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Appraisal of wheat germplasm for adult plant resistance against stripe rust

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Abstract: The resurgence of wheat stripe rust is of great concern for world food security. Owing to resistance breakdown and the appearance of new virulent high-temperature adapted races of *Puccinia striiformis* f. sp. *tritici* (Pst), many high yielding commercial varieties in the country lost their yield potential. Searching for new sources of resistance is the best approach to mitigate the problem. Quantitative resistance (partial or adult plant) or durable resistance is reported to be more stable than race specific resistance. In the current perusal, a repertoire of 57 promising wheat lines along with the KLcheck line Morocco, developed through hybridisation and selection of local and international lines with International Maize and Wheat Improvement Center (CIMMYT) origin, were evaluated under natural field conditions at Nuclear Institute for Agriculture and Biology (NIAB) during the 2012–2013 and 2013–2014 time periods. Final rust severity (FRS), the area under the rust progress curve (AURPC), the relative area under the rust progress curve (rAURPC), and the coefficient of infection (CI) were unraveled to infer the level of quantitative resistance. Final rust severity was recorded when the susceptible check exhibited 100% severity. There were 21 lines which were immune (no disease), 16 which were resistant, five moderately resistant, two resistant-to-moderately resistant, one moderately resistant-to-moderately susceptible, 5 moderately susceptible-to-susceptible, one moderately susceptible, and six exhibited a susceptible response. Nevertheless, 51 lines exhibited a high level of partial resistance while the three lines, NW-5--1212-1, NW-7-30-1, and NW-7-5 all showed a moderate level of partial resistance based on FRS, while 54 lines, on the basis of AURPC and rAURPC, were identified as conferring a high level of partial resistance. Moreover, adult plant resistance was conferred by 47 wheat lines, based on CI value. It was striking that, 13 immune lines among 21 were derived from parents of CIMMYT origin. Cluster analysis was executed to determine the diversity among the wheat genotypes based on stripe rust resistance and yield parameters. All genotypes were grouped into nine clusters exhibiting a high level of diversity at a 25% linkage distance. There were 29 wheat lines resistant to stripe rust that were grouped into the first three clusters, while 4 high-yielding lines were in Cluster VIII. The susceptible check, Morocco, was separated from rest of lines and fell in the last cluster i.e. Cluster IX. Based on the results, inter-crossing immune/resistant lines is recommended, and with high yielding lines – it is also recommended that cultivars with improved disease resistance and yield potential be developed.

Key words: durable resistance, food security, pathogen, stripe rust, wheat

Introduction

Stripe rust of wheat, caused by *Puccinia striiformis* f. sp. *tritici* (Pst), incurred serious wheat-yield loss around the globe (Singh *et al*. 2005; Ali *et al*. 2009a; Hovmoller *et al*. 2010; Hovmoller *et al.* 2011; De Vallavieille-Pope *et al*. 2012). The pathogen is present in wheat growing areas of temperate, moist, and cooler regions of the world, except the Antarctica (Chen 2005). In the case of highly susceptible varieties, loss may escalate up to 80% (Kolmer 1996; Beard *et al*. 2007). The causal pathogen Pst, reduces the total photosynthetic area, utilises plant assimilates, and interrupts the normal growth of the host, which leads to the shriveling of the grains. Moreover, the physical and biochemical quantitative characteristics of wheat are negatively influenced. Newly spreading, high-temperature adapted races of Pst were reported from many countries which made this pathogen even more notorious (Ali *et al*. 2014). Pakistan faced four major stripe rust epidemics. The intensity of the epidemics exceeded 20% in 1973 (35%), 1978 (55%), 1995 (37.5%), and 2003 (20%). Stripe rust intensity has never gone below 8% in the country's history (Ahmad 2004). Pst infects the wheat crop in winter or early spring or at cooler areas. Generally, the temperature for stripe rust urediniospores to geminate and for disease epidemics ranges from 2 to 15°C (Hogg *et al*. 1969). Development of yellow stripes of urediniospores on wheat leaves is the characteristic symptom of Pst in comparison with leaf and stem rust. Once yellow uredinia stripes appear on flag leaf, the uredinia can cover the whole leaf within a short time under favourable weather conditions.

New and more effective fungicides like Tilt® (propiconazole), Quadris® (azoxystrobin), Stratego™ (propiconazole + trifloxystrobin), and Quilt™ (azoxystrobin +

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propiconazole) are available to control stripe rust. The use of these fungicides, however, is uneconomical and ecologically unsafe (Chen 2005). Growing wheat cultivars which have resistant genes against Pst, remains the most inexpensive, efficient, environmentally friendly, and sustainable approach to manage the disease. Screening of varieties against stripe rust is a regular activity due to the dynamic evolutionary nature of the pathogen. New races of Pst evolved quickly through mutation and somatic hybridisation (Stubbs 1985). Due to their air-borne dispersal mechanism and rapid multiplication, local races can migrate to other areas and quickly become regionally and often globally predominant. Thus, virulence has been reported for many *Yr* genes worldwide (Singh *et al*. 2002). Two types of resistance have been identified in several cereal-rust pathosystems: hypersensitive or qualitative (race specific) and quantitative (race-nonspecific) resistance. Race-specific resistance is highly effective but not durable because cultivars with such a type of resistance often become susceptible within a few years of release either as a consequence of selecting previously rare races or as a consequence of the development of new races (Wellings and McIntosh 1990; Line and Qayoum 1992; Chen 2005, 2007). Conversely, race-nonspecific resistance is mainly polygenic and is known to be long-lasting and more durable (Herrera-Fossel *et al*. 2007).

