **Table S1. Colocalization and position switching events of homologous centromeres**

12

13 **Table S2. Strain genotypes**

14

15 **Table S3. Strains used in figures**

16

17 **Movie S1. Centromere and SPB dynamics at metaphase I in a wild-type cell**

18 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) in a meiotic
19 wild-type cell exhibiting Cut2 spindle localization. The rate of the movie is 5 frames per
20 second.

21

22 **Movie S2. Centromere and SPB dynamics at metaphase I in a *rec12Δ* cell**

23 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) in a meiotic
24 *rec12Δ* cell exhibiting Cut2 spindle localization. The rate of the movie is 5 frames per
25 second.

26

27 **Movie S3. Centromere and SPB dynamics at metaphase I in a *mad2Δ* cell**

28 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) in a meiotic
29 *mad2Δ* cell exhibiting Cut2 spindle localization. The rate of the movie is 5 frames per
30 second.

31

32 **Movie S4. Centromere and SPB dynamics at metaphase I in a *mad2Δ rec12Δ* cell**

1 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) in a meiotic
2 *mad2Δ rec12Δ* cell exhibiting Cut2 spindle localization. The rate of the movie is 5 frames
3 per second.

4

5 **Movie S5. Centromere and SPB dynamics at metaphase I in an *ark1-so* cell**

6 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) in a meiotic
7 *ark1-so* cell exhibiting Cut2 spindle localization. The rate of the movie is 5 frames per
8 second.

9

10 **Movie S6. Centromere and SPB dynamics at metaphase I in an *ark1-so rec12Δ* cell**

11 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) in a meiotic
12 *ark1-so rec12Δ* cell exhibiting Cut2 spindle localization. The rate of the movie is 5 frames per
13 second.

14

15 **Movie S7. Centromere and SPB dynamics at metaphase I in a *dam1Δ* cell**

16 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) in a meiotic
17 *dam1Δ* cell exhibiting Cut2 spindle localization. The rate of the movie is 5 frames per
18 second.

19

20 **Movie S8. Centromere and SPB dynamics at metaphase I in a *dam1Δ rec12Δ* cell**

21 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) in a meiotic
22 *dam1Δ rec12Δ* cell exhibiting Cut2 spindle localization. The movie plays at rate of 5
23 frames per second.

24

25 **Movie S9. Centromere and spindle dynamics at anaphase I in a wild-type cell**

26 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) and an
27 *mCherry*-visualized spindle in a meiotic wild-type cell exhibiting Cut2 spindle
28 localization. The rate of the movie is 3 frames per second.

29

30 **Movie S10. Centromere and spindle dynamics at anaphase I in a *dam1Δ* cell**

1 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) and an
2 *mCherry*-visualized spindle in a meiotic *dam1Δ* cell. The rate of the movie is 3 frames
3 per second.

4

5 **Movie S11. Centromere and spindle dynamics at anaphase I in a *dam1Δ rec12Δ* cell**

6 The movie shows time-lapse images of GFP-visualized *cen2* and SPB (Sid4) and an
7 *mCherry*-visualized spindle in a meiotic *dam1Δ rec12Δ* cell. The rate of the movie is 3
8 frames per second.

