Maize meiotic mutants with improper or nonhomologous synapsis due to problems in pairing or synaptonemal complex formation

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Supplementary material

Figure S1. LE/AE behavior of wild type A344 line

DAPI staining of chromosomes (blue in merged images) and immunostaining with antibodies against ASY1 (green) and AFD (red) are shown for leptotene (A-D), zygotene (E-H), and pachytene (I-L). AFD1 staining is not shown for zygotene; rather, FISH with a telomere probe showing bouquet is shown in G (red in H). Note that ASY1 signal in pachytene (J) is masked on the synapsed chromosomes and only apparent on an unsynapsed loop (lower right). Bar = 5 μm.

Figure S2. Amino acid sequence of ZYP1 and western blot analysis

(A) Diagram of ZYP1 protein with marked positions of coiled-coil domains and a region used to raise ZYP1 antibody. The arrows indicate the corresponding positions and orientations of primers used in cloning. (B) Amino acid sequence of maize ZYP1 protein. (C) Western blot analysis of ZYP1 in wild-type tissues.

Figure S3. Axial and central element behavior in *asynaptic1*

Immunostaining with antibodies against ASY1 or ZYP1 (green in merged images) and AFD1 (red) and DAPI staining of chromatin (blue) demonstrates that AE and CE behavior is similar to wild type.

(A-D) In early zygotene, both ASY1and AFD1 are loaded onto chromosome cores, and ASY1 is brighter than AFD1.

(E-H) In zygotene, ASY1 and AFD1 are equally bright and appear as green on unsynapsed chromosomes and red in synapsed regions as the ASY1 signal is masked at synapsis.

(I-L) In zygotene, AFD1 is fully loaded onto chromosomes, but ZYP1 is installed only on synapsed regions. Bar = 5 μm.

Figure S4. Retarded synapsis in *mei*N2415*

Immunostaining of zygotene (A-D) and pachytene (E-H) nuclei with AFD1 (red in merged images) and ZYP1 (green) and DAPI stained chromosomes (blue) show retarded synapsis at zygotene and almost complete synapsis at late pachytene.

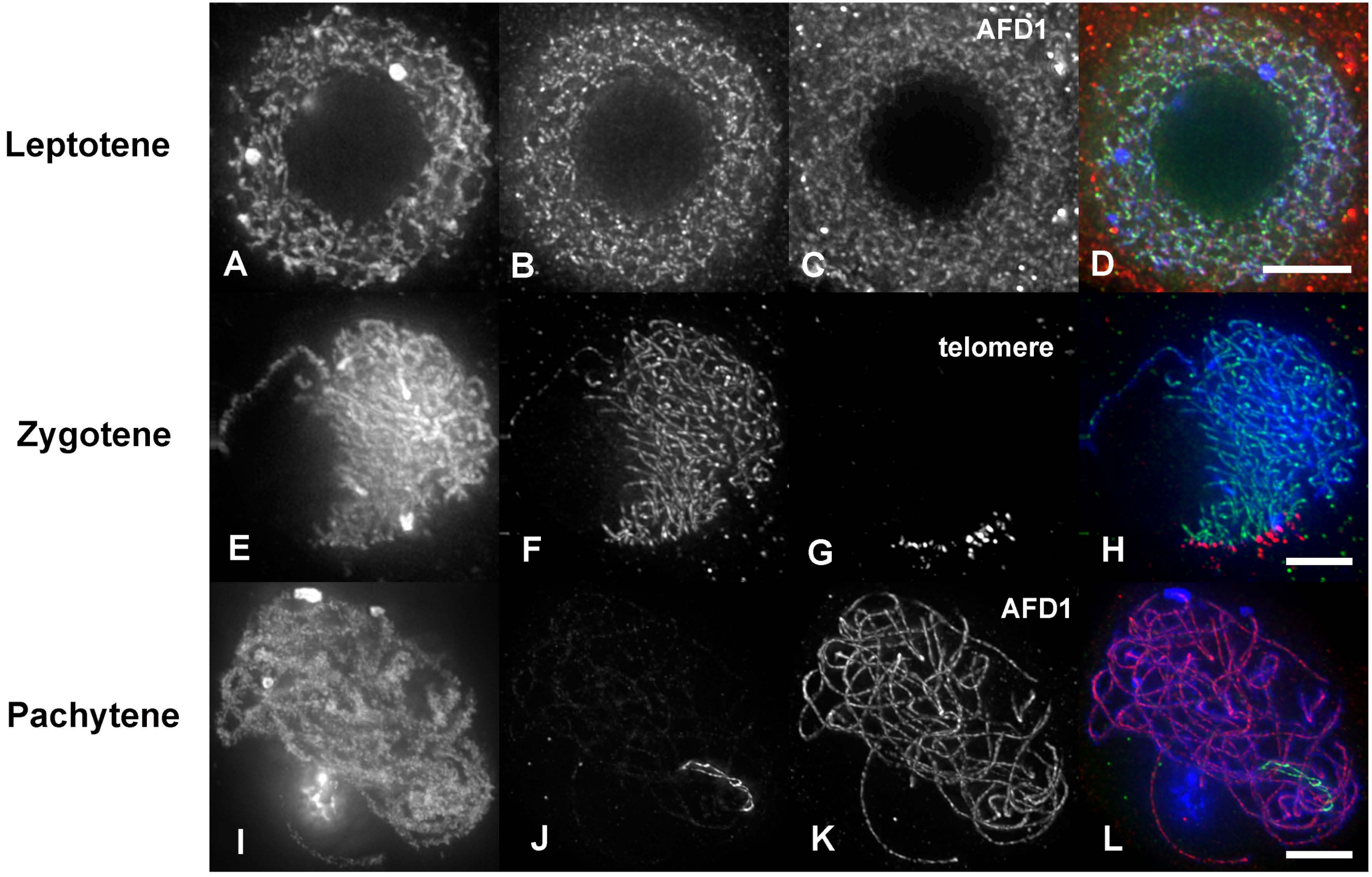
(A-D) Immunostaining of zygotene nucleus shows that ZYP1 is loaded onto synapsed regions. In desynaptic bubbles where chromosome axes are far apart (arrowhead), axes are stained only by AFD1 but in synapsed regions the chromosomes are stained with both AFD1 and ZYP1 and in the merged image appear yellow.

