Theor Appl Genet (2004) 109: 1133–1138
DOI 10.1007/s00122-004-1734-5

ORIGINAL PAPER

Hong-Qing Ling · Jiwen Qiu · Ravi P. Singh · Beat Keller

Identification and genetic characterization of an *Aegilops tauschii* ortholog of the wheat leaf rust disease resistance gene *Lr1*

Received: 3 March 2004 / Accepted: 25 May 2004 / Published online: 16 July 2004

Abstract *Aegilops tauschii* (goat grass) is the progenitor of the D genome in hexaploid bread wheat. We have screened more than 200 *Ae. tauschii* accessions for resistance against leaf rust (*Puccinia triticina*) isolates, which are avirulent on the leaf rust resistance gene *Lr1*. Approximately 3.5% of the *Ae. tauschii* accessions displayed the same low infection type as the tester line Thatcher *Lr1*. The accession Tr.t. 213, which showed resistance after artificial infection with *Lr1* isolates both in Mexico and in Switzerland, was chosen for further analysis. Genetic analysis showed that the resistance in this accession is controlled by a single dominant gene, which mapped at the same chromosomal position as *Lr1* in wheat. It was delimited in a 1.3-cM region between the restriction fragment length polymorphism (RFLP) markers ABC718 and PSR567 on chromosome 5DL of *Ae. tauschii*. The gene was more tightly linked to PSR567 (0.47 cM) than to ABC718 (0.79 cM). These results indicate that the resistance gene in *Ae. tauschii* accession Tr.t. 213 is an ortholog of the leaf rust resistance gene *Lr1* of bread wheat, suggesting that *Lr1* originally evolved in diploid goat grass and was introgressed into the wheat D genome during or after domestication of hexaploid wheat. Compared to hexaploid wheat, higher marker polymorphism and recombination frequencies were observed in the region of the *Lr1* ortholog in *Ae. tauschii*. The identification of $Lr1^{Ae}$, the orthologous gene of wheat *Lr1*, in *Ae. tauschii* will allow map-based cloning of *Lr1* from this genetically simpler, diploid genome.

Communicated by G. Wenzel

Hong-Qing Ling and Jiwen Qiu have contributed equally to this work

H.-Q. Ling (✉) · J. Qiu
State Key Laboratory of Plant Cell and Chromosome Engineering, Institute of Genetics and Developmental Biology, Chinese Academy of Sciences,
Datun Road,
Chaoyang District, Beijing 100101, China
e-mail: hqling@genetics.ac.cn
Tel.: +86-10-64860377
Fax: +86-10-64860377

H.-Q. Ling · B. Keller
Institute of Plant Biology, University of Zurich,
Zollikerstrasse 107,
8008 Zurich, Switzerland

R. P. Singh
International Maize and Wheat Improvement Center (CIMMYT),
Apdo. Postal 6-641, 06600 Mexico, DF, Mexico

Introduction

Leaf or brown rust caused by the fungal pathogen *Puccinia triticina* is one of the most common diseases affecting wheat production worldwide. It occurs mainly on the leaf blades where uredinia rapidly develop following the infection of susceptible wheat cultivars under favorable environmental conditions. Epidemics of this disease can lead to severe losses of grain yield and a decrease in nutritional quality. An effective, economical and ecological method to control epidemics of leaf rust disease is the cultivation of resistant wheat cultivars. More than 50 resistance genes have been found in the germplasm of wheat and related species (Knott 1989; McIntosh et al. 1995; http://www.cdl.umn.edu/res_gene/wlr.html) and of these, 46 leaf rust resistance genes have been mapped to specific chromosomes and given official designations (*Lr1–Lr51*) according to the standards set forth in the Catalogue of Gene Symbols of Wheat (Kolmer 1996). These 46 resistance genes have been intensively used in wheat breeding (Johnson and Lupton 1987). Of these 46 leaf rust resistance genes 26 have been detected in hexaploid wheat cultivars, whereas the remaining 20 were introgressed into the wheat genome by interspecific crosses from lower-ploidy relatives of hexaploid wheat within the tribe Triticeae (Kolmer 1996). The two leaf rust resistance genes *Lr10* and *Lr21* have recently been isolated from the hexaploid wheat genome by map-based cloning (Feuillet et al. 2003; Huang et al. 2003).

