Root and Crown Rot Fungi Associated with Spring, Facultative, and Winter Wheat in Turkey

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ABSTRACT

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The objective of this study was to determine the distribution frequency of the fungi associated with wheat (*Triticum aestivum*) crowns and roots in cereal producing areas of Turkey through a targeted survey of 518 commercial fields over a 2-year period. More than 26% of the fields had one or more of the fungal species commonly reported as part of the dryland root rot complex, *Fusarium culmorum* (14%) > *Bipolaris sorokiniana* (10%) > *F. pseudograminearum* (2%). The fungi considered to be part of the high rainfall root rot complex were found at very low frequencies: 2% for *Gaeumannomyces graminis* and 3% for *Pythium* spp. Species of *Rhizoctonia* were found in 22% of the fields. Several *Fusarium* species considered to be less or nonpathogenic to cereals were also found in high frequencies at 11% (*F. oxysporum*, *F. chlamydosporum*), 10% (*F. sporotrichioides*), and 8% (*F. avenaceum* and *F. solani*). The mostly random distribution of cereal root-rotting species across the survey area suggests the fungi are not distributed in any distinct agroecological relationship. As a result, the relative economic importance of a given species on wheat will be determined by a number of factors, such as their fungal pathogenicity, host susceptibility/tolerance, and the seasonal conditions. Results from this study suggest that there are a wide range of fungal species associated with root and crown tissues of wheat.

Turkey is among the 10 largest wheat (*Triticum aestivum* L.) producers worldwide with annual production varying between 16 and 21 million tonnes, including 4 to 6 million tonnes of durum wheat (*Triticum durum* L.). Turkey's climate is extremely diverse (10). Southern and western coastal regions have Mediterranean, hot and dry summers with mild, rainy winters. The northern coast is a high rainfall region. Central Anatolia has continental, dry, hot summers and cold, low rainfall and low humidity winters. Eastern Anatolia has extremely cold winters. The European part of Marmara region has moderately cold winters and humid summers. National average grain yield is approximately 2 t ha⁻¹ from the total wheat production area of 9.5 Mha; however, due to diverse agroecological conditions it varies greatly from 1 t ha⁻¹ in the eastern region

of the country to 3 t ha⁻¹ in the European part of Marmara region (10).

Guler et al. (27) classified Turkey into seven agroecological wheat zones (Fig. 1 and Table 1), which define the wheat type in relation to climatic zones. Scarcity and poor distribution of rainfall is the major constraint for wheat production zones 1 to 3, 4B, and 5C. Variable rainfall distribution can cause sporadic drought or water logging in the higher rainfall zones 6 and 7. Higher elevations (over 800 m) are associated with the production of winter wheat, which requires a vernalization period over the winter months, while lower, mostly coastal elevation zones are associated with the production of spring wheat. The transitional zones between the coast and higher elevation usually grow facultative wheat.

Approximately 90% of this wheat area is grown under rainfed or semi-supplementary irrigation conditions, most of which can be considered drought stressed, particularly postanthesis (10). The cropping system is a wheat-fallow rotation, although limited grain legume rotations are practiced in some regions.

Root and crown rots are important diseases of cereals worldwide and are found wherever cereal-based farming systems

predominate. Invasion of crown and root tissues by root and crown rotting fungi diminishes the plant's capacity for efficient nutrient and water uptake. Damage caused by these pathogens is difficult to assess from year to year due to differences in location, crop management, and climatic factors. Recognition of the role of soil-borne pathogens as a limiting factor for wheat production potential in most areas is problematic due to a focus on more visible foliar and head diseases. Identification and quantification of root diseases involves more laborious procedures than the simple visual observations needed to detect and quantify the presence of foliar and head pathogens (43,51). In addition, there are usually many species of root rotting fungi associated with root disease and yield loss; however, their economic importance is predominately determined by plant tolerance and the associated environmental conditions.

