Sister chromatid separation and monopolar spindle organization in the first meiosis as two mechanisms of unreduced gametes formation in wheat-rye hybrids

Plant Reproduction

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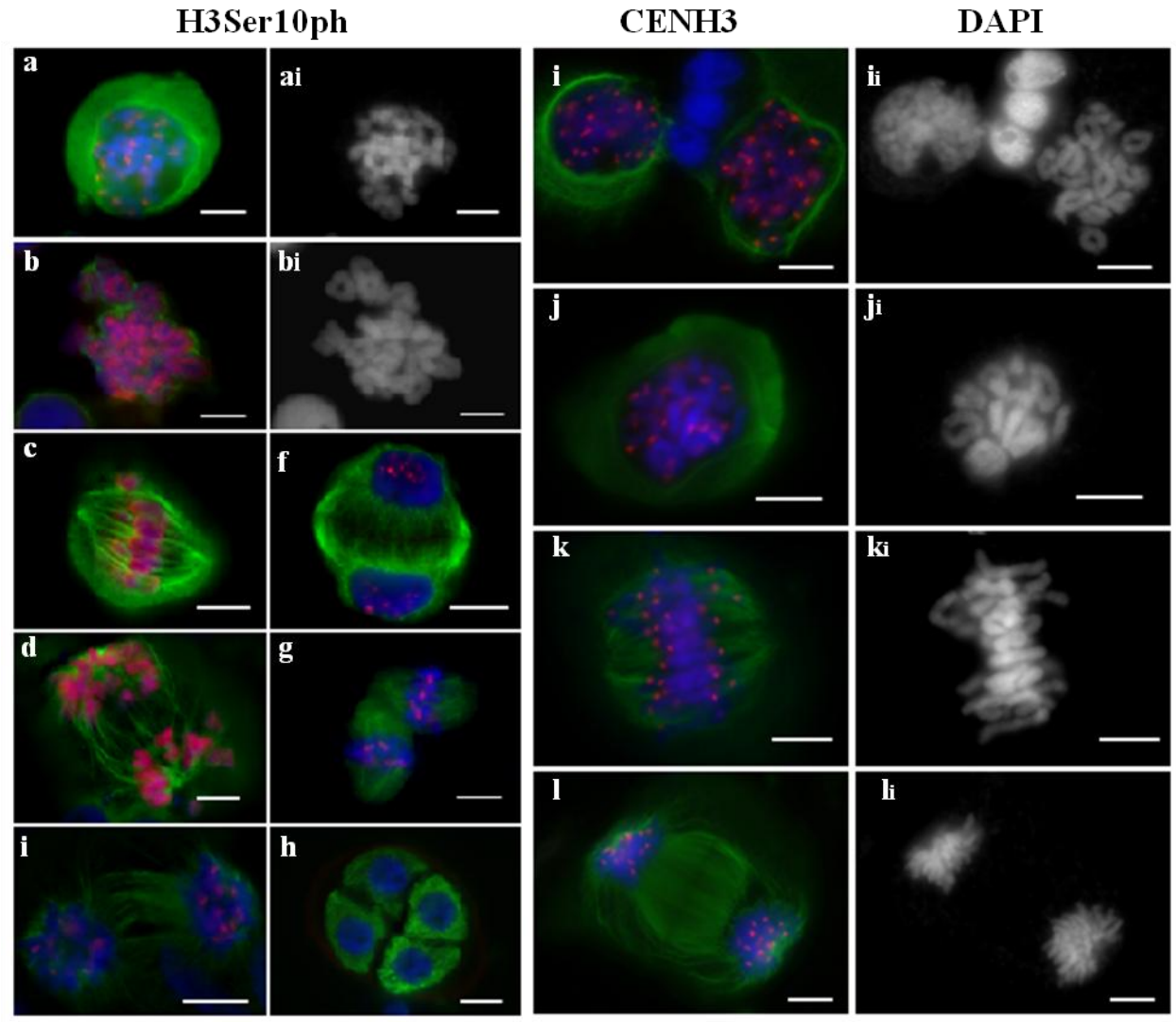