Up till now, more than 50 genes for stripe rust resistance have been identified and named while many other genes or quantitative trait loci (QTL) have been tentatively designed (McIntosh *et al*. 2008; Chen 2013). The majority of these *Yr*-genes are race specific, though, and become ineffective due to the development of new races. The average lifetime of the genes conferring race specific resistance is estimated to be five years on a global basis (Kilpatrick 1975). Pathogen also keeps its momentum to evolve in rapid ways, therefore there is a dire need to investigate new sources of resistance against Pst. Keeping in mind the role of wheat as a staple food, the present study was conducted to identify the resistance potential in some commercial varieties and promising wheat lines developed at the Nuclear Institute for Agriculture and Biology (NIAB), Faisalabad, Pakistan.

Materials and Methods

Plant material

Winter wheat germplasm comprising 57 lines, along with the susceptible check Morocco (Table 1) were screened for stripe rust resistance under natural conditions at the field area of NIAB, Faisalabad for two years (2012–2013 and 2013–2014). Each entry was planted in two rows (the rows were 2 m in length) spaced at 30 cm. Morocco was planted as every 10th row and as a spreader border around the nursery to ensure uniform infection. The material was screened in natural conditions at the booting stage and no artificial inoculation was carried out. The experiment was conducted using a randomised complete block design with three replications. Recommended cultural practices were used for trial management.

Disease assessment

Stripe rust was scored four times with one week interval, when leaves of the susceptible check Morocco acquired up to a 50% severity of the disease. Observation of the host response against stripe rust was recorded following Roelfs *et al*. (1992), while severity was recorded as the % of infection on the plants according to modified Cobb's Scale (Peterson *et al*. 1948). The coefficient of infection (CI) was derived by multiplying disease severity (DS) and constant values of infection type (IT). The constant values for infection types were: R = 0.1, MR = 0.25, M = 0.5, MS = 0.75, S = 1 (Pathan and Park 2006).The area under the rust progress curve (AURPC) and the relative area under rust progress curve (rAURPC) was derived through the formula described by Milus and Line (1986):

$$\text{AURPC} = [N_1(X_1 + X_2)/2] + [N_2(X_2 + X_3)/2] + \\ + [N_2(X_3 + X_4)/2],$$

where: X_1, X_2, X_3, X_4 – the rust intensities recorded on the first, second, third, and fourth dates; N_1 – the interval day between X_1 and X_2, and N_2 – the interval day between X_2 and X_3.

$$\text{rAURPC} = [\text{line AUDPC/susceptible AUDPC}] \times 100.$$

Statistical analysis

The mean disease scores were subjected to analysis of variance (ANOVA) in order to detect differences between individual wheat lines. The difference between the mean disease parameters of Morocco and the mean disease scores of the individual wheat lines were calculated using the *t*-test. Cluster analysis of mean data was conducted by using Minitab software, version 16.

Results

Parameters used as criteria to appraise the level of partial resistance in tested germplasm of bread wheat were final rust severity (FRS), AURPC, rAURPC, and CI, which are detailed below.

Final rust severity (FRS)

When the susceptible check showed 100% FRS, all the tested lines were observed for final stripe-rust severity. Significant differences (p = 0.001) were observed among the mean score of 57 wheat lines compared to the check Morocco for FRS (Table 2). The maximum FRS of up to 100% was recorded for Morocco, followed by line 10-C (70%), 5-C (63.3%), NW-7-30-1 (40%), NW-5-1212-1 (40%), and 14-C (40%). Twenty wheat lines (37%) were immune with 0 FRS throughout the season. The evaluated genotypes of wheat were categorised into three groups based on their partial resistance i.e. high, moderate, low level, having 1–30%, 31–50%, and 51–70% FRS, respectively.

Table 1. Parentage of wheat lines evaluated for adult plant resistance during the 2012–2013 and 2013–2014 time period

