The identification of DNA markers tightly linked to pollen-fertility restoration genes in rye (*Secale cereale* L.) with CMS Pampa using RIL mapping population

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RYE HYBRID BREEDING based on cms Pampa started in the seventies of the XX century. However, the first hybrid variety was released ca 30 years ago (Miedaner 2004). With the introduction of the cms pampa based rye hybrids, molecular studies on the molecular basis of the phenomenon (Miedaner 2000) started. It was demonstrated that pollen fertility restoration trait is covered by many genes located on the chromosomes 1R, 3R, 4R, 5R, and 6R. Two of the genes, the one on 1R and the second on 4R chromosomes cover most of the phenotypic variance of the trait. However, the gene on 4R is considered to be the most welcome one and is usually unavailable in polish materials.

MATERIALS AND METHODS

Recently, a prospective pollen fertility restoration line based on cms Pampa was detected and used for the construction of RIL mapping population consisting of 190 F2 individuals. The population was genotyped with DArTseq and silicoDArTs resulting in a highly dense genetic map with a few small gaps (MultiPoint Ultra Dense software). The cms pampa non-restorer line (S 305P / 00) was crossed to RILs (F7) ((S64 / 04/01): S305N / 00 x S037R / 05) and the BC1F7: S 305P / 00 x [RIL 7 S64 / 04/01): S 305N / 00 x S037R / 05], population was used to evaluate pollen fertility based on Geiger's visual scale. The data were combined with genetic maps allowing composite interval (WinQTL Cartographer) and association mapping (Tassel) approach.

Table 1. The arrangement of markers identified within the QTL regions.

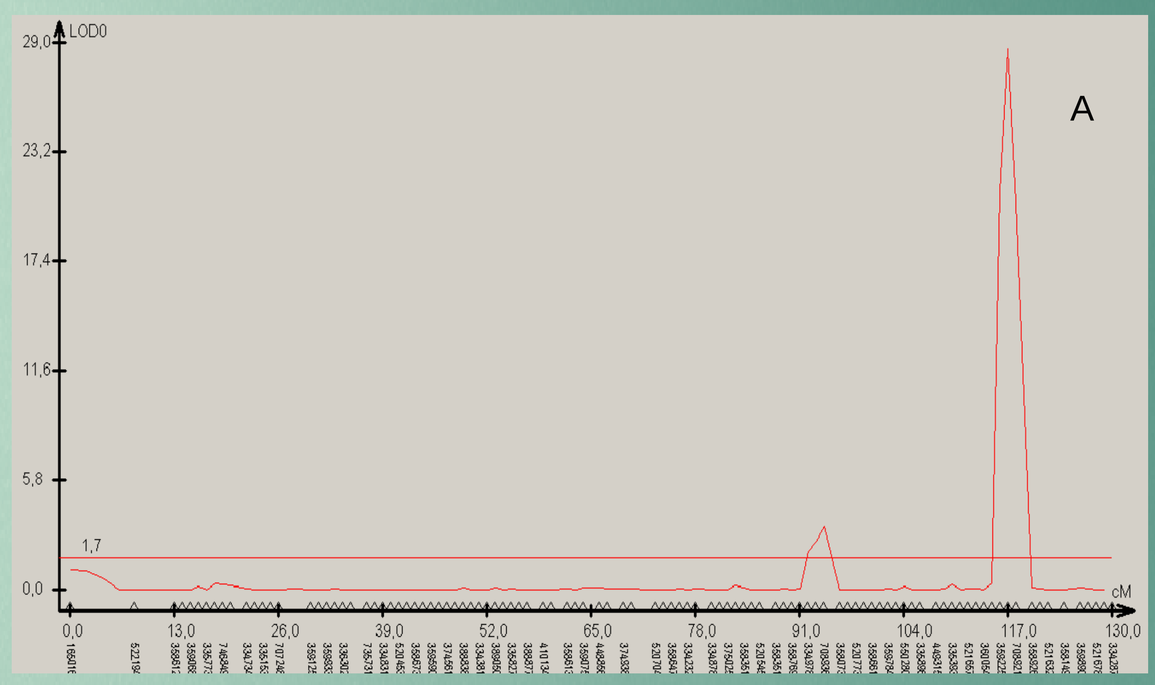
QTL	QTL1	QTL2	QTL3
QTL assigmnent	LG2(4R)	LG2(4R)	LG8(5R)
QTL maximum (cM)	94,2	117,0	117,2
LOD score	3,35	28,66	3,1
Range of QTL (cM)	3,1	4,7	3,0
Skeleton marker within QTL maximum*	70893	37303	-
Skeleton markers within QTL**	55041_9:G>A	35922_15:G>A	38858
	35807	70892_9:T>C	52274_46:G>T

*It should be emphasized that the skeletal markers linked to pollen fertility restoration QTLs have some redundant and/or interpolated markers.
**The sequences of markers linked/associated to/with pollen fertility restoration trait are available from the Authors if requested

RESULTS

Three QTLs located on 4R (two QTLs) and 5R chromosomes were detected using composite interval mapping (Figure 1). The most significant QTL on the 4R chromosome explained up to 66% of variance and was highly saturated with silicoDArT and DArTseq markers. The QTL spanned over 3cM. The remaining QTLs were of less importance. All associated markers were within the 4R QTL or its vicinity (Table 1).
Association mapping allowed the identification of 90 DArTseq and 813 silicoDArT markers that passed the Bonferroni test. Those markers had association value (R2) ranging from 0.63 to 0.14 at p = 0.01. A comparison of the results of composite and association mapping demonstrated that the associated markers mapped to the 4R chromosome within the QTL LG3-2.

Figure 1. Composite interval mapping. A and B – QTLs on the chromosome 4R and 5R, respectively.



SUMMARY

Our biparental RIL mapping population was based on parental inbred rye lines that were exploited for rye hybrid breeding purposes. As advanced breeding materials were used, the lack of drag effect was expected. Advanced recombinant lines used for map construction resulted in a densely saturated genetic map. Based on the collinearity phenomenon linkage groups were successfully assigned to rye chromosomes. Composite interval mapping allowed the identification of three pollen fertility restoration QTLs mapped to the 4R and 5R chromosomes. The QTLs were limited to a few cM and were represented by numerous markers. Association mapping indicated that markers associated with pollen fertility were present within QTLs or their nearest vicinity. The identified markers need to be tested on a broad range of materials to verify whether they could be used for marker assisted selection purposes or marker based backcrossing.

REFERENCES

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