Phytopathologia Mediterranea (2012) 51, 3, 577–586

Research Papers

Pathotype diversity among Algerian isolates of *Pyrenophora teres* f. *teres*

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Summary. Barley net blotch, caused by *Pyrenophora teres* f. *teres (Ptt)*, is one of the most important foliar diseases in north western Algeria. The disease causes appreciable yield losses under favorable environmental conditions. Studies on pathogen variability and distribution of virulence in *Ptt* are essential to identify effective sources of resistance to this disease. In this study, pathogenic variability in 48 isolates of *Ptt*, collected from different barley-growing areas of north-western Algeria, were evaluated using 22 barley genotypes as differential hosts. Trials carried out under controlled conditions confirmed the large variation in the virulence of this fungus. The genotypes exhibited variability in response ranging from complete resistance to high susceptibility. This is the first report of pathogenic variability in *Ptt* in north western Algeria. The 48 isolates tested were further differentiated into 12 pathotypes. Eight differential cultivars were resistant to all isolates tested, but others were susceptible to one or more of the isolates. The variability in the pathogen and the resistance identified in some genotypes used in this study are being investigated further to develop superior, adapted germplasm for use in barley breeding programs in north western Algeria.

Key words: *Hordeum vulgare*, *Drechslera teres*, pathogenic variability, virulence testing, disease resistance.

Introduction

Barley net blotch. caused by the Ascomycete fungus *Pyrenophora teres* f. *teres (Ptt)* Smedeg. [Anamorph: *Drechslera teres* (Sacc.) Shoem. f. *teres* Smedge.], is one of the most widely distributed foliar diseases of barley (*Hordeum vulgare* L. emend Bowden) and can cause substantial yield losses (Shipton *et al*., 1973; Steffenson *et al*., 1991). This disease most severe in temperate regions of high rainfall and humidity, but epidemics have occurred in low rainfall areas as well (Steffenson and Webster, 1992). Two types of leaf symptoms are associated with net blotch: the net type, caused by *Ptt*, which causes horizontal and vertical crisscrossed dark brown venation that sometimes turns chlorotic; and the spot type, caused by *P. teres* f. *maculata*, which causes dark brown circular or elliptical spots accompanied by chlorosis of the surrounding leaf tissue (Smedegård-Petersen, 1971). Both pathogens are present in north-western Algeria, but the *teres* form is more common (Boungab *et al*., 2009) and can cause an average loss of 40 % of the annual value of the barley crop in North Africa (Algeria, Tunisia and Morocco) (Sayoud *et al*., 1999).

The life cycle of *Ptt* involves both asexual and sexual stages. The asexual component includes overwintering of the pathogen as mycelium in seed or in crop debris. The sexual pseudothecia form in barley straw after harvest in autumn. *Ptt* is a heterothallic haploid fungus that produces pseudothecia which contain ascospores. Ascospores are discharged early in the growing season, and are the primary inocu-

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ISSN (print): 0031-9465
ISSN (online): 1593-2095

lum for net blotch induction. During most of its life cycle, *Ptt* produces haploid asexual conidia that are the secondary inoculum. Conidia are dispersed by wind and rain to produce disease throughout the growing season when environmental conditions are favorable (Smedegård-Petersen, 1972; Shipton *et al*., 1973). Infection on barley seedlings is caused by *D. teres* conidia or *P. teres* ascospores, and occurs at cool temperature (10–15°C). The role of ascospores in the life cycle of *Ptt* is considered more as a source of novel variation than as an important source of primary inoculum (Shipton *et al*., 1973). In Algeria, the most severe damage caused by seed-borne infection of *Ptt* occurs in dry soils at temperature of 12°C (Youcef-Benkada *et al*., 1994).

In cell culture, *Ptt* produces two types of toxins: low molecular weight compounds (LMWCs) derived from aspergillomarasamine and proteinaceous toxins (Sarpeleh *et al.,* 2007). The LMWCs cause chlorosis in plant tissue regardless of species (Sarpeleh *et al*., 2009), whereas the protein mixture causes necrosis on some cultivars of barley and not others, suggesting this contains host-specific toxins (Sarpeleh *et al*., 2008).

