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PJBS

ISSN 1028-8880

Pakistan Journal of Biological Sciences

ANSInet
Asian Network for Scientific Information
308 Lasani Town, Sargodha Road, Faisalabad - Pakistan

Pakistan Journal of Biological Sciences 3 (3): 469-472, 2000

Evaluation of Annual Wild Grass Species for Leaf Rust Resistance

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Abstract: Annual wild grass species of wheat i.e. *Aegilops tauschii, Ae. geniculata, Ae. neglecta, Ae. variables, Ae. speltoids and Ae. triunciallis* were screened against leaf rust *Puccinia recondita* Roberge ex Desmaz.f.sp. *tritici* (Eriks. & E.henn.) D.M. Henderson. The frequency of immune and resistant accessions was fairly high in *Ae. geniculata, Ae. variables* and *Ae. tauschii* while low in the accessions of *Ae. neglecta*. No accession of *Ae. geniculata* were found to be susceptible. All the tested accessions of *Ae. variables, Ae. speltoides* and *Ae. triunciallis* were immune to the prevalent races of leaf rust in this region.

Key words: Annual grasses, Leaf rust, *Aegilops* species, *Puccinia recondita*

Introduction

Of the approximately 325 wild species in the tribe Triticeae, about 250 are perennial and 75 are annuals (Dewey, 1984). The annual species, which includes many forage grasses, have the potential to serve as vital genetic reservoir for the improvement of the wheat. Leaf rust caused by *Puccinia recondita* Roberge ex Desmaz.f.sp. *tritici* (Eriks. & E.henn.) D.M. Henderson, is one of the major constrain in the wheat production of Pakistan and all over the world. Its importance varies with the cultivars, regions, seasons, biotic and abiotic stresses. Leaf rust disease sometimes becomes epidemic over a wide area when favorable environmental conditions, such as high moisture and relatively high temperature are present.

Wild relatives of wheat with its diverse range of accessions and distribution provides a unique opportunity for exploiting novel genetic variability for wheat improvement associated with biotic/abiotic stress factors. Varying number of accessions of *Aegilops* species were evaluated against prevalent races of yellow, brown rust and Karnal bunt and identified excellent sources of resistance (Khem *et al*., 1987).

Most of the annual and perennial species have been screened against salinity (Farooq *et al.,* 1988, 1989) and the identified species are now being used as donor of stress tolerance genes to cultivated wheat varieties (Farooq *et al.*, 1994, 1995). In the present study, some of the results obtained on the screening of annual species against rust resistance are being presented.

Materials and Methods

Material used in this study consisted of different accessions of annual *Aegilops* species (Table 1). Seeds of *Aegilops* species were obtained from Plant Breeding Institute (PBI) Cambridge U.K., ICARDA Syria and USDA , ARS, USA . All the species are being maintained under natural environmental conditions in the net house at NIAB Faisalabad, Pakistan and shifted in the month of January 1994 for the screening to the Leaf Rust Screening nursery of Wheat Research Institute, Ayub Agriculture Research Institute Faisalabad, Pakistan.

Maintenance of the species: Seeds of each accession/species were dusted with fungicide (Vitavax Ciba-Giegy, Switzerland) and placed in a petri-plate lined with moist filter paper in the mid of October. After 24 hours at room temperature (24 ± 2°C), the seeds were given cold shock for one week in a refrigerator (4°C) to break the dormancy, and then placed at room temperature. Germinated seeds were transplanted in plastic pots (10 cm diameter) containing sterilized mixture of sand and soil (1:1) and transferred in a net house under natural conditions. The annual grasses were harvested at the end of April and seeds were stored in the refrigerator at 4°C to maintain the viability of seeds.

Evaluation for leaf rust resistance: The seedlings of 59 accessions of *Ae. tauschii*, 17 of *Ae. geniculata*, 33 of *Ae. neglecta,* two of *Ae. speltoides* and one each of *Ae. variables* and *Ae .triunciallis* were grown in plastic pots (10 cm diameter). The pots were surrounded by highly susceptible wheat varieties (Balochi Local White, Morocco, Yecora, SA-42) which acted as spreader for the disease. Two pots of each accession with three seedlings per pot were artificially inoculated at the end of January with freshly collected uredospores of the *Puccinia recondita tritici* and also against prevalent leaf rust races present in this region. The data on rust severity were recorded at seedling, tillering and maturity stages according to the modified Cobb scale (Saari and Prescott, 1975).

