

STUDY OF FUSARIUM HEAD BLIGHT RESISTANCE IN WHEAT USING MICROSATELLITE MARKERS

Katalin PUSKÁS¹ - Gyula VIDA¹ - Judit KOMÁROMI¹ - Hermann BÜRSTMAYR² - Marc LEMMENS² - Zoltán BEDO¹ - Ottó VEISZ¹

¹Agricultural Research Institute of the Hungarian Academy of Sciences, Martonvásár, Hungary

²IFA-Tulln, Institute for Agrobiotechnology, Tulln, Austria

Introduction

Investigations on Fusarium head blight (FHB) in wheat are gaining importance throughout the world. This can be attributed to the fact that *Fusarium* species not only cause yield losses, but also produce mycotoxins in infected plant tissues, the accumulation of which makes the grain unsuitable for both human and animal consumption (Larsen *et al.* 2004).

Nowadays spring genotypes of Far Eastern origin are considered to have the best resistance, so these are generally used as basic material in studies on the genetic background of resistance. Several types of resistance have been described in wheat on the basis of the plant's defence mechanisms (Mesterházy *et al.* 1999), but in most cases genetic analysis is focussed on Type II resistance, i.e. resistance to the spread of the fungus within the spike, since the use of point inoculation allows it to be examined separately from Type I resistance, which prevents the penetration of the fungus into the spike tissue. (Schroeder and Christensen 1963). The other method used to investigate FHB resistance is spray inoculation (Buerstmayr *et al.* 2003, Schmolke *et al.* 2005), but the infection data only allow conclusions to be drawn on field resistance, i.e. on the joint effect of the two main types of resistance (Miedaner *et al.* 2003). Although this inoculation method is used less frequently in the phenotypic analyses required for genetic research, its role should not be underestimated. The present experiments involved the genetic analysis of field resistance in the moderately resistant Chinese line Ning8331 and the moderately susceptible variety Martonvásári 17.

Materials and methods

Field resistance trial

A population of 228 F₅-derived recombinant inbred lines (RILs) from the wheat cross Ning8331/Martonvásári 17 was used in the examinations. The parents and the RILs were sown in Martonvásár in three experimental years. As each line flowered, the heads were sprayed with *Fusarium culmorum* spore suspension (for multiplication method, see Buerstmayr *et al.* 2000) on three occasions, at two-day intervals. High humidity was ensured during the trial by a mist irrigation system. Fusarium head blight symptoms were evaluated on the spikelets 26 days after inoculation (except in 2003). In July ten ears/RIL were harvested and threshed. The degree of kernel infection was determined and the resulting yield loss was analysed on the basis of changes in the kernel weight per head, test weight and thousand kernel weights.

Microsatellite analysis

DNA extraction and PCR amplification were carried out in Tulln, and fragment analysis in both Tulln and Martonvásár, as described by Buerstmayr *et al.* (2002). A total of 150 microsatellite markers were tested for polymorphism between the parental DNAs, and the DNA of the RILs has so far been tested using 63 primer pairs, evenly distributed over the wheat chromosomes (Röder *et al.* 1998).

Statistical analysis

Correlation analysis and simple regression analysis were used to calculate correlation coefficients between pairs of experiments and coefficients of determination (R^2) for the markers. Both were performed using Microsoft Excel. Two years of data were available for all the phenotypic traits studied in the experiment, and these were averaged for use in the analyses.

Results and discussion

In the course of the laboratory analysis special emphasis was placed on the analysis of loci on chromosomes 3B and 5A, since the vast majority of research on the genetics of FHB resistance, particularly the testing of genotypes of *Sumai3* origin, has suggested that these are the main carriers of resistance genes (Buerstmayr *et al.* 2003).

Table 1 lists the markers that gave the closest correlations, or were found to correlate with several traits. As expected, loci on the short arm of chromosome 3B were found to be most closely related to FHB resistance, while chromosome 5A was only found to have a minor effect. A substantial gene effect was also observed on the short arm of chromosome 2B, in agreement with the findings of Chen *et al.* (2003) in connection with Type II FHB resistance. The phenotypic studies, carried out over several years, confirmed earlier results (Puskás *et al.* 2002) showing an effect contributing to field resistance, linked to the Gwm169 marker on chromosome 6A. However, the linkage was far weaker than for the gene effects on chromosomes 3B and 2B. Other minor effects for several traits were identified on chromosomes 5B and 6D. One product of the Gwm614 marker, with smaller fragment size, exhibited a correlation with yield loss parameters, but its chromosomal location has not yet been determined. In addition, minor effects were demonstrated on chromosomes 4B and 7A for resistance to spike infection, on 2D for kernel infection, and on 1D, 2D, 3A and 6B for various components of yield loss. All the effects listed here were derived from the *Ning8331* parent.

Table 1. Coefficients of determination (R^2) and P values for the SSR markers shown by the analysis to be most closely associated with FHB resistance traits in the *Ning8331/Martonvásári 17* population

Chromosome	Marker	Resistance against	R^2 (%)	P
3B	Gwm493	spike infection	6.5	1.4×10^{-4}
		kernel infection	14.7	2.7×10^{-7}
	Gwm389	loss in kernel weight/head	14.7	6.0×10^{-5}
		loss in test weight	15.1	3.8×10^{-5}
		loss in thousand kernel weight	17.7	7.2×10^{-6}
2B	Gwm429	kernel infection	8.3	1.9×10^{-4}
		loss in test weight	9.7	1.0×10^{-3}
	Gwm257	loss in kernel weight/head	14.0	4.3×10^{-4}
		loss in thousand kernel weight	8.9	4.9×10^{-3}