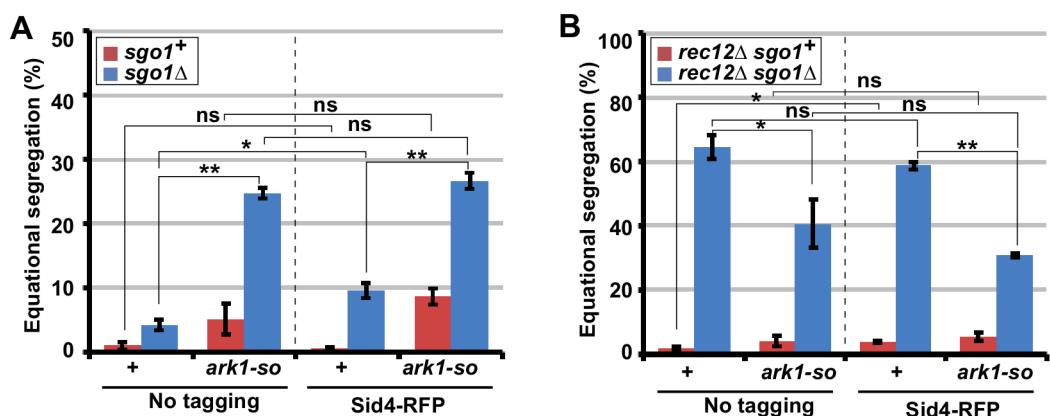
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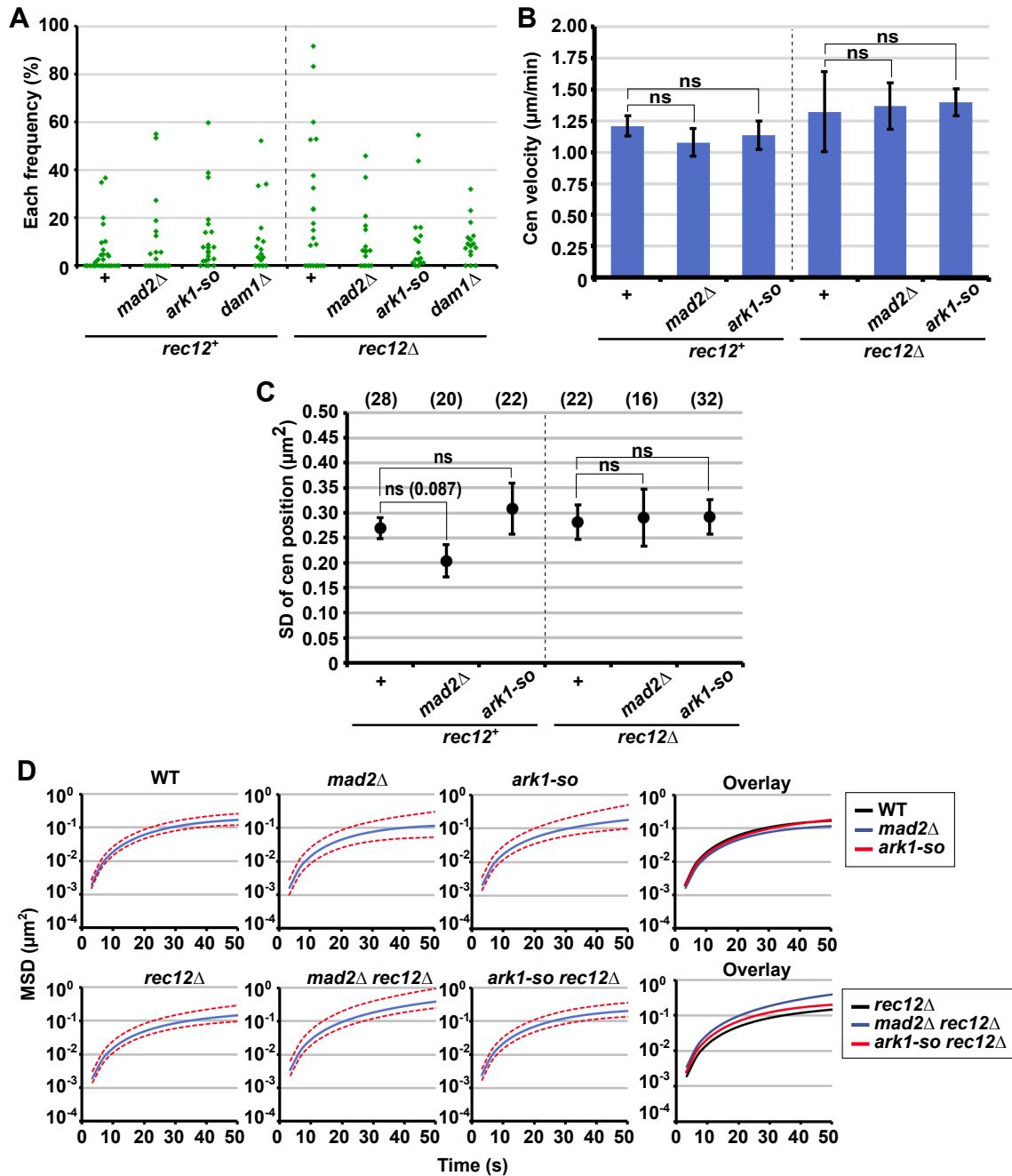
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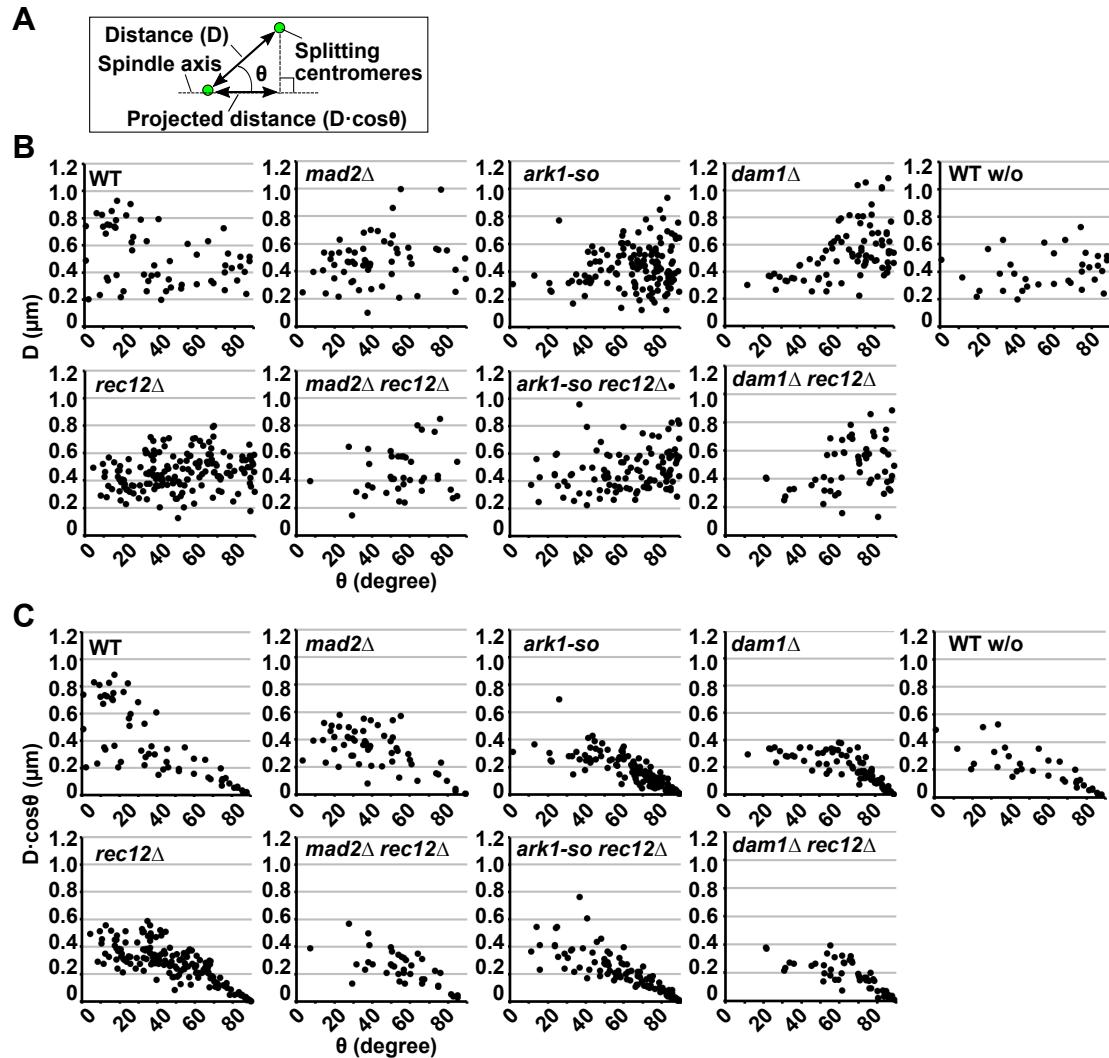
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1 Fig S3