Leaf rust disease resistance gene 1 (*Lr1*) was first described by Ausemus et al. (1946) in the hexaploid wheat cultivar Malakoff (Dyck and Samborski 1968). It is a single, dominant gene that was located on the long arm of chromosome 5DL by monosomic analysis (McIntosh et al. 1965). The *Lr1* gene is present in a number of wheat cultivars (Knott 1989; McIntosh 1988), and plants containing the *Lr1* resistance gene show a typical hypersensitive reaction after infection with an avirulent pathogen race. Feuillet et al. (1995) characterized the *Lr1* locus using restriction fragment length polymorphism (RFLP) markers and wheat deletion lines and showed that *Lr1* is localized at the distal end of wheat chromosome 5DL. Ling et al. (2003) constructed a high-resolution genetic map in the region of the *Lr1* locus by saturation mapping of two large segregation F_2 populations, and *Lr1* was delimited in a 0.16-cM region between RFLP markers ABC718 and PSR567.

Bread wheat (*Triticum aestivum* L. em Thell.) is an allohexaploid containing the three subgenomes A, B and D. It originates from two independent hybridization events. The first hybridization occurred between the wild diploid wheat *T. urartu* as the A-genome donor and an unknown species containing the B genome approximately half a million years ago, resulting in the tetraploid ancestor of modern *Triticum* species, *T. turgidum* (Feldman et al. 1995; Huang et al. 2002). *T. aestivum* (bread wheat) then arose from the hybridization of *T. turgidum* with *Aegilops tauschii* (the D-genome donor) 8,000 year ago (Dvorak et al. 1998; Kihara 1944; McFadden and Sears 1946). Therefore, *T. aestivum* is an evolutionary young member of the Poaceae family. The large size of the bread wheat genome (16×10^9 bp/1C, Bennett and Smith 1976), the high content of repetitive DNA (more than 80%, Gill and Gill 1994; Kam-Morgan et al. 1989) as well as the presence of three homologous genomes make gene isolation difficult. The use of diploid progenitors, both for genetic or genomic studies, greatly simplifies gene cloning (Feuillet et al. 2003; Huang et al. 2003). The diploid *Ae. tauschii* has been successfully used for the cloning of the *Lr21* resistance gene, which was derived from this species (Huang et al. 2003).

In the ideal case, an ortholog of the target gene in bread wheat can be identified in a diploid, and the mapping and cloning can be exclusively done in the diploid species. We report here the identification and genetic characterization of an ortholog of leaf rust disease resistance gene *Lr1* in *Ae. tauschii*.

Materials and methods

Screening of *Aegilops tauschii* accessions for leaf rust resistance

Studies in Mexico

Two hundred accessions of *Ae. tauschii* were screened with nine races of *Puccinia triticina* in the greenhouse. The nomenclature and the avirulence/virulence formula of these races are described in Singh (1991). Between six and eight seeds of each line were sown as hills in flat trays (48 entries/tray). Two-week-old seedlings were inoculated by spraying with a urediniospore-lightweight oil suspension. Inoculated plants were placed in a dew chamber for 16 h at about 20°C and then transferred to a greenhouse maintained at 18–23°C. Infection type displayed by the plants was recorded 10 days after inoculation using a 0–4 scale for leaf rust similar to that described by Stakman et al. (1962).

Studies in Switzerland

Six accessions of *Ae. tauschii* (RL5688, TA1704, TA2397, TA2567, Tr.t. 213 and AUS18913) were screened with six leaf rust isolates BRW-96258, BRW-95032, BRW-95279, BRW-98015, BRW-98017, BRW-90035. For screening, nine seeds of each *Ae. tauschii* accession were sown in three Jiffy pots (three seeds in each 4×5-cm pot; Jiffy A/S, Denmark) in plastic plates (each plate contained 25 Jiffy pots). Six seeds of Thatcher and Arina (two susceptible wheat cultivars as a negative control) and Thatcher *Lr1* (a resistant wheat cultivar as a positive control) were also sown in the same plate. Two-week-old seedlings were artificially infected as described by Schachermayr et al. (1995). The phenotypes (resistant or susceptible) were evaluated 10 days after infection.

For genetic analysis and mapping of the resistance gene in diploid *Ae. tauschii*, the resistant accession Tr.t. 213 was crossed with the susceptible accession TA1704 and a segregating population containing 318 F_2 plants developed. Evaluation of resistance in F_2 and F_3 plants was carried out at the seedling stage using the leaf rust isolate 90035, which is avirulent for the resistance gene *Lr1*, as described by Ling et al. (2003).

