Hierarchical Regulation of Centromeric Cohesion Protection by Meikin and Shugoshin during Meiosis I

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The kinetochore is the key apparatus regulating chromosome segregation. Particularly in meiosis, unlike in mitosis, sister kinetochores are captured by microtubules emanating from the same spindle pole (mono-orientation), and sister chromatid cohesion mediated by cohesin is protected at centromeres in the following anaphase. Shugoshin, which localizes to centromeres depending on the phosphorylation of histone H2A by Bub1 kinase, plays a central role in protecting meiotic cohesin Rec8 from separase cleavage. Another key meiotic kinetochore factor, Moa1 (meikin), which was initially characterized as a mono-orientation factor in fission yeast, also regulates cohesin protection. Moa1, which associates stably with CENP-C during meiosis I, recruits Plk1 (polo-like kinase) to the kinetochores and phosphorylates Spc7 (KNL1), inducing the persistent accumulation of Bub1 at kinetochores. The meiotic Bub1 pool ensures robust Sgo1 (shugoshin) localization and cohesin protection at centromeres by cooperating with heterochromatin protein Swi6, which binds and stabilizes Sgo1. Further, molecular genetic analyses reveal a hierarchical regulation of centromeric cohesin protection by meikin and shugoshin during meiosis I.

For the proper transmission of the genetic information, faithful chromosome segregation is essential in all organisms. During the cell cycle, sister chromatid cohesion is established in S phase dependent on the cohesin complex, and is maintained until metaphase. Sister chromatid cohesion at centromeres is essential to establish chromosome biorientation, in which sister kinetochores are captured by spindle microtubules from opposite poles. In the transition from metaphase to anaphase, the anaphase-promoting complex (APC) triggers the degradation of securin, an inhibitory chaperone for separase that cleaves cohesin and removes cohesin along the entire chromosome (Uhlmann et al. 1999; Onn et al. 2008; Peters et al. 2008; Nasmyth and Haering 2009). During meiosis, however, one round of DNA replication is followed by two consecutive nuclear divisions, resulting in the production of four haploid nuclei or gametes. In meiosis I, sister chromatids are captured from the same pole (mono-orientation), whereas homologous chromosomes (homologs) connected by chiasmata are captured by spindle microtubules emanating from the opposite poles (Fig. 1). At the onset of anaphase I, cohesin is cleaved by separase along the arm regions but protected at centromeres until metaphase II (Buonomo et al. 2000; Kitajima et al. 2003a; Tachibana-Konwalski et al. 2010). Thus, mono-orientation and the protection of centromeric cohesin are two hallmarks of the regulation of meiotic chromosome segregation (Moore and Orr-Weaver 1998; Petronczki et al. 2003; Brar and Amon 2008; Watanabe 2012; Duro and Marston 2015).

COHESION PROTECTION

Cohesin complexes are modified in meiosis. Especially, the Rad21(Scc1) subunit is largely replaced by its meiotic counterpart, Rec8 (Klein et al. 1999; Watanabe and Nurse 1999). During anaphase of meiosis I, Rec8 is cleaved only along the chromosome arms by separase, whereas centromeric Rec8 is preserved until meiosis II. If Rec8 is replaced by Rad21 during meiosis, sister chromatid cohesion, but not protection at the centromeres, is restored, leading to the separation of sister chromatids at meiosis I. Therefore, an intrinsic property of the Rec8 subunit absent from Rad21 contributes to centromeric protection at meiosis I (Toth et al. 2000; Yokobayashi et al. 2003). Pericentric heterochromatin plays a crucial role in enriching cohesin complexes and, thereby, strengthens centromeric cohesion in mitosis (Bernard et al. 2001b; Nonaka et al. 2002; Fukagawa et al. 2004). This might be applicable in meiosis, because the localization of the Rec8 complex is reduced from the pericentric regions in heterochromatin mutants (Kitajima et al. 2003b).