Wheat germplasm	Parentage/Pedigree	Origin	Wheat germplasm	Parentage/Pedigree	Origin
23-C	SUPER KAUZ/KS94U215// SUPERKAUZ	CIMMYT	NW-1-20	NARC-234/NARC-241-20	Pakistan
NIAB-1	Chirya-3 / Opata // 2 * Parula /3/ Rohtas 90	CIMMYT	15-C	BABAX/LR43//BABAX	CIMMYT
NW-10-4	NARC 241/BHITTAI-4	Pakistan	NW-10-1111-3	NR-241/Bhittai-1111-3	Pakistan
NW-5-1212-1	NARC-234/Bhitaai-3341-7	Pakistan	14-C	SARSABZ/NARC-234-116-1	Pakistan
4-C	NARC-234/SARSABZ-15-1	Pakistan	17-C	PASTOR/KAUZ	CIMMYT
NW-10-1111-7	NACR-241/Bhittai-1111-7	Pakistan	NW-2-8	NARC-234/NESSER-2242-8	CIMMYT
NW-10-18	NARC 241/BHITTAI-18	Pakistan	NW-7-5	NARC-241/NESSER-6123-5	CIMMYT
3-C	NARC-234/NARC-241-11-48	Pakistan	2-C	NARC-234/NARC-241-4-26	Pakistan
19-C	SUNSU	CIMMYT	26-C	W462/WEERY/KOEL/3/PEREGRINE /MAIORAL/BUCKBUCK	CIMMYT
NIAB-2	Weebil /4/ Sabuf *2 / Weebil // Pavon 76 /3/ MH 97	CIMMYT	NW-7-30-1	NARC-241/Nesser-30-1	CIMMYT
5-C	NARC-234/Bhitaai-20-1	Pakistan	11-C	NESSER/BHITTAI-111-3	CIMMYT
NW-10-1111-37	NARC-241/Bhittai-1111-37	Pakistan	1-C	NARC-234/NARC-241-2-21	Pakistan
NW-1-9-47	NARC-234/NR-241-9-47	Pakistan	13-C	NESSER/BHITTAI-115-6	CIMMYT
NW-17-5	SARSABZ/NARC-241-5	Pakistan	NW-1-47	NARC-234/NARC-241-47	Pakistan
NW-1-39	NARC-234/NARC-241-8183-39	Pakistan	NW-1-52	NARC-234/NR-241-8188-52	Pakistan
NW-1-38	NARC-234/NR-241-4343-38	Pakistan	NW-10-45	NARC-241/BHITTAI-45	Pakistan
21-C	SUPER KAUZ*2/ FLYCATCHER	CIMMYT	NW-10-16	NARC-241/BHITTAI-3349-16	Pakistan
10-C	NESSER/SARSABZ-106-13	CIMMYT	28-C	BOBWHITE/MOR/BAGULA	CIMMYT
NW 2-2	NARC-234/NESSER-2247-2	CIMMYT	7-C	NARC-241/BHITTAI-56-1	Pakistan
NW-7-28	NARC-241/NESSER-28	CIMMYT	29-C	SITTA CM77091-14Y-04M-06Y-3B-1Y-0B	CIMMYT
NW-1-8183-8	NARC-234/NR-241-8183-8	Pakistan	24-C	SIRKKU	CIMMYT
6-C	NARC-241/NESSSER-37-17	CIMMYT	NW-3-3341-7	NARC-234/Sarsabz-3341-7	Pakistan
18-C	TOROCAHUI S2004	CIMMYT	27-C	OTUS	CIMMYT
22-C	KAUZ/PASTOR	CIMMYT	NW-10-1111-5	NARC-241/Bhittai-1111-5	Pakistan
20-C	WEAVER/4/NACOZARI F 76/ TH.ACUTUM//3*PAVON F 76/3/MIRLO/BUCKBUCK	CIMMYT	25-C	IRENA/KAUZ	CIMMYT
NW-1-35	NARC-234/NARC-241-9561-35	Pakistan	NW-10-19	NARC-241/BHITTAI-19	Pakistan
NW-7-14	NARC-241/NESSER-6123-14	CIMMYT	8-C	NARC-241/BHITTAI-57-2	Pakistan
NW-2-4	NARC-234/NESSER-5445-4	CIMMYT	MOROCCO	SUSCEPTIBLE CHECK	Pakistan
NW-1-36	NARC-234/NR-241-8193-36	Pakistan			

Table 2. Mean comparison of final rust severity, infection type, area under rust progress curve, relative area under rust progress curve, and coefficient of infection in promising wheat lines to stripe rust