Several studies of pathotypes of *Ptt* have focused on the variability of virulence in the pathogen in field populations (Tekauz, 1990; Steffenson and Webster, 1992; Jonsson *et al*., 1997; Gupta and Loughman, 2001; Cromey and Parkes, 2003; Wu *et al*., 2003). Based on the resistance genes present in the differential sets used, high levels of variability were identified in worldwide collections of *Ptt,* indicating that many different virulence factors are present in the populations. These factors could be either virulence genes used to induce infection, avirulence genes recognized by the host to induce resistance or a combination of these mechanisms. Based on the common differential lines used, it is estimated that as many as 20 resistance genes could be present in cultivated barley germplasm (Lai *et al*., 2007).

The genetic structure of fungal pathogen populations is a key indicator of how rapidly a pathogen is evolving, and this can be used to predict how long a control measure or resistance source is likely to be effective (Campbell *et al*., 2002; MacDonald and Linde, 2002; Serenius *et al*., 2007).

Little is known about the genetic complexity of *Ptt* in Algeria, and understanding the pathotypes of the pathogen is essential to guide the development of appropriate strategies for disease management that will enhance durability of the resistance. Determination of both host specificity and genetic diversity in pathogen population are prerequisites to breeding for durable resistance.

Several studies have documented high variability in the pathogenicity of the fungus, and different pathotypes have been reported from many countries using a range of differential barley lines (Harrabi and Kamel, 1990; Tekauz, 1990; Steffenson and Webster, 1992; Afanasenko *et al*., 1995; Cromey and Parkes, 2003; Wu *et al*., 2003).

The objective of the present study was to determine the virulence spectrum of Algerian populations of *Ptt,* and to identify new sources of resistance within differential cultivars that may constitute effective material for breeding against barley net blotch in Algeria.

Materials and methods

Survey and collection of isolates

In spring of 2008, 2009 and 2010, leaves with characteristic net blotch symptoms were collected from barley fields distributed across all barley growing regions of north western Algeria. Information on the origin, year of collection and host source of each isolate used is listed in Table 1. The infected leaves from each location were placed in paper envelopes, air-dried at room temperature and stored at 20 to 25°C until used to isolate the pathogen. One hundred isolates of *Ptt* were collected from barley in the major cereal-producing regions. Based on sporulation and pathogenicity tests on cv. Saida (highly susceptible Algerian cultivar), 48 of the isolates were screened, to characterize the virulence spectrum of *Ptt*.

Differential barley genotypes

A set of 22 barley lines was used in this study (Table 2), provided by the USDA World Barley Collection; they originated from various areas of the world, and contained different resistance genes, e.g. the Ethiopian 2-rowed barley line CIho9819 carries two independent major genes for net blotch resistance: *Rpt5*, active against net-type isolates; and *Rpt6*, active against specific spot-type isolates (Manninen *et al*., 2006). Bockelman *et al*. (1977) identified a resistance gene, *Rptla*, on chromosome 3 in barley cv. Tifang and a resistance gene, *Rpt3d*, on chromosome 2 in

Table 1. Geographical origin and pathotypes of the *Pyrenophora teres* f. *teres* isolates obtained from diseased barley from different departments in north-western Algeria.

