Emir. J. Food Agric. 2011. 23 (5): 413-423
http://ejfa.info/

Expression of the Wheat Leaf Rust Resistance Gene *Lr34* alone and in combination with other Leaf Rust Genes against UK isolates of *Puccinia triticina*

S. D. Abdul*

John Innes Centre for Plant Science Research, Research Park Colney, Norwich, NR4 7UH, England

Abstract: Wheat cultivars and 'Thatcher' backcross lines with the resistance gene *Lr34* alone and in combination with different genes for leaf rust resistance were assessed for their reaction to UK isolates of *Puccinia triticina.* The experiments were conducted in controlled environments and in the field. The seedlings were grown at 5 and 20^0C and as adult plants at 12 and 26^0C. The temperature control was used to identify the *Lr34* gene. In the field, all lines and cultivars with *Lr34* were resistant to leaf rust. In controlled environments, seedlings with *Lr34* alone gave variable infection types (ITs) of ; to 3 at 5^0C and 2 to 3- at 20^0C. This indicates that, temperature control is not effective at identifying the *Lr34* gene with these UK isolates. Based on similarities of reaction types in the field, the wheat cultivars Pavon 76, Ciano 67, Chris, Era and Marco Juarez Inta were postulated as having the resistance gene *Lr34.*

Key words: Leaf rust, *Puccinia triticina*, wheat, *Triticum aestivum*, durable resistance, Lr34.

تعبير جين *Lr34* مقاومة صدأ ورق القمح بمفرده و بالإشتراك مع جينات أخرى لصدأ الورق ضد عزلات بريطانية من *Puccinia triticina*

س. د. عبدول*

مركز جون انس لابحاث علوم النبات ، ساحة البحوث، كولني نورفيدش ، انجلترا . NR4 7UH

الملخص:تم تقييم اصناف من القمح و سلالات تلقيح رجعي بها جين *Lr34* مقاومة صدأ ورق القمح بمفرده و بالإشتراك مع جينات أخرى لصدأ الورق من حيث تفاعلها مع عزلات بريطانية من *Puccinia triticina*. أجريت التجارب في بيئات صناعية محكمة وفي الحقل. زرعت البادرات في درجة حرارة 5 و20^0C و النباتات الكاملة في12و 26^0C. تم التحكم في درجات الحرارة للتعرف علي جين *Lr34*. كانت كل النباتات مقاومة لصدأ الورق في الحقل. وفي البيئات المحكمة أظهرت البادرات المحتوية على جين *Lr34* بمفرده تباين في انواع الإصابة (ITs) من: ١-٣ عند 5^0C و٢-٣ عند 20^0C. هذا يوضح أن التحكم في درجة الحرارة غير فعال في تعريف جين *Lr34* في هذه السلالات البريطانية. بناءً على التشابه في انواع التفاعلات في الحقل، يفترض أن سلالات القمح Pavon 76 Ciano 67, Chris, Era and Marco Juarez Inta تحتوي علي جين المقاومة *Lr34*.

*Corresponding Author, *Email*: sdanabdul2003@yahoo.com
Received 03 December 2009; Revised 24 March 2011; Accepted 24 March 2011

Introduction

The resistance gene *Lr34* either alone or in combination with other leaf rust resistance gene(s) is implicated in conferring to wheat durable resistance against leaf rust disease. The leaf rust resistance genes *Lr13* and *Lr34*, which condition resistance mainly at the adult plant stage, have the longest history of effectiveness **(**Kolmer and Liu, 2002). The excellent adult plant resistance (APR) against leaf rust of the wheat cv. Chinese Spring is partially due to the resistance gene *Lr34* (Dyck, 1991). In addition Chinese Spring has *Lr31*, one of a complementary pair of *Lr31* + *Lr27* (Singh and McIntosh, 1984), both of which are required for the expression of resistance, and the APR gene *Lr12* (McIntosh and Baker, 1966; Dyck and Kerber, 1971; Dyck, 1991). Other wheat cultivars and lines known to have *Lr34* in combination with the *LrT3* gene include PI 321999, CIRC 26809-68 and CIRC 32125-70 from Turkey; PI 197249 and 72 Hills 175 from Ethiopia; and Frontana, Terenzio and Lageadinho (Dyck and Samborski, 1982). Also Dyck et al. (1966) identified *Lr34*, *LrT3* and the APR gene *Lr13* in Frontana. The resistance genes *Lr34* and *LrT3* individually provide some protection, but together they interact in a complementary manner giving increased levels of resistance (Dyck and Samborski, 1982).

The resistance gene *Lr34* is widespread among wheat cultivars as a result of the use of Frontana in many breeding programmes (Dyck and Samborski, 1982). Shang et al. (1986) identified this gene in seven out of nine wheat cultivars that were originally collected from the Mediterranean region and Middle East to China following extensive screening for leaf rust response. It was also identified in wheat cultivars Sturdy, Roblin, PI 250413, PI 58548 (from China) and the Russian wheat Bezostaja 1, which was widely grown in the USSR, Eastern Europe and Turkey for many years (Dyck and Samborski, 1979; Dyck and Samborski, 1982; Dyck, 1991; Dyck, 1992; McIntosh, 1992).