Wheat lines	FRS [%] the mean±SE	IT	AURPC [%] the mean±SE	rAURPC [%] the mean±SE	CI the mean±SE	APR
23-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NIAB-1	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW-10-4	5.0±0.0***	R	17.5±0.0***	1.0±0.0***	0.5±0.0***	high
NW-5-1212-1	40.0±0.0***	R-MR	333.7±16.3***	19.9±1.0***	12.0±0.0***	high
4-C	5.0±0.0***	R	38.5±0.0***	2.3±0.0***	0.5±0.0***	high
NW-10-1111-7	5.0±0.0***	R	17.5±0.0***	1.0±0.0***	0.5±0.0***	high
NW-10-18	10.0±0.0***	MR	105.0±0.0***	6.3±0.0***	2.5±0.0***	high
3-C	10.0±0.0***	R	128.3±11.7***	7.6±0.7***	1.0±0.0***	high
19-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NIAB-2	20.0±0.0***	R	210.0±0.0***	12.5±0.0***	2.0±0.0***	high
5-C	63.3±3.3**	MS	805.0±0.0***	47.9±0.0***	47.5±2.5**	low
NW-10-1111-37	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW-1-9-47	30.0±0.0***	MR	175.0±0.0***	10.4±0.0***	7.5±0.0***	high
NW-17-5	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW-1-39	10.0±0.0***	MR	60.7±4.7***	3.6±0.3***	2.5±0.0***	high
NW-1-38	10.0±0.0***	R	105.0±0.0***	6.3±0.0***	1.0±0.0***	high
21-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
10-C	70.0±2.9**	S	606.7±11.7***	36.1±0.7***	70.0±2.9**	low
NW 2-2	30.0±0.0***	MS-S	119.0±98.0**	7.1±5.8**	29.0±1.0***	moderate
NW-7-28	10.0±0.0***	R	70.0±0.0***	4.2±0.0***	1.0±0.0***	high
NW-1-8183-8	5.0±0.0***	R	52.5±0.0***	3.1±0.0***	0.5±0.0***	high
6-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
18-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
22-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
20-C	10.0±0.0***	R	35.0±0.0***	2.1±0.0***	1.0±0.0***	high
NW-1-35	30.0±0.0***	MS-S	385.0±0.0***	22.9±0.0***	25.5±1.5***	moderate
NW-7-14	28.3±1.7***	S	281.2±5.1***	16.7±0.3***	28.3±1.7***	moderate
NW-2-4	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW-1-36	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW1-20	11.7±1.7***	MR	99.2±5.8***	5.9±0.3***	2.9±0.4***	high
15-C	20.0±0.0***	MS-S	280.0±0.0***	16.7±0.0***	18.0±0.0***	high
NW-10-1111-3	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
14-C	40.0±0.0***	MS-S	420.0±0.0***	25.0±0.0***	36.0±0.0***	moderate
17-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW-2-8	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW-7-5	36.7±3.3**	S	210.0±0.0***	12.5±0.0***	36.7±3.3**	moderate
2-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
26-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW-7-30-1	40.0±0.0***	S	423.5±3.5***	25.2±0.2***	40.0±0.0***	moderate
11-C	16.7±3.3***	S	140.0±0.0***	8.3±0.0***	16.7±3.3***	high
1-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
13-C	30.0±0.0***	MS-S	315.0±0.0***	18.8±0.0***	27.0±0.0***	moderate
NW-1-47	30.0±0.0***	R	245.0±0.0***	14.6±0.0***	3.0±0.0***	high
NW-1-52	10.0±0.0***	R	70.0±0.0***	4.2±0.0***	1.0±0.0***	high
NW-10-45	10.0±0.0***	R	35.0±0.0***	2.1±0.0***	1.0±0.0***	high
NW-10-16	20.0±0.0***	MR-MS	140.0±0.0***	8.3±0.0***	12.0±0.0***	high
28-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
7-C	8.3±1.7***	R	71.2±1.2***	4.2±0.1***	0.8±0.2***	high
29-C	30.0±0.0***	R-MR	315.0±0.0***	18.8±0.0***	9.0±0.0***	high
24-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW-3-3341-7	5.0±0.0***	R	17.5±0.0***	1.0±0.0***	0.5±0.0***	high
27-C	5.0±0.0***	R	52.5±0.0***	3.1±0.0***	0.5±0.0***	high
NW-10- 1111-5	10.0±0.0***	R	56.0±0.0***	3.3±0.0***	1.0±0.0***	high
25-C	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
NW-10-19	0.0±0.0***	I	0.0±0.0***	0.0±0.0***	0.0±0.0***	high
8-C	10.0±0.0***	MR	70.0±0.0***	4.2±0.0***	2.5±0.0***	high
MOROCCO	100.0±0.0	S	1680.0±0.0	100±0.0	100.0±0.0	low

Classification: IT = infection type; I = immune; R = resistant; MR = moderately resistant; R-MR = resistant to moderately resistant; MR-MS = moderately resistant to moderately susceptible; MS-S = moderately susceptible to susceptible; S = susceptible; FRS = final rust severity; AURPC = area under rust progress curve; rAUPRC = relative area under rust progress curve; CI = coefficient of infection; PR = partial resistance; APR = adult plant resistance

Mean values of disease parameters are significantly different at p = 0.01 (**) and at p = 0.001 (***) from Morocco control

Coefficient of infection (CI)

Wheat lines having CI values of 0–20, 21–40, 41–60 were regarded as possessing high, moderate, and low levels of adult plant resistance (APR). The CI value of the susceptible check Morocco, was maximum (100) which shows high disease pressure. Forty-seven (82%) wheat lines possessed the CI value of 0–20 which conferred a high level of adult plant resistance, while Morocco (100), followed by 10-C (70) and 5-C (47.5), exhibited a low level of APR (Table 2). Only seven tested lines (12%) exhibited a moderate level of APR.

Area under rust progress curve (AURPC)

Wheat germplasm was also categorised into three groups of partial resistance based on AURPC value. Wheat lines having AURPC values up to 500 were grouped as having a high level of partial resistance, while those having AURPC values from 500–800 were categorised as having a moderate level of partial-resistance conferring lines. Similarly, lines having AURPC ranging from 800–1,100, were marked as having a low level of partial resistance. The susceptible check Morocco exhibitedthe maximum value of AURPC i.e. 1,680 lying at the lowest level of partial resistance followed by line 5-C having AURPC value of 805, while 10-C exhibit moderate level of partial resistance with AURPC values of 607. Fifty-four wheat lines (95%) had high levels of partial resistance (Table 1). Wheat lines falling under a moderate level of partial resistance first showed rust infection and sporulation but the final host reaction was characterized as chlorotic and necrotic strips (MR and/or MS infection types). Subsequently, rust development remained slower and restricted.