Region	Isolate	Department	Location of sampling area	Host cultivar	Date of collection	Pathotype
Costal plains	1	Ain temouchent	Chentouf	Saida	2008	0 a
	2	Ain temouchent	El Amria	Saida	2008	22
	3	Ain temouchent	Tadmaya 1	Saida	2008	22
	4	Ain temouchent	Tadmaya 2	Saida	2009	20-22
	5	Ain temouchent	Hassasna	Saida	2009	20-22
	6	Ain temouchent	Ain Tolba 1	Saida	2009	3-9-10-22
	7	Ain temouchent	Ain Tolba 2	Saida	2009	2-6-13-16-18-20
	8	Ain temouchent	Ain Kihal	Saida	2009	2-3-6-10-13-16-18-20
	9	Mostaganem	Sidi Lakhdar	Saida	2008	0
	10	Mostaganem	Oued El Kheir 1	Saida	2008	3-9-15
	11	Mostaganem	Oued El Kheir 2	Saida	2008	3-9-15
	12	Mostaganem	Achaacha	Saida	2008	3-9-15
	13	Mostaganem	Bouguirat	Saida	2008	3-9-10-22
	14	Mostaganem	Bouguirat	Saida	2010	3-9-10-22
	15	Mostaganem	Khadra	Saida	2010	3-10-15-19-21
	16	Mostaganem	Sidi Ali	Saida	2010	3-10-15-19-20-21
Interior plains	17	Mascara	Ferme expérimentale	Tichedrett	2008	0
	18	Mascara	Maoussa	Saida	2008	0
	19	Mascara	Khalouia	Saida	2009	22
	20	Mascara	Tighennif	Saida	2009	22
	21	Mascara	El Bordj	Saida	2009	22
	22	Mascara	Mamounia	Saida	2009	3-9-10-22
	23	Mascara	Ain Fares	Saida	2010	3-10-15-19-21
	24	Mascara	Oued El Abtal	Saida	2010	3-10-15-19-21
	25	Sidi-belabes	ITGC Sidi-belabes	Rihane 03	2008	10-20-22 (1)
	26	Sidi-belabes	ITGC Sidi-belabes	Saida	2008	3-9-10-22
	27	Sidi-belabes	Sfisef	Saida	2009	3-9-10-22
	28	Sidi-belabes	Sfisef	Saida	2009	3-9-10-22
	29	Sidi-belabes	Lamtar	Saida	2009	3-10-15-19-21
	30	Sidi-belabes	Tellagh	Saida	2009	3-10-15-19-21
	31	Sidi-belabes	Belarbis	Saida	2010	3-10-15-19-21
	32	Sidi-belabes	Chettoune	Saida	2010	3-10-15-19-20-21

(*Continued*)

Table 1. *Continues.*

Region	Isolate	Department	Location of sampling area	Host cultivar	Date of collection	Pathotype
High plateau	33	Saida	ITGC Saida	Saida	2009	15
	34	Saida	ITGC Saida 1	Rihane 03	2009	15
	35	Saida	ITGC Saida 2	Tichedrett	2009	15
	36	Saida	Sidi Boubekeur	Saida	2010	22
	37	Saida	Sidi Ahmed	Saida	2010	22
	38	Saida	Maamora	Saida	2010	22
	39	Saida	El Hassasna 1	Saida	2010	3-9-10-22
	40	Saida	El Hassasna 2	Saida	2010	3-9-10-22
	41	Tiaret	El Malah	Saida	2008	20-22
	42	Tiaret	Dahmouni	Saida	2008	20-22
	43	Tiaret	Mahdia	Saida	2009	10-11-22
	44	Tiaret	Rahouia	Saida	2009	10-11-22
	45	Tiaret	Frenda	Saida	2009	3-9-10-22
	46	Tiaret	Hamadia	Saida	2009	3-9-10-22
	47	Tiaret	Takhemaret	Saida	2009	3-10-15-19-20-21
	48	Tiaret	Tamda	Saida	2009	3-10-15-19-20-21

[a] Pathotype designations are given in Table 3.

CIho7584. These hosts have been used as differentials to evaluate the pathogenic variability among isolates of *Ptt* by previous authors (Steffenson and Webster, 1992; Cromey and Parkes, 2003; Wu *et al*., 2003).

Five to eight seeds of each cultivar were surface disinfected with 2% sodium hypochlorite for 5 min, then rinsed in twice in sterile distilled water. The seeds were then sown in 15 cm diam. plastic pots in a pasteurized soil mix (two parts river sand and one peat moss with nutrients and trace elements, pH 6.8 to 7). Plants were grown in controlled environment chambers at 20°C and 12 h photoperiod for 2 weeks or until the second leaf on plants were fully opened.