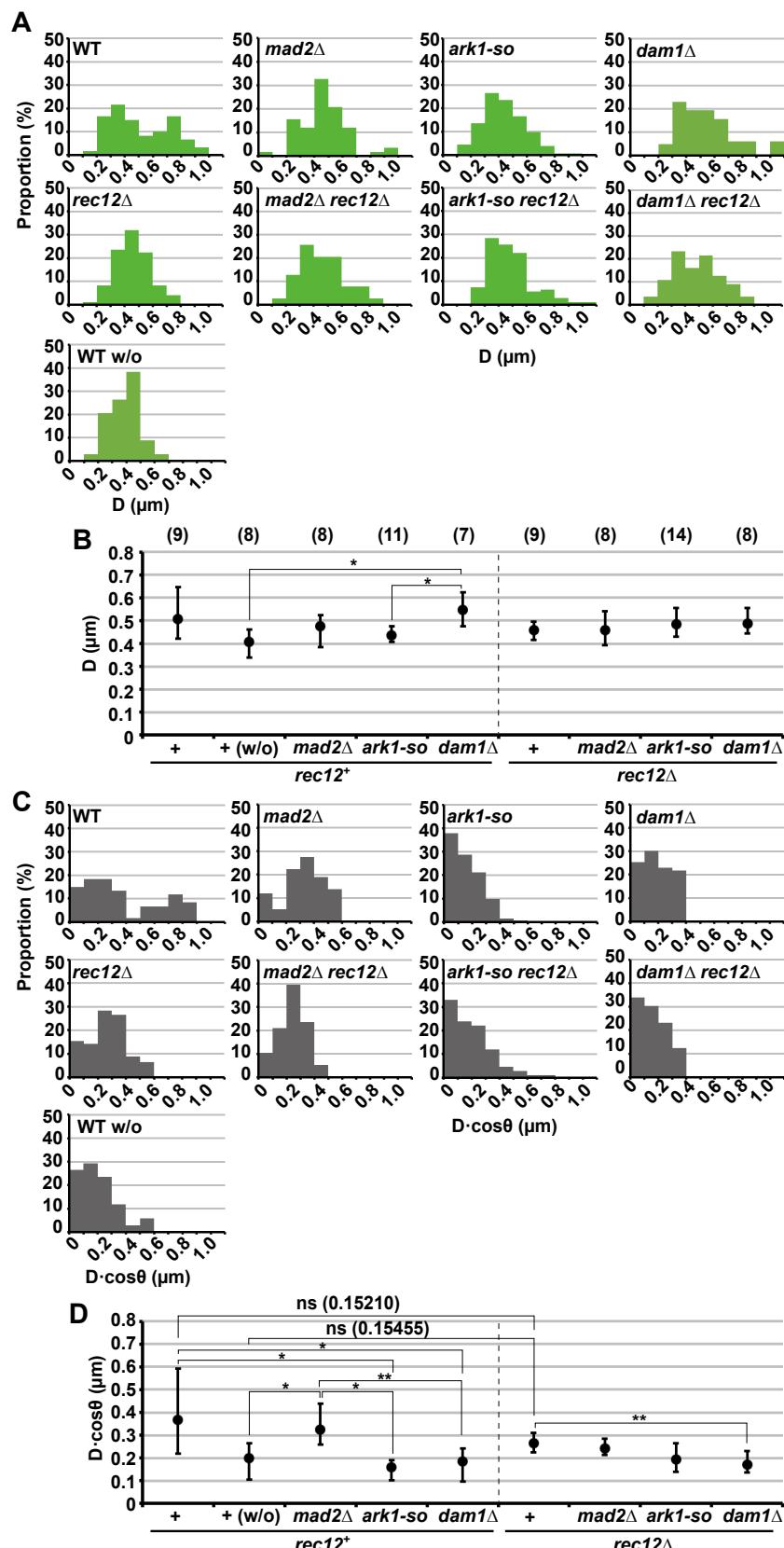


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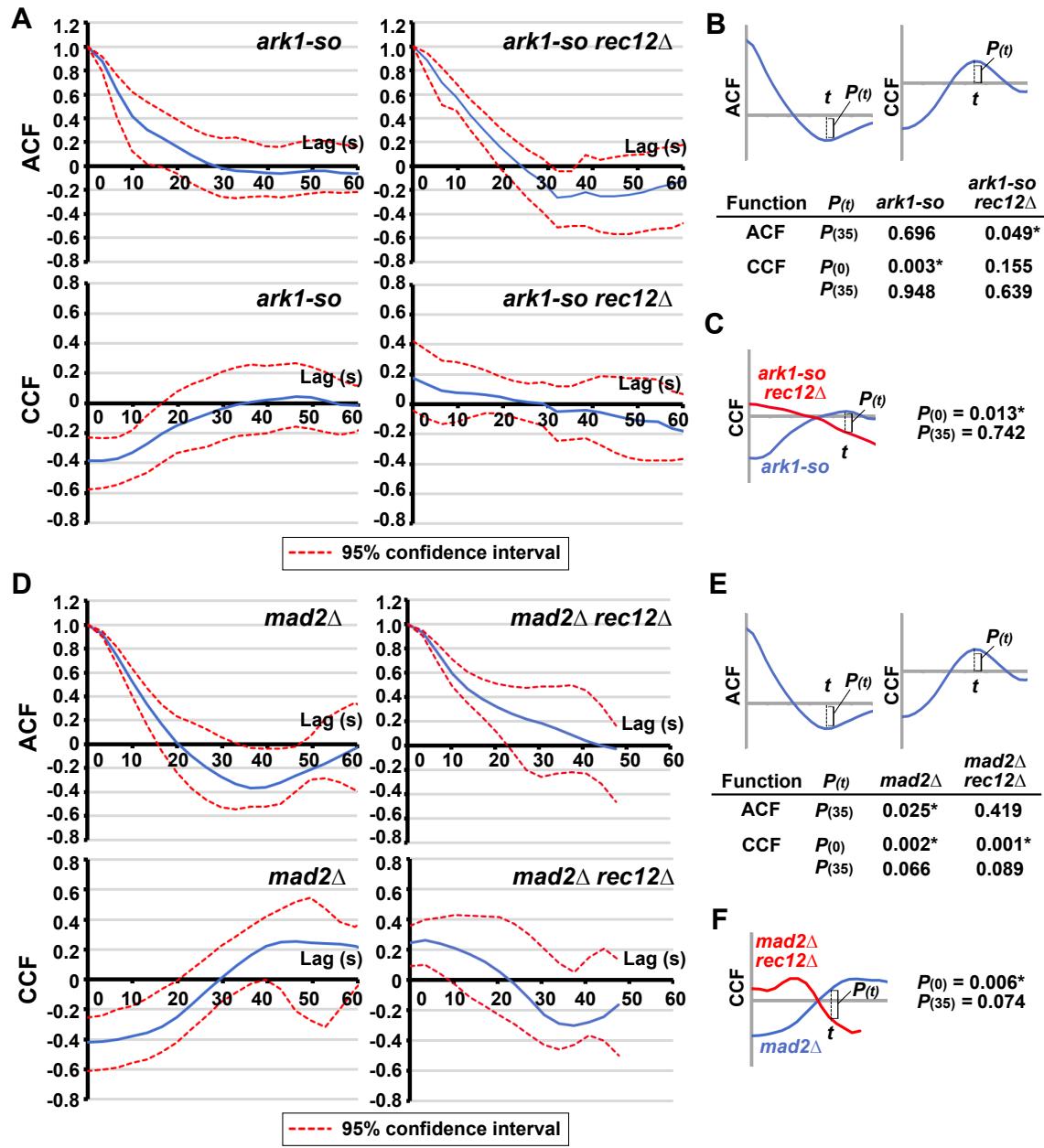
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1 Fig S4



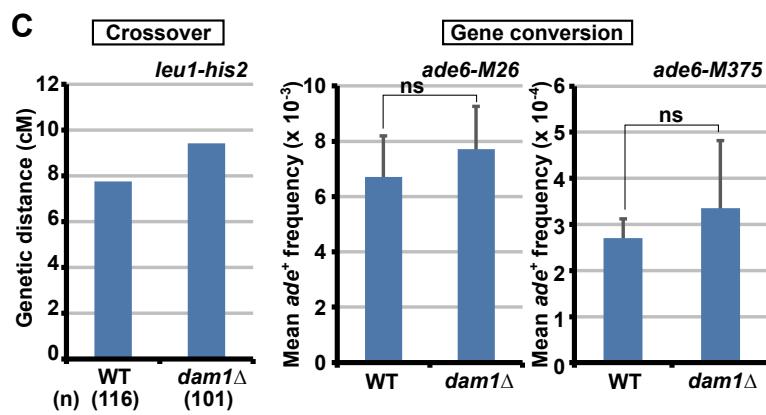
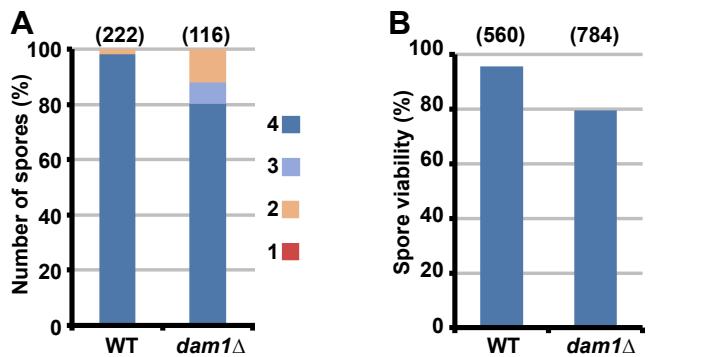
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1 Fig S5