Restriction fragment length polymorphism analysis

RFLP markers were screened for polymorphism in the mapping parents. About 5 g of leaf tissue was harvested from 6-week-old plants of *Ae. tauschii* accessions Tr.t. 213 and TA1704 and frozen in liquid nitrogen. Genomic DNA from leaf tissue was extracted following the protocol described by Graner et al. (1990). Six restriction endonucleases (*Bam*HI, *Dra*I, *Eco*RI, *Eco*RV, *Hin*dIII and *Xba*I) were used for digestion of genomic DNA. A 20-μg aliquot of total DNA was incubated with 50 U of an endonuclease in a final volume of 35 μl at 37°C for 5 h. The cleaved DNA fragments were separated on a 0.75% agarose gel and transferred to Hybond-N^+ membranes (Amersham, Germany) according to the manufacturer's instructions. The membranes were baked at 80°C for 1 h for fixation of DNA.

For the RFLP analysis of the segregating F_2 population, genomic DNA extraction from F_2 plants, DNA cleavage, fragment separation by agarose gel electrophoresis and

DNA transfer from gel to nylon membranes as well as Southern hybridization were performed as described by Ling et al. (2003).

Linkage analysis

Linkage estimation of RFLP markers and the resistance gene $Lr1^{Ae}$ was based on the recombination frequency (defined as the number of recombination events among the total number of gametes analyzed). The recombination frequency was converted to map units [centiMorgans (cM)] using the Kosambi function.

Results

Screening of *Ae. tauschii* accessions with wheat leaf rust isolates

Studies in Mexico

Of the 200 accessions of *Ae. tauschii* tested, seven displayed a low infection type—0 (tiny chlorotic flecks difficult to see by the naked eye)—that resembled the low infection type pattern displayed by the Thatcher *Lr1* tester line. In Table 1, we present infection type data for eight accessions together with those displayed by Thatcher and its *Lr1*-bearing near-isogenic line. Seven of these accessions were postulated to carry gene *Lr1*, indicating that *Lr1* occurred in 3.5% of the *Ae. tauschii* accessions. Three accessions, Tr.t. 308, Tr.t. 309 and Tr.t. 310, are probably reselections of the same accessions as all were numbered consecutively and displayed the same infection type pattern. If we take this into consideration, then *Lr1* was in fact present in 2.5% of the accessions. Based on the infection type response it also appeared that four accessions, Tr.t. 213, Tr.t. 308, Tr.t. 309 and Tr.t. 310, had only *Lr1*, whereas the remaining three accessions, Tr. t. 224, Tr.t. 225 and Tr.t. 327, carried additional gene(s) that conferred intermediate infection types to the six *Lr1* virulent races. Thatcher and *Ae. tauschii* accession Tr. t. 254 were susceptible to all nine races used.

Studies in Switzerland

The resistance of six *Ae. tauschii* accessions was first evaluated by artificial infection with wheat leaf rust isolate BRW-90035, which is avirulent on the resistance gene *Lr1* in wheat (Ling et al. 2003). After 10 days of infection, strong uredinia formation was observed on the inoculated leaves of accessions RL5688, TA2397, TA2567 and TA1704 as well as on the susceptible wheat lines Thatcher and Arina, whereas no uredinia were observed on the infected leaves of accessions Tr.t. 213, AUS18913 and the resistant wheat line Thatcher *Lr1* (see Table 2). The resistance of the six accessions was then investigated with five additional leaf rust isolates. All *Ae. tauschii* accessions and the resistant (Thatcher *Lr1*) and susceptible (Thatcher and Arina) controls showed susceptibility to isolates BRW-96258, BRW-95032 and BRW-95279, whereas Tr.t. 213 and AUS18913 as well as Thatcher *Lr1* were resistant against the isolates BRW-98015 and BRW-98017 (Table 2). These results demonstrate that the isolates BRW-96258, BRW-95032 and BRW-95279 were virulent on the resistance gene(s) in Tr.t. 213, AUS18913 and the *Lr1* gene in wheat, while BRW-98015 and BRW-98017 were avirulent for the resistance gene(s) in Tr.t. 213 and AUS18913 and for *Lr1* in wheat. Among the six *Ae. tauschii* accessions, Tr.t. 213 and AUS18913 showed identical resistance reactions as Thatcher *Lr1* with six leaf rust isolates. These data suggest that Tr.t. 213 and