Inoculum production

Leaf tissue with net blotch lesions was cut into 5 to 10 mm diam. fragments, surface-disinfected in 2% sodium hypochlorite solution for 2 min, and then rinsed twice in sterile deionized water for 1 min. Fragments were blotted dry, and aseptically transferred to moist chambers and incubated at 20°C with a 12 h photoperiod to induce sporulation (Tekauz, 1990). After 2 to 3 d, a single conidium representing each collected sample was transferred, using a needle under a microscope to plates of 17.7% V8 juice agar (177 mL of vegetable juice, 3 g $CaCO_3$ and 16 g agar L^{-1} of distilled water) and incubated for 10–14 d under the conditions described above (Steffenson and Webster, 1992). The virulence and sporulation of *P. teres* can vary greatly, especially after successive sub-culturing (McDonald, 1967). To reduce such variation, and to confirm the pathogenicity of the fungus, seedlings of cv. Saida at the two to three leaf stage were inoculated with a conidium suspension from each of the isolates (see method below). Infected leaves were collected approx. 2 weeks after inoculation, dried and used to produce further inoculum.

Inoculation and disease scoring

For plant inoculation, inoculum was prepared by washing conidia and conidiophores from 14-d-

Table 2. Barley genotypes used as differential lines to evaluate the pathogenic variability of isolates of *Pyrenophora teres* f. *teres*.

No.	Genotype	CIho number[a]	Country of origin
1	Tifang	14373	China, Heilongjiang
2	Canadian Lake Shore	2750	United States, Wisconsin
3	Atlas	4118	United States, California
4	Rojo	5401	United States, California
5	Coast	2235	United States, Idaho
6	Manchurian	739	Canada, Ontario
7	Ming	4797	China, Heilongjiang
8	CIho9819	9819	Ethiopia, Welo
9	Algerian	1179	Algeria
10	Kombar	15694	United States, California
11	CIho11458	11458	Poland
12	CIho5791	5791	Unknown
13	Harbin	4929	China, Manchuria
14	CIho7584	7584	United States, Tennessee
15	Prato	15815	United States, California
16	Manchuria	2330	United States, Minnesota
17	CIho5822	5822	Unknown
18	CIho4922	4922	China, Heilongjiang
19	Hazera	12673	Israel
20	Cape	1026	Australia, New South Wales
21	Beecher	6566	United States, Colorado
22	Rika	8069	Sweden, Malmohus

[a]Ciho, Cereal investigation *Hordeum.*

old V8 juice agar cultures by adding sterile distilled water and scraping each colony surface with a metal spatula. The suspension was homogenized for 1–2 min in a Waring Blendor, filtered through gauze and adjusted to a concentration of 2×10^4 conidia per mL using a haemacytometer (Brown *et al*., 1993; Gupta *et al*., 2003; Jalli, 2011). A drop of Tween 20 per 500 mL of suspension was added as a wetting agent (Douiyssi *et al*., 1998).

The inoculum was applied to plants at the three-leaf stage using an airbrush sprayer. In each experiment, as an experimental control, an uninoculated set of plants was sprayed with sterile distilled water. The plants were incubated in the dark for 48 h at 20°C and at 100% RH. They were then returned to the previous conditions for symptom development. The tests were conducted in three replicates.

Infection responses were assessed based on the ten point scale of Tekauz (1985) for net blotch. This scale is quantitative and is based on lesion size and morphology. Ratings from 1 to 5 were classified as low infection responses and those from 6 to 10 as high infection responses. Infection responses on the second leaves of plants were scored 9 d after inoculation.

Pathotypes designations

The nomenclatural system used for pathotype designations in this study followed the system described by Steffenson and Webster (1992) and Wu *et al.*, (2003). Each number in a pathotype designation corresponds to the numbered genotype for which the isolate induced severe symptoms. e.g. a pathotype designated 10-11-22 indicates that this isolate was virulent on cv. Kombar (No. 10), CIho1458 (No. 11) and cv. Rika (No. 22). Pathotype 15 was virulent only on cv. Prato (No. 15). Isolates exhibiting low infection responses on all the differential barley genotypes were designated as pathotype 0.