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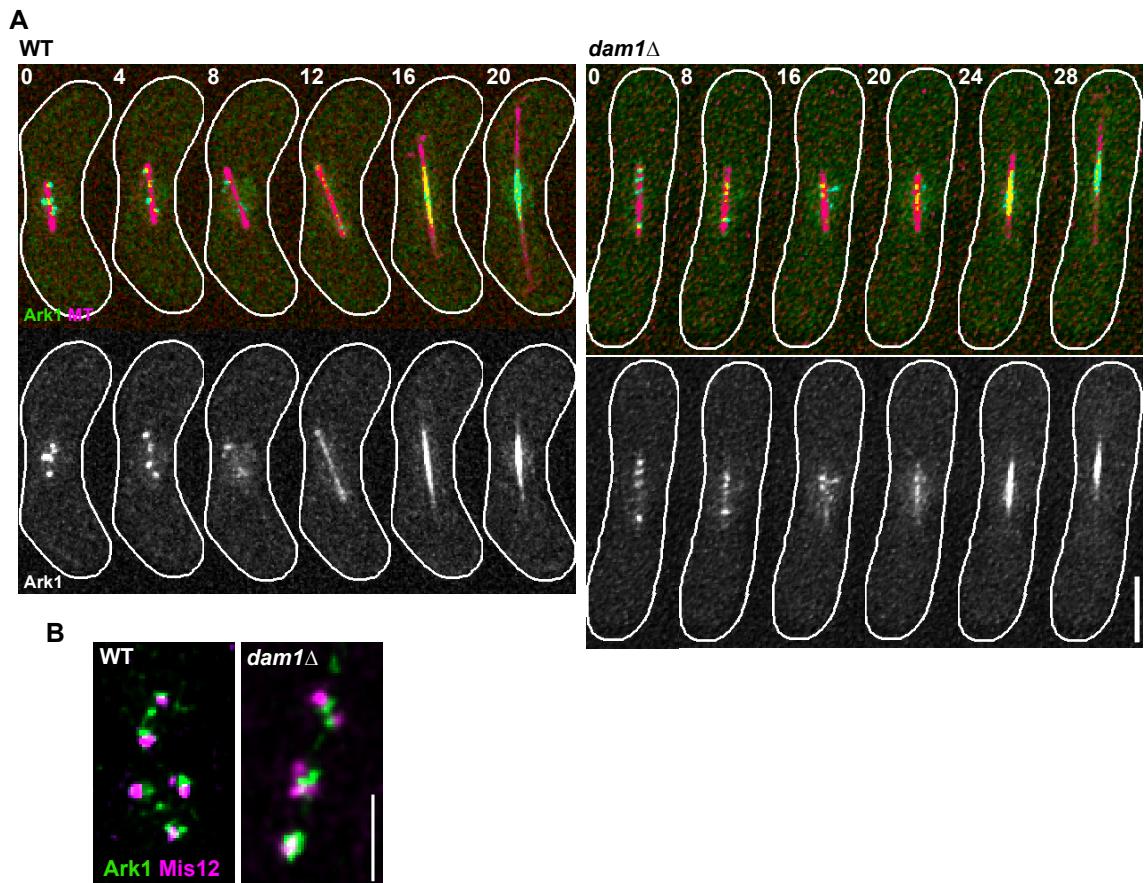
1 Fig S6



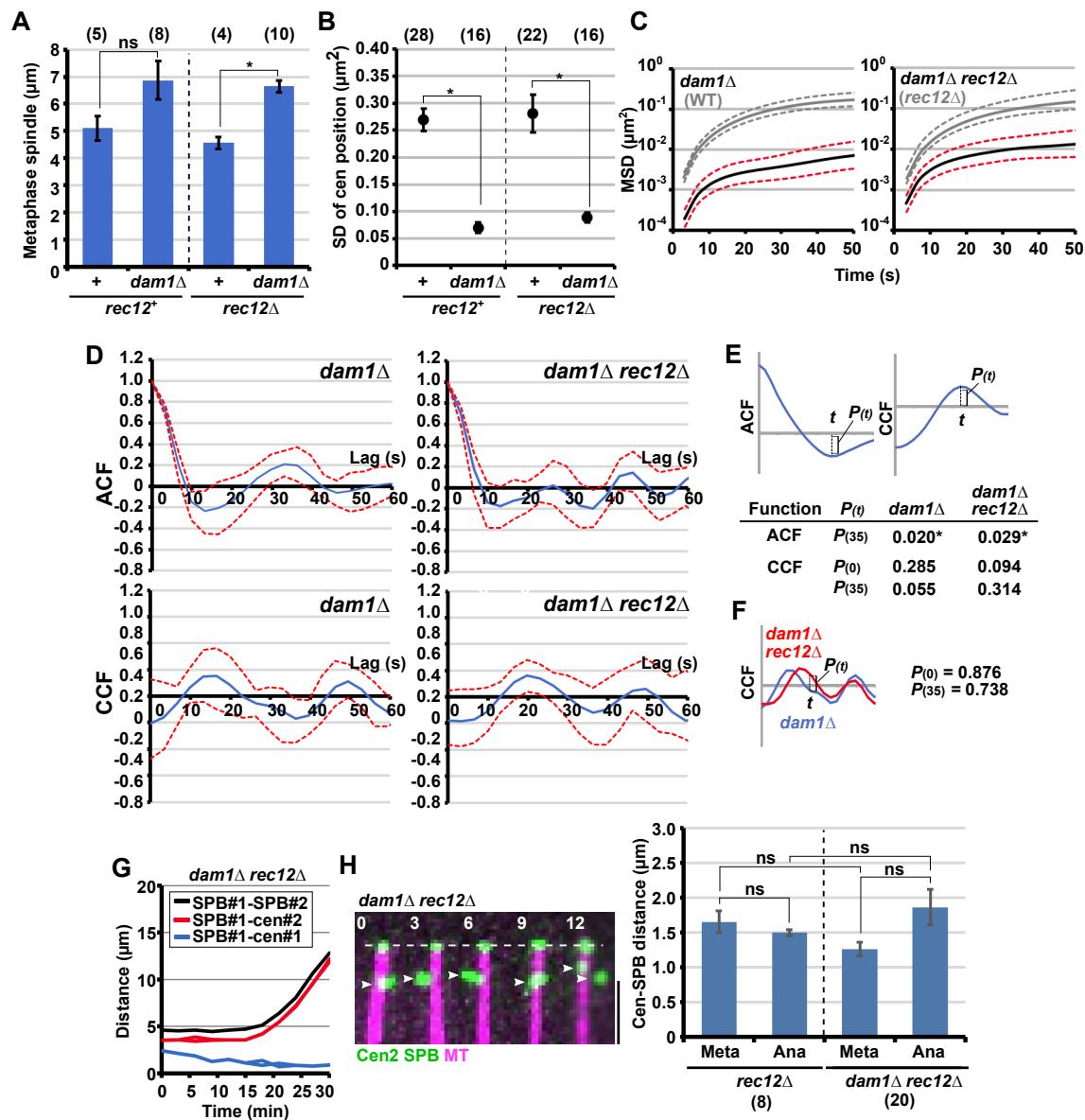
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1 **Fig S7**