Fig.1 Immunolocalization of H3Ser10ph (**a – h**) or CENH3 (**i – l**) and α-tubulin during meiosis in *Triticum aestivum* L. **a** Diplotene. $\mathbf{a_i}$ The same cell as **a**, DAPI counterstained. **b** Diakinesis. $\mathbf{b_i}$ the same cell as **b**, DAPI counterstained. **c** Metaphase I. **d** Anaphase I. **i** Late anaphase I. **f** Telophase I. **g** Metaphase II. **h** Telophase II. **i** Pachytene and diakinesis. $\mathbf{i_i}$ The same cell as **i**, DAPI counterstained. **j** Diakinesis. $\mathbf{j_i}$The same cell as **i**, DAPI counterstained. **k** Metaphase I. **k l** Telophase I. DNA *blue*, H3Ser10ph and CENH3 labeling *red*, α-tubulin labeling *green*. Bars represent 10 μm

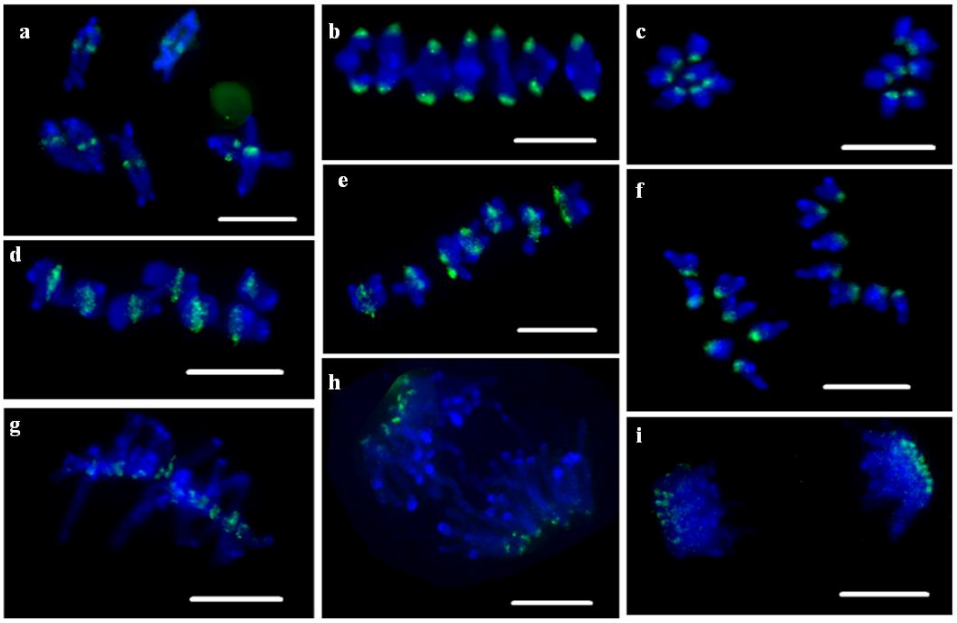

Fig.2 *In situ* hybridization using the centromeric pAet6-09 probe (*green*) to meiotic (**a-f**) and mitotic (**g-i**) chromosomal spreads of rye. **a** Single dense hybridization sites at diplotene. **b** Metaphase I. **c** Anaphase I**. d** Metaphase II, stretched diffuse hybridization sites. **e** Late metaphase II, pAet6-09 signals appear as dense twin spots indicating sister centromere separation. **f** Anaphase II, single dense hybridization sites. **g** Metaphase, stretched diffuse pAet6-09 hybridization sites. Telophase (**h**) and anaphase (**i**), single dense spots of pAet6-09 hybridization sites. Chromatin was counterstained with DAPI (*blue*). Bars represent 10 μm

