Pathogenicity of *Puccinia recondita* f. sp. *tritici* in Morocco During 1985, 1988, 1990, and 1992

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ABSTRACT

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Surveys of pathogenic races of *Puccinia recondita* f. sp. *tritici* in Morocco in 1985, 1988, 1990, and 1992 indicated the presence of 12 unified numeration (UN) races. Virulence frequencies were high to *Lr*1, 2a, 3, 10, 16, and 17 and low to *Lr*3ka, 9, 24, and 26. The virulence frequency to *Lr*2a declined in 1990 and 1992. Isolates from durum wheats were generally less virulent than those from bread wheats to the designated *Lr* genes in a bread wheat background. The virulence diversity levels were higher in 1990 and 1992 than in 1985 and 1988. The populations with the highest phenotypic diversity were obtained from durum wheat cultivars and the susceptible bread wheat cultivar Siete Cerros. Durum wheat is often grown in marginal areas where the alternate host *Anchusa italica* occurs, while Siete Cerros is grown under irrigation in the south.

Both bread (*Triticum aestivum* L.) and durum (*T. turgidum* L.) wheats are cultivated in Morocco under a wide range of environmental and cultural conditions. They occupy on average 1 million and 1.5 million hectares, respectively. A number of cultivars are grown, including older local cultivars and semidwarf cultivars from the International Maize and Wheat Improvement Center (CIMMYT), El Batan, Mexico.

Leaf rust caused by *Puccinia recondita* Roberge ex Desmaz. f. sp. *tritici* (Eriks. & E. Henn.) D.M. Henderson is among the foliar diseases of wheat that occur annually in Morocco. The disease can be severe in some areas (4,6). The bread wheat cultivars are not fully resistant to leaf rust. Nesma, postulated to possess the gene *Lr*1 for resistance to leaf rust (5), was widely cultivated and occupied about 75% of total area grown to bread wheat until 1987. Since then, the area occupied by Nesma has declined sharply because of the cultivar's susceptibility to Hessian fly and diseases, including leaf rust. The newly released cultivars are not resistant to all races of the pathogen found in Morocco.

Among the durum wheats, Kyperounda (2777), introduced from Cyprus in the 1950s, is still the dominant cultivar in nonirrigated areas throughout Morocco. The cultivars introduced during the last 20 yr from CIMMYT, the International Center for Agricultural Research in the Dry Areas (ICARDA), Aleppo, Syria, and the Arab Center for the Study of Aridzones and Drylands (ACSAD), Damascus, Syria, are either moderately resistant or fully susceptible to leaf rust (*unpublished*).

The study of the pathogen variation by means of the race surveys is an effort to enhance the usefulness of resistances to be incorporated in commercial cultivars. Roelfs (13) has described the objectives of the cereal rust surveys. Surveys were conducted in Morocco in 1985, 1988, 1990, and 1992 to determine the distribution of pathogen virulence in relation to the host species and to the geographic area and to evaluate the temporal changes of race frequencies of the leaf rust pathogen.

MATERIALS AND METHODS

Field surveys. Collections of wheat leaf rust were made from commercial field areas of Morocco in 1985, 1988, 1990, and 1992. The surveyed region was subdivided into six agroecological areas (Fig. 1). Area 1 is the Sous, is a coastal irrigated plain surrounded by the High and the Low Atlas mountains. The main cultivar grown here is Siete Cerros; other cultivars of bread and durum wheats are grown mainly for seed production. Area 2 is the central or coastal Atlantic plains, where the climate is mild and both bread and durum wheats are grown. The wheat crop can be either irrigated or not irrigated under marginal cultural conditions. Infected plants of the alternate host, *Anchusa italica* Retz., are common. Area 3 is the Tadla, an irrigated plain at the foot of the southern part of the Middle Atlas mountains. The dominant cultivar Siete Cerros was replaced in 1990 by the bread wheat cultivar Potam and by durum wheat cultivars. *A. italica* plants are rarely observed in this area. Area 4 is the Gharb, a partially irrigated coastal plain north of Rabat where some fields are irrigated and where durum wheats predominate. *A. italica* plants are present but rarely infected. Area 5 is the Sais, a nonirrigated fertile lowland at the foot of the northern part of the Middle Atlas mountains where both durum and bread wheats are cultivated. *A. italica* plants are common but rarely infected. Area 6 is the Jbala, which is hilly and has a high rainfall. Traditional cultivars predominate, and infected plants of *A. italica* are common.