6A	Gwm169	spike infection	2.8	1.1×10^{-2}
		kernel infection	6.5	6.1×10^{-4}
		loss in kernel weight/head	7.2	3.8×10^{-3}
		loss in test weight	4.6	2.2×10^{-2}
		loss in thousand kernel weight	7.1	4.0×10^{-3}
6D	Gwm325	spike infection	2.6	2.0×10^{-2}
		kernel infection	4.8	5.2×10^{-3}
		loss in kernel weight/head	4.4	3.3×10^{-2}
		loss in test weight	4.5	3.0×10^{-2}
5A	Gwm154	kernel infection	3.4	1.8×10^{-2}
		loss in kernel weight/head	6.3	1.4×10^{-2}
	Gwm304	loss in test weight	4.4	3.8×10^{-2}
		loss in thousand kernel weight	4.8	3.0×10^{-2}
5B	Gwm604	kernel infection	2.6	4.6×10^{-2}
		loss in kernel weight/head	4.5	3.3×10^{-2}
		loss in test weight	6.1	1.2×10^{-2}
		loss in thousand kernel weight	7.0	7.4×10^{-3}

For some phenotypic traits, minor effects linked to resistance could also be demonstrated from the more susceptible parental genotype, *Martonyásári 17*. Linkage was observed between milder spike infection and marker Gwm186 (chromosome 5A), kernel infection and marker Gwm793, and lower yield loss and markers at the end of the long arm of chromosome 2B.

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References

- Buerstmayr, H., Lemmens, M., Hartl, L., Doldi, L., Steiner, B., Stierschneider, M., Ruckebauer, P., 2002. Molecular mapping of QTLs for Fusarium head blight resistance in spring wheat. I. Resistance to fungal spread (Type II resistance). *Theor. Appl. Genet.* 104: 84-91.
- Buerstmayr, H., Steiner, B., Hartl, L., Griesser, M., Angerer, N., Lengauer, D., Miedaner, T., Schneider, B., Lemmens, M., 2003. Molecular mapping of QTLs for Fusarium head blight resistance in spring wheat. II. Resistance to fungal penetration and spread. *Theor. Appl. Genet.* 107: 503-508.
- Buerstmayr, H., Steiner, B., Lemmens, M., Ruckebauer, P., 2000. Resistance to Fusarium head blight in winter wheat: heritability and trait associations. *Crop Sci.* 40: 1012-1018.
- Chen, J., Griffey, C.A., Saghai Maroof, M.A., Wilson, J.A., Nabati, D., Biyashev, R. M., 2003. Molecular mapping and marker assisted selection of QTLs for Fusarium head blight resistance in Chinese wheat line W14. In: Canty, S. M., Lewis, J., Ward, R. W. (Eds.), *Proc. National Fusarium Head Blight Forum; 2003 Dec 13-15; Bloomington, MN, Michigan State University*, 7-9.
- Larsen, J. C., Hunt, J., Perrin, I., Ruckebauer, P., 2004. Workshop on trichothecenes with a focus on DON: summary report. *Toxicology Letters* 153: 1-22.
- Mesterházy, Á., Bartók, T., Mirocha, C. G., Komoróczy, R., 1999. Nature of wheat resistance to *Fusarium* head blight and the role of deoxynivalenol for breeding. *Plant Breeding* 118: 97-110.
- Miedaner, T., Moldovan, M., Ittu, M., 2003. Comparison of spray and point inoculation to assess resistance to Fusarium head blight in a multienvironment wheat trial. *Phytopathology* 93: 1068-1072.
- Puskás, K., Vida, G., Veisz, O., Buerstmayr, H., Bedő, Z., 2004. Analysis of Fusarium head blight resistance QTLs in the 'Ning 8331' x 'Martonvásári 17' population. In: J. Vollmann, H. Grausgruber, P. Ruckebauer (eds.) *Genetic variation for plant breeding. Proceedings of the 17th EUCARPIA General Congress. 8-11 September 2004, Tulln, Austria*, 189-194.
- Röder, M. S., Korzun, V., Wendehake, K., Plaschke, J., Tixier, M.-H., Leroy, P., Ganal, M. W., 1998. A microsatellite map of wheat. *Genetics* 149: 2007-2023.
- Schmolke, M., Zimmermann, G., Buerstmayr, H., Schweizer, G., Miedaner, T., Korzun, V., Ebmeyer, E., Hartl, L., 2005. Molecular mapping of Fusarium head blight resistance in the winter wheat population Dream/Lynx. *Theor. Appl. Genet.* 111: 747-756.
- Schroeder, H. W., Christensen, J. J., 1963. Factors affecting resistance of wheat to scab caused by *Gibberella zeae*. *Phytopathology* 53: 831-838.
- Kádár, I. (2004): Szennyvíziszap-adagok hatása a növény (*Triticum vulgare* L.)-talaj rendszer néhány mikrobiológiai és biokémiai tulajdonságára (Impact of sludge treatments on some microbiological and biochemical properties of plant (*Triticum vulgare* L.)-soil system. – in *Hungarian*). *Agrokémia és Talajtan.* 53. 3-4. 331-342 pp
- Svetková, K.; Henselová, M.; Morvová, M. (2005): Effects of a carbonization product as additive on the germination, growth and yield parameters of agricultural crops. *Acta Agron. Hun.* 53. (3). 241-250 pp
- Tanács, L., Matuz, J., Gerő, L., Petrőczy, I.M. (2005): Effect of NPK Fertilizers and Fungicides on the Quality of Bread Wheat in Different Years. *Cereal Res. Commun.* 33 (2-3): 627-634 pp