Results

Pathogenic variation of *Pyrenophora teres* f. *teres* isolates

The 48 isolates tested exhibited large variation in their pathogenic reactions, ranging from 1 to 10. Only a few isolates showed identical reactions to each other on the differential set. The mode and range of infection responses elicited by each pathotype on the differential genotypes are shown in Table 3. All tested isolates produced typical net blotch symptoms on susceptible plants 9 days after inoculation. Disease development progressed rapidly, showing the effectiveness of the inoculation method used, and symptoms differed mainly in the extent of lesions. Disease symptoms were observed first on the most susceptible cultivars (cv. Rika, Atlas and Kombar) within 48 h after inoculation. Control plants did not develop any symptoms. After comparing the reactions of the differentials host, the isolates of *Ptt* could be divided into 12 pathotypes according to their reactions (Table 3). The most common pathotype (3-9-10-22) comprised 11 (23%) of the 48 isolates tested. This pathotype caused disease on four genotypes among the 22 tested. Pathotypes 22 and 3-10-15-19-21 were the next most prevalent with frequencies of 17 and 23%, respectively. The other pathotypes were found in frequencies of less than 9%.

Two pathotypes (15 and 22) were virulent on only one differential genotype. Four isolates were virulent on cv. Cape and Rika (pathotype 20-22). Pathotypes 10-11-22, 3-9-15 and 10-20-22 were each virulent on three cultivars. One isolate was the most complex pathotype (2-3-6-10-13-16-18-20), with sensitivity reactions on eight of the differentials, while pathotype 0 was avirulent on all of the differentials.

Reaction of barley cultivars to *Pyrenophora teres* f. *teres*

According to the reactions of the genotypes tested against the different isolates of *Ptt* under controlled conditions, differences in the levels of resistance between cultivars became apparent. Eight of the 22 differential barley genotypes tested were resistant to all of the isolates: including cv. Coast', CIho9819, CIho5791, CIho7584, CIho5822, and cv. Ming, Tifang and Rojo.

The genotype cv. Rika was the most susceptible of the differentials, attacked by 26 isolates (54%) of the 48 tested, followed by cv. Atlas and Kombar which were susceptible to 52% of the isolates. Cultivars Prato and Algerian were susceptible to 33 and 29% of the isolates, respectively. The next most susceptible group of hosts included cv. Cape susceptible to 23% of the isolates, and cv. Hazera and Beecher which gave high infection responses to 21% of the investigated isolates (Figure 1).

During the experiment, the genotypes: cv. Canadian Lake Shore, Manchurian, CIho11458, Harbin, Manchuria and CIho4922 showed the same level of resistance, and developed disease from only two isolates (4%).

Occurrence and distribution of virulence in *Pyrenophora teres* isolates

Results showed clearly that *Ptt* isolates from different Algerian regions differed in virulence. Variations within department were also evident. Based on the virulence test, isolates from the coastal plains were the most diverse, as isolates from this region comprised the greatest number of pathotypes. The interior plains and the high plateau were intermediate in the number of pathotypes. The most complex pathotypes, 2-6-13-16-18-20 and 2-3-6-10-13-16-18-20, were present only in Ain Temouchent (coastal plains) at a low frequency. Pathotypes 15 and 10-11-22 were specific to the high plateau (region 3) where barley is grown extensively, while other pathotypes were found in different regions. The most common pathotype 3-9-10-22 occurred throughout the surveyed region, and 11 isolates were of this pathotype (Table 1).

Discussion

The 48 *Ptt* isolates tested in this study showed the presence of 12 pathogenicity groups according to their reactions on the differential set of hosts. In

Table 3. Pathotype designations for isolates of *Pyrenophora teres* f. *teres* collected from barley growing areas in north western Algeria.