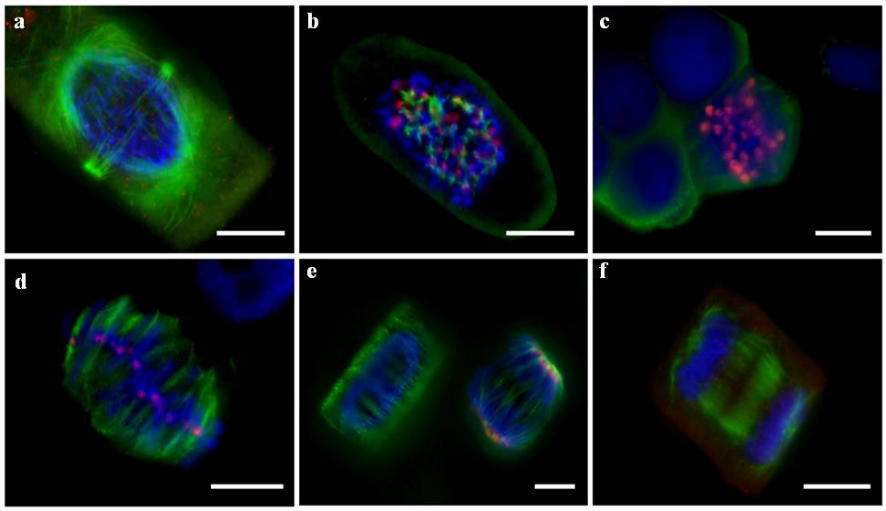

Fig.3 Immunolocalization of H3Ser10ph (*red*) and α-tubulin (*green*) during mitosis in *Triticum aestivum* L. **a** Preprophase band. **b** Prophase. **c** Prometaphase. **d** Metaphase. **e** Anaphase. **f** Telophase. Chromatin was counterstained with DAPI (*blue*). Bars represent 10 μm

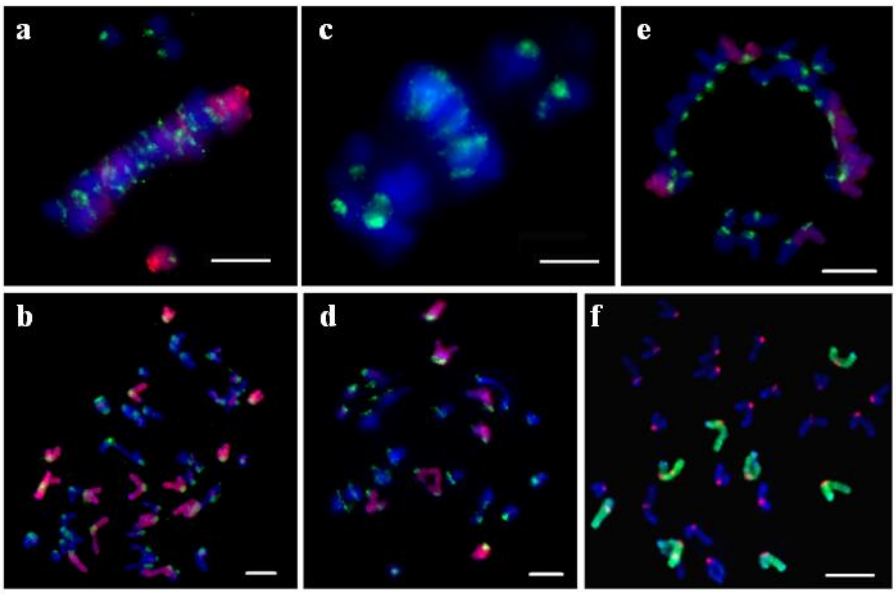

Fig.4 *In situ* hybridization using the centromeric pAet6-09 probe (*green*) and rye genomic DNA probe (*red*) to meiotic chromosomal spreads of wheat-rye amphihaploids. Equational division of chromosomes (**a, b**). **a** Metaphase I, all chromosomes at equatorial plate have stretched diffuse hybridization sites. **b** Anaphase I, separation of sister chromatids**.** Equational+reductional division of chromosomes (**c, d**). **c** Metaphase I, chromosomes at the equatorial plate with stretched diffuse hybridization sites, chromosomes outside of the equatorial plate with single dense hybridization sites. **d** Early anaphase I, sister centromere separation at the equatorial plate, appearance of two local hybridization sites. **e** Chromosomes form a ring with their centromeres facing the center, and arms oriented outwards. **f** Reductional division of chromosomes. The centromeric pAet6-09 probe (*red*) and rye genomic DNA probe (*green*). Metaphase I-anaphase I, chromosomes with single dense hybridization sites. Chromatin was counterstained with DAPI (*blue*). Bars represent 10 μm