A functional screening of fission yeast identified the Rec8 protector as a gene that causes the disjunction of chromosomes, and thus it is toxic during mitotic growth only when co-expressed with Rec8 but not with Rad21 (Kitajima et al. 2004). This gene encodes a meiosis-specific protein named shugoshin (Sgo1), which means “guardian spirit” in Japanese. Sgo1 localizes exclusively at pericentric heterochromatin regions, the site at which

Figure 1. Moa1–Plp1 promotes cohesion protection at centromeres. (A) Schematic image of chromosome segregation during meiosis. (B) The green dot represents an *imr1-GFP*-labeled centromere. In normal meiosis I (MI), sisters are captured by spindles from the same pole, and heterozygous *imr1-GFP* signals move to one side (reductional segregation). The majority of *imr1-GFP* signals in prometaphase II zygote are observed as one dot (cohered) because centromeric cohesion is protected, whereas some cells exhibit two dots (separated) because of the failure of cohesion protection. If mono-orientation is compromised at meiosis I, some sisters would segregate to opposite sides (equational segregation). (C) The chromosome segregation pattern during meiosis I and the splitting of centromeres were counted by observing heterozygous *imr1-GFP* signals in prophase II arrest (by the *mes1-B44* mutation) in the indicated cells. Error bars, SD, $n > 150$ cells, three independent experiments. (D) The frequency of the splitting of centromeres was measured by observing heterozygous *imr1-GFP* signals in prometaphase II (prometa II) arrest (*mes1*) in the indicated cells. WT, wild type. (Modified, with permission, from Miyazaki et al. 2017, © John Wiley & Sons, Inc.)

Rec8 was predicted to play a role in the centromeric protection at meiosis I (Kitajima et al. 2003b). Independent knockout screening in fission yeast and budding yeast also identified the *sgo1/SGO1* gene (Marston et al. 2004; Rabitsch et al. 2004) as a Rec8 protector in meiosis. Remarkably, it turns out that shugoshin shares a hitherto unperceived limited similarity to MEI-S332, a *Drosophila* protein that was previously shown to be required for the persistence of centromeric cohesion during meiosis I (Davis 1971; Kerrebrock et al. 1995; Lee and Orr-Weaver 2001). Analyses in several eukaryotic organisms indicate that meiotic cohesin protection at centromeres is mediated by the centromeric protein shugoshin (Sgo1 in fission yeast). Shugoshin forms a complex with protein phosphatase 2A (PP2A) at the centromeres (Kitajima et al. 2006; Riedel et al. 2006; Tang et al. 2006; Lee et al. 2008; Llano et al. 2008) and antagonizes Rec8 phosphorylation, a prerequisite for cleavage by separase in meiosis I (Ishiguro et al. 2010; Katis et al. 2010).

Bub1, a well-conserved spindle checkpoint kinase, is required to preserve centromeric protection during meiosis I in fission yeast (Bernard et al. 2001a). Indeed, Sgo1 fails to localize at centromeres in *bub1* mutants (Kitajima et al. 2004). A biochemical approach in fission yeast identified serine 121 of histone 2A (H2A) as a Bub1 substrate (Kawashima et al. 2010). Crucially, Sgo1 binds nucleosomes including phosphorylated H2A-S121, accounting for the mechanism of Sgo1 localization at centromeres (Kawashima et al. 2010). Sgo1 localization is also pro-

moted by pericentric heterochromatin protein Swi6 (HP1), which binds directly to Sgo1 (Yamagishi et al. 2008). In mitosis, Bub1 as a complex with Bub3 localizes to kinetochores through its interaction with Spc7 (KNL1) only when Spc7 is phosphorylated by Mph1 (MPS1). Mph1 is a conserved protein kinase required for the spindle assembly checkpoint (SAC) locating at unattached or misaligned kinetochores (Yamagishi et al. 2012; London and Biggins 2014; Musacchio 2015; Sacristan and Kops 2015). Therefore, Bub1 is largely released from kinetochores when chromosomes are aligned during metaphase in mitotic cells. Recent reports reveal that KNL1 is also phosphorylated by PLK1 in humans and *Caenorhabditis elegans* (Espeut et al. 2015; von Schubert et al. 2015), suggesting the existence of a versatile regulation of Bub1 localization.

MONO-ORIENTATION VERSUS BIORIENTATION

The geometric aspect of kinetochores has been long recognized in vertebrates (Östergren 1951). The staining of human interphase nuclei with anticentromere antibodies revealed that the centromere is duplicated and resolved by the end of interphase (Brenner et al. 1981). This physical separation or resolution of sister centromeres would be important for the back-to-back assembly of sister kinetochores, thus facilitating bipolar attachment to microtubules in mitosis. In contrast to mitosis, cytological

analyses of several animal germ cells have shown that sister kinetochores orient side by side and fuse in meiosis I (Goldstein 1981; Moore and Orr-Weaver 1998; Lee et al. 2000; Parra et al. 2004). The molecular mechanism underlying the regulation of kinetochore geometry has been studied recently (Watanabe 2012).