Using a molecular marker Kolmer et al. (2008) traced the origin of the *Lr34* gene in wheat cultivars from North and South America, CIMMYT, Australia, and Russia back to the cultivars Mentana and Ardito developed in Italy by Nazareno Strampelli in the early 1900s.

Dyck (1991) suggested that it may have been spread through Chinese Spring and line PI 58548, an introduction from China (Dyck, 1977). Tracing the history of Chinese Spring, initially named Chinese White, Sears (1988) reported that it was obtained by the Plant Breeding Institute, Cambridge, now Plant Breeding International, from China in about 1900. A few years later, it was introduced and probably used in breeding programme in South America by an Argentinean wheat breeder. This might have been responsible for its presence in wheat cultivars of South American origin. Roelfs (1988b) suggested that Alfredo Chaves, a land cultivar found in Brazil about 1921 and Americano 44D, selected in 1918 from a land cultivar in Uruguay, may be the sources of genes *Lr34*, *Lr13* and/or *Lr12***.** Dyck (1991) concluded that the wide distribution of *Lr34* among wheat cultivars and lines may have arisen from its introduction from a number of sources.

Dyck (1987) located *Lr34* in chromosome 7D of wheat, and in association with this gene is a non-suppressor allele for increased resistance to stem rust (*Puccinia graminis*) in Thatcher. In the presence of the suppressor allele the level of resistance to stem rust in Thatcher is reduced. The non-suppressor allele is said to be the same as or closely linked to the *Lr34* gene (Dyck, 1987).

Although *Lr34* can be detected at the seedling stage using low temperatures and low light intensity, its effects can be more easily recognized on adult plants (Dyck and Samborski, 1982; Dyck, 1987). A cultivar with *Lr34* can give IT ;1- to 3 with the same isolate depending on temperature and light. Also the genetic background of both host and pathogen may affect the IT. Drijepondt et al. (1991) were able to identify *Lr34* in seedlings of the Thatcher backcross line (TBL) RL6058 at 7°C, but not at 15°C. The same line was found to express APR against all three isolates in their studies at 15°C including that with corresponding virulence at the seedling stage. Variable influences of environment and

location on the expression of APR conferred by *Lr34* have also been observed in Mexico (Singh and Gupta, 1992; Singh, 1992a).

This study was carried out to determine the field responses of TBLs with *Lr34* alone or in combination with other genes to some UK leaf rust isolates, as well as to identify wheat cultivars having or suspected of having *Lr34*, and also to determine the effect of temperature and genetic background in the expression of *Lr34* at the seedling and adult plant growth stages.

Materials and methods

Evaluation of adult plants in the field

TBLs with *Lr34* alone or *Lr34* in combination with other genes obtained from Dr. P.L. Dyck, Agriculture and Agri-food Canada, Winnipeg, Canada along with wheat cultivars known or suspected to have the *Lr34* gene, were evaluated with the leaf rust isolates WBRP 85-20, WBRP 85-31 and WBRP 90-12 in the field in 1993 at Morley, near Norwich (Table 1). Thatcher and Armada were included as susceptible controls, and line *Lr19* as a resistant control. Also included were some wheat cultivars suspected of having the resistance gene *Lr34*. All the TBLs and the wheat cultivars were sown in the spring of 1993 due to their spring habit. However, the winter wheat Armada, Bezostaja, Cappelle Desprez, Nord Desprez and Bersée were initially grown and vernalized in a controlled environment before being transplanted into the field in spring. The test was carried out in hill plots of 30 by 30 cm. The wheat cultivars and lines were allocated to the hill plots at random and at regular intervals a spreader plot of the susceptible Armada was included. As a result of delays in disease build up the spreader plot technique of inoculation was supplemented by direct spraying of wheat plants with their respective isolates in odourless kerosene using micro ulva sprayer (Micon sprayers Ltd, Bromyard, England). Disease scores were taken in the months of June and July, 1993 as percent Leaf Area Infection (LAI) and ITs. The ITs were scored as Very Resistant (VR) = ;, Resistant (R) = 1&1+, Moderately Resistant (MR) = 2-&2, Moderate (M) = 2+&3-, Moderately Susceptible (MS) = 3 and Susceptible (S) = 3+&4. Each value of the percent LAI was taken as the mean of two replicates and the three isolates were evaluated in separate blocks.

Evaluation of adult plants in controlled environment

Duplicate adult plants of the TBLs and Chinese Spring, Chinese Spring ditelosomic 7DL, Chinese Spring ditelosomic 7DS, Bezostaja, Cappelle Desprez, Nord Desprez and Bersée (Table 2) were tested with isolates WBRP 85-31 and WBRP 90-12 in controlled environments set at 12°C and 26°C. Plants were first grown in containment glasshouse supplied with filtered air up to growth stage 39-52 (Zadoks et al., 1974), when at least one quarter of the ear had emerged, before inoculation. The seeds of the cultivars Bezostaja, Cappelle Desprez, Nord Desprez and Bersee were vernalized at 4°C before been sown. Mildew infection was controlled by a single spray of Bayleton (Triadimefon).