No.	Genotype	Pathotype (Mode/Range)											
		0[a]	15	22	20-22	10-11-22	3-9-15	10-20-22	3-9-10-22	3-10-15-19-21	3-10-15-19-20-21	2-6-13-16-18-20	2-3-6-10-13-16-18-20
1	Tifang	1/1-3	2/1-3	2/1-4	2/1-3	3/2-3	2/1-3	1/1-2	2/1-3	2/1-3	2/1-3	1/1-2	1/1-3
2	Canadian Lake Shore	1/1-3	1/1-3	2/1-3	3/2-4	4/3-4	2/1-3	2/1-3	3/2-5	3/2-5	4/2-5	7/7-8	6/6-7
3	Atlas	3/1-4	4/3-4	3/3-5	3/2-5	5/4-5	9/7-9	3/2-3	8/7-9	9/7-10	9/8-10	2/1-3	9/8-10
4	Rojo	1/1-2	1/1-2	1/1-2	2/1-3	1/1-2	2/2-3	1/1-2	2/1-3	1/1-3	1/1-2	1/1-2	1/1-2
5	Coast	1/1-3	1/1-3	1/1-3	1/1-2	2/1-3	2/1-2	1/1-2	2/1-3	1/1-3	2/1-3	1/1-2	2/2-3
6	Manchurian	2/1-3	2/2-3	2/2-4	2/2-3	2/2-4	2/1-3	3/2-3	2/1-3	3/2-5	3/2-3	8/7-9	7/7-8
7	Ming	1/1-2	2/1-3	2/1-3	2/1-3	3/3-5	3/2-5	2/1-2	2/2-3	2/2-3	3/2-3	2/1-3	2/2-3
8	CIho9819	1/1-2	2/1-2	1/1-3	2/1-2	1/1-2	2/1-2	2/2-3	2/1-3	2/2-5	2/1-3	1/1-3	1/1-3
9	Algerian	3/2-5	3/2-4	3/2-4	3/1-4	4/3-5	7/5-8	4/3-4	7/6-9	3/2-3	4/2-5	2/2-3	3/2-4
10	Kombar	3/1-4	4/3-5	4/3-5	4/2-5	7/6-9	5/4-5	9/9-10	9/8-10	8/8-9	9/8-10	4/3-4	9/8-10
11	CIho11458	2/2-4	2/1-3	2/1-4	1/1-3	6/6-8	3/2-3	2/2-3	2/2-3	2/2-5	2/1-3	1/1-3	2/1-3
12	CIho5791	1/1-2	1/1-3	2/1-3	2/1-3	2/1-2	2/1-2	2/1-2	2/1-3	2/1-3	2/1-3	1/1-2	1/1-2
13	Harbin	1/1-3	2/2-4	2/1-3	2/2-3	3/2-4	2/2-3	2/1-2	3/2-5	3/2-3	2/2-5	7/7-8	6/6-7
14	CIho7584	1/1-3	2/1-2	2/1-4	2/1-2	3/2-3	2/1-2	1/1-2	2/2-5	3/2-5	2/2-5	2/1-3	1/1-2
15	Prato	3/2-5	7/6-7	4/3-5	3/2-5	4/3-4	8/7-9	4/4-5	4/3-6	8/7-9	8/8-9	2/2-3	3/2-3
16	Manchuria	1/1-2	2/2-3	2/2-4	2/1-3	3/2-3	2/2-3	2/1-2	3/2-5	2/1-3	3/2-3	7/7-8	7/7-8
17	CIho5822	1/1-4	2/1-3	2/2-4	2/1-3	2/1-3	2/2-3	1/1-2	2/2-5	2/1-3	2/1-3	1/1-3	2/1-3
18	CIho4922	1/1-2	2/1-3	2/1-3	2/1-2	2/1-2	2/1-2	2/1-2	3/2-5	2/2-3	3/2-3	6/6-7	7/7-8
19	Hazera	2/1-5	3/1-4	3/2-4	3/2-4	3/2-3	2/2-3	3/2-3	4/2-5	7/6-9	8/7-9	2/2-3	3/2-4
20	Cape	2/2-5	2/1-3	3/2-4	7/5-8	2/1-3	3/2-3	8/8-9	3/2-4	3/2-5	8/8-9	9/9-10	9/9-10
21	Beecher	3/1-4	3/2-4	4/3-5	3/2-5	4/3-5	3/2-4	3/2-3	4/2-5	7/7-9	7/6-9	3/2-3	4/3-4
22	Rika	3/2-5	4/3-4	8/7-10	8/7-9	9/8-10	5/4-5	9/9-10	9/8-10	4/3-4	4/4-5	2/2-5	1/1-3
Total number of isolates/%		4/8	3/6	8/17	4/8	2/4	3/6	1/2	11/23	6/13	4/8	1/2	1/2