Spo13-MONOPOLIN IN BUDDING YEAST

In budding yeast a set of proteins called monopolin, which are required for mono-orientation, have been identified and extensively studied. Monopolin includes Csm1 (chromosome segregation in meiosis protein 1), Lrs4 (loss of rDNA silencing protein 4), Mam1 (monopolar microtubule attachment during meiosis I protein 1), and CK1, and localizes to centromeres specifically in meiosis I (Toth et al. 2000; Rabitsch et al. 2003). The structural analysis of Csm1–Lrs4 suggests that this complex forms a V shape with two pairs of kinetochore-binding domains that indeed bind kinetochore component Dns1. These results suggest that Csm1–Lrs4 may bring kinetochores together, which favors the clamp model (Corbett et al. 2010; Corbett and Harrison 2012). Evidence suggests that the enrichment of CK1 activity at kinetochores, which depends on the presence of Csm1–Lrs4, might be an ultimate requirement for the establishment of mono-orientation in budding yeast (Petronczki et al. 2006). However, which CK1 substrates are required for mono-orientation and the mechanism by which it occurs remain elusive. Budding yeast Spo13 (sporulation-specific protein 13), another factor that is required for mono-orientation as well as cohesion protection, associates with the Polo-like kinase Cdc5 and acts to recruit or stabilize the monopolin complex at centromeres (Clyne et al. 2003; Katis et al. 2004, 2010; Lee et al. 2004; Monje-Casas et al. 2007). In budding yeast, paired sister centromeres assemble a single kinetochore and bind only one microtubule (Sarangapani et al. 2014). Therefore, monopolin would conjoin two microtubule attachment sites and thereby make them into a one “point centromere.”

Rec8 IS REQUIRED FOR MONO-ORIENTATION IN FISSION YEAST

In fission yeast, homologs of Csm1 and Lrs4 (named Pcs1 and Mde4, respectively) are dispensable for mono-orientation in meiosis I. However, in mitosis, *psc1* or *mde4* mutant cells show merotelic attachment, in which a single kinetochore is attached by microtubules emanating from both spindle poles (Gregan et al. 2007). Indeed, Pcs1–Mde4 recruits condensin, which may act to clamp together adjacent microtubule attachment sites, although the meiosis I–specific mono-orientation function is not conserved (Tada et al. 2011).

The finding that a *rec8* mutation in fission yeast causes equational, rather than reductional, division at meiosis I raises the possibility that cohesin complexes regulate kinetochore orientation. Similarly, plant and worm Rec8 have been shown to play an essential role in establishing

monopolar attachment at meiosis I (Yu and Dawe 2000; Chelysheva et al. 2005; Severson et al. 2009). In fission yeast, a mitotic cohesin complex that includes Rad21 accumulates preferentially at the pericentromeric heterochromatin, whereas the meiotic Rec8–cohesin complex accumulates additionally at the core centromere, the region where the kinetochore assembles. When Rec8 is removed and replaced by Rad21 in meiosis I, the Rad21–cohesin complex accumulates at the pericentromeric region, but much less at the central core region, causing equational rather than reductional division at meiosis I. When Rec8 is inactivated specifically only at the core centromere, but its other functions are preserved, kinetochores become bioriented at meiosis I, proving the essential role of Rec8 at the central core region for mono-orientation (Yokobayashi and Watanabe 2005).

Based on the above evidence, it has been proposed that physical attachment of sister chromatids or cohesion at the centromeric core conjoins the two kinetochore domains at meiosis I, whereas the core regions open to opposite sides when not establishing this cohesion at mitosis and meiosis II. A direct observation of cohesion at the core centromere in fission yeast was enabled by popping out this DNA region from the neighboring chromosomal domains during prophase I, which is before the attachment of kinetochores to spindle microtubules (Sakuno et al. 2009). This analysis revealed that cohesion at the core centromere is indeed established and maintained, particularly during meiosis I, whereas this cohesion is lost in *rec8Δ* or *moa1Δ* (see below), which is defective in mono-orientation. Importantly, cohesion at the central core region is not detected during mitosis or meiosis II in wild-type cells, whereas cohesion at the pericentromeric region is intact. Finally, when a proteinous artificial tether is introduced at the core centromere, monopolar attachment is restored in meiotic *rec8Δ* cells and even in normal mitotic cells. These results imply that mono-orientation of kinetochores is promoted ultimately by conjoining DNA duplexes underlying the kinetochores rather than the action of a kinetochore protein itself (Sakuno et al. 2009).