Evaluation of seedlings in controlled environment

Seedlings of TBLs RL6058 (*Lr34* from line PI 58548), RL6091 (*Lr34* from Chinese Spring), 91RN514 (*Lr34* from Bezostaja), 86RN122 (*Lr34* from Glenlea) and RL6050 (*Lr34* and *LrT3* from Terenzio) as well as those of Chinese Spring, Cappelle Desprez, Nord Desprez and Bersée were tested with 16 rust isolates (Table 3) at 5°C and 20°C (±2°C). Armada and Thatcher were used as susceptible controls, while line *Lr19* was used as temperature insensitive resistant control.

Inoculation

Inoculation was done using urediospores that were dispersed in talc at a rate 1 part of spores to 20 parts talc. This was used to inoculate sets of wheat seedlings in a laminar flow bench with an isolate. The laminar flow bench was cleaned between inoculations with different isolates using alcohol to avoid contamination between rust isolates. Adult plants were inoculated in separate glasshouse compartments with different isolates. Inoculated plants were incubated at 15°C (±2°C) and high relative humidity for 18h in cabinets. They were

thereafter transferred to their respective temperatures in controlled environments.

Disease score

Seedlings and adult plant reactions were recorded using the scale described by Stakman et al. (1962). Cultivars and lines having Infection Types (ITs) ;, 0, 1, 2 and X were considered resistant, whereas 3 and 4 were considered susceptible. ITs separated by a slash (/) suggest heterogeneity among seedlings of the affected cultivar or line, while plants with different ITs at the proximal and distal part of a leaf are represented by the two ITs in the same sequence.

Results

All the TBLs having *Lr34* alone or in combination with different genes were resistant to isolates WBRP 85-20, WBRP 85-31 and WBRP 90-12 in the field. Only a few of the backcross lines had LAI above 5% (Table 1). The highest score of 30MR-M was recorded on line 91RN514 (*Lr34*) at the second scoring date about mid-July with isolate WBRP 90-12. With the same isolate, lines RL6070 (*Lr34* and *LrT3*) had 14M, RL6112 (*Lr34* and *Lr10*) had 14MR-MS, and RL6113 (*Lr34* and *Lr11*) had 10MR-M by the second scoring date.

Table 1. Percent LAI of field evaluated wheat cultivars including lines having *Lr34* alone and *Lr34* in combination with different genes to different leaf rust isolates.

***Lr* line/ Cultivar**	***Lr* gene**	**WBRP 85-20**		**WBRP 85-31**		**WBRP 90-12**	
		Score1*	**Score2**	**Score1**	**Score2**	**Score1**	**Score2**
RL6058	*34*	0.35M-MS*	0.3MR-M	0.1VR	0.25MR-M	0.3M-MS	4M-MS
RL6091	*34*	0.25R-MR	1.5MR-M	0.1VR	0.15VR-M	0.1VR	3.5R-MR
91RN514	*34*	1.65MR-M	4MR	0.1VR	0.2VR-M	1.6MS	30MR-M
91RN516	*34*	0.15VR-R	0.3R-MR	0.1VR	0.15VR-MR	0.25MR-M	2.7MR-MS
86RN122	*34*	0.25R-M	0.4MR	0.1VR	0.15VR-MR	0.25R-MR	0.3R-MR
86RN127	*34*	0.25M	2MR-M	0.1VR	0.2R-MS	0.2M-MS	1MR-R
90RN1220	*34*	0.1VR	0.3R	0.1VR	0.15VR-R	0.15VR-MS	0.3R
RL6050	*34, T3*	0.2M	2MR-M	0.1VR	0.2R-M	0.15VR-R	0.3MR-MS
RL6069	*34*, *T3*	0.2VR-MR	0.2R	0.1VR	0.1VR	0.1VR	0.25R-MR
RL6070	*34, T3*	0.4M	4.5MR	0.3MR-MS	1.75R-MR	0.25MR-M	14M
RL6106	*34*, *T3*	0.2VR-MS	0.5R-M	0.1VR	0.15VR-MR	0.2R-MR	0.4R-MR
RL6108	*34*, *1*	0.1VR	0.2VR-R	0.1VR	0.1VR	0.1VR	0.25R
RL6109	*34*, *2a*	0.4R-M	0.4MR	0.1VR	0.15VR-R	0.25MR	0.3MR-M
RL6110	*34*, *3*	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR
RL6111	*34*, *3ka*	0.1VR	0.2VR-R	0.1VR	0.15VR-R	0.1VR	0.1VR
RL6112	*34*, *10*	1.05VR-S	0.65M	0.1VR	0.3MR	12.65M-MS	14MR-MS
RL6113	*34*, *11*	0.15VR-R	0.5MR-M	0.15VR-MS	0.25R-MR	0.3M	10MR-M
RL6114	*34, 13*	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.2R-M
RL6115	*34, 16*	0.4M	1.5MR	0.1VR	0.15VR-M	0.15VR-M	0.45R-M
RL6116	*34*, *17*	0.14VR-M	0.3R-M	0.15VR-MR	0.25R	0.2VR-MR	0.45R-M
RL6117	*34*,*18*	0.1VR	0.1VR-R	0.1VR	0.1VR	0.1VR	0.1VR
RL6118	*34*, *21*	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR
RL6119	*34*, *26*	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR
RL6120	*34*,*B*	.15VR-M	0.4R-MR	0.1VR	0.2R-MR	0.3R-MR	0.75MR
RL6121	*34*,	0.15VR-M	0.3R-M	0.1VR	0.1VR	0.15VR-	0.65MR-M

