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Production and identification of new structural chromosome mutations in barley (Hordeum vulgare L.)

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Abstract A total of 52 reciprocal translocations and 9 pericentric inversions were induced and identified in both standard and cytologically marked barley karyotypes using gamma-rays as the clastogenic agent. An analysis based upon Giemsa N-banding patterns and arm length measurements of the reconstructed chromosomes enabled a rather precise cytological localization of intra- and interchange breakpoints. This analysis was significantly facilitated and improved, especially for the identification of pericentric inversions, when the reconstructed karyotype T-1586 was used as starting material. The majority, if not all, of the aberration breakpoints proved to be localized in interband regions or in medial and terminal parts of the chromosomes, i.e., in regions which are deficient in constitutive heterochromatin. A great number of the structural mutations produced in this study contain specific cytological markers covering nearly all of the chromosomes of barley karyotype. This material might be of considerable interest in solving various problems of barley cytogenetics and chromosome engineering and especially in constructing a physical map of barley genome.

Key words Reciprocal translocations · Pericentric inversions · Giemsa N-banding · Hordeum vulgare

Introduction

Chromosomal rearrangements are one of the most frequently produced class of mutations (gene or point mutations are the other important class of genetic changes) that result from the action of both physical and chemical mutagenic agents. The different aspects of chromosome reconstruction have been thoroughly dis-
mutants was the metaphase analysis of the Feulgen-stained root-tip metaphase chromosomes of M1 plants that showed well-expressed partial sterility. For this purpose, the examination of Feulgen squashes of the root tips of about 4–5 seeds of such M1 plants proved to be quite adequate. About 30 seeds of each M1 plant showing karyotype reconstructions of interest were grown in a glasshouse or in the field and the plant allowed to self-fertilize; homozygous structural mutations were then isolated among the fertile M2 plants.

The location of intra- and interchange breakpoints along the chromosomes was established using both arm ratio measurements of Feulgen-stained reconstructed chromosomes and their Giemsa banding patterns (up to ten samples of each chromosome type, the number being dependent on the position of the breakpoints). Further, the newly produced structural mutants were test-crossed onto the respective original lines ('Freya' or T-1586) or, when necessary, onto the suitable tester set of translocations, and the meiotic metaphase I of F1 plants was examined to specify the reconstruction of the karyotype as a whole.

All data concerning both conventional and Giemsa banding cytological techniques have been given in previous paper (Gecheff 1978, 1989).

Results

Figure 1 shows a schematic representation of the chromosomal localization of the breakpoints of gamma-induced pericentric inversions and reciprocal translocations in standard variety 'Freya'. It was observed that only about 11% of the partially sterile M1 plants of this variety showed some alterations in the morphology of their root-tip metaphase chromosomes. Obviously, partial sterility observed in the first generation may be due to reasons other than the induction of heterozygous chromosome mutations (cf. KüNZel et al. 1984). The low selective capacity of the karyogramme analysis applied in this case might also be due to the fact that a great portion of the induced translocations have resulted in the exchange of approximately equal chromosome segments. This type of interchange can be easily identified using a traditional approach (Hagberg 1960). However, since this study was aimed at the production of structural mutants containing specific cytological markers in different chromosomes, the technique used here proved to be the most suitable for the purposes of the investigation.

The identification of induced chromosomal rearrangements was significantly facilitated when the cytologically marked karyotype T-1586 (Fig. 2), which contains a single reciprocal translocation between chromosomes 3 and 4 (marked as T-8 in Fig. 1), was used as starting material. As a result of this translocation all chromosome pairs of the karyotype became easily indistinguishable, and the resolution power of the karyogramme analysis was improved. This was an important step in the development of efficient techniques for detection of chromosome rearrangements, especially pericentric inversions. More than 26% of the total number of partially sterile M1 plants contained some kind of karyotype reconstruction. What is interesting in this investigation is that at least one-fifth of the induced chromosomal rearrangements in T-1586 were found to be pericentric inversions.

As expected, the reciprocal translocations and pericentric inversions proved to be the main types of...