Results

Maintenance of annual *Aegilops* species: Fungicide treated seeds did not show seed borne diseases like smuts and bunts. The seeds of annual grasses were harvested when the seed was of normal size, fully matured with moisture contents of 6 ± 1 percent. The threshed seeds were treated with fungicide Vitavax-200 and sealed in aluminum foil packets and stored at a temperature of -21 ± 1°C. To get better seed germination, the seeds were given cold shock treatment for one week at 4°C. This helped to break the seed dormancy.

Evaluation for leaf rust resistance: The frequency of immune and resistant accessions was fairly high in *Ae. geniculata*, *Ae. variables* and *Ae. tauschii* (Fig. 1) while low in the accessions of *Ae. neglecta*. Out of 59 accessions of *Ae. tauschii*, 36 (61%) accessions were immune, 3 (8.3%) accessions resistant, 8 (13.6%) accessions moderately resistant and 12 (20.3 percent) accessions were susceptible (Table 2). Among 33 accessions of *Ae. neglecta,* 11 (33.3%) accessions were found to be immune, only one (3.1%) accession resistant, 7 (21.2%) accessions moderately resistant and 14 (42.2%) accessions were susceptible (Table 3). From 17 accessions of *Ae. geniculata,* 13 (76.5%) accessions were immune, 2 (12%) accessions resistant and 2 (12 percent) accessions were found to be moderately resistant (Table 4). No accession of *Ae. geniculata* were found to be susceptible. All the tested accessions of *Ae. variables, Ae. speltoides* and *Ae. triunciallis* were immune to the prevalent races of leaf rust in this region.

Discussion

Maintenance of annual *Aegilops* species: Wild seeds need to be retained for more than one season or a year for further use, If necessary protection against heat, moisture and pest is not maintained they lose their viability. Moisture content of seed is one of the most important factors affecting seed longevity, the drier the seed the greater is longevity (Van der Maesen, 1984). It is well established fact that the seed with proper moisture germinate easily. Our earlier study showed that seeds harvested

Table 1: The genomes and ploidy levels of different goat grass species

Aegilops species	Genome	Choromosome Number	No. of accessiions used
Ae. tauschii	DD (Lilienfeld, 1951)	14	59
Ae. neglecta	CCDD (“”)	28	33
Ae. geniculata	UUMM (Mujeeb-Kazi and Hettel, 1987)	28	17
Ae. triunciallis	UUCC (Kimber and Sears, 1983)	28	1
Ae.variables	UUS’S’ (Mujeeb-Kazi and Hettel, 1987)	28	1

Table 2: Different accessions of *Aegilops tauschii,* with their origin and reaction to leaf rust grown in the field

Species	Accession No.	Origin	Rust severity (percent)	Species	Accession No.	Origin	Rust severity (percent)
Ae. tauschii	A**	Pakistan	0	*Ae. tauschii*	“20	“	5R
	B**	“	40 MS-S		“21	“	5R
	C**	Afganistan	0		“22	“	25
	D**	“	0		“23	“	0
	E**	“	0		“24	“	0
	Af-8A**	Turkey	0		“25	“	20S
	Af-9A**	Turkey	0		“26	“	0
	Af-10**	Turkey	40 MR-MS		“27	“	40S
	Af-15**	Turkey	0		“28	“	30S
	Af-16**	“	R-MR		“30	Japan	0
	Ciae 1*	Pakistan	60S		“50	Maryland USA	0
	“ 2	Afganistan	0		“51	“	40S
	” 3	“	30S		“68	“	40S
	“ 4	“	20S		“71	Turkey	0
	” 5	“	0		“72	USA Missouri	0
	“ 6	“	30S		PI 210987*	USA California	10MR-MS
	” 7	“	0		PI 220331	Afghanistan	20S
	“ 8	Iran	0		PI 276975	USSR	20MR-MS
	” 9	“	0		PI 317392	Afghanistan	30S
	“ 10	“	0		PI 349037	Azerbaijan	0
	” 11	“	0		PI 428560	“	0
	“ 12	“	0		PI 428561	“	0
	” 13	“	0		PI 428564	“	0
	“ 14	“	0		PI 431598	Turdmenistan	0
	” 15	“	20-30S		PI 431599	Azerbaijan	0
	“ 16	“	0		PI 431600	Russia Fed	0
	” 17	“	20S		PI 431601	“	0
	“ 18	“	0		PI 431602	Turkmenistan	0
	” 19	“	0		PI 452130	China	30S
					PI 452131	China	30S

0-Immune, 1-10 Resistant, 11-25 Moderately Resistant, Above 25- Susceptible Received from: *USDA, ARS, USA.
**Plant Breeding Institute, Cambridge, U.K.