1 Fig S8

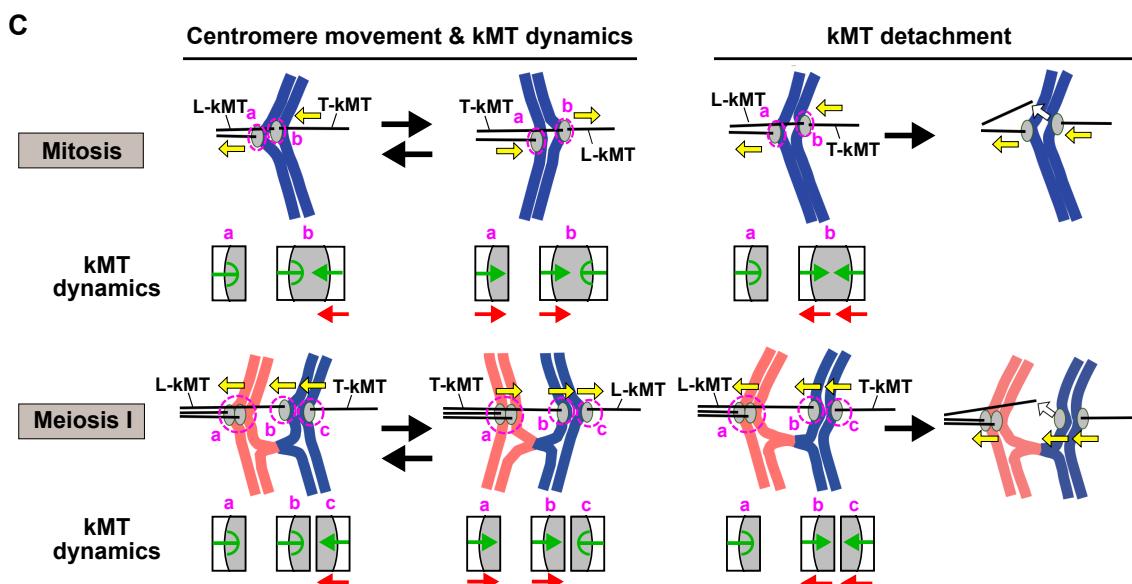
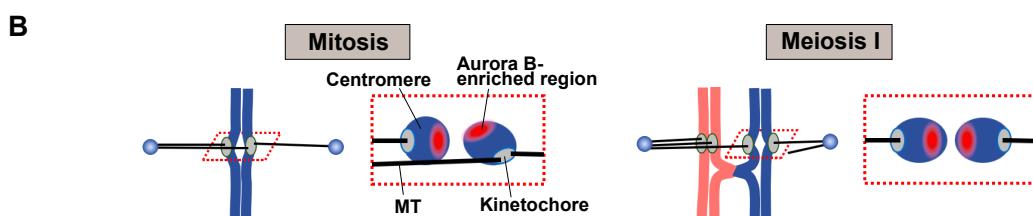
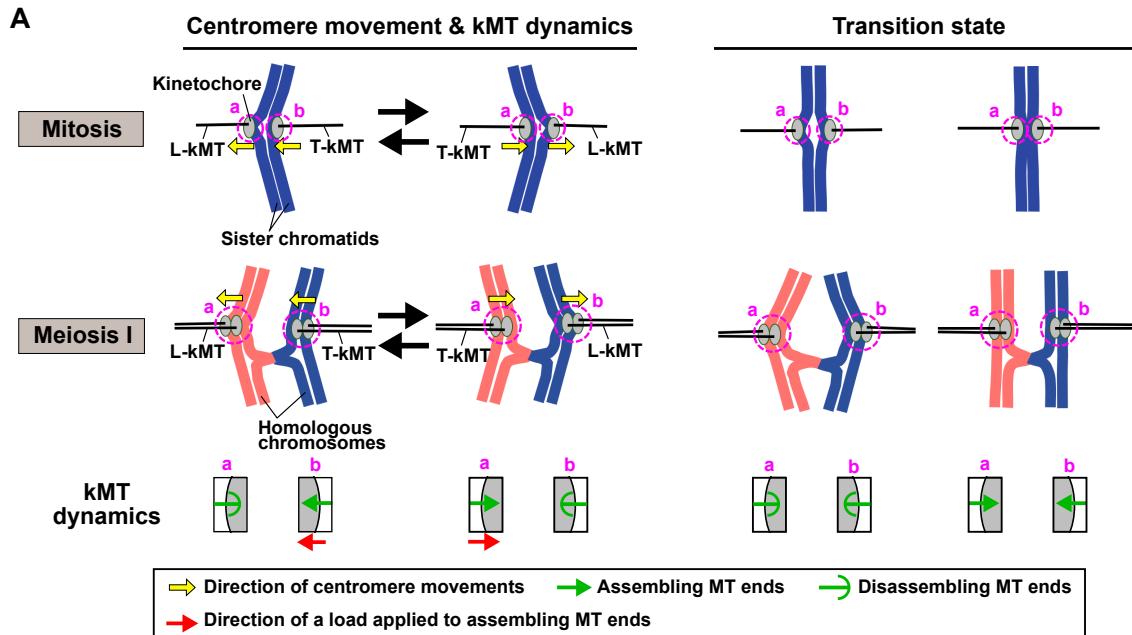


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1 Fig S9



2

1 **Table S1. Colocalization and position switching events of homologous centromeres**

Cell #	WT			<i>mad2Δ</i>			<i>ark1-so</i>		
	Co-localization time points	Switching events	Observation time points	Co-localization time points	Switching events	Observation time points	Co-localization time points	Switching events	Observation time points
1	0	0	41	0	0	14	0	2	38
2	0	0	34	0	0	24	10	0	50
3	0	0	38	0	0	18	3	0	36
4	0	0	28	0	0	17	0	2	28
5	0	0	30	0	0	11	0	0	53
6	0	0	23	0	0	15	0	0	49
7	0	0	31	0	0	35	20	0	52
8	0	0	23	0	0	69	12	0	51
9	0	0	21	0	0	40	1	0	46
10	0	0	17	0	0	35	0	0	57
11	0	0	68				0	2	38
12	0	0	40						
13	0	0	52						
14	0	0	40						
Total time points/events	0	0	486	0	0	278	46	6	498
	Co-localization frequency	Total switching events	Total observation time* (s)	Co-localization frequency	Total switching events	Total observation time* (s)	Co-localization frequency	Total switching events	Total observation time* (s)
Total	0.00%	0	1506.6	0.00%	0	917.4	9.24%	6	1643.4

2 *Total observation time: calculated from total time points and time intervals (3.1 s for WT and 3.3 s for *mad2Δ* and *ark1-so* cells).