CONSERVED MEIOTIC KINETOCHORE FACTOR MEIKIN (Moa1, Spo13, MEIKIN)

Genetic screening to search for factors that regulate mono-orientation has identified a meiosis-specific kinetochore protein, Moa1 (monopolar attachment) in fission yeast (Yokobayashi and Watanabe 2005). Moa1 interacts with the conserved kinetochore protein Cnp1 (CENP-C homolog) and localizes exclusively at the central core of the centromere from prophase I to metaphase I but disappears in anaphase I (Tanaka et al. 2009). Moa1 also interacts with Rec8 and plays a role in establishing cohesion at the core centromere regions (mono-orientation) and some cohesion protection in pericentric regions. Taking advantage of the knowledge that Moa1 binds to conserved kinetochore protein CENP-C (Cnp3), two-hybrid screening using CENP-C as bait identified a meiosis-specific kinetochore protein MEIKIN in mice (Kim et al. 2015). Although there is no significant sequence homology

between MEIKIN and Moa1, significant biochemical and functional similarities were identified between these two factors. Both MEIKIN and Moa1 recruit polo-like kinase (PLK) to kinetochores and the kinase activity of PLK is crucial for mono-orientation and cohesion protection (also see below). It turned out that these meiotic functions are reminiscent of those of budding yeast Spo13. Thus, the conserved meiosis-specific kinetochore regulator, meikin (MEIKIN in vertebrates; Moa1 in fission yeast; Spo13 in budding yeast) and its associated PLK play a crucial role in promoting mono-orientation and, at least partly, cohesion protection.

FISSION YEAST MEIKIN AFFECTS COHESION PROTECTION

Although fission yeast Moa1 was identified as a mono-orientation factor, a role in cohesion protection was also implicated (Yokobayashi and Watanabe 2005). Indeed, in *moa1Δ* cells, although a small population of cells undergo equational segregation at meiosis I (because of defects in mono-orientation), the majority undergo reductional segregation because of the presence of chiasmata and tension exerted across homologs (Fig. 1B). Strikingly, 22% of these “reductional” *moa1Δ* cells showed the separation of the GFP-marked centromere (*imr1*-GFP) in prometaphase II (Fig. 1B). This separation value is significantly higher than in wild-type cells (<5%), although lower than in *sgo1Δ* cells (50%), in which cohesion protection is completely abolished (Fig. 1B). These results suggest that *moa1Δ* cells show partial defects in cohesion protection during reductional division at meiosis I.

Moa1 associates with Plo1 (PLK1 homolog), and the centromeric Plo1 is required for reductional segregation at meiosis I (Kim et al. 2015). Here we examined whether Plo1 is responsible for defects in cohesion protection at prometaphase II, as is observed in *moa1Δ* cells. For this purpose, we first analyzed the *moa1-T101A* mutant, in which Moa1 localizes at kinetochores but fails to recruit Plo1 (Kim et al. 2015). Indeed, the separation of *imr1*-GFP in prometaphase II was observed in *moa1-T101A* cells to the same extent as in *moa1Δ* cells (Fig. 1C). These results indicate that Plo1 recruited to kinetochores by Moa1 is responsible for the protection of centromeric cohesion during anaphase I.

Bub1 REGULATES COHESION PROTECTION

Bub1, a well-conserved spindle checkpoint kinase, is required to preserve centromeric protection during meiosis I (Bernard et al. 2001a). Indeed, Sgo1 fails to localize at centromeres in *bub1* mutants (Kitajima et al. 2004; Fernius and Hardwick 2007). A biochemical approach in fission yeast identified serine 121 of histone 2A (H2A) as a Bub1 substrate (Kawashima et al. 2010). Crucially, Sgo1 binds nucleosomes, including phosphorylated H2A-S121, accounting for the mechanism of Sgo1 localization at centromeres (Kawashima et al. 2010). Sgo1 localization is also promoted by the pericentric heterochromatin protein Swi6

(HP1), which binds directly to Sgo1 (Yamagishi et al. 2008). In mitosis, Bub1 as a complex with Bub3 localizes to kinetochores through its interaction with Spc7 (KNL1) only when Spc7 is phosphorylated by Mph1 (MPS1) at unattached or misaligned kinetochores. Therefore, Bub1 is largely released from kinetochores when chromosomes are aligned during metaphase in mitotic cells.