(E-H) At pachytene, synapsis is complete. In the merged image (H), the axes are stained in yellow due to the close association of ZYP1 (green in F) and AFD1 (red in G). Bar = 5 μm.

Figure S5. Less condensed heterochromatin in *mtm99-25*

DAPI staining of pachytene chromosomes in wilde-type (A) and *mtm99-25* (B). Compare the sizes of heterochromatic knobs in wild-type (arrow) and in *mtm99-25* (double-arrow). Bar = 5 μm.

Fig S1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Merged
Leptotene
Zygotene
Pachytene
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telomere
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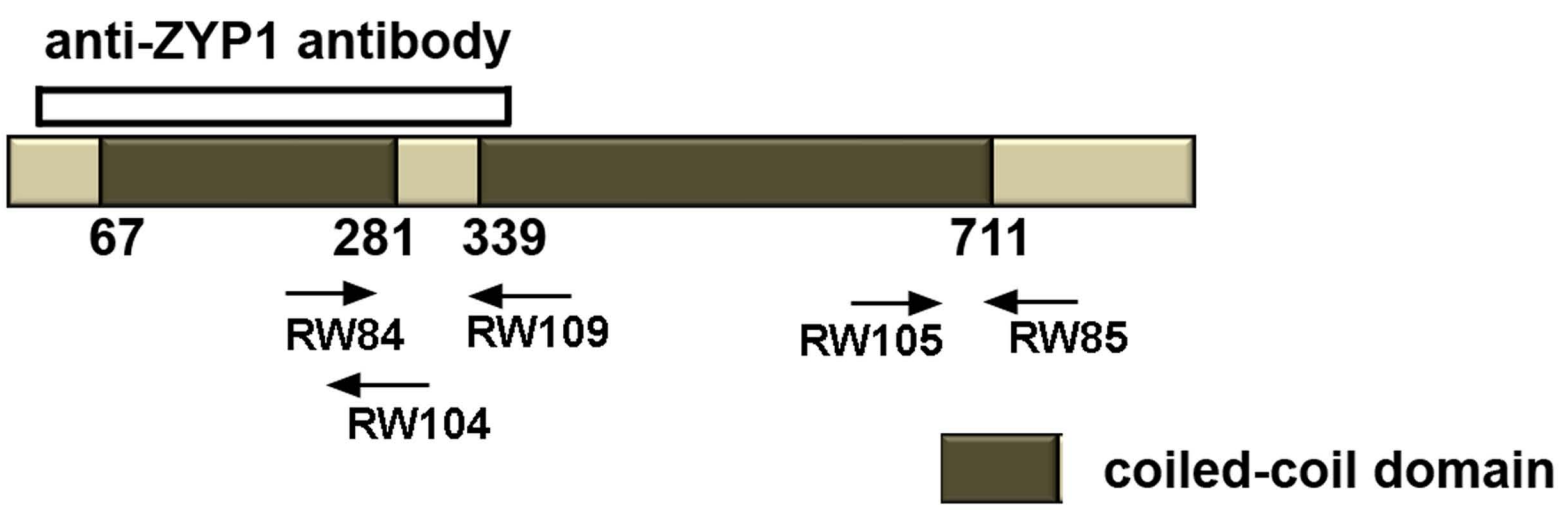
A
B
C
D
E
F
G
H
I
J
K
L