Table S2. Strain genotypes

Strain name	Genotype
SY16-31	<i>h⁻ leu1-32 lys1-131 ura4-D18 his7⁺::Pdis1-GFP-lacI-NLS</i>
SY6-59	<i>h⁺ ade6-210 leu1-32 ura4-D18 lys1⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH129-3C	<i>h⁻ leu1-32 ura4-D18 mad2::ura4⁺ his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH129-2D	<i>h⁺ leu1-32 ura4-D18 lys1⁺::lacO mad2::ura4⁺ his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH317-13B	<i>h⁻ leu1-32 ura4-D18 lys1⁺::lacO ark1-so::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH257-2B	<i>h⁺ leu1-32 lys1-131 ura4-D18 ark1-so::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH91-14B	<i>h⁻ leu1-32 lys1-131 ura4-D18 sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH91-2-11B	<i>h⁺ leu1-32 ura4-D18 sgo1::kan^r lys1⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH215-3A	<i>h⁻ leu1-32 ura4-D18 mad2::ura4⁺ sgo1::kan^r lys1⁺::lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH215-1C	<i>h⁺ lys1-131 leu1-32 ura4-D18 mad2::ura4⁺ sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYI15-16B	<i>h⁻ leu1-32 lys1-131 ura4-D18 ark1-so::kan^r sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYI15-19B	<i>h⁺ leu1-32 ura4-D18 ark1-so::kan^r sgo1::kan^r lys1⁺::lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH129-16D	<i>h⁻ leu1-32 ura4-D18 rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH129-3B	<i>h⁺ leu1-32 ura4-D18 rec12-152::LEU2 lys1⁺::lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH129-6D	<i>h⁻ leu1-32 ura4-D18 mad2::ura4⁺ rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH129-3D	<i>h⁺ leu1-32 ura4-D18 mad2::ura4⁺ rec12-152::LEU2 lys1⁺::lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH257-1B	<i>h⁻ leu1-32 lys1-131 ura4-D18 rec12-152::LEU2 ark1-so::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH317-16A	<i>h⁺ leu1-32 ura4-D18 rec12-152::LEU2 ark1-so::kan^r lys1⁺::lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH91-1C	<i>h⁻ leu1-32 lys1-131 ura4-D18 rec12-152::LEU2 sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH91-15B	<i>h⁺ leu1-32 ura4-D18 rec12-152::LEU2 sgo1::kan^r lys1⁺::lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH215-2A	<i>h⁻ leu1-32 ura4-D18 mad2::ura4⁺ rec12-152::LEU2 sgo1::kan^r lys1⁺::lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH215-6A	<i>h⁺ leu1-32 ura4-D18 mad2::ura4⁺ rec12-152::LEU2 sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SVV89-2D	<i>h⁻ leu1-32 lys1-131 ura4-D18 ark1-so::kan^r rec12-152::LEU2 sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYI15-20B	<i>h⁺ leu1-32 ura4-D18 ark1-so::kan^r rec12-152::LEU2 sgo1::kan^r lys1⁺::lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH19-6A	<i>h⁻ leu1-32 lys1-131 ura4-D18 his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH19-4C	<i>h⁺ leu1-32 lys1-131 ura4-D18 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SY14-48	<i>h⁻ ade6-210 leu1-32 ura4-D18 mad2::ura4⁺ his7⁺::Pdis1-GFP-lacI-NLS</i>
SY20-27	<i>h⁺ ade6-210 leu1-32 lys1-131 ura4-D18 mad2::ura4⁺ cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH258-16A	<i>h⁻ ade6-216 leu1-32 lys1-131 ura4-D18 ark1-so::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH257-2B	<i>h⁺ leu1-32 lys1-131 ura4-D18 ark1-so::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH18-2-6A	<i>h⁻ leu1-32 ura4-D18 sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH18-2-8B	<i>h⁺ leu1-32 lys1-131 ura4-D18 sgo1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYI18-15D	<i>h⁺ lys1-131 ura4-D18 mad2::ura4⁺ sgo1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>

SYH215-1C	h^+ <i>leu1-32 lys1-131 ura4-D18 mad2::ura4⁺ sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SVV89-12A	h^- <i>leu1-32 ura4-D18 ark1-so::kan^r sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SVV89-2C	h^+ <i>leu1-32 ura4-D18 ark1-so::kan^r sgo1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SY16-43	h^- <i>leu1-32 lys1-131 ura4-D18 rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SY15-26	h^+ <i>leu1-32 lys1-131 rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH44-1-6B	h^- <i>leu1-32 lys1-131 ura4-D18 mad2::ura4⁺ rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH44-1-16A	h^+ <i>leu1-32 lys1-131 ura4-D18 mad2::ura4⁺ rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH257-1B	h^- <i>leu1-32 lys1-131 ura4-D18 ark1-so::kan^r rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH277-1B	h^+ <i>leu1-32 lys1-131 ura4-D18 ark1-so::kan^r rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH18-2-6D	h^- <i>leu1-32 ura4-D18 sgo1::kan^r rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH18-2-1C	h^+ <i>leu1-32 lys1-131 ura4-D18 sgo1::kan^r rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH226-6B	h^- <i>leu1-32 lys1-131 ura4-D18 mad2::ura4⁺ sgo1::kan^r rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH215-6A	h^+ <i>leu1-32 lys1-131 ura4-D18 mad2::ura4⁺ sgo1::kan^r rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SVV89-2D	h^- <i>leu1-32 lys1-131 ura4-D18 ark1-so::kan^r sgo1::kan^r rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SVV89-11C	h^+ <i>leu1-32 lys1-131 ura4-D18 ark1-so::kan^r sgo1::kan^r rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
HM4501-2	h^- <i>ura4-D18 lys1⁺::mat-Pc pat1-114 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYIT30-3	h^+ <i>ura4-D18 mad2::ura4⁺ aur1⁺::aur1^r-mat-Mc pat1-114 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYIT18-2	h^+ <i>ura4-D18 ark1-so::kan^r aur1⁺::aur1^r-mat-Mc pat1-114 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYH193-2A	h^- <i>ura4-D18 aur1⁺::aur1^r-mat-Pc pat1-114 sgo1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYIT7-1	h^- <i>ura4-D18 mad2::ura4⁺ lys1⁺::mat-Pc pat1-114 sgo1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYIT17-1	h^- <i>ura4-D18 ark1-so::kan^r lys1⁺::mat-Pc pat1-114 sgo1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SMW43-22B	h^- <i>leu1-32 lys1-131 ura4-D18 his7⁺::Pdis1-GFP-lacI-NLS sid4⁺::RFP-kan^r</i>
SMW43-9A	h^- <i>leu1-32 lys1-131 ura4-D18 sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS sid4⁺::RFP-kan^r</i>
SKY20-11B	h^- <i>leu1-32 lys1-131 ura4-D18 ark1-so::kan^r sgo1::kan^r sid4⁺::RFP-kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SVV89-11B	h^+ <i>leu1-32 lys1-131 ura4-D18 ark1-so::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SMW43-20D	h^- <i>leu1-32 ura4-D18 rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS sid4⁺::RFP-kan^r</i>
SMW43-9B	h^- <i>leu1-32 ura4-D18 sgo1::kan^r rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS sid4⁺::RFP-kan^r</i>
SKY20-11A	h^+ <i>leu1-32 lys1-131 ura4-D18 ark1-so::kan^r sgo1::kan^r rec12-152::LEU2 sid4⁺::RFP-kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SVV89-16D	h^- <i>leu1-32 lys1-131 ura4-D18 ark1-so::kan^r sgo1::kan^r rec12-152::LEU2</i>