Table 1 Seedling reactions of eight *Aegilops tauschii* and two hexaploid wheat lines when tested with nine Mexican *Puccinia recondita tritici* races

Plant lines	Accession no.	*Puccinia recondita tritici* race and infection type[a]								
		BBB/BN	LCJ/BN	KBB/JP	TBB/JP	CBJ/QL	MCD/SM	TBD/TM	TCB/TD	TCB/TD
Thatcher	CI10003	3+	3+	3+	3+	3+	3	3+	3+	3+
Thatcher *Lr1*	RL6003	0;	3+	0;	3+	0;	3+	3+	3+	3+
Tr.t. 213	CI11	0;	3+	0;	3+	0;	3	3+	3+	3+
Tr.t. 308	TA2453	0;	3	0;	3+	0;	3	3+	3	3+
Tr.t. 309	TA2454	0;	3	0;	3+	0;	3	3+	3	3+
Tr.t. 310	TA2455	0;	3	0;	3+	0;	3	3+	3	3+
Tr.t. 224	CI25	0;	3−	0;	X−	0;	3	X	X	3C
Tr.t. 256	TA2400	0;	1+3C	0;	3C	0;	1+3−	1+3−	1+3−C	1+3C
Tr.t. 327	TA2476	0;	X	0;	3−C	0;	3−C	;1+	;3−	3−C
Tr.t. 254	TA2397	3	3	3+	3+	3+	3	3+	3	3+

[a]Infection types are based on the 0–4 scale (Stakman et al. 1962). 0 = no uredinia or other macroscopic signs of infection; ";" = no uredinia, but hypersensitive, necrotic or chlorotic flecks of varying sizes present; 1 = small uredinia surrounded by necrosis; 2 = small to medium uredinia surrounded by green islands; X=random distribution of variable-sized uredinia on single leaf with a pure culture; 3 and 4=medium and large uredinia, respectively, without chlorosis or necrosis; + and − = uredinia somewhat larger and smaller, respectively, than normal for the infection type; C= more chlorosis than normal for the infection type. More than one designation represents a range of infection types

AUS18913 contain a gene with a very similar specificity as the bread wheat *Lr1* gene.

The resistance of the *Lr1* gene in bread wheat is caused by a block of pathogen growth resulting from a hypersensitive reaction after infection with an avirulent isolate. Lines containing the *Lr1* gene show chlorotic spots on the infected leaves, whereas uredinia were formed on leaves of susceptible plants. Chlorotic spots were also observed in the resistant accessions Tr.t. 213 and AUS18913. However, the chlorotic spots were significantly smaller than those formed on leaves of Thatcher *Lr1* and were hardly visible by eye.

Genetic analysis of leaf rust disease resistance in *Ae. tauschii* accession Tr.t 213

For genetic analysis of the resistance against leaf rust isolate BRW-90035 in *Ae. tauschii*, accession Tr.t. 213 was crossed with the susceptible accession TA1704. F_2 plants were evaluated for phenotypic segregation of resistance after artificial infection with the leaf rust isolate BRW-90035. Among the 318 F_2 plants, 255 plants were resistant and 63 were susceptible (χ^2=4.57, P=0.033). The observed segregation of leaf rust resistance in the population deviates slightly from the expected one, but it shows that resistance is controlled by a single gene with a probability of 0.03.

In order to identify whether the resistance in Tr.t. 213 is controlled by a gene located at an orthologous position of *Lr1* in hexaploid wheat, a small segregating population (51 F_2 plants) of Tr.t. 213×TA1704 was tested with the RFLP marker ABC718, which is tightly linked to *Lr1* on chromosome 5DL of bread wheat (Ling et al. 2003). We found that the leaf rust resistance gene in Tr.t. 213 cosegregated with the single-copy marker ABC718, indicating that the leaf rust resistance gene in Tr.t. 213 is possibly an orthologous gene of wheat *Lr1* and is located on chromosome 5 of *Ae. tauschii*. To establish a complete genetic map around the *Lr1* ortholog in *Ae. tauschii*, we mapped seven RFLP markers (PSR567, ABC718, TAG621, BCD1421, MWG602, CDO348 and KsuG14) from chromosome 5DL of wheat and polymorphic between Tr.t. 213 and TA1704. A segregating population containing 318 F_2 plants of the cross Tr.t. 213 × TA1704 was analyzed with the seven polymophic RFLP markers. Figure 1 shows the resulting genetic map. The resistance gene in Tr.t. 213 was mapped between the RFLP markers ABC718 (0.78 cM, five recombinants) and PSR567 (0.47 cM, three recombinants).