Fig S2

A



B

Zm ZYP1 amino acid sequence

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ETNRKDVNLPGILQSPISNMLRKVEKVSQDIPKHRKVTHHEYEVETANGR
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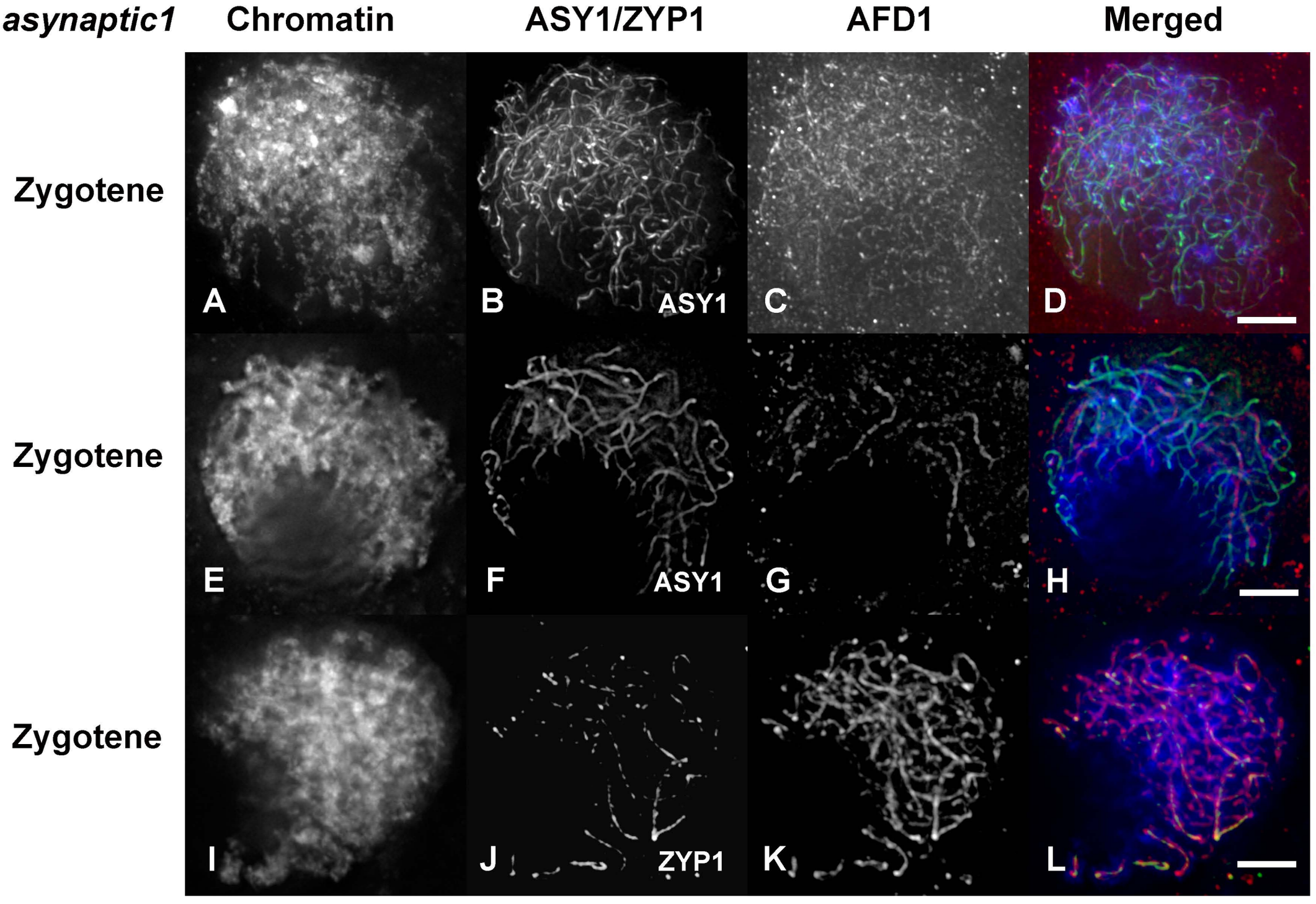
C