The same roads were taken on each annual field survey. When wheat fields were grouped in an area, stops were made every 10 km; when wheat fields were scattered, stops were made at the first one after 10 km. Sometimes leaf rust samples were taken from neighboring fields of durum and bread wheats. Surveys were conducted during late March and early April in areas 1, 2, and 3 and during late April and early May in areas 4, 5, and 6. The hosts were between the anthesis and milk growth stages at the time of collection.

Pathogenicity analysis. Isolates from uredinia collected in the field were inoculated to 7-day-old seedlings of Fertas, a susceptible bread wheat cultivar. Single-uredinium isolates were made and reinoculated to Fertas to increase the inoculum. Purified and increased uredinial inoculum of each isolate was tested on differential hosts. These were near-isogenic lines containing the genes *Lr*1, 2a, 2c, 3a, 3ka, 9, 10, 11, 16, 17, 24, and 26. The lines with the genes *Lr*11 and 26 were tested in 1990 and 1992.

The inoculated seedlings were held at 100% RH at 20–25 C in an illuminated chamber. Spore collection and inoculum techniques used were those described by Browder (3). Rust reactions on the differential lines were scored after 12 days by the method described by Stakman et al (14). Infection types 0, 1, and 2 corresponded to a resistant host reaction, and infection types 3 and 4 corresponded to a susceptible reaction.

Data analysis and presentation. Virulence frequencies on individual *Lr* genes were calculated. The combination of virulence and avirulence to the genes *Lr*1, 2a, 2c, and 3, known as unified numeration (UN) races (1,12), was used as a basis of comparison of cultures between years,

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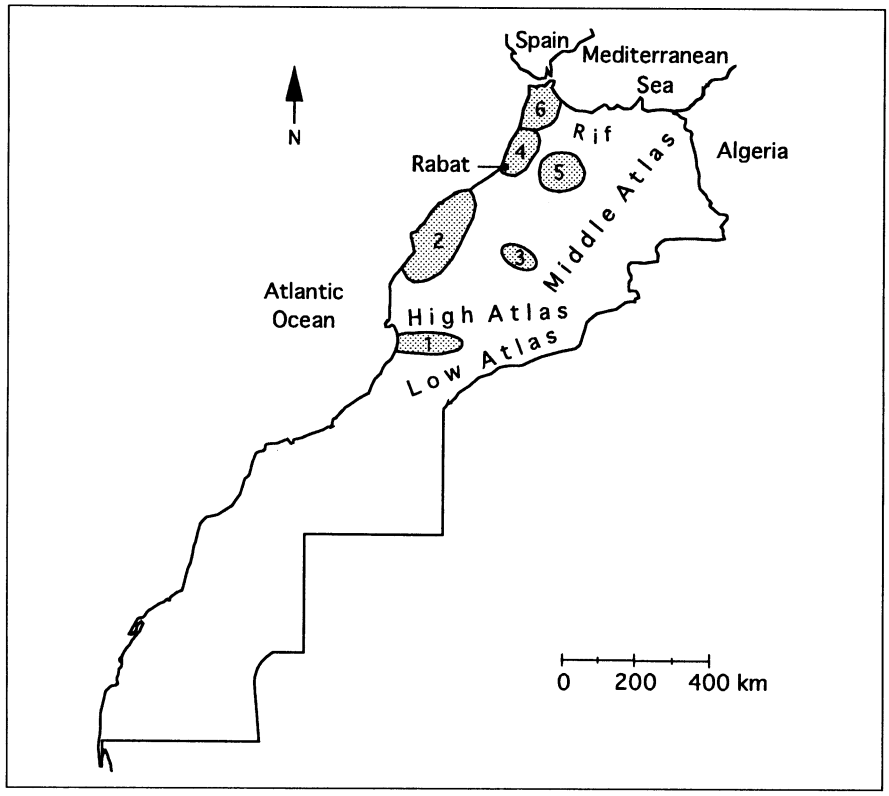

Fig. 1. Agroecological areas of cereal production in Morocco showing the geographic spacing and mountainous regions. Area 1 = Sous, 2 = central plains, 3 = Tadla, 4 = Gharb, 5 = Sais, and 6 = Jbala.

host species, and agroecological areas. The Shannon index of diversity as reported by Groth and Roelfs (8) was used to compare the intraspecific diversity of races in populations of *P. r. tritici* occurring on bread and durum wheats and in different agroecological areas.