Moa1–Plo1 REGULATES MEIOTIC Bub1 LOCALIZATION

KNL1 is also phosphorylated by PLK1 in humans and *C. elegans* (Espeut et al. 2015; von Schubert et al. 2015), suggesting the existence of a versatile regulation of Bub1 localization. It is reasonable to speculate that Moa1–Plo1, which is required for cohesion protection, may play a role in Bub1 enrichment during meiosis I. In fission yeast mitosis, Bub1 is enriched at kinetochores only when Mph1 (MPS1 homolog) accumulates at unattached kinetochores and phosphorylates the MELT repeats of the kinetochore protein Spc7 (KNL1) (Fig. 2A; Yamagishi et al. 2012). Curiously, however, meiotic Bub1 signals are retained at kinetochores throughout metaphase I until anaphase I in wild-type cells, and the signals persist even in *mph1Δ* cells (Fig. 2B). Instead, centromeric Bub1 signals decline around late metaphase I in *moa1Δ* cells and largely disappear in *mph1Δ moa1Δ* cells (Fig. 2B). Thus, Moa1-associated Plo1 may play a key role in the accumulation of Bub1 at kinetochores in a redundant capacity with Mph1. The time-lapse live cell imaging of Bub1-GFP indicates that the duration of metaphase I is shortened in *moa1Δ*

Figure 2. Bub1 is dispersed in *mph1Δ moa1Δ* cells as in *spc7-12A* cells. (A,B) The indicated cells expressing Bub1-GFP and mCherry-Atb2 (tubulin) were observed by time-lapse imaging during mitosis (A) or meiosis I (B) at 1-min intervals. A representative example of indicated cell is shown. The red arrowhead denotes the onset of spindle elongation at anaphase. WT, wild type. (Modified, with permission, from Miyazaki et al. 2017, © John Wiley & Sons, Inc.)

mph1Δ cells (Fig. 2B), suggesting that Moa1–Plo1, like Mph1, has a function in SAC activation in meiotic cells.

LIKE Mph1, Moa1–Plo1 PHOSPHORYLATES Spc7

Is Moa1–Plo1 indeed responsible for the phosphorylation of MELT repeats in Spc7? Bacterially purified Plo1, like Mph1, phosphorylates the amino-terminal domain of Spc7 (Spc7-N), which contains MELT repeats, but not other domains that lack MELT repeats (Fig. 3A). Consistently, centromeric Bub1 signals are largely dispersed in *spc7-12A* cells as is seen in *mph1Δ moa1Δ* cells during meiosis I (Fig. 2B). It is also shown that centromere-tethering Plo1 enables Bub1 recruitment to kinetochores even in mitotic interphase, and this is also the case in *mph1Δ* cells (Miyazaki et al. 2017). These results strongly support the notion that meiosis-specific Bub1 recruitment to kinetochores relies on the phosphorylation of the MELT repeats on Spc7, a process mediated by Moa1–Plo1 and Mph1.

KINETOCHORE-BOUND Bub1 AND CENTROMERIC HETEROCHROMATIN REDUNDANTLY SUSTAIN Sgo1 LOCALIZATION

Notably, centromeric Sgo1 signals show an ~70% reduction in *spc7-12A* cells, in which Bub1 is largely dispersed from kinetochores, whereas few defects in cohesion protection are observed in *spc7-12A* cells (Fig. 4A). Moreover, the expression of the Bub1 kinase catalytic

domain, which cannot bind kinetochores, allows Sgo1 localization at centromeres depending on the heterochromatin protein Swi6 (Kawashima et al. 2010). Therefore, the residual Sgo1 localization (~30%; yet functional) in *spc7-12A* cells might be produced by centromeric Swi6 together with dispersed Bub1 kinase activity, which would mildly phosphorylate histone H2A along the whole chromosomes including centromeres (Kawashima et al. 2010). Indeed, the cohesion protection defect in *spc7-12A* cells is much enhanced by introducing the *sgo1-VE* mutation, which renders Sgo1 unable to interact with Swi6 (Fig. 4A; Yamagishi et al. 2008). Thus, Bub1 enrichment mediated by Mph1 and Moa1–Plo1 (Spc7 phosphorylation) plays a crucial role in Sgo1 localization and cohesion protection in a redundant capacity with centromeric heterochromatin (Fig. 4B).