[a] Pathotype nomenclature is according to Steffenson and Webster (1992) and Wu *et al*. (2003).
[b] The mode represents the most common reaction type induced by an isolate within a specific pathotype.
[c] The range represents the virulence phenotypes (low - high virulence) of isolates on 22 barley differentials.

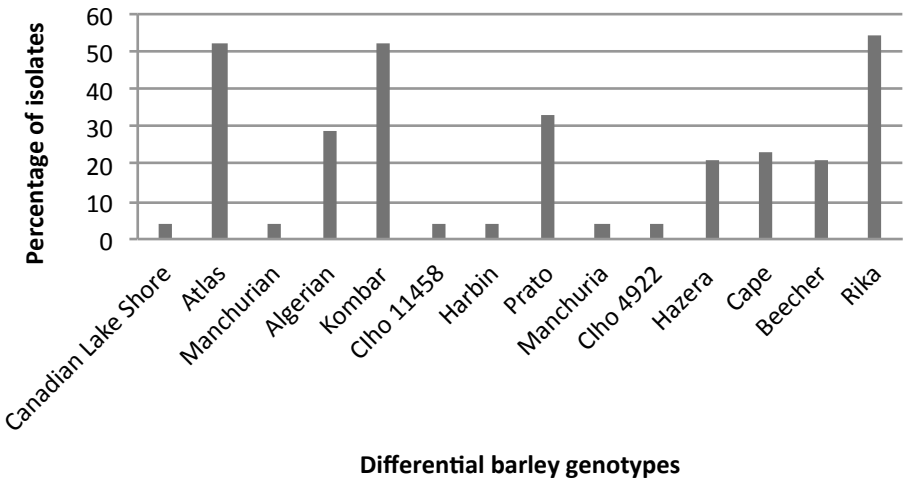

Figure 1. Proportions of tested isolates of *Pyrenophora teres* f. *teres* with virulence to barley differentials.

a similar study using the same host range (Steffenson and Webster, 1992), California *Ptt* isolates were differentiated into 13 pathotypes. However, as in the present study, the most common pathotype was not particularly virulent.

Considerable variation in the pathogenicity of different isolates of *Ptt* has been reported by researchers. Most studies have not used the same, or the same number, of differential genotypes, or have applied different environmental conditions, so comparison of results between studies is difficult. The environmental factors, temperature, light and humidity, were stable in all trials and their effects were not examined in the present study. However, it has been shown that even the slight changes in environmental condition can significantly affect the expression of plant infection (Jalli, 2010).

The differentials lines or cultivars used in this study have been used quite frequently, suggesting that they are particularly useful for evaluating pathogenic variability in various parts of the world. Tekauz (1990) identified 45 *Ptt* pathotypes using a set of nine barley lines, since this level of variability had not been found in previous studies. Jonsson *et al*., 1997, reported 14 pathotypes of *Ptt* using 18 barley lines in Sweden. In New Zealand, 11 pathotypes were characterized amongst 29 isolates tested against 31 differential genotypes (Cromey and Parkes, 2003), 22 of the 31 cultivars were the same as used in the present study. Wu *et al*., (2003) reported 15 pathotypes from a collection of 23 *Ptt* isolates, whereas the Moroccan population of *Ptt* comprised ten pathotypes (Jebbouj and El Yousfi, 2010).