RESULTS

The effectiveness of individual host genes for resistance was evaluated by examining the percentage of isolates virulent to near isogenic lines (Table 1). Virulence to the genes *Lr*1, 2c, 3a, 10, 16, and 17 was high during the 4 yr of the study. The virulence frequency to *Lr*2a was about 50% in 1985 and 1988 but decreased to about 39% in 1990 and to about 36% in 1992. *Lr*24 and 26 were resistant to all isolates evaluated, and *Lr*3ka was effective against more than 90% of the population. The virulence frequency to *Lr*9 varied from low to medium.

Virulence frequencies were also calculated according to the host species from which isolates were collected (Table 2). The virulence frequency *Lr*1, 2a, 2c, and 3 was lower among isolates from durum wheats than among those from bread wheats. Virulences to *Lr*10, 16, and 17 were equally frequent among isolates

Table 1. Number and percentage of *Puccinia recondita* f. sp. *tritici* isolates virulent to selected leaf rust resistance genes from collections made in Morocco in 1985, 1988, 1990, and 1992

	1985		1988		1990		1992	
Lr gene[a]	No.	%	No.	%	No.	%	No.	%
1	116	85.9	79	67.0	102	68.9	109	80.7
2a	101	74.8	77	65.3	57	38.5	48	35.6
2c	114	84.4	106	89.8	116	78.4	85	63.0
3a	122	90.4	91	77.0	127	85.8	107	79.3
3ka	1	0.7	0	0.0	3	2.0	12	8.9
9	25	18.5	3	2.5	11	7.4	50	37.0
10	129	95.6	82	69.0	124	83.8	97	71.9
11[b]	...	...	...	...	79	53.4	52	38.5
16	113	83.7	96	81.4	133	89.9	117	86.7
17	108	80.0	63	53.4	126	85.1	104	77.0
No. of collections	135		118		148		135	

[a]No virulence was observed to *Lr*24 and 26.
[b]Not tested in 1985 and 1988.

Table 2. Frequency (%) of virulence of *Puccinia recondita* f. sp. *tritici* to selected leaf rust resistance genes in relation to wheat crop from which collection was made

	1985		1988		1990		1992	
Lr gene	Bread	Durum	Bread	Durum	Bread	Durum	Bread	Durum
1	94.3	71.4	73.3	55.8	76.2	59.4	86.7	60.0
2a	86.0	55.1	73.3	51.2	51.2	21.9	41.0	16.7
2c	95.2	65.3	96.0	79.1	86.9	67.2	68.6	43.3
3a	98.8	75.5	82.7	67.4	91.7	78.1	87.6	50.0
3ka	1.2	0.0	0.0	0.0	1.2	3.1	10.5	3.3
9	24.2	8.2	4.0	0.0	10.7	3.1	41.0	23.3
10	98.8	89.8	76.0	58.1	83.3	84.4	73.3	66.7
11	0.0	0.0	0.0	0.0	58.3	46.9	40.0	33.3
16	91.9	69.4	90.7	65.1	89.3	90.6	87.6	83.3
17	90.7	61.2	65.3	32.6	85.7	84.4	76.2	80.0
No. of isolates	86	49	75	43	84	64	105	30

Table 3. Distribution (%) of unified numeration (UN) races of *Puccinia recondita* f. sp. *tritici* identified in Morocco

Year / Wheat crop	UN race 1	2	3	5	6	9	10	11	12	13	14	17
1985	6.7	3.0	4.4	3.0	5.2	0.0	0.0	3.0	0.0	74.8	0.0	0.0
Bread	0.0	2.3	3.5	1.2	5.8	0.0	0.0	1.2	0.0	86.0	0.0	0.0
Durum	18.4	4.1	6.1	6.1	4.1	0.0	0.0	6.1	0.0	55.1	0.0	0.0
1988	5.9	4.2	12.7	0.0	5.9	8.5	4.2	0.0	2.5	50.8	1.7	3.4
Bread	1.3	2.7	12.0	0.0	4.0	8.0	5.3	0.0	1.3	60.0	1.3	4.0
Durum	14.0	7.0	14.0	0.0	9.3	9.3	2.3	0.0	4.7	34.9	2.3	2.3
1990	4.1	6.1	16.9	8.1	19.6	3.4	2.0	3.4	0.0	33.1	1.4	2.0
Bread	2.4	4.8	11.9	6.0	21.4	3.6	2.4	0.0	0.0	45.2	0.0	2.4
Durum	6.3	7.8	23.4	10.9	17.2	3.1	1.6	7.8	0.0	17.2	3.1	1.6
1992	9.6	3.0	1.5	17.0	24.4	1.5	2.2	5.2	0.0	31.9	1.5	2.2
Bread	3.8	1.9	1.9	19.0	26.7	1.9	1.9	3.8	0.0	37.1	0.0	1.9
Durum	30.0	6.7	0.0	10.0	16.7	0.0	3.3	10.0	0.0	13.3	6.7	3.3

