Genetic Map of Rapeseed – the Basis for Localization of QTLs for Seed Glucosinolate and Erucic Acid Content

Marcin Matuszczak, Jan Krzymanski, Iwona Bartkowiak-Broda,
Plant Breeding and Acclimatization Institute, Department of Oilseed Crops
Strzeszynska 36, 60-479 Poznan, Poland, e-mail: marmat@nico.ihar.poznan.pl

ABSTRACT

Two winter rapeseed DH lines which vary in respect of chemical characters and molecular markers were chosen to develop the mapping population. The DH-JN-86 line has low erucic acid and high glucosinolate content in seeds whereas the DH-ER_2-13/1 line has high erucic acid and low glucosinolate content in seeds. After the crossing of parental lines DH-JN-86 × DH-ER_2-13/1, from the resulted F_1 hybrid, 320 dihaploid lines were obtained using microspore-derived embryos.

The subset of these lines, as well as two parental lines, was analyzed using RAPD and AFLP methods. These analyses gave the matrix of marker data (51 markers for 36 DH lines) that was the basis for preparation of the genetic map of rapeseed. The preliminary statistical analyses of these data resulted in construction of the map comprising 38 loci. 24 of the loci are assembled into 8 linkage groups covering 169 Haldane cM. The remaining 14 loci are unlinked. The map is under construction and will be enriched with new markers.

The analyses of seed glucosinolates and fatty acids content were also performed in that population. The amount of each glucosinolate and fatty acid was measured using gas chromatography. These analyses showed the high degree of phenotypic variation of all glucosinolates and of erucic acid. Adding of new markers to the genetic map together with the use of chemical characters data collected for studied population will let us localize the QTLs for important quality traits of winter rapeseed.

Key words: rapeseed – genetic mapping – RAPD – AFLP – glucosinolates and fatty acids

INTRODUCTION

The genetic map is the basis for further studies of genome and is also useful for searching of important molecular markers that can accelerate the breeding of new prospective varieties of common crops.

This paper reports on the present stage of development of winter rapeseed genetic map that will serve as the basis for localization of QTLs for seed glucosinolate and erucic acid content. The parental lines which have genetic background representing the breeding material from the Department of Oilseed Crops were chosen for these studies. It gives us the opportunity to find the QTLs and particular markers that will be useful for breeding projects in our Institute.

MATERIALS AND METHODS

Two parental lines were chosen. The DH-JN-86 line has low erucic acid and high glucosinolate content in seeds whereas the DH-ER_2-13/1 line has high erucic acid and low glucosinolate content in seeds. The DH-JN-86 line was derived from the Jet Neuf variety and the DH-ER_2-13/1 line comes from the breeding materials of Department of Oilseed Crops (Matuszczak and Krzymanski 1999). After the crossing of parental lines DH-JN-86 × DH-ER_2-13/1, from the resulted F_1 hybrid, 320 dihaploid lines were obtained using microspore-derived embryos.

To obtain molecular markers the subset of 36 dihaploid lines, as well as two parental lines, was analyzed using RAPD (Welsh and McClelland 1990; Williams et al. 1990) and AFLP (Vos et al. 1995) methods. RAPD analyses were performed using Operon 10-mer primers. The names of markers are based on the symbols of primers. AFLP analyses were done using Gibco BRL AFLP Analysis System I Kit. The coding system for primer combinations that was used to name AFLP markers was previously described (Lombard and Delourme 2001).

The genetic linkage map (fig. 1) was constructed using MAPMAKER/EXP 3.0. The linkage groups were established with the minimum LOD score of 2.0 and a maximum recombination frequency of 0.4 (maximum distance: 80.5 Haldane cM). Markers within each linkage group were ordered using “Compare” command and then the distances between markers were counted using “Map” command and expressed with the Haldane cM.

The analyses of seed glucosinolates and fatty acids content were performed using gas chromatography. These analyses were done for the whole population of DH lines (320 lines). To minimize the environmental influences on seeds all the plants were grown at the same field and at the same time.

