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Inter- and intragenomic spontaneous translocations in multigeneric hybrids of *Triticeae* tribe

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SUMMARY – Interspecific hybridisation has been used in order to transfer interesting agronomic characteristics from wild species to wheat. This event could induce structural rearrangements such as spontaneous translocations. We performed *in situ* hybridisation on mitotic and meiotic chromosome spreads of F1 bread wheat × tritordeum hybrids with genomic DNA from *Hordeum chilense* and the rDNA sequence pTa71, as probes. All PMCs at metaphase I indicated a spontaneous intergenomic translocation wheat/H^{ch} and one wheat univalent with two rDNA *loci*. This intragenomic translocation (1B/6B) was also present in all mitotic cells of the same plant. The occurrence of wheat/ H^{ch} translocations might be useful for the introgression of *H. chilense* chromatin in wheat.

Introduction

The *Triticeae* tribe incorporates a huge genetic diversity that could be exploited by plant breeders in order to produce new allopolyploid species by interspecific hybridisation. This strategy allows the transfer of useful agronomic characteristics from wild species to wheat. Some authors considered the allopolyploidy a revolutionary rather than an evolutive mechanism (Ozkan *et al.*, 2001). Hybridisation followed by polyploidy can induce structural rearrangements at the chromosome level (Leitch and Bennett, 1997), DNA sequence (Song *et al.*, 1995), gene expression (Comai *et al.*, 2000) or elimination of highly repetitive DNA sequences (Ozkan *et al.*, 2001). Spontaneous translocations could be a consequence of chromosome rearrangements involved in the production of multigeneric hybrids. Several hybrids involving bread wheat and tritordeum have been obtained at CGB-UTAD (Lima-Brito *et al.*, 2006). We aim to discriminate the parental genomes and to analyse the chromosome pairing behaviour of those hybrids by using *in situ* hybridisation.

Material and methods

Fifteen F1 multigeneric hybrid plants ($AABBDH^{ch}$, 2n = 42) were obtained at CGB-UTAD from crosses between the bread wheat 'Barbela' (AABBDD; 2n = 42) with tritordeum ($H^{ch}H^{ch}AABB$; 2n = 42). Root-tips and anthers containing pollen mother cells (PMCs) at metaphase I were fixed on ethanol:acetic acid (3:1) for chromosome spreads preparations, following Lima-Brito *et al.* (1996). *In situ* hybridisation was performed on mitotic and PMC chromosome spreads with genomic DNA from *H. chilense* and the rDNA sequence pTa71 (Gerlach and Bedbrook, 1979), as probes, both labelled by nick translation, with biotin or digoxigenin (Roche). For blocking, we used sheared genomic DNA from durum wheat. Detection was performed using streptavidin-FITC (Vector Laboratories) and anti-digoxigenin-rhodamine (Roche) antibodies.

Results and discussion

In situ hybridisation was successfully used for discrimination of parental genomes and to detect chromosome rearrangements in mitotic and meiotic metaphase cells of F1 multigeneric hybrid plants (Fig. 1). A 1B/6B intragenomic translocation was detected at mitosis (Fig. 1a) and in all PMCs at metaphase I (MI) as univalent in heterozygotic condition (Figs 1b and 1c).

No hybridisation signal was observed when we reprobed the same chromosome spreads with genomic DNA from *Triticum tauschii* (DD) (data not shown). Thus, the wheat chromatin involved in the

intergenomic rearrangement is from A or B genomes, and it was classified as wheat/H^{ch} (Figs 1b and 1c). Most of the PMCs (85.7%) showed the intergenomic translocation as univalent and in heterozygotic condition (Fig. 1c) and the remaining presented it in homozygous condition sometimes involved in open bivalents (Fig. 1b). The spontaneous translocations detected here appear to result from Robertsonian translocations (Fig. 1). A high frequency of wheat and H^{ch} univalents was previously reported for these hybrids (Lima-Brito *et al*., 2006) and it could induce centric-breakage-fusions of homoeologous chromosomes, producing intergenomic translocations. The occurrence of the intragenomic translocation could also be explained by the mechanism of centric-breakage of the satellite chromosomes 1B and 6B and subsequent fusion of their short arms bearing the two major rDNA *loci* of B wheat genome, resulting in the 1BS/6BS rearrangement. Our results suggest that newly formed multigeneric hybrids experiment rapid structural rearrangements (Ozkan *et al.*, 2001) and constitute useful material for the production of translocation lines. The occurrence of intergenomic translocations in the multigeneric hybrids analysed here could ensure the transfer of useful agronomic characteristics from *H. chilense* to wheat by recombination.

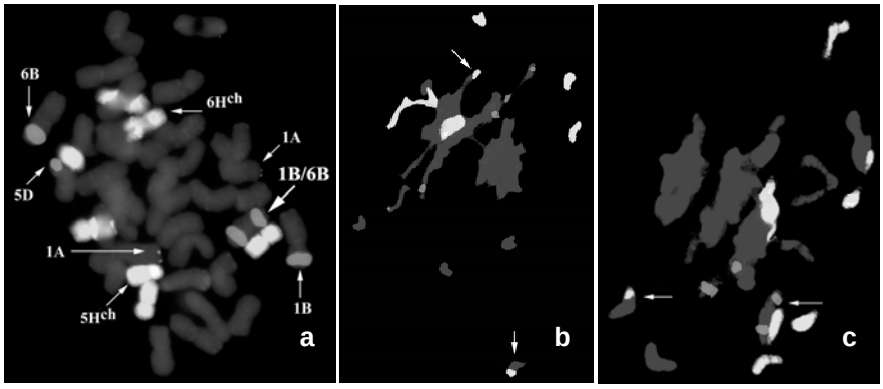

Fig. 1. Mitotic metaphase cell (a) and two PMCs at MI (b-c) of 'Barbela' × tritordeum F1 hybrids probed with genomic DNA of *H. chilense* (white) and pTa71 (light grey). Wheat chromosomes were counterstained with DAPI (dark grey). a) pTa71 identified all satellite chromosomes and the wheat chromosome with two rDNA *loci* (1B/6B; arrow); b) Wheat/H^{ch} translocation in homozygous state (arrows) and c) in heterozygous state (left arrow) as well as the 1B/6B translocation (right arrow).

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