Compared to the genetic map at the *Lr1* locus in hexaploid wheat (Ling et al. 2003), the order of mapped markers in Tr.t. 213 was completely identical (Fig. 1). The genetic distance of the flanking RFLP markers (ABC718, PSR567 and TAG621) to the resistance gene in *Ae. tauschii* (0.78, 0.47, and 3.58 cM, respectively) is about six- to sevenfold larger than in wheat (0.12, 0.04 and 0.52 cM, respectively), indicating that recombination occurred more frequently in the region of the resistance gene in the segregating population of Tr.t. 213 × TA1704 than in the F_2 population of Thatcher *Lr1* × Thatcher. The mapping results strongly indicate that the resistance gene in Tr.t. 213 is an orthologous gene of leaf rust resistance gene *Lr1* of wheat. We called this gene $Lr1^{Ae}$.

Table 2 Resistance test of *Ae. tauschii* lines using leaf rust isolates virulent and avirulent on the leaf rust disease resistance gene *Lr1* (*R* resistant, *S* susceptible)

Plant lines	Leaf rust isolates					
	96258	95032	95279	98015	98017	90035
Thatcher	S	S	S	S	S	S
Thatcher*Lr1*	S	S	S	R	R	R
RL5688	S	S	S	S	S	S
Tr.t. 213	S	S	S	R	R	R
TA2397	S	S	S	S	S	S
TA2567	S	S	S	S	S	S
TA1704	S	S	S	S	S	S
AUS18913	S	S	S	R	R	R
Arina	S	S	S	S	S	S

Discussion

The wheat D genome originated from *Ae. tauschii* by spontaneous hybridization between the tetraploid *T. turgidum* ssp. *dicoccon* (AABB) and the diploid goat grass *Ae. tauschii* ssp. *strangulata* (DD) 8,000 years ago in southwestern Asia (Dvorak et al. 1998; Huang et al. 2002).

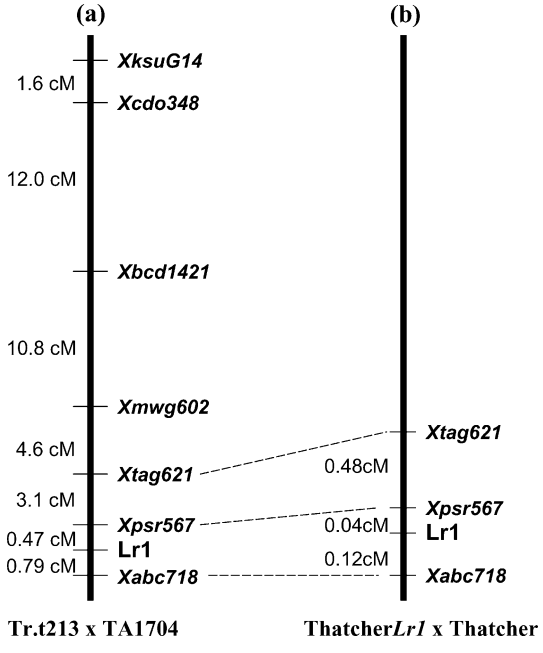

Fig. 1a,b Comparative genetic maps of the resistance gene $Lr1^{Ae}$ in *Ae. tauschii* and *Lr1* in wheat. **a** Genetic map of $Lr1^{Ae}$ in *Ae. tauschii*, resulting from the analysis of 318 F_2 plants of the cross Tr. t. 213 × TA1704. **b** High-resolution genetic map at the locus of the leaf rust disease resistance gene *Lr1* in wheat (data from Ling et al. 2003)