Moa1–Plo1 MAY ENHANCE Sgo1 FUNCTION IN ADDITION TO ITS LOCALIZATION

A remarkable observation is that protection defects are prominent in *moa1Δ* cells but not in *spc7-12A* cells (Fig. 4C), whereas the opposite is true for Bub1 reduction (Fig. 2B). Therefore, in addition to the enhancement of Bub1 localization, Moa1 may play another key role in cohesion protection. This role of Moa1 might differ from the heterochromatin pathway because *moa1Δ* and *sgo1-VE* show additive defects in cohesion protection (Fig. 4C). Given that *moa1Δ* shows no additive defects with *sgo1Δ* (Fig. 4C), it is reasonable to speculate that Moa1 may somehow enhance Sgo1 function directly as well as by its localization (Fig. 4B).

CONCLUSION

The mono-orientation of sister kinetochores and protection of centromeric cohesion are two hallmarks of the regulation of meiotic chromosome segregation that are widely conserved among eukaryotic organisms. Meikin is a recently emerging protein family that may regulate both mono-orientation and cohesion protection. Although fission yeast Moa1 was initially identified as a mono-orientation factor, it regulates cohesion protection similarly to mouse MEIKIN and budding yeast Spo13. Especially, in fission yeast, Moa1–Plo1 together with Mph1 play a crucial role in the enrichment of Bub1 at kinetochores throughout meiosis I. This Bub1 pool ensures robust Sgo1 localization and cohesion protection at centromeres by cooperating with heterochromatin protein Swi6, which binds and stabilizes Sgo1. Although the meiosis-specific Bub1–Sgo1 enrichment mechanism is conserved in mouse (Miyazaki et al. 2017), the contribution of heterochromatin to Sgo1 stabilization is not yet proven in mammalian meiosis. In summary, accumulating evidence indicates that not only shugoshin but also meikin contribute to the meiosis I–specific cohesion protection mechanism. Previous studies suggest that the ectopic or physiological localization of shugoshin at centromeres in meiosis II cannot protect cohesin from separase cleavage at the onset of

Figure 3. Spc7 phosphorylation at MELT sequence by Plo1 and Mph1 promotes Spc7 and Bub1/Bub3 binding. (A) Schematic depiction of Spc7 fragments used in the in vitro phosphorylation assay. (B) Recombinant GST-fused Spc7 fragments were incubated with recombinant GST-Plo1 (left) or GST-Mph1ΔN (kinase domain only) (right) in the presence of [γ - 32 P] ATP. The incorporation of radioactive phosphate groups was visualized by autoradiography (32 P) and compared with protein levels (Coomassie brilliant blue [CBB]). The asterisk indicates Mph1ΔN autophosphorylation. (Modified, with permission, from Miyazaki et al. 2017, © John Wiley & Sons, Inc.)

Figure 4. Moa1–Plo1 enhances Sgo1 function in addition to its localization. (A,C) The frequency of centromere splitting was measured by observing heterozygous *imr1*-GFP signals in prometaphase II (prometa II) arrest (*mes1*) in the indicated cells. Error bars, SD from three independent experiments. Total cells: $n > 140$. n.s.; not significant; **, $P < 0.01$; ***, $P < 0.005$; ****, $P < 0.001$; one-way ANOVA with Bonferroni's multiple comparisons test. (B) Schematic depiction of Moa1–Plo1 function in the regulation of cohesion protection at centromeres during meiosis I. Moa1–Plo1 facilitates Sgo1 localization at centromeres through Spc7 phosphorylation in a redundant capacity with Mph1. Swi6 stabilizes Sgo1 localization. Moa1–Plo1 also regulates another pathway required for cohesion protection in meiosis I. (Modified, with permission, from Miyazaki et al. 2017, © John Wiley & Sons, Inc.)

anaphase II (Moore et al. 1998; Rabitsch et al. 2004). This could be accounted for at least in part, by the fact that meikin is absent from kinetochores in meiosis II.

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