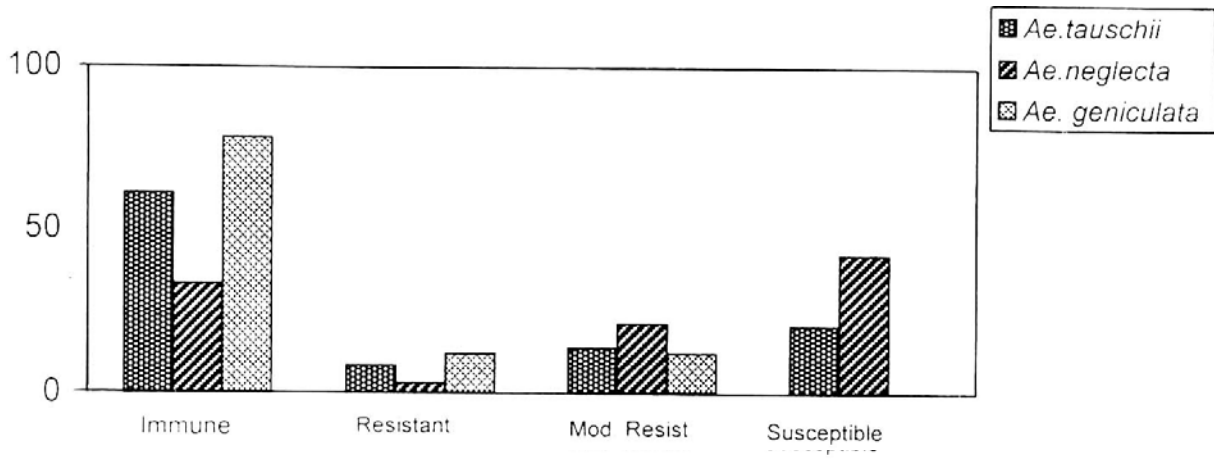

Fig. 1: Leaf rust resistance (%) in *Aegilops* species

Table 3: Different accessions of *Aegilops neglecta* and reaction to leaf rust shown in the field

Species	Accession #	Rust severity (%)	Species	Accession	Rust severity (%)
Aegilops neglecta	A	0	*Aegilops neglecta*	PI 486238	15S
	D	0		PI 486241	40MR-MS
	E	30MS-S		PI 486242	20S
	F	30S		PI 486246	35S
	G	80S-100S		PI 487222	0
	PI 171469	0		PI 487225	35S
	PI 179160	0		PI 487226	0
	PI 172357	35S		PI 491427	0
	PI 172358	40S		PI 491432	0
	PI 172681	35S		PI 491434	0
	PI 172683	0		PI 491435	0
	PI 176853	25S		PI 502242	0
	PI 210987	80MS-S		PI 502243	20S
	PI 228333	20S		PI 504261	0
	PI 254864	30S		PI 524430	20S
	PI 266814	0		PI 554205	25S
	PI 266816	0		PI 554208	5MR-MS
	PI 276974	15S		PI 554209	0
	PI 276976	35S		PI 554216	0
	PI 298891	0		PI 554217	40S
	PI 298893	60S		PI 554219	35S
	PI 314406	30		PI 554222	30S
	PI 330484	0		PI 554225	15S
	PI 344778	35		PI 554230	0
	PI 349035	35S		PI 560509	20S
	PI 374332	15S		PI 560516	25S
	PI 374345	20S		PI 560518	30S
	PI 374353	5MR-MR		PI 560519	20S
	PI 388753	20 R-MR		PI 560737	40S
	PI 383530	0		PI 560739	40S
	PI 392331	30S		PI 568162	40S
	PI 428561	0		PI 573367	40S
	PI 486236	30S		PI 574461	40S

0-Immune, 1-10 Resistant, 11-25 Moderately Resistant, Above 25- Susceptible Received from: USDA, ARS, USA. Plant Breeding Institute, Cambridge, U.K.