To date, 19 leaf rust resistance genes have been localized on the D genome (Knott 1989; Kolmer 1996), seven of which have been described in hexaploid wheat, whereas the other 12 have been introduced into the bread wheat D genome from *Ae. tauschii* (eight genes) and *Agropyron elongatum* (three genes) as well as *A. intermedium* (one gene) by intergeneric crosses (Kolmer 1996; http://www.cdl.umn.edu/res_gene/wlr.html). It is unknown whether the seven leaf rust resistance genes (including *Lr1*) that were directly identified in the wheat D genome had evolved before or after hybridization of the AB genomes with the D genome. Based on our data, we speculate that *Lr1* in hexaploid wheat is originally derived from the diploid goat grass. It possibly evolved in *Ae. tauschii* and was later integrated in hexaploid wheat by hybridization or genetic exchanges of the wheat D genome with the *Ae. tauschii* genome by spontaneous outcrossing between bread wheat and goat grass after domestication of hexaploid wheat. Alternatively, the *Lr1* gene might have evolved independently in *Ae. tauschii* and bread wheat. This is rather unlikely as there are few examples of an independent evolution of orthologous resistance gene loci. Resistance genes are usually not conserved even between relatively closely related species. An exception to this general observation is the finding of a functional ortholog of the *Lycopersicon esculentum* (tomato) *Pto* gene in the distantly related wild tomato *L. hirsutum* (Riely and Martin 2001). The future isolation of the *Lr1* and $Lr1^{Ae}$ gene from hexaploid wheat and *Ae. tauschii*, respectively, will reveal the evolutionary history of the two genes: if they evolved independently, we expect a lower degree of homology in comparison to a recent introgression from *Ae. tauschii* to hexaploid wheat. The case of these two genes will thus allow us to study at the molecular level the evolution of leaf rust resistance genes in wheat and one of its progenitors.

The *Ae. tauschii* (D) genome is approximately one-fourth the size of the bread wheat genome (4.024 Mbp/1C, Arumuganathan and Earle 1991). Thus, it is even smaller than the D genome of wheat (4.7 Mbp/1C, Lee et al. 1997). Our genetic mapping analysis indicates that the D genome of *Ae. tauschii* has high levels of polymorphism among different accessions. In our case, more than 60% (7/11) of tested RFLP markers were polymorphic between Tr.t. 213 and TA1704, whereas fewer than 30% (5/19) of RFLP markers showed polymorphisms in wheat (Ling et al. 2003). Our mapping results also revealed that the region of $Lr1^{Ae}$ in the segregating population of Tr. t. 213 × TA1704 underwent recombination much more frequently than the *Lr1* locus in wheat. A similar observation was made in chromosome 1DS of *Ae. tauschii* where very high recombination frequencies were observed (Spielmeyer et al. 2000). Obviously, recombination frequency and marker polymorphism are higher in *Ae. tauschii* than in wheat. This will simplify map-based cloning of the resistance genes, in our case the *Lr1* gene, from diploid *Ae. tauschii* compared to hexaploid wheat.

Acknowledgements The authors thank Beatrice Senger for technical assistance. We would also like to thank Philipp Streckeisen for providing leaf rust isolates of *Lr1*, Evans Lagudah (CSIRO Plant Industry, Australia) for the *Ae. tauschii* accession AUS18913 and Catherine Feuillet for critical reading of the manuscript. This work was supported by the Chinese National Science Foundation (grant nr. 30170493), the Chinese Academy of Sciences (grant nr. KSCX2-SW-304), the Swiss Ministry of Sciences (Grant BBW no. 97-0208 in the frame of the EU-project EGRAM) and the Swiss National Science Foundation (grant nr. 3100-65114).

References

Arumuganathan K, Earle ED (1991) Nuclear DNA content of some important plant species. Plant Mol Biol Rep 9:208–218

Ausemus ER, Harrington JB, Reitz LP, Worzella WW (1946) A summary of genetic studies in hexaploid and tetraploid wheats. J Am Soc Agron 38:1083–1099

Bennett MD, Smith JB (1976) Nuclear DNA amounts in angiosperms. Philos Trans R Soc London Ser B Biol Sci 274:227–274

Dvorak J, Luo M-C, Yang Z-L, Zhang H-B (1998) The structure of the *Ae. tauschii* gene pool and the evolution of hexaploid wheat. Theor Appl Genet 97:657–670

Dyck PL, Samborski DJ (1968) Genetics of resistance to leaf rust in common wheat varieties Webster, Loros, Brevit, Carina, Malakoff and Centenario. Can J Genet Cytol 10:7–17

Feldman M, Lupton FGH, Miller TE (1995) Wheats. In: Smartt J, Simmonds NW (eds) Evolution of Crops, 2nd edn. Longman Scientific, London, pp 184–192

Feuillet C, Messmer M, Schachermayr G, Keller B (1995) Genetic and physical characterization of the *Lr1* leaf rust resistance locus in wheat (*Triticum aestivum* L.). Mol Gen Genet 248:553–562