Relative area under rust progress curve (rAURPC)

As a parameter of partial resistance of slow stripe rusting, the rAURPC was also elucidated. The evaluated wheat germplasm was categorised into two distinct groups based on rAURPC value. When there were rAURPC values of up to 30% ofthe susceptible check Morocco, the lines were regarded as better slow rusting lines, while wheat lines having rAURPC values of up to 70% of Morocco, were marked as moderately slow rusting. The two wheat lines 5-C and 10-C, were marked as moderately slow rusting with rAURPC values of 48% and 36%, respectively. The rest of the lines were characterised as better slow rusting. Wheat lines in the first groups were composed of cultivars with varying degrees of slow rusting (Table 1). In lines exhibiting moderate levels of slow rusting, rust initiated and sporulated but the final MR/MS infection types subsequently led to slow rust development.

Yield potential and partial resistance

Significant variation was observed for yield and yield contributing parameters among the studied wheat germplasm. Maximum yield (kg · ha^{-1}) was recorded for the line NW-10-1111-3 (6,146) followed by NW-1-9-47 (6,125) and 14-C (6,048), having a relative AURPC value of 0, 10.4, and 25%, respectively (Table 3). Nevertheless, NW-1-9-47 and 14-C showed MR and MS-S reaction types. The susceptible check Morocco, having maximum disease severity and relative AURPC, produced the minimum grain yield (1,037) followed by wheat lines 6-C (2,788 kg · ha^{-1}). Yield difference within the repertoire of wheat lines was narrow but broader in comparison with Morocco. In addition to yield, a 1,000 grain weight was also determined to elucidate the effect of disease on the grain weight. NW-1-35 produced the maximum 1,000 grain weight (49.7 g) followed by 2-C (49.6 g) and 17-C (48.3 g) having a rAURPC value of 23 and 0%, respectively. The genotypes 2-C and 17-C were immune to stripe rust. Morocco, likewise carried a minimum 1,000 grain weight (21.59 g) followed by 29-C (30.7 g). A very minute difference in the 1,000 grain weight existed within the tested lines but the difference was quite large in comparison to Morocco.

Association between partial resistance parameters and yield components

Partial resistance in field conditions was assessed through FRS, AURPC, rAURPC, and CI. The most commonly used parameter to describe this association is CI. The relationship between these partial resistance parameters was elucidated for 57 wheat genotypes in this study, along with the susceptible check, at p = 0.001 (Table 4). A significant positive correlation of CI was observed with FRS (r = 0.93) and with both AURPC (r = 0.92) and rAURPC (r = 0.90). Moreover, a strong positive correlation was observed between FRS and AURPC and rAURPC (r = 0.94). Similarly, AUPRC and rAURPC had a complete correlation (r = 1). On the other hand, a negative correlation was observed between the partial resistance component and yield components. There was a negative correlation for 1,000 grain weight both AURPC and rAURPC (r = 0.39), followed by CI (r = 0.36), and FRS (r = 0.33). Likewise, yield per hectare was also negatively correlated with AURPC (23%) and rAURPC (23%).

Diversity among wheat genotypes

For selecting high yielding lines with disease resistance traits, cluster analysis was carried out. It is evident from the dendrogram (Fig. 1), that a considerable level of diversity was observed for the level of partial resistance and yield components among wheat breeding lines. All of the genotypes were divided into nine clusters at a 25% linkage distance according to their resistance potential (mean values of FRS, AURPC, rAURPC, CI), yield, and 1,000 grain weight. There were 9, 6, 14, 5, 9, 2, 7, 4, and one wheat lines in Cluster I, II, III, IV, V, VI, VII, VIII, and IX, respectively (Table 5). Genotypes with optimum stripe rust resistance were grouped in Cluster I, II, and III while four wheat lines (14-C, NW-10-1111-3, NW-1-35, and NW-7-5) with a maximum yield and grain weight fall in Cluster VIII. Wheat lines in Cluster I, II, and III exhibited the maximum potential for adult plant resistance, while Morocco (the susceptible check) was separated from the rest of the genotypes and grouped in Cluster IX.

Table 3. Analysis of variance for yield and 1,000 grain weight of evaluated wheat lines