	<i>cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SKKT100-a	<i>h⁻ ade6(-210 or -216) lys1-131 ura4-D18 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS sid4⁺::GFP-kan^r aur1⁺::aur1^r-Pcut2-cut2⁺-mCherry</i>
SY15-12	<i>h⁺ ade6-210 leu1-32 lys1-131 ura4-D18 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SKKT102-a	<i>h⁻ leu1-32 lys1-131 ura4-D18 rec12-152::LEU2 sid4⁺::GFP-kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS aur1⁺::aur1^r-Pcut2-cut2⁺-mCherry</i>
SY15-26	<i>h⁺ leu1-32 lys1-131 rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SENT25-1	<i>h⁻ ade6(-210 or -216) leu1-32 lys1-131 ura4-D18 mad2::ura4⁺ sid4⁺::GFP-kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS aur1⁺::aur1^r-Pcut2-cut2⁺-mCherry</i>
SY20-27	<i>h⁺ ade6-210 leu1-32 lys1-131 ura4-D18 mad2::ura4⁺ cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SENT26-1	<i>h⁻ ade6(-210 or -216) leu1-32 ura4-D18 mad2::ura4⁺ rec12-152::LEU2 sid4⁺::GFP-kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS aur1⁺::aur1^r-Pcut2-cut2⁺-mCherry</i>
SYH144-1-16A	<i>h⁺ leu1-32 lys1-131 ura4-D18 mad2::ura4⁺ rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SMW19-3A	<i>h⁻ leu1-32 lys1-131 ura4-D18 ark1-so::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SMW21-5A	<i>h⁺ leu1-32 ura4-D18 ark1-so::kan^r sid4⁺::GFP-kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS aur1⁺::aur1^r-Pcut2-cut2⁺-mCherry</i>
SYH277-15D	<i>h⁻ leu1-32 lys1-131 ura4-D18 rec12-152::LEU2 ark1-so::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SENT28-1	<i>h⁺ leu1-32 lys1-131 ura4-D18 rec12-152::LEU2 ark1-so::kan^r sid4⁺::GFP-kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS aur1⁺::aur1^r-Pcut2-cut2⁺-mCherry</i>
SAH5-1C	<i>h⁺ leu1-32 lys1-131 ura4-D18 dam1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SAH8-3D	<i>h⁻ leu1-32 lys1-131 ura4-D18 dam1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SHM114-6D	<i>h⁺ leu1-32</i>
SHM115-6D	<i>h⁻ his2-245</i>
SKY7-1A	<i>h⁻ his2-245 dam1::kan^r</i>
SKY7-5B	<i>h⁺ leu1-32 dam1::kan^r</i>
SY5-67	<i>h⁻ ade6-L469</i>
SHM148-11A	<i>h⁺ ade6-M26</i>
SHM230-2C	<i>h⁺ ade6-M375</i>
SMW37-9A	<i>h⁻ ade6-L469 dam1::kan^r</i>
SMW34-23D	<i>h⁺ ade6-M26 dam1::kan^r</i>
SMW35-22D	<i>h⁺ ade6-M375 dam1::kan^r</i>
SYH19-2-9A	<i>h⁻ leu1-32 lys1-131 ura4-D18 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SAH5-1D	<i>h⁻ leu1-32 lys1-131 ura4-D18 dam1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SAH4-5D	<i>h⁺ leu1-32 lys1-131 ura4-D18 dam1::kan^r sgo1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SAH8-2D	<i>h⁻ leu1-32 lys1-131 ura4-D18 dam1::kan^r sgo1::kan^r his7⁺::Pdis1-GFP-lacI-NLS</i>
SAH5-2D	<i>h⁺ leu1-32 lys1-131 ura4-D18 dam1::kan^r rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SAH8-8B	<i>h⁻ leu1-32 lys1-131 ura4-D18 dam1::kan^r rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SAH4-6D	<i>h⁺ leu1-32 lys1-131 ura4-D18 dam1::kan^r sgo1::kan^r rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>

SAH8-9B	<i>h⁻ leu1-32 lys1-131 ura4-D18 dam1::kan^r sgo1::kan^r rec12-152::LEU2 his7⁺::Pdis1-GFP-lacI-NLS</i>
SMW27-4D	<i>h⁻ ura4-D18 dam1::kan^r lys1⁺::mat-Pc pat1-114 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SAK20-12A	<i>h⁻ ura4-D18 dam1::kan^r lys1⁺::mat-Pc pat1-114 sgo1::kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SHM193-2C	<i>h⁹⁰ ark1⁺::GFP-hyg^r lys1⁺::Pnda3-mCherry-atb2⁺</i>
SMW41-2D	<i>h⁻ leu1-32 lys1-131 ura4-D18 dam1::kan^r ark1⁺::GFP-hyg^r</i>
SMW41-6A	<i>h⁺ leu1-32 ura4-D18 dam1::kan^r ark1⁺::GFP-hyg^r lys1⁺::Pnda3-mCherry-atb2⁺</i>
FY20781	<i>h⁻ leu1-32 ura4-D18 mis12⁺-RFP::LEU2</i>
SMW36-9A	<i>h⁺ leu1-32 ark1⁺::GFP-hyg^r mis12⁺-RFP::LEU2</i>
SMW39-6A	<i>h⁻ leu1-32 lys1-131 ura4-D18 dam1::kan^r ark1⁺::GFP-hyg^r mis12⁺-RFP::LEU2</i>
SMW39-7D	<i>h⁺ leu1-32 ura4-D18 dam1::kan^r ark1⁺::GFP-hyg^r mis12⁺-RFP::LEU2</i>
SYS2-2D	<i>h⁻ ade6(-210 or -216) ura4-D18 dam1::kan^r sid4⁺::GFP-kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS aur1⁺::aur1'-Pcut2-cut2⁺-mCherry</i>
SMWT8-1	<i>h⁺ leu1-32 lys1-131 ura4-D18 dam1::kan^r rec12-152::LEU2 sid4⁺::GFP-kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS aur1⁺::aur1'-Pcut2-cut2⁺-mCherry</i>
SAH4-10C	<i>h⁻ leu1-32 lys1-131 ura4-D18 dam1::kan^r rec12-152::LEU2 cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SAH43-1D	<i>h⁻ leu1-32 ura4-D18 lys1⁺::Pnda1-mCherry-atb2⁺ cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SYI32-20B	<i>h⁺ ade6(-210 or 216) leu1-32 lys1-131 ura4-D18 sid4⁺::GFP-kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SENT59-1	<i>h⁻ leu1-32 ura4-D18 dam1::kan^r lys1⁺::Pnda1-mCherry-atb2⁺ cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SEN50-25C	<i>h⁺ ade6(-210 or 216) leu1-32 dam1::kan^r sid4⁺::GFP-kan^r cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SMWT7-1	<i>h⁻ leu1-32 ura4-D18 dam1::kan^r rec12-152::LEU2 lys1⁺::Pnda1-mCherry-atb2⁺ cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>
SMWT6-1	<i>h⁺ leu1-32 ura4-D18 dam1::kan^r rec12-152::LEU2 lys1⁺::Pnda1-mCherry-atb2⁺ cen2::kan^r-ura4⁺-lacO his7⁺::Pdis1-GFP-lacI-NLS</i>