	Columbus					MR	
Besostaja	*34,3,* +	-	-	-	0.15VR-R	-	0.2MS
Chinese Spring	*34*, *12*	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.15VR-R
CS-Ditelosomic 7DS		0.1VR	0.15VR-R	0.1VR	0.1VR	0.1VR	0.25R
CS-Ditelosomic 7DL		0.1VR	0.15VR-R	0.1VR	0.1VR	0.1VR	0.3VR-M
Pavon 76		0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR
Ciano 67		0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR
Chris		0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR
Era		0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR
Marco Juarez Inta		0.1VR	0.1VR	0.1VR	0.1VR	0.1VR	0.1VR
Cappelle Desprez		-	-	-	2R-MR	-	1M
Nord Desprez		-	-	-	70MS-S	-	45MS
Bersee		-	-	-	27.5M	-	16.5R-MS
Armada		2MS	20M-MS	2.5MS	5M	2MS	2.5M
Thatcher		47.5S	70MS-S	1.5M	25MR-M	45S	70MS
Lr19	19	0.1VR	0.1VR	0.15VR-R	0.1VR	0.15VR-R	0.1VR

* Score1 and Score2 are readings taken on 30.06.93 and 12.07.93 respectively.

* These letters represent infection types, where VR = Very resistant; R = Resistant; MR = Moderately resistant; M = Mixed reaction; MS = Moderately susceptible; S = Susceptible.

Pavon 76, Ciano 67, Chris, Era and Marco Juarez Inta all from CIMMYT, Chinese Spring, Chinese Spring ditelosomics 7DL and 7DS just like the *Lr34* lines were resistant to all three isolates in the field. Of the two susceptible controls, Armada was resistant to all the three isolates, while Thatcher was susceptible only to isolates WBRP 85-20 and WBRP 90-12. The resistant control *Lr19* had only traces of infection from the three isolates.

Table 2 shows the flag leaf reaction of wheat cultivars and lines known or suspected of having *Lr34* alone or in combination with other gene(s) in controlled environments set at 12°C and 26°C. All cultivars and lines with *Lr34* demonstrated some form of resistance to isolates WBRP 85-31 and WBRP 90-12 at both/either 12°C and/or 26°C. TBLs expressed higher levels of resistance against isolate WBRP 85-31 that were mostly characterized by IT ;. Isolate WBRP 90-12 had variable ITs of ; to 2+;.on all the Thatcher backcross lines. This variability was further demonstrated by the TBLs having *Lr34* and *LrT3* genes at the two temperature conditions. Lines RL6050 (*Lr34* and *LrT3*) and RL6070 (*Lr34* and *LrT3*) were more resistant at 26°C (ITs ;1+ and ;) than at 12°C (ITs ;2 and 2;), while line RL6069 (*Lr34* and *LrT3*) was more resistant at 12°C (IT ;1+) and less so at 26°C (IT ;2+), and line RL6106 (*Lr34* and *LrT3*) had IT 2+ at both temperatures.

Table 2. Flag leaf ITs of wheat cultivars and lines known or suspected of having *Lr34* alone or *Lr34* in combination with different genes at different temperatures.

***Lr* line/ Cultivar**	***Lr* gene**	**Seedlings ITs**			
		WBRP 85-31		**WBRP 90-12**	
		12°C	**26°C**	**12°C**	**26°C**
RL6058	*34*	;	;	;	;1
RL6091	*34*	;	;	;2	;1
91RN514	*34*	;	;	;2	;2+
91RN516	*34*	;	;	;2+	2+;
86RN122	*34*	;	;1-	;3	;2
86RN127	*34*	;	;	2+;	2+;

90RN1220	*34*	;	;	;2	;2+
RL6050	*34, T3*	;	;	;2	;1+
RL6069	*34, T3*	;	;1	;1+	;2+
RL6070	*34, T3*	;2	2;	2;	;
RL6106	*34, T3*	;	;	2+	2+
RL6108	*34, 1*	;	;	;	;
RL6109	*34, 2a*	;	;	;	;1-
RL6110	*34, 3*	;	;	;	;
RL6111	*34 ,3ka*	;	;	;	;
RL6112	*34 ,10*	;	;	;2	2;
RL6113	*34, 11*	;	;1-	;2	;2+
RL6114	*34, 13*	;	;	;2	2;
RL6115	*34 ,16*	;	;	2;	3-;
RL6116	*34 ,17*	;	;	2;	;1
RL6117	*34 ,18*	;	;	;	3-;
RL6118	*34, 21*	;	;	;1	1;
RL6119	*34, 26*	;	;	;2+	;
RL6120	*34, B*	;	;	2+;	3-;
RL6121	*34,* Columbus	;	;	2+;	2;
Bezostaja	*34, 3, +*	;	;	;	;
Chinese Spring	*34, 12, +*	;	X;	;2+	;14
CS-Ditelosomic 7DS		;1	;23+	;1+	;14-
CS-Ditelosomic 7DL		;	;2+	;1	;14-
Cappelle Desprez		;2+	23;	;2	;2
Nord Desprez		3-;	23+;	3+;	;4-
Bersée		;2+	31;	3+;	42;
Armada		;3	;1+	2+;	;1
Thatcher		1-;	;1-	1-	;1-
Lr19	*19*	;	;	;	;