Wheat germplasm	Yield [kg · ha-1]	1,000 Grain weight [g]	Wheat germplasm	Yield [kg · ha-1]	1,000 Grain weight [g]
23-C	4,883 fghi	44.97 defgh	NW-1-20	4,706 hijk	40.1 pqrstu
NIAB-1	3,946 pqr	39.1 rstuv	15-C	5,244 ef	39.9 qrstu
NW-10-4	4,448 klmno	40.2 pqrst	NW-10-1111-3	6,146 a	44.3 efghi
NW-5-1212-1	5,123 fg	42.8 ijklmn	14-C	6,048 abc	40.6 pqrs
4-C	3,906 qrs	41 mnopqr	17-C	4,225 mnopq	48.3 ab
NW-10-1111-7	4,575 ijklm	43 hijklm	NW-2-8	3,485 t	44 fghij
NW-10-18	4,638 hijkl	42.9 ijklmn	NW-7-5	5,256 ef	45.1 defg
3-C	4,869 ghij	45.5 cdef	2-C	3,927 pqrs	49.6 a
19-C	3,881 qrs	38.1 uvwx	26-C	4,169 nopq	35.2 A
NIAB-2	3,675 rst	40.9 nopqr	NW-7-30-1	4,684 hijk	35.3 A
5-C	4,894 fghi	43.4 ghijk	11-C	4,192 nopq	36.4 xyzA
NW-10-1111-37	5,104 fg	42.1 jklmnop	1-C	4,281 lmnop	40.7 opqr
NW-1-9-47	6,125 ab	37.5 vwxyz	13-C	4,933 fghi	35.5 zA
NW-17-5	3,644 rst	35.7 yzA	NW-1-47	5,817 abcd	36.6 wxyzA
NW-1-39	4,165 nopq	46.3 bcde	NW-1-52	4,998 fgh	43.9 fghij
NW-1-38	5,169 fg	46.3 bcde	NW-10-45	5,181 fg	40.2 pqrst
21-C	3,562 st	34.9 A	NW-10-16	3,887 qrs	43.4 ghijk
10-C	3,596 rst	36.57 wxyzA	28-C	3,469 t	35 A
NW 2-2	5,169 fg	38.3 tuvwx	7-C	5,188 efg	44.2 fghi
NW-7-28	3,656 rst	32.5 B	29-C	3,496 t	30.7 B
NW-1-8183-8	4,500 jklmn	41.4 klmnopq	24-C	5,744 cd	41.3 lmnopq
6-C	2,788 u	37.7 vwxy	NW-3-3341-7	5,556 de	47.3 bc
18-C	3,887 qrs	32.5 B	27-C	5,758 bcd	37.7 vwxy
22-C	3,656 rst	35.4 A	NW-10-1111-5	4,977 fgh	42.7 ijklmno
20-C	4,169 nopq	34.8 A	25-C	4,394 klmno	38.6 stuvw
NW-1-35	5,177 fg	49.7 a	NW-10-19	5,669 d	46.9 bcd
NW-7-14	3,467 t	38.3 tuvwx	8-C	4,087 opq	41.3 lmnopq
NW-2-4	3,475 t	43.1 ghijkl	MOROCCO	1,037 v	21.59 C
NW-1-36	4,396 klmno	46.7 bcd			

The mean experimental values within the same columns are significant ($p < 0.05$) if they do not share the same letter (due to minute difference between values after the "yz" the capital letters were used)

Table 4. Linear correlation coefficients between partial resistance parameters and yield components for stripe rust among 57 lines and a susceptible check

		Yield	1,000 gw	AURPC	rAURPC	FRS	CI
Yield	1	–					
1,000 gw	2	0.50**	–				
AURPC	3	–0.23	–0.39**	–			
rAURPC	4	–0.23	–0.39**	1**	–		
FRS	5	–0.07	–0.33**	0.94**	0.94**	–	
CI	6	–0.20	–0.36**	0.92**	0.92**	0.93**	–

Gw = grain weight; AURPC = area under rust progress curve; rAURPC = relative area under rust progress curve; FRS = final rust severity; CI = coefficient of infection
**significant at $p = 0.01$ level of probability

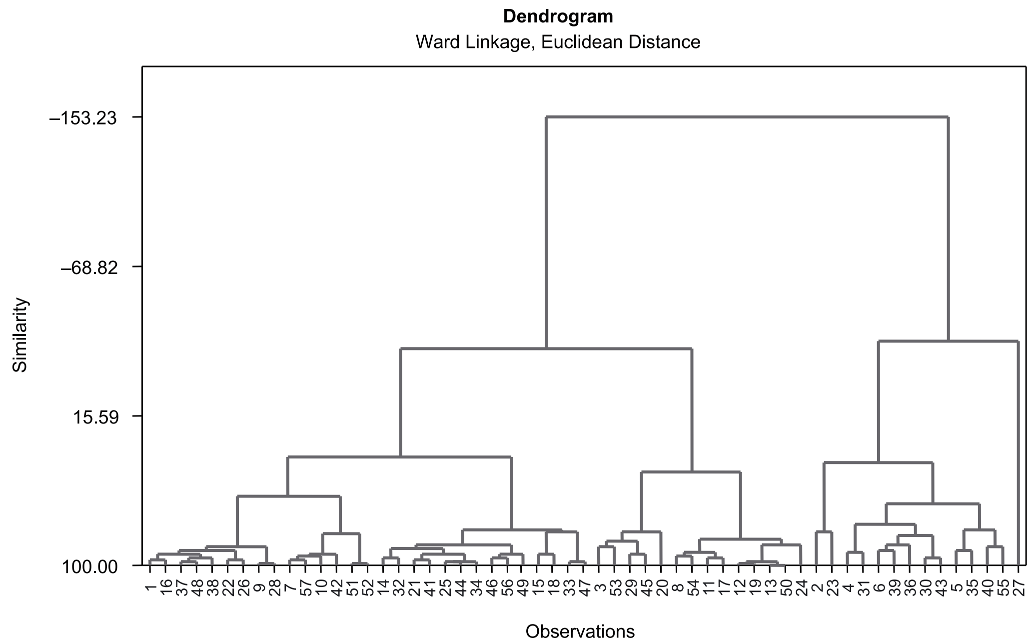

Fig. 1. Cluster analysis of 57 wheat lines based on stripe-rust parameters and yield components among 57 lines and a susceptible check

