

Dissecting the Barley Genome to Chromosome Arms by Flow Sorting

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Abstract: The analysis and mapping of the nuclear genome of barley is hampered by its large size (1C ~ 5100 Mbp). In our previous work, we have demonstrated that in some crops, purification of mitotic chromosomes using flow cytometry may be used to dissect nuclear genomes into small and well-defined parts. This approach is especially useful in plants with complex genomes, where it offers a targeted and hence economical approach to genome analysis and gene cloning. DNA of flow-sorted chromosomes has been used for the isolation of molecular markers from specific genome regions, physical mapping using PCR and FISH, integration of genetic and physical maps and for construction of chromosome-specific DNA libraries. Until now, chromosome analysis and sorting using flow cytometry (flow cytogenetics) found little application in barley ($2n = 14$) due to the inability to discriminate and sort individual chromosomes, except the smallest chromosome 1H, and some translocation chromosomes whose DNA content differed from the remaining chromosomes. To overcome this bottleneck, we have analyzed wheat-barley telosome addition lines. Our results revealed that the addition lines may be used to sort any of the fourteen barley chromosome arms. The identity of sorted arms was verified using chromosome-specific markers; FISH was used to assess the purity in sorted fractions. Furthermore, our results indicate a possibility to sort sub-arm chromosomal segments that can be generated in the wheat-barley chromosome addition lines by the gametocidal system. These advances make barley flow cytogenetics an attractive tool that may greatly facilitate its genome mapping.