Bezostaja had high level of resistance against both isolates across temperatures with IT ; to ;1. With isolate WBRP 85-31, Chinese Spring and its ditelosomics 7DL and 7DS were more resistant at 12°C than at 26°C. Similar trends were observed with isolate WBRP 90-12 on these lines. Adult plants of both Cappelle Desprez and Bersée were resistant to isolate WBRP 85-31, at 12°C and less so at 26°C. Cappelle Desprez was also resistant to isolate WBRP 90-12, at both temperatures. Nord Desprez had slight resistance against both isolates at 26°C, but not at 12°C. The flag leaf of the susceptible controls Armada and Thatcher expressed resistance against the two isolates across temperatures, with Thatcher having IT ;1-. The resistant control, line *Lr19* had IT ; across temperatures with both isolates. Table 3 has the seedling ITs of wheat cultivars and lines known or suspected to have *Lr34* alone or in combination with other gene(s) at 5°C and 20°C with various isolates. TBLs having *Lr34* alone had variable ITs at 5°C. Line RL6058 (*Lr34*) the original standard single gene line for *Lr34* had ITs ; to 3 with different isolates at 5°C. The same line had mostly ITs 2 and 2+ with the occasional IT 3- at 20°C. Similar response patterns were observed on lines RL6091, 91RN514 and 86RN122 all with *Lr34* from different sources. The TBL, RL6050 with *Lr34* and *LrT3* had IT ; to ;1- at 5°C and IT 2+ to 3- at 20°C with all the 16 isolates. Also Chinese Spring had IT ; at 5°C with all isolates and variable resistant reaction at 20°C with different isolates. Cappelle Desprez, Nord Desprez and Bersée were susceptible to most isolates, apart from the resistance of Cappelle Desprez to isolates WBRP 90-24, WBRP 85-31 WBRP 90-10, WBRP 90-11 and WBRP 90-12; Bersée to WBRP 90-24, WBRP 90-10 and WBRP 90-12; Nord Desprez to WBRP 90-10, WBRP 90-11 and WBRP 90-12.

Table 3. Seedling leaf rust reactions of wheat cultivars and lines having or suspected of having *Lr34* alone or *Lr34* in combination with other genes at different temperatures.

***Lr* line/ Cultivar**	***Lr* gene**	**WBRP 61-37**		**WBRP 76-1**		**WBRP 76-2**		**WBRP 80-1**		**WBRP 81-2**		**WBRP 81-5**		**WBRP 82-1**		**WBRP 90-24**	
		5°C	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**
RL6058	*34*	;1+	2+	;	2+	;2	2+	2+	2+	2+	2+	;1+	2	1+;	2+	;1	2
RL6091	*34*	2+;	2+	;	2+	2+;	2+	3-	3-	3-	3-	2+	2	3-	2+	1+;	2+
91RN514	*34*	2+;	2+	2+;	2+	3-	2+	2	2+	3-	3-	2	2+	2+	2+	;1-	2+
86RN122	*34*	2;	2+	;	2+	2+	2+	2	3-	2+	2+	2	2+	2;	2+	1+	2+
RL6050	*34, T3*	0i	2+	0i	2+	;	2+	;	3-	;	2+	;	2+	;	2+	;	2+
Chinese Spring	*34 ,12, +*	;	X-;	0i	;	;	2;	;	;	;	X;	;	2+	;	2+;	;	X;
Cappelle Desprez		3	3	3-	3	3-	3-	3+	3	3+	3	3-	3-	3-	3-	2	2+
Nord Desprez		3+	3	3-	3	3	3	3+	3+	3+	3	3-	3-	3	3-	3-	3-
Bersée		3	3	3-	3	3	3	3	3	3+	3+	3-	3-	3	3-	2+	3-
Armada		3	3	3	3+	3+	3	3+	3	3+	3	3	3-	3	3	3-	3-
Thatcher		3	3	3-	3	3-	3	3+	3	3+	3	3-	3-	3	3	2	3-
Lr19	*19*	;	;	;	;	;	;	;	;	;	;	;	;	;	;	;	;
***Lr* line/ Cultivar**	***Lr* gene**	**WBRP 90-25**		**WBRP 85-20**		**WBRP 85-31**		**WBRP 86-8**		**WBRP 90-26**		**WBRP 90-10**		**WBRP 90-11**		**WBRP 90-12**	
		5°C	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**	**5°C**	**20°C**
RL6058	*34*	3	2	2+	3-	2;	2+	2;	2+	2	2+3-	;	2+	;1	2+	;	2+
RL6091	*34*	3	2+	2	2+	2;	2+	2;	2+	3-	3-	1+;	2+	;12+	2+	1+;	2+
91RN514	*34*	3	2+	2+	2+	2+	2+	2+3-	3-	3	3-	1;	2+	2+	2	1+;	2+
86RN122	*34*	3	2+	2+	2+	2+;	2+	2;	2	2+3-	3-	1;	2+	;1	2	1+;	2+
RL6050	*34, T3*	;	2+	;	2+	;	2+	;	2+	;	3-	;	2	;1-	22+	;	2+
Chinese Spring	*34, 12, +*	;	;X+	;	X;	;	;	;	X;/3-;	;	3-	;	2+	;	3-;	;	2+/3-;
Cappelle Desprez		3	3-	3	3-	3-	2+	3-	3	33+	3+	22+	3-	2+	3-	2	3-
Nord Desprez		3+	3+	3	3	3	3	3	3	3+	3+	2+	3-	2+3-	3-	2+	3-
Bersée		4	3+	3-	3	3	3	3	3	4-	3	2+	3-	3-	3-	2+	3
Armada		4-	3+	3	3	3+	3	3	3	4	3+	3-	3-	3-	3	3-	3
Thatcher		3	3+	3-	3	3+	3	3	3	3+	3	2+	3-	2+	3-	2+	3
Lr19	*19*	;	;	;	;	;	;	;	;	;	;	;	;	;	;	;	;