Fig S3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ASY1/ZYP1
AFD1
Merged
Zygotene
Zygotene
Zygotene
A
B
ASY1
C
D
E
F
ASY1
G
H
I
J
ZYP1
K
L

Fig S4

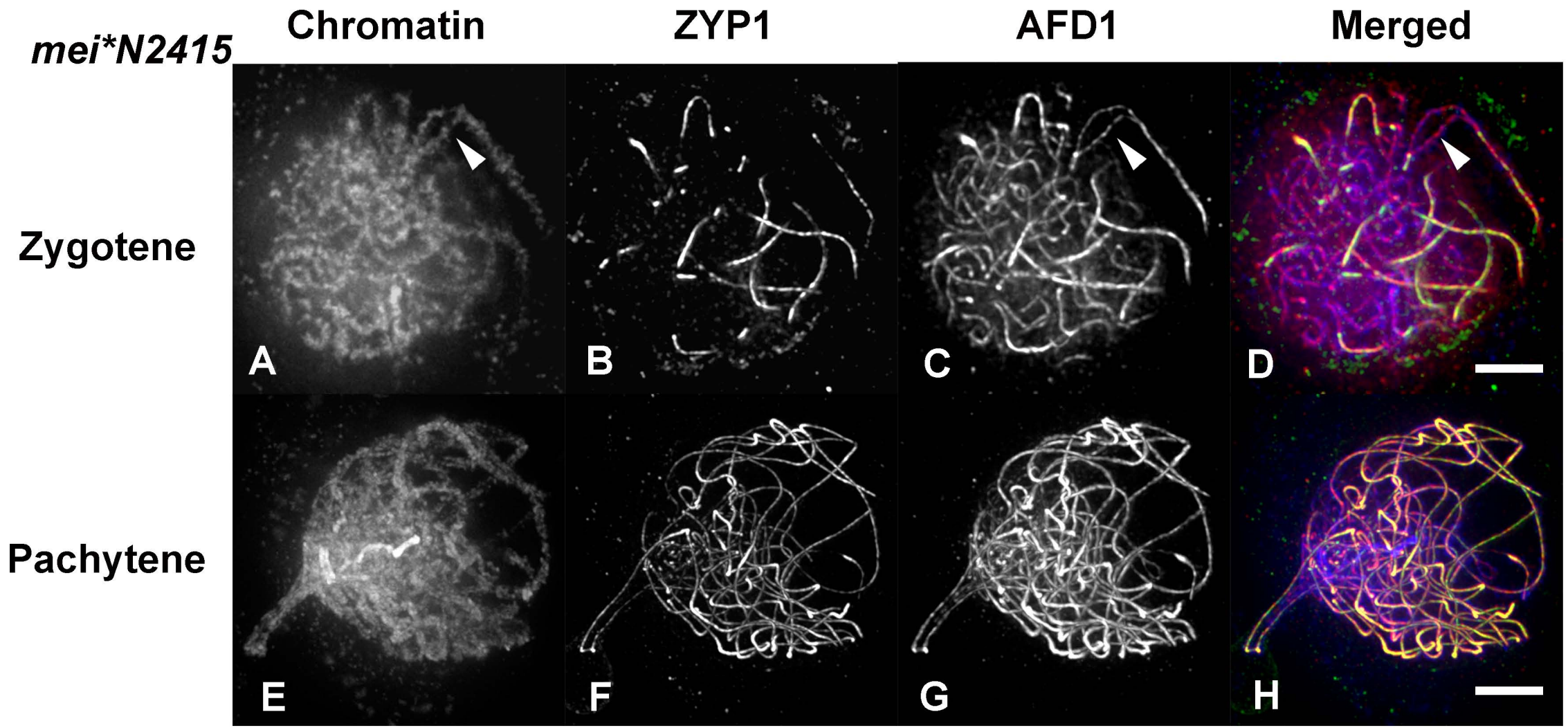
mei*N2415
Chromatin
ZYP1
AFD1
Merged
Zygotene
Pachytene
A
B
C
D
E
F
G
H

Fig S5