Table 4. Number of isolates of unified numeration (UN) races of *Puccinia recondita* f. sp. *tritici* annually by geographic area of Morocco

Year / Area	UN race 1	2	3	5	6	9	10	11	12	13	14	17	Total
1985													
Sous	—[a]	2	6	—	3	—	—	—	—	2	—	—	13
Central plains	1	1	3	1	—	—	—	2	—	41	—	—	49
Tadla	—	—	—	—	—	—	—	—	—	13	—	—	13
Gharb	—	—	1	—	—	—	—	—	—	11	—	—	12
Sais	—	—	—	—	—	—	—	—	—	8	—	—	8
Jbala	8	1	—	3	—	—	—	2	—	26	—	—	40
1988													
Sous	—	4	5	—	1	1	1	—	—	4	—	—	16
Central plains	—	—	—	5	2	1	1	—	1	23	1	1	35
Tadla	—	1	4	—	1	—	2	—	—	4	—	—	12
Gharb	1	—	—	—	—	3	—	—	1	13	—	1	19
Sais	—	—	—	—	3	1	—	—	—	10	1	1	16
Jbala	6	—	1	—	—	4	1	—	1	7	—	1	21
1990													
Sous	2	3	11	3	7	—	—	1	—	7	—	—	34
Central plains	2	4	9	8	11	3	1	4	—	27	2	1	72
Tadla	—	—	1	1	3	—	—	—	—	6	—	1	12
Gharb	—	1	2	—	2	1	—	—	—	5	—	—	11
Sais	—	—	—	—	1	—	—	—	—	3	—	—	4
Jbala	2	1	2	5	1	—	2	—	—	1	—	1	15
1992													
Sous	1	2	—	11	9	—	—	2	—	3	—	1	29
Central plains	10	1	1	6	13	2	3	1	—	23	—	2	62
Tadla													
Gharb	2	—	—	3	7	—	—	3	—	8	2	—	25
Sais	—	—	1	3	2	—	—	—	—	5	—	—	11
Jbala	—	1	—	—	2	—	—	1	—	4	—	—	8

[a] — = Not detected.

from bread and durum wheats. The virulence frequency to *Lr*3ka and *Lr*9 was higher among isolates from bread wheats than among those from durum wheats, even though *Lr*9 does not occur in North African cultivars.

UN races 1, 2, 3, 5, 6, 9, 10, 11, 12, 13, 14, and 17 were identified (Table 3). Race UN13 was the most common during the 4 yr, although its frequency declined from 75% in 1985 to 32% in 1992 and it was consistently more frequent on bread than on durum wheats. UN13 was the only race present in each area and year (Table 4). In 1990 and 1992, UN5 and UN6 were widely distributed and were second in frequency to UN13. UN2 and UN3 were isolated from bread and durum wheats annually but at low frequencies. UN1 was rare and was isolated mainly from durum wheat. UN1 and UN3 were found primarily in coastal areas 1, 2, and 3, and UN10, UN11, UN12, UN14, and UN17 were rare.

The Shannon index values for virulence diversity ranged from 0.98 in 1985 to 1.93 in 1992 (Table 5). Diversity values in 1990 and 1992 were similar and differed significantly from those in 1985. The diversity indices of *P. r. tritici* isolates from durum wheats were consistently higher than those from bread wheats. The overall diversity indices for the 4 yr were 2.08 for durum wheats and 1.53 for bread wheats. The isolates from durum wheats were generally more diverse, except those from the Sous area where the susceptible bread wheat Siete Cerros was grown under irrigation (Table 6).