Of the two susceptible controls, Armada was susceptible (ITs 3- to 4) to all isolates at the two temperatures, while Thatcher was resistant (ITs 2 to 2+) to isolates WBRP 90-24, WBRP 90-10, WBRP 90-11 and WBRP 90-12 at 5°C only. The resistant control, line *Lr19* had IT ; with all 16 isolates across temperatures.

Discussion

In this study all the TBLs with *Lr34*, either alone or in combination with different genes were resistant in the field to isolates WBRP 85-31, WBRP 85-20, and WBRP 90-12. However the level of resistance depended on the source and gene combination of the line.

In the field, of the two susceptible control cultivars, Armada appeared more resistant, this resistant reaction is attributed to a combination of factors. At the time of scoring, Armada plants were at growth stage 20-29 (Zadoks et al., 1974), having a rosette appearance with a relatively large number of tillers and a main shoot in some instances. Due to the winter habit of Armada, seedlings were vernalized in growth chambers set at 4°C for 45 days before being transplanted into the field in the spring. The vernalization treatment might have been inadequate, resulting in the vegetative growth. The second susceptible control, Thatcher, resulted sensitive to isolates WBRP 85-20 and WBRP 90-12, and resistant to WBRP 85-31. The resistant reaction of Thatcher against WBRP 85-31 is probably due to Lr22b identified in Thatcher (Dyck, 1979). The avirulence of isolate WBRP 85-31 on the *Lr22b* is further supported by the increased flag leaf resistance of TBLs having *Lr34* either alone or in combination with different genes in controlled environments to the isolate.

Temperature controls are often used to identify resistance genes that are sensitive to environmental changes, and use similarities in their trend with known resistance genes to postulate the presence of such genes. In this study despite having the resistance genes *Lr34* and *LrT3* adult plants of lines RL6050, RL6069, RL6070 and RL6106 had variable resistant reactions against isolates WBRP 85-31 and WBRP 90-12 at 12°C and 26°C.On the whole the resistance gene *Lr34* alone or in combination with different genes was effective against these two UK isolates. However there was no consistency in the expression of the *Lr34* gene in either of the two temperature conditions across the different genetic backgrounds. More examples of the complex interactions of the *Lr34* resistance with both its genetic and physical environments are displayed by the TBLs RL6115 (*Lr34* and *Lr16*), RL6117 (*Lr34* and *Lr18*) and RL6120 (*Lr34* and *LrB*) reacting with isolate WBRP 90-12 in controlled environments and the field. These lines showed a shift from resistance at 12°C to susceptible IT at 26°C. The resistance gene *Lr34* alone is known to be more effective at low temperature (Dyck and Samborski, 1982), while *Lr16* and *LrB* in Thatcher background were ineffective against isolate WBRP 90-12 in the field (Abdul, 1994). Also the resistance gene *Lr18* is known to be effective at low but not at high temperature. As expected, a combination of any of these genes with *Lr34* in Thatcher background resulted in a susceptible reaction at 26°C. However, the same lines were resistant to this isolate in the field.

The flag leaves of Bezostaja were resistant in the controlled environments. Bezostaja is known to have the seedling resistance gene *Lr3* (Bartoš, 1969) in addition to the *Lr34* gene. No virulence has been detected for the *Lr3* gene among UK isolates (Abdul, 1994).

Flag leaves of Cappelle Desprez, Nord Desprez and Bersée expressed varying levels of resistance in the controlled environments, however only Cappelle Desprez and Bersée maintained this reaction in the field.

The resistance reaction expressed by the flag leaves of Armada and Thatcher against isolates WBRP 85-31 and WBRP 90-12 in controlled environments was not effective in the field. This flag leaf resistance may be due to temperature and light conditions within the controlled environment.