Table 4: Accessions of *Aegilops geniculata*, *Ae. Speltoides, Ae. triuciallis* and *Ae. variables,* with leaf rust resistance grown in the field

Species	Accession No.	Origin	Rust severity (Percent)
Ae.geniculata	F		0
	Ciae 43	Canada	0
	Ciae 53	USA Maryland	0
	Ciae 63	"	10
	Ciae 65	"	0
	PI 276978	Japan	0
	PI 279578	Japan	R-MR
	PI 330487	U.K.	0
	PI 361880	Romania	0
	PI 368181	Romania	0
	PI 369576	Argentina	20S
	PI 369577	"	0
	PI 369578	"	0
	PI 369580	"	0
	PI 388754	Morocco	20S-MS
	PI 388756	"	0
	PI 289578	U.K.	0
Ae. speltoides	PI 393494	Israil	0
	PI 393495	Israil	0
Ae. triuncialis	PI 172682	Turkey	0
Ae.variables	E		0

with proper moisture contents, have good germination (Unpublished data). In contrast to these seeds with more than 12 percent moisture contents may be attacked by fungi or insects (Van der Maesen, 1984), the results of this study are in agreement with the above findings. The results of present study showed that the use of Fungicide Vitavax-200 can be recommended for seed treatment as it did not alter the viability and longevity of the seeds (Anonymous, 1990). This type of Fungicide treatment to seeds is relatively cheap and poses less threat to the environment. In this investigation, dormancy of annual grasses was broken by removing the seed coat over the embryo with a needle and transferred the seeds just above

freezing to room temperature daily the same was also recommended by Tosun *et al*. (1980). For long term storage of wild seeds were stored in air tight containers with 5 ± 1 percent moisture content at -18°C or less as suggested by Roberts (1972). This type of storage provided good results in case of seed viability and longevity. All the above mentioned germplasm has been maintained and stored for further utilization as gene source for the improvement of salt and rust resistance of bread wheat.

Leaf rust Resistance: It was observed that *Aegilops* species have higher potential for leaf rust resistance (Kerber and Dyck, 1969, 1979). As *Ae. tauschii* (ancestor of wheat with diploid chromosomes) have also possessed resistance for leaf and stem rust so it can be used in the breeding program to evolve the leaf and stem rust resistant wheat varieties which is necessary for the climatic conditions of Pakistan. *Ae. geniculata* possesses resistance against powdery mildew (Gill *et al*., 1989), yellow rust, brown rust. powdery mildew and Karnal bunt (Dhaliwal *et al*., 1986; Warham *et al*., 1986). Khem *et al*. (1987) also confirmed the above findings as they screened 11 different *Aegilops* species (1763 accessions) against yellow rust, brown rust and Karnal bunt and 407 different accessions were identified to be resistant. It means that the wild relatives of wheat possessed a large gene pool which is useful to create genetic variation necessary for varietal improvement. Screening of the closely related genus i.e. *Aegilops speltoides* has also identified for having resistance to powdery mildew (Miller and Reader, 1987). In the present study, different accessions of *Ae. speltoides, Ae. triunciallis* and *Ae. variables* also showed good resistance for rust and can be used for wheat improvement.

Ae. geniculata ($2n = 4x = 28$, UU M^oM^o) is a species of particular interest for genetic and breeding research as donor of genes for complex disease resistance (Bochev *et al*., 1982). Farooq *et al*. (1990a) observed that crossability of *Ae. geniculata* with *Triticum aestivum* and *T. turgidum* is very high compared to that of *Ae. tauschii* and of *Ae. neglecta*. Since *Ae. geniculata* shares genome U with *Ae. umbellulata*, so it can be anticipated as a donor of agronomically important gene(s) to wheat.

Ae. variables ($2n = 4x = 28$, UUS^VS^V) is also an annual, bushy, tetraploid member of the section Polyeides of the genus *Aegilops* has resistance against leaf rust which is confirmed by our study. Beside the above resistant genes it also possesses pairing promoters genes which further increase the crossability to tetraploid and hexaploid wheat cultivars (Farooq *et al*., 1990b).

Specialized cytogenetic techniques are now available to make successful genetic transfer from distantly located genomes and it is anticipated that genes for resistance to leaf rust and salt tolerance could be transferred to wheat from resistant accessions of *Ae. geniculata, Ae. tauschii., Ae. neglecta, Ae. triunciallis, Ae. speltoides* and *Ae. variables.*

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