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Table S3. Strains used in figures

Figure	Characteristics/genotype		Strains
Figs 1D, 1E and S1 (no tagging)	Cen1	WT	SY6-59, SY16-31
		<i>mad2Δ</i>	SYH129-2D, SYH129-3C
		<i>ark1-so</i>	SYH317-13B, SYH257-2B
		<i>sgo1Δ</i>	SYH91-2-11B, SYH91-14B
		<i>mad2Δ sgo1Δ</i>	SYH215-3A, SYH215-1C
		<i>ark1-so sgo1Δ</i>	SYI15-19B, SYI15-16B
		<i>rec12Δ</i>	SYH129-3B, SYH129-16D
		<i>mad2Δ rec12Δ</i>	SYH129-3D, SYH129-6D
		<i>ark1-so rec12Δ</i>	SYH317-16A, SYH257-1B
		<i>rec12Δ sgo1Δ</i>	SYH91-15B, SYH91-1C
		<i>mad2Δ rec12Δ sgo1Δ</i>	SYH215-2A, SYH215-6A
		<i>ark1-so rec12Δ sgo1Δ</i>	SVV89-2D, SYI15-20B
	Cen2	WT	SYH19-4C, SYH19-6A
		<i>mad2Δ</i>	SY20-27, SY14-48
		<i>ark1-so</i>	SYH258-16A, SYH257-2B
		<i>sgo1Δ</i>	SYH18-2-8B, SYH18-2-6A
		<i>mad2Δ sgo1Δ</i>	SYH215-1C, SYI18-15D
		<i>ark1-so sgo1Δ</i>	SVV89-2C, SVV89-12A
		<i>rec12Δ</i>	SY15-26, SY16-43
		<i>mad2Δ rec12Δ</i>	SYH44-1-16A, SYH44-1-6B
		<i>ark1-so rec12Δ</i>	SYH277-1B, SYH257-1B
		<i>rec12Δ sgo1Δ</i>	SYH18-2-1C, SYH18-2-6D
		<i>mad2Δ rec12Δ sgo1Δ</i>	SYH215-6A, SYH226-6B
		<i>ark1-so rec12Δ sgo1Δ</i>	SVV89-2D, SVV89-11C
Fig 1F	<i>pat1-114</i>		HM4501-2
	<i>mad2Δ pat1-114</i>		SYIT30-3
	<i>ark1-so pat1-114</i>		SYIT18-2
	<i>sgo1Δ pat1-114</i>		SYH193-2A
	<i>mad2Δ sgo1Δ pat1-114</i>		SYIT7-1
	<i>ark1-so sgo1Δ pat1-114</i>		SYIT17-1
Fig S1 (Sid4-RFP)	WT		SMW43-22B, SYH19-4C
	<i>sgo1Δ</i>		SMW43-9A, SYH18-2-8B
	<i>ark1-so</i>		SKY20-11B, SVV89-11B
	<i>ark1-so sgo1Δ</i>		SKY20-11B, SVV89-2C
	<i>rec12Δ</i>		SMW43-20D, SY15-26
	<i>rec12Δ sgo1Δ</i>		SMW43-9B, SYH18-2-1C
	<i>ark1-so rec12Δ</i>		SYH277-15D, SKY20-11A
	<i>ark1-so rec12Δ sgo1Δ</i>		SVV89-16D, SKY20-11A
Figs 2–4, and S2 – S5, and Table S1	WT		SKKT100-a, SY15-12
	<i>rec12Δ</i>		SY15-26, SKKT102-a
	<i>mad2Δ</i>		SY20-27, SENT25-1
	<i>rec12Δ mad2Δ</i>		SYH44-1-16A, SENT26-1
	<i>ark1-so</i>		SMW19-3A, SMW21-5A
	<i>ark1-so rec12Δ</i>		SYH277-15D, SENT28-1
	Fig S6A and S6B	WT	SYH19-4C, SYH19-6A
		<i>dam1Δ</i>	SAH5-1C, SAH8-3D
Fig S6C (Crossover)	WT		SHM114-6D, SHM115-5D
	<i>dam1Δ</i>		SKY7-5B, SKY7-1A
Fig S6C (Gene Conversion)	WT	<i>ade6-M26</i>	SHM148-11A, SY5-67
		<i>ade6-M375</i>	SHM230-2C, SY5-67

	<i>dam1Δ</i>	<i>ade6-M26</i>	SMW34-23D, SMW37-9A
		<i>ade6-M375</i>	SMW35-22D, SMW37-9A
Fig 5A	WT		SYH19-2-9A, SYH19-4C
	<i>dam1Δ</i>		SAH5-1C, SAH5-1D
Fig 5B	<i>dam1Δ</i>		SAH5-1C, SAH8-3D
	<i>dam1Δ sgo1Δ</i>		SAH4-5D, SAH8-2D
	<i>dam1Δ rec12Δ</i>		SAH5-2D, SAH8-8B
	<i>dam1Δ rec12Δ sgo1Δ</i>		SAH4-6D, SAH8-9B
Fig 5C	<i>dam1Δ</i>		SMW27-4D
	<i>dam1Δ sgo1Δ</i>		SAK20-12A
Fig S7A	WT		SHM193-2C
	<i>dam1Δ</i>		SMW41-2D, SMW41-6A
Fig S7B	WT		FY20781, SMW36-9A
	<i>dam1Δ</i>		SMW39-7D, SMW39-6A
Figs 5D – 5I, 6B, 6C, and S8A–S8F	<i>dam1Δ</i>		SYS-2D, SAH5-1C
	<i>dam1Δ rec12Δ</i>		SMWT8-1, SAH4-10C
Figs 6A, 6D, 6E, S8G and S8H	WT		SAH43-1D, SYI32-20B
	<i>dam1Δ</i>		SEN50-25C, SENT59-1
	<i>dam1Δ rec12Δ</i>		SMWT6-1, SMWT7-1

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