Table 5. Grouping of wheat genotypes based on the dissimilarity between the means of the disease and the yield parameters

Groups	Wheat genotypes	AURPC	rAURPC	FRS	CI	TSW	GY
Cluster I	1, 9, 16, 22, 26, 28, 37, 38, 48	21.8	1.3	3.3	0.5	40.4	4,224
Cluster II	7, 10, 42, 51, 52, 57	10.1	0.6	1.7	0.4	46.3	3,946
Cluster III	14, 15, 18, 21, 25, 32, 33, 34, 41, 44, 46, 47, 49, 56	52.8	3.1	6.4	0.9	43.3	5,174
Cluster IV	3, 20, 29, 45, 53	217.2	12.9	23.0	13.6	37.9	3,743
Cluster V	8, 11, 12, 13, 17, 19, 24, 50, 54	11.7	0.7	2.2	0.2	34.9	3,667
Cluster VI	2, 23	705.9	42.0	66.7	58.8	40.0	4,245
Cluster VII	4, 6, 30, 31, 36, 39, 43	270.2	16.1	31.4	19.5	38.0	5,299
Cluster VIII	5, 35, 40, 55	393.8	23.4	39.2	32.0	44.9	5,657
Cluster IX	27	1,680	100	100	100	21.593	1,037

AURPC = area under rust progress curve; rAURPC = relative area under rust progress curve; FRS = final rust severity; CI = coefficient of infection; TSW = thousand seed weight; GY = grain yield

Discussion

The use of resistant varieties not only ensures protection against disease but also saves time, energy, and money compared to the chemical control of rust which is expensive and impractical. Dominant or major resistance genes are deployed to confer vertical resistance against one or a few races of the pathogen. However, because resistance is based on recognition of a single pathogen-derived molecular pattern, these narrow spectrum genes are usually readily overcome by evolving virulent races of the pathogen. In contrast, horizontal resistance, also known as adult plant resistance, partial, durable, and quantitative resistance, in which more than one gene; usually minor genes, are deployed to reduce the rate of sporulation and infection. Quantitative resistance to stripe rust hampered the fungus development at different stages of the infection cycle. The extent to which fungus is hampered at the adult plant stage can be accessed through the study of the FRS, CI, AURPC, and rAURPC components of resistance.

To ascertain resistance potential against stripe rust at the adult plant stage, the current investigation was carried out at the experimental area of NIAB, Faisalabad, for two years. Fifty-seven selected wheat lines along with Morocco, were accessed for partial or quantitative resistance under natural stripe rust epidemic conditions. Diversity in the germplasm response was observed from immune to highly susceptible with the FRS range of 0 (immune) to 100 (susceptible). Twenty-one wheat lines (37%) were immune and exhibited the race specific resistance against the local stripe rust race with epidemic disease conditions in Morocco. These immune lines were obtained through hybridisation in NIAB and performed well for two years. Among the immune lines, 13 were derived from crossingwhich included one or more parent of CIMMYT origin, postulated to contain major resistant genes against stripe rust. Based on the categorisation of these lines through AURPC, rAURPC, and CI, a high level of adult plant resistance was observed in the previous studied lines (Ali *et al*. 2007; Rizwan *et al*. 2010; Shah *et al*. 2010). At the adult plant stage, rAURPC denotes the disease progress at a given time and provides a clue of the resistance level in the host germplasm. In 21 immune lines, the rAURPC remained zero throughout the season, which is well in agreement with previous studies (Ma *et al*. 1995; Shah *et al.* 2010). Based on the infection type, diversity was observed in germplasm with a variable number of R (resistant), MR (moderately resistant), R-MR (resistant to moderately resistant), MR-MS (moderately resistant to moderately susceptible), MS-S (moderately susceptible to susceptible), and S (susceptible) reactions. Nineteen wheat lines with a R-type reaction exhibited the acceptable level of AURPC, rAURPC, and CI, and conferred a high level of partial resistance, while twelve wheat lines with a MR, R-MR, and MR-MS reaction also exhibited a high level of partial resistance and it can be postulated that they contain *Yr*18 or unknown partial resistance genes. In most of Pakistani wheat cultivars, *Yr*18 is well documented to confer partial resistance (Bux *et al*. 2011; Qamar *et al*. 2012). Rust appears on these lines but with an increase in temperature and maturity, rust stops or ends with a MR or MS type of infection. Such a kind of resistance is durable and controlled by minor or a combination of major and minor genes for resistance (Brown *et al*. 2001; Singh *et al.* 2005).