DISCUSSION

The leaf rust population in Morocco is highly diverse, with 12 UN races occurring. These races include all genetically possible combinations of virulence and avirulence on genes *Lr*1, 2a, 2c, and 3 (12). UN13, virulent to *Lr*1, 2a, 2c, and 3, was commonly isolated in 1985 and

Table 5. Shannon diversity indices for unified numeration (UN) races of *Puccinia recondita* f. sp. *tritici*

Year / Wheat crop	Index	SE
1985	0.98	0.10
Bread	0.57	0.12
Durum	1.41	0.14
1988	1.69	0.10
Bread	1.44	0.13
Durum	1.94	0.12
1990	1.93	0.07
Bread	1.64	0.10
Durum	2.10	0.09
1992	1.84	0.07
Bread	1.68	0.09
Durum	1.99	0.10
Total (4 yr)		
Bread	1.53	0.06
Durum	2.08	0.08

Table 6. Shannon diversity indices for unified numeration races of *Puccinia recondita* f. sp. *tritici* in relation to crop and ecological area of Morocco

Year / Area	Bread wheat				Durum wheat			
	No. of isolates	No. of races	Shannon index	SE	No. of isolates	No. of races	Shannon index	SE
1985								
Sous	9	4	1.31	0.11	4	2	0.56	0.23
Central plains	26	4	0.58	0.17	23	4	0.64	0.20
Tadla	8	4	...	...	5	1	...	...
Gharb	10	1	...	...	2	2	...	...
Sais	7	1	...	...	1	1	...	...
Jbala	26	2	0.16	0.12	14	14	0.22	0.22
1988								
Sous	10	5	1.41	0.18	5	5	0.95	0.23
Central plains	19	4	0.61	0.23	17	17	1.44	0.17
Tadla	10	5	1.50	0.10	2	2	...	...
Gharb	13	3	0.68	0.20	6	6	1.24	0.20
Sais	12	5	1.09	0.28	4	4	0.69	...
Jbala	11	7	1.66	0.24	9	9	0.93	0.16
1990								
Sous	21	6	1.63	0.10	10	5	1.47	0.16
Central plains	39	7	1.31	0.16	33	10	2.17	0.08
Tadla	5	3	1.05	0.12	7	4	1.15	0.26
Gharb	7	3	0.79	0.27	4	3	1.03	0.17
Sais	3	2	0.63	...	1	1	...	...
Jbala	6	5	1.56	...	11	5	1.52	0.13
1992								
Sous	26	7	1.46	0.17	26	5	1.60	...
Central plains	51	9	1.60	0.13	51	5	1.15	0.28
Tadla	...	...	...	...	...	...	...	...
Gharb	16	5	1.40	0.13	9	6	0.17	0.11
Sais	9	4	1.20	0.18	2	2	0.70	...
Jbala	5	3	0.90	0.23	3	3	1.00	...

1988, but the frequency declined thereafter. UN13 represented 3.3% of the isolates tested in Morocco during 1967–1970 (2); UN3 was the most prevalent during that period, comprising 46.3% of the isolates. In our study, UN13 was commonly isolated from the bread wheat cultivar Nesma, which was widely grown until 1987. The dominance of this race reduced the diversity levels, even though a wide range of races was present annually. Diversity levels varied among years because of the decrease in the area where Nesma was grown.

The diversity for virulence was higher among isolates from durum wheats than among those from bread wheats. Durum wheat cultivars grown mainly in the marginal nonirrigated areas along the Atlantic coast (areas 2 and 6) support a diverse population of *P. r. tritici*. In these areas, infected plants of the alternate host *A. italica* were commonly observed in traditional agricultural fields. However, diversity was significant in irrigated areas of the Sous maintained on the nonselective, fully susceptible bread wheat cultivar Siete Cerros. These diversity levels appear to be higher than those reported by Kolmer (10) and Leonard et al (11) for North America. Race UN1, avirulent to most of the genes tested, was mainly found on durum wheats in areas where *A. italica* was infected naturally by *P. r. tritici* (7). Other studies have shown that the races found on durum wheats are very different from those obtained from bread wheats worldwide (9).

The coexistence of extensive wheat production of local cultivars and intensive production of the semidwarf susceptible cultivars maintains a high diversity of leaf rust races in Morocco. This diversity is generated annually from the alternate host *A. italica*, which occurs in most of the major cereal-growing areas (7). Coordination of efforts to introduce resistance to leaf rust in both bread and durum wheats is necessary. In Morocco, the durum wheat cultivars are not necessarily more resistant to leaf rust than the bread wheat cultivars. Although leaf rust in not an important disease of durum wheat in many areas of the world, it is in Morocco.

Long-term pathogenicity studies are also important to understand the response of rust fungi to changes in cultural practices, such as the shift from extensive low-input to intensive high-input agriculture. In Morocco, such a shift would greatly reduce the occurrence of *A. italica* and thereby the diversity generated by the sexual cycle of *P. r. tritici*.

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