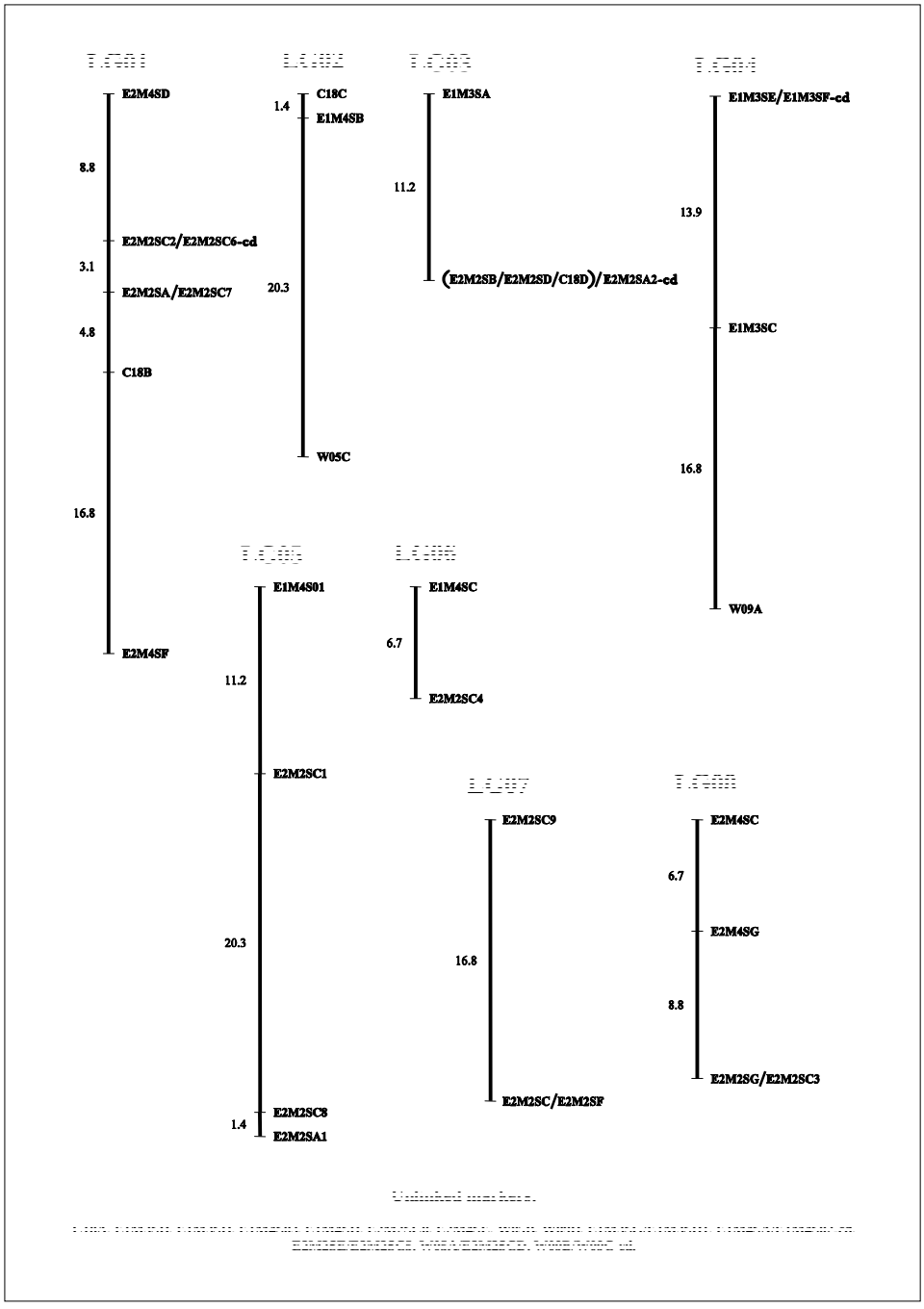

Fig.1. Linkage map of *Brassica napus* L. constructed with 51 markers on the 36 DH lines of the studied population. Distances are expressed in Haldane cM.

RESULTS

The RAPD and AFLP analyses performed for 36 DH lines resulted in the discovery of many polymorphic bands. Only 51 of them were scored as reproducible markers. 11 markers (22%) were obtained using RAPD method and 40 markers (78%) using AFLP method. The matrix of marker data (51 markers for 36 DH lines) was the basis for preparation of the genetic map of winter rapeseed.

The preliminary statistical analyses of collected data resulted in construction of the map comprising 38 loci (fig. 1). 24 of the loci were assembled into 8 linkage groups covering 169 Haldane cM. The remaining 14 loci were unlinked. Some pairs of markers were identical or behaved like a codominant marker (in the latter case markers were indicated with “cd” letters).

Parallel chromatographic analyses of seeds showed the high degree of phenotypic variation of all glucosinolates and of erucic acid in the studied population. These results make possible the localization of the QTLs for important quality traits of winter rapeseed.

DISCUSSION

The results presented here are not the final summary of the work but rather the introductory remarks. The main goal of that paper was to announce what kind of studies are conducted and what benefits can be expected. The genetic map presented still comprises too small number of markers. There are many unlinked loci and it is difficult to establish any linkage groups with the minimum LOD score of 3.0 or more. The result is that the number of linkage groups does not reflect the number of chromosomes and the percentage of genome covered by linkage groups is very low. The conclusion is that a lot of work has to be done yet with that population. However, the use of DH population makes that work possible and in the future the studies on that material in order to add new characters and markers to the map can bring more benefits. It would be of great value to take opportunity and map QTLs using well characterized and stable DH population that exhibits segregation of important quality traits. It is also important for us to compare as well as integrate our genetic map and markers with the results obtained in other laboratories (Foisset et al. 1996; Lombard and Delourme 2001) – to contribute in the international collaborative work on the rapeseed genome.

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