Wheat lines NW-7-13, NW-2-2, NW-7-14, NW-1-35, 15-C, 14-C, NN-7-30-1, and 13-C behaved as moderately susceptible to susceptible lines but their FRS values were 30, 30, 50, 30, 20, 40, 40, and 30, respectively. While observing the partial resistance parameters of these lines, all the lines conferred high levels of partial resistance, except NW-7-14, 14-C, and NN-45, which incurred low and moderate levels of partial resistance, respectively. Such lines are believed to contain slow rusting or high temperature adult plant resistance (HTAP) genes and these lines are recommended to be utilised in achieving durable resistance because the acceptable degree of slow rusting restricts the evolution of new virulent races. These results are in agreement with the results of Singh *et al*. (2005). Consequently, the reaction of NW-5-1212-1, NW-3-2, and NW-10-1111-3 is resistant to moderately resistant or moderately susceptible but the level of partial resistance is of a moderate to low level. The presence of the major resistance genes can be hypothesised in these lines. The two wheat lines, evaluated in this study, 5-C and 10-C, were most susceptible after Morocco, and marked as having a low level of quantitative resistance (FRS, AURPC, rAURPC, and CI). These lines may not be used in a future breeding program. The high level of diversity in these wheat lines was observed regarding the response against stripe rust. An immune to susceptible reaction was observed in investigations by many researchers (Ahmad *et al*. 2006; Afzal *et al*. 2008; Ali *et al*. 2009b).

Disease always has a negative impact on crop yield, so control measures need to be adopted. Owing to the devastating nature of the stripe rust pathogen, the yield per hectare and 1,000 grain weight was determined and compared with Morocco. There was a variation in the yield and the 1,000 grain weight, and both of these parameters were negatively correlated with disease. The shriveling of grains and ayield reduction was well-documented in many previous studies (Afzal *et al.* 2007, 2008; Safavi and Afshari 2012). Moreover, the correlation between FRS, CI, AURPC, and rAURPC was determined and found positive at $p = 0.01$ which is in agreement with the results of other researchers on cereal-rust pathosystems (Sandoval-Islas *et al*. 2007; Safavi *et al*. 2010; Shah *et al*. 2010). The significant correlation of rAURPC with quantitative resistance components i.e. latent period and infection frequency, was also reported by Sandoval-Islas *et al*. (2007) while a negative correlation between rAURPC and yield loss was also documented (Ochoa and Parlevliet 2007). A low rAURPC value along with a low CI is the preferred criteria in field selection of lines, conferring partial resistance among the repertories of germplasm, in situations where greenhouse facilities are inadequate (Singh *et al*. 2007). Due to a strong negative correlation of rAURPC with the yield and 1,000 grain weight, it is suggested that rAURPC may be preferred to monitor the influence of disease on yield as compared to other epidemiological parameters.All the partial resistance components are positively correlated with each other, therefore we used FRS, CI, and high yield potential for the selection of durable resistance wheat lines. The most appropriate parameters to identify partial resistance were FRS and CI (Safavi and Afshari 2012), while FRS increase the yield loss proportionally against leaf rust (Salman *et al*. 2006).

Cluster analysis was executed to determine diversity among wheat lines. Based on partial resistance parameters, yield, and grain weight, nine groups/clusters were formed. Wheat lines in Cluster I, II, and III exhibited optimum adult plant resistance while lines in Cluster VIII attained the maximum yield and 1,000 grain weight. Keeping in mind that the objective was toimprove the wheat group at NIAB, wheat genotypes from Cluster I, II, and III may be inter-crossed with each other and also with the genotypes in Cluster VIII to get stripe rust resistance and high yielding varieties. Previously, the presence of varying degrees of partial resistance to stripe rust in breeding lines from Pakistan was reported by Ali *et al*. (2007). In another study, on the basis of overall host reaction, 38 varieties were clustered into six groups (Ali *et al*. 2009), although this finding was different from our study because yield parameters were also considered in the current study. Some wheat lines producing maximum yield under high disease pressure may have less grain weight, as observed in this study. We found that the NW-10-1111-3 line produced the maximum yield but the 1,000 grain weight was maximum for NW-1-35. Both the yield and grain weight are positively correlated but a reduction in the grain weight in some lines is less – even under high disease pressure. It was the same case with NW-1-35, which exhibited a MS-S reaction type with a FRS of 30% but with the maximum grain weight, and was the 13th highest yielding line. Attaining different grain weights in different lines may be due to different genes controlling this trait independently from stripe rust resistance. In spite of the infection type, NW-1-35 conferred a high level of partial resistance which is most probably due to minor or a combination of minor and major resistance genes with high yield potential. Similarly, in a previous work oncultivars, the inqulab-91 variety exhibited a minimum reduction in the 1,000 grain weight in spite of a maximum disease severity (Afzal *et al*. 2008).

Conclusion

Durability and effectiveness of resistance against stripe rust was demonstrated by growing wheat cultivars for adult plant resistance. Targeted lines were identified phenotypically by necrotic stripes under relatively high temperature, in comparison with susceptible plants. In this investigation, those immune, resistant, and moderately resistant wheat lines conferring a high level of partial and adult plant resistance, need to be utilized for the development of rust-resistant wheat varieties so that farmers can play a part in the uphill battle of feeding an ever-increasing world population. A high level of diversity was observed which helped to select the genotypes with stripe-rust resistance and better yield. However, further studies for the development of diagnostic molecular markers to assess the APR gene are still needed for genotypic characterization and field evaluations in the current era. Wheat lines, selected on the basis of resistance and yield parameters, could be used in a hybridization program to develop new rust resistant and high yielding cultivars and may also be evaluated at multiple locations to fulfill the requirements for the direct release of wheat cultivars.

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