The resistance gene *Lr34* is located in chromosome 7D (Dyck, 1987), however its arm location is yet to be determined. In this study assessment of ditelosomic 7DL and 7DS of Chinese Spring, known to have *Lr34* (Dyck, 1991) failed to reveal any differences in their

reaction to different isolates in controlled environments as well as in the field. However, in addition to the *Lr34* gene, Chinese Spring is known to have the resistance gene *Lr31*, one of a complementary pair (*Lr31* + *Lr27*) required for the expression of resistance, located on chromosome 4Ab (Singh and McIntosh, 1984a) and the APR *Lr12*. It is possible that the resistance of the ditelosomics was due to *Lr12* or interactions between *Lr12* and other unidentified gene(s).

Temperature controls have been used to detect adult plant genes with avirulent isolates at the seedling stage. Using this procedure Pretorius et al. (1984) detected *Lr13* at 25.5°C and *Lr34* at 7°C (Drijepondt et al., 1991). In this study seedling of TBLs having *Lr34* alone, demonstrated variations in their reaction at a given temperature. At 5°C the ITs ; to 3 were recorded and at 20°C ITs 2 to 3- were obtained depending on the isolate. Since all the lines were evaluated at the same time, under similar conditions, the variation in the ITs must be due to the genetic background of both the host lines and pathogen isolates. The *Lr34* gene was transferred into the Thatcher background from different sources and Stam and Zeven (1981) reported that often linked with such transfers are genes that are closely associated with the marker locus that may contribute to the differences between backcross lines. Similar genetic interactions leading to variable expression of the *Lr34* gene at the seedling stage have been reported by Singh (1992a) and German and Kolmer (1992). In addition (Kolmer 1999a, 2001) reported that the resistance conditioned by *Lr34* is nonspecific, since isolates that are fully virulent to this gene have not yet been detected.

Additive genetic interaction between *Lr34* and *LrT3* is likely to be responsible for the high level of resistance recorded on seedlings of lines with this gene combination at low temperature. A similar phenomenon has been reported by Drijepondt et al. (1991). They reported that the resistance genes *Lr34* and *LrT3* individually are more effective at low temperature and in combination they produced IT that is lower than their individual ITs with the same isolate.

The data obtained show that the expression of the *Lr34* gene is subject to environmental changes, both genetic and physical (temperature). Temperature control did not give a fixed and repeatable IT in the interaction between the leaf rust resistance gene *Lr34* and different UK isolates of *Puccinia triticina*, but it appeared to be more effective at low temperatures. In most instances the resistance gene *Lr34* alone and in combination with other genes was effective against all the UK isolates used in this investigation. The *Lr34* gene was equally postulated in the wheat cultivars Pavon 76, Ciano 67, Chris, Era and Marco Juarez Inta based on similarity in their reactions in the field. Low temperatures at both seedling and adult plant growth stages in controlled environment could be used in conjunction with the leaf tip necrosis marker (Dyck, 1991; Singh, 1992c) and other molecular markers for the identification of the *Lr34* that confer durable resistance to leaf rust disease of wheat.

Acknowledgement

I wish to thank the Cambridge Commonwealth Trust, The British Society for Plant Pathology and the Cambridge Laboratory, John Innes Centre for Plant Science Research, Norwich, England for their financial assistance during the course of this study. I also wish to thank Mr. A.J. Worland of the Cambridge Laboratory, John Innes Centre for Plant Research for providing me with the substitution lines.

References

Abdul, S. D. 1994. *Brown rust of wheat: Temperature sensitivity, genetic analysis and pathogen variation.* Ph. D. Thesis. University of Cambridge, Cambridge, UK.

Bartoš, P., D. J. Samborski and P. L. Dyck. 1969a. Leaf rust resistance of some European varieties of wheat. Canadian J. Bot. 47:543-546.

Drijepondt, S. C., Z. A. Pretorius and F. H. J. Rijkenberg. 1991. Expression of two

wheat leaf rust gene combinations involving *Lr34*. Plant Dis. 75:526-528.

Dyck, P. L., D. J. Samborski and R. G. Anderson. 1966. Inheritance of adult plant leaf rust resistance derived from the common wheat varieties Exchange and Frontana. Canadian J. Gen. Cytol. 8:665-671.

Dyck, P. L. and E. R. Kerber. 1971. Chromosome location of three genes for leaf rust resistance in common wheat. Canadian J. Gen. Cytol. 13:480-483.

Dyck, P. L. 1977. Genetics of leaf rust reaction in three introductions of common wheat. Canadian J. Gen. Cytol. 19, 711-716.

Dyck, P. L. 1979. Identification of the gene for adult-plant leaf rust resistance in Thatcher. *Canadian Journal of Plant Science* 59, 499-501

Dyck, P. L. and D. J. Samborski. 1979. Adult-plant leaf rust resistance in PI 250413, an introduction of common wheat. Canadian J. Plant Sci. 59:329-332.

Dyck, P. L. and D. J. Samborski. 1982. The inheritance of resistance to *Puccinia recondita* in a group of common wheat cultivars. Canadian J. Gen. Cytol. 24:273-283.

Dyck, P. L. 1987. The association of a gene for leaf rust resistance with the chromosome 7D suppressor of stem rust resistance in common wheat. Genome 29:467-469.