The results presented here indicate that the barley differential lines have different genes for net blotch resistance. The majority of resistant varieties originated from Ethiopia and Manchuria, with those from Ethiopia providing a higher degree of resistance (Khan and Boyd, 1969). Afanasenko *et al*. (1995) found CIho5791, CIho9819, and cv. Tifang to be the most resistant lines against Russian, German, Czech, and Slovak *Ptt* isolates. Khan (1982) also found barley lines CIho5791 and CIho7584 were resistant pathotypes present in Western Australia, and the same lines were resistant to all Algerian isolates tested in the present study. This indicates that the resistances effective against Australian isolates were also effective against a wide range of virulence. The cv. Coast, Rojo and CIho5822 were also found to be resistant in an international study (Wu *et al*., 2003).

On the other hand, the present study suggests the existence of wide variation in the reaction of barley cultivars to the pathogen around the world. The cv. Beecher, Canadian Lake Shore, Hazera, Manchuria and CIho4922 were resistant to all the pathotypes in New Zealand (Cromey and Parkes, 2003), whereas they were susceptible to some pathotypes in the present study, indicating that the virulence types in New Zealand are different from those in Algeria. The Algerian cultivar which was recorded as resistant in California and in other countries (Steffenson and Webster, 1992; Gupta and Loughman, 2001) was

sensitive to 14 isolates in our study. In addition, cv. Rika, which was the most sensitive in New Zealand (Cromey and Parkes, 2003), was resistant to Algerian isolates, as well as those from Western Australia (Gupta and Loughman, 2001).

Differences between results from the present study and those of the other authors may be due to the occurrence of different fungal populations in Australia, California, New Zealand and Algeria, or to differences in experimental conditions applied in the different studies.

There was a difference in pathotype diversity and complexity among locations in Algeria. Ain Temouchent and Mostaganem sites (region 1) yielded the greatest number of pathotypes including the most complex one. pathotype diversity was greater in the rainy coastal regions than in the interior plains and high plateau with low and irregular rainfall, where environmental conditions are not optimal for net blotch development. Similar results were observed by Steffenson and Webster (1992), who confirmed distinct geographical differences in the distribution of *Ptt* pathotypes in California.

Some pathotypes were specific to a single region while others were found in various regions. The most common pathotype (3-9-10-22) was recorded throughout all the regions. This might be due to seed transmission of *Ptt*, since infected barley is important for natural spread of the pathogen (Peever and Milgroom, 1994). In Algeria, the transfer of barley seed from one region to another is common and uncontrolled.

Isolates originating from one location showed similar levels of variation in pathogenicity to those collected from different locations across north western Algeria. However, localized populations of *Ptt* can exist in some regions because the fungus survives in crop stubble and spreads only short distances within a field or to nearby fields via wind- and rain-borne spores. Moreover, infected volunteer plants of barley or wild *Hordeum* species, which occasionally survive between cropping seasons, may also serve as sources of primary inoculum for newly sown crops. The net form of the net blotch pathogen can attack a number of different grasses (Shipton *et al.*, 1973; Brown *et al.*, 1993), but wild *Hordeum* species are the most likely to play roles in the epidemiology of the disease (Brown *et al.*, 1993).

This study has demonstrated that the population of *Ptt* in Algeria was highly variable from region to region. This might be due to differences in the barley cultivars grown in different regions, which may have exerted different selective pressures on the pathogen (Tekauz, 1990). In accordance, qualitative resistance was reported among Nordic barley genotypes and the Finnish *Ptt* isolates differed in virulence according to the host barley (Robinson and Jalli, 1996).

In conclusion, the results suggest the presence of high variability and virulence in the Algerian population of *Ptt*. Furthermore, eight resistant genotypes were uniform in their responses to the range of isolates tested, and these may provide valuable sources of resistance to net blotch. Incorporation of useful resistance sources effective against diverse pathotypes into adapted genotypes will be necessary to minimize future losses due to net blotch. Effective seed hygiene is also very important in order to prevent spread and introduction of highly virulent strains of the pathogen into new areas.

Acknowledgements

We thank Harold E. Bockelman, Supervisory Agronomist and Curator National Small Grains Collection, USDA, USA, for providing seed barley differential genotypes.

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Accepted for publication: April 16, 2012