Feuillet C, Travella S, Stein N, Albar L, Nublat A, Keller B (2003) Map-based isolation of the leaf rust disease resistance gene *Lr10* from the hexaploid wheat (*Triticum aestivum* L.) genome. Proc Natl Acad Sci USA 100:15253–15258

Gill KS, Gill BS (1994) Mapping in the realm of polyploidy: the wheat model. BioAssays 16:841–846

Graner A, Siedler H, Jahoor A, Hermann RG, Wenzel G (1990) Assessment of the degree and type of polymorphism in barley (*Hordeum vulgare*). Theor Appl Genet 80:826–832

Huang S, Sirikhachornkit A, Su X, Faris J, Gill B, Haselkorn R, Gornicki P (2002) Genes encoding plastid acetyl-CoA carboxylase and 3-phosphoglycerate kinase of the *Triticum*/*Aegilops* complex and the evolutionary history of polyploid wheat. Proc Natl Acad Sci USA 99:8133–8138

Huang L, Brooks SA, Li W, Fellers JP, Trick HN, Gill BS (2003) Map-based cloning of leaf rust resistance gene *Lr21* from the large and polyploid genome of bread wheat. Genetics 164:655–664

Johnson R, Lupton FGH (1987) Breeding for disease resistance. In: Lupton FGH (ed) Wheat breeding. Its scientific basis. Chapman and Hall, Cambridge, pp 369–418

Kam-Morgan LNW, Gill BS, Muthukrishnan S (1989) DNA restriction fragment length polymorphisms: a strategy for genetic mapping of the D genome of wheat. Genome 32:724–732

Kihara H (1944) Discovery of the DD-analyzer, one of the ancestors of *vulgare* wheats. Agric Hortic 19:889–890

Knott DR (1989) The wheat rusts—breeding for resistance. Monographs on theoretical and applied genetics, vol 12. Springer, Berlin Heidelberg New York

Kolmer JA (1996) Genetics of resistance to wheat leaf rust. Annu Rev Phytopathol 34:435–455

Lee J-H, Yen Y, Arumuganathan K, Baenziger PS (1997) DNA content of wheat monosomics at interphase estimated by flow cytometry. Theor Appl Genet 95:1300–1304

Ling H-Q, Zhu Y, Keller B (2003) High-resolution mapping of the leaf rust disease resistance gene *Lr1* in wheat and characterization of BAC clones from the *Lr1* locus. Theor Appl Genet 106:875–882

McFadden ES, Sears ER (1946) The origin of *Triticum spelta* and its free-threshing hexaploid relatives. J Hered 37:81–89

McIntosh RA (1988) Catalogue of gene symbols for wheat. In: Miller TE, Koeber RMD (eds) Proc 7th Int Wheat Genet Symp. Bath Press, Bath, p 1273

McIntosh RA, Baker EP, Driscoll CJ (1965) Cytogenetical studies in wheat. I. Monosomic analysis of leaf rust resistance in the cultivars Uruguay and transfer. Aust J Biol Sci 18:971–977

McIntosh RA, Wellings CR, Park RF (1995) Wheat rust: an atlas of resistance genes. CSIRO/Kluwer, Australia/Dordrecht

Riely BK, Martin GB (2001) Ancient origin of pathogen recognition specificity conferred by the tomato disease resistance gene *Pto*. Proc Natl Acad Sci USA 98:2059–2064

Schachermayr GM, Messmer MM, Feuillet C, Winzeler H, Winzeler M, Keller B (1995) Identification of molecular markers linked to the *Agropyrum elongatum*-derived leaf rust resistance gene *Lr24* in wheat. Theor Appl Genet 30:982–990

Singh RP (1991) Pathogenicity variations of *Puccinia recondita* f. sp. *tritici* and *P. graminis* f. sp. *tritici* in wheat-growing areas of Mexico during 1988 and 1989. Plant Dis 75:790–794

Spielmeyer W, Moullet O, Laroche A, Lagudah ES (2000) Highly recombinogenic regions of seed storage protein loci on chromosome 1DS of *Ae. tauschii*, the D-genome donor of wheat. Genetics 155:361–367

Stakman EC, Stewart DM, Loegering WQ (1962) Identification of physiologic races of *Puccinia graminis* var. *tritici*. US Agric Res Serv E-617 (revised)