Dyck, P. L. 1991. Genetics of adult-plant leaf rust resistance in `Chinese Spring' and `Sturdy' wheats. Crop Sci. 31:309-311.

Dyck, P. L. 1992. Inheritance of leaf rust and stem rust resistance in `Roblin' wheat. Genome 36:289-293.

German, S. E. and J. A. Kolmer. 1992. Effect of *Lr34* in the enhancement of resistance to leaf rust of wheat. Theor. App. Gen. 84:97-105.

Johnson, R. and C. N. Law. 1975. Genetic control of durable resistance to yellow rust (*Puccinia striiformis*) in the wheat cultivar Hybrid de Bersée Annal. App. Biol. 81:385-391.

Johnson, R. 1994. Developments in prospects of breeding for durable resistance to the three rust diseases of wheat. In: Developing Sustainable Wheat Production Systems. The 8th Regional Wheat Workshop for Eastern, Central and Southern Africa. Addis Ababa, Ethiopia: CIMMYT. pp 123-140.

Kolmer, J. A. 1999. Physiologic specialization of *Puccinia triticina* in Canada in 1997. Plant Dis. 83:194–197.

Kolmer, J. A. 2001. Physiologic specialization of *Puccinia triticina*in Canada in 1998. Plant Dis. 85:155–158.

Kolmer, J. A. and J. Q. Liu 2002. Inheritance of leaf rust resistance in the wheat cultivars AC Majestic, AC Splendor, and AC Karma. Can. J. Plant Pathol. 24:327–331

Kolmer, J. A., R. P. Singh, D. F. Garvin, L. Viccars, H. M. William, J. Huerta-Espino, F. C. Ogbonnaya, H. Raman, S. Orford, H. S. Bariana and E. S. Lagudah. 2008. Analysis of the *Lr34/Yr18* rust resistance region in wheat germplasm. Crop Sci. 48:1841–1852.

Law, C. N., R. C. Gaines, R. Johnson and A. J. Worland. 1978. The application of aneuploid techniques to a study of stripe rust resistance in wheat. In: Ramanujam (Ed.) pp. 427-436, Proceedings of the fiveth Internation Wheat Genetics Symposium, New Delhi, India.

McIntosh, R. A., Baker EP, 1966. Chromosome location of mature plant leaf rust resistance in Chinese Spring wheat. Australian J. Biol. Sci. 19:943-944.

McIntosh, R. A. 1992. Close genetic linkage of genes conferring adult-plant resistance to leaf rust and stripe rust in wheat. Plant Pathol. 41:523-527.

Pretorius, Z. A., R. D. Wilcoxson, D. L. Long and J. F. Schafer. 1984. Detecting wheat

leaf rust resistance gene *Lr13* in seedlings. Plant Dis. 68:585-586.

Roelfs, A. P. 1988b. Resistance to leaf and stem rusts in wheat. In: Breeding Strategies for Resistance to the Rusts of Wheat, CIMMYT, Mexico City. pp. 10-22.

Sears, E. R. 1988. History of the Chinese Spring aneuploids. In: T. E. Miller and R. M. D. Koebner (Eds.), Proceedings of the Seventh International Wheat Genetics Symposium, 13-19 July, IPSR, Cambridge, England.

Shang, H. S., P. L. Dyck and D. J. Samborski. 1986. Inheritance of resistance to *Puccinia recondita* in a group of resistant accessions of common wheat. Canadian J. Plant Pathol. 8:123-131.

Singh, R. P. and R. A. McIntosh. 1984a. Complementary genes for reaction to *Puccinia recondita tritici* in *Triticum aestivum*: I. Genetic and linkage studies. Canadian J. Gen. Cytol. 26:723-735.

Singh, R. P. 1992a. Expression of wheat leaf rust resistance gene *Lr34* in seedlings and adult plants. Plant Dis. 76:489-491.

Singh, R. P. 1992b. Genetic association of leaf rust resistance gene *Lr34* with adult plant resistance to stripe rust in bread wheat. Phytopathol. 82:835-838.

Singh, R. P. 1992c. Association between gene *Lr34* for leaf rust resistance and leaf tip necrosis in wheat. Crop Sci. 32:874-878.

Singh, R. P. and A. K. Gupta. 1992. Expression of wheat leaf rust resistance gene Lr34 in seedlings and adult plants. Plant Dis. 76:489-491.

Singh, R. P. and S. Rajaram. 1992. Genetic of adult plant resistance of leaf rust in `Frontana' and three CIMMYT wheats. Genome 35:24-31.

Stakman, E. C., D. M. Stewart and W. Q. Loegering. 1962. Identification of physiologic races of *Puccinia graminis* var. tritici. Washington, USA: United States Department of Agriculture: US, Agric. Res. Serv. E-617 (revised), 1-53.

Stam, P. and A. C. Zeven. 1981. The theoretical proportion of the donor genome in near-isogenic lines of self-fertilizers bred by backcrossing. Euphytica 30:227-238.

Zadoks, J. C., T. T. Chang and C. F. Konzak. 1974. A decimal code for the growth stages of cereals. Weed Res. 14:415-421.