The dryland root rot complex (DLRRC) is known by a variety of names including dryland foot rot, *Fusarium* foot rot, crown rot, dryland root rot, and common root rot (41). Pathogens associated with the complex include *Bipolaris sorokiniana* (Sacc.) Shoemaker (Syn. = *Helminthosporium sativum*, *H. sorokinianum*, teleomorph *Cochliobolus sativus* ((Ito & Kurib.) Drech. ex Dast.), *Microdochium nivale* (Fr.:Fr.) Samuels & I.C. Hallett (= *Fusarium nivale* (Fr.) Sorauer), and several *Fusarium* spp. The two most frequently reported *Fusarium* species are *F. pseudograminearum* O'Donnell & Aoki (formerly *F. graminearum* Group 1, teleomorph *Gibberella coronicola*) and *F. culmorum* W.G. Smith, while several others such as *F. acuminatum*, *F. avenaceum*, *F. oxysporum*, and *F. crookwellense* have also been reported (12,25,47). *Fusarium* spp. are a diverse group of fungi which damage small-grain cereals by rotting the seed, seedlings, roots, crowns, basal stems, and in some cases, infect the heads or spikes (41). Damage to spring and winter cereals often goes unnoticed until prematurely ripening heads (whiteheads) appear shortly before the crops mature (41). *B. sorokiniana* causes a soilborne disease of

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seedlings similar to that of *Fusarium* spp., and in particular causes browning of the subcrown internode. However it is not associated with the “white-head” or premature death of wheat tillers. *Microdochium nivale* is an important pathogen in cool temperate zones and causes snow mold of winter cereals. It also causes seedling infection and foot rot during cool

temperatures. In the cooler areas of the subtropics, this organism causes leaf blotches on wheat, triticale, and rye (54). These different species may occur singly, but they often coexist in the same fields and even within individual plants (47).

Once plants are infected, the root rotting complex is most damaging under moisture-restricted conditions (6,12,16,40–42).

Although many species are associated with the DLRRRC, *F. pseudograminearum*, *F. culmorum*, and *B. sorokiniana* are the most pathogenic and economically important pathogens (12,41,46). In the Pacific Northwest of the United States, this complex causes latent infections with the measurable effects on yield becoming most apparent when the cereals are subjected to

Fig. 1. Survey sites across geographical regions and agroecological wheat field growing zones of Turkey where wheat fields were sampled in 2001 and 2003. Most sites included more than one field. Geographical regions and agroecological zones were described by Braun et al. (10) and Guler et al. (27), respectively.

Table 1. Agroecological zone and geographical regions of wheat production in Turkey with respect to climate, production area, yield, and wheat growth habit as reported by Guler et al. (27) and Braun et al. (10)

Agroecological zone ^a	Wheat growth habit ^b	Rainfall 2000 (mm)	Rainfall 2001 (mm)	Annual rainfall (mm) (1994-98)	Mean temp (°C) 2000	Mean temp (°C) 2001	Mean temp (°C) 1994-98	Area (ha)	Ave yld (t/ha)	National production (%)
1	W			700			8	536	1.13	3
2	W			435			8	715	1.53	6
3A	W, F			350			11	3,650	1.85	38
3B	W, F			380			13	249	1.46	2
4A1	W, F			480			14	402	3.06	7
4A2	F			490			14	582	1.40	5
4B	F			320			22	678	1.50	6
5A1	S			745			17	303	2.30	4
5A2	S, F			700			15	638	2.70	10
5B	S, F			750			15	486	2.32	6
5C	S			420			18	403	1.75	4
6	S			799			18	109	3.04	2
7	S			540			19	566	2.66	8
Geographical region ^c										
Cent. Anatolian Plateau	W	330	362	358	11	13	12			
Aegean	S	670	682	616	17	18	17			
Marmara	S	695	727	752	15	15	14			
South East Anatolia	S, F	516	550	536	17	17	17			
Black Sea	S	520	432	548	13	15	14			

^a Agroecological zone. After Guler et al. (27) given in Figure 1.

^b Winter (W), facultative (F), spring (S).

^c Geographical region as illustrated in Figure 1.

water stress later in the growing season and the temperatures are high (41). Burgess et al. (12) reviewed crown rot and noted that infection by *F. pseudograminearum* only occurs in relatively moist soils with optimum water potential of -0.3 to -0.6 MPa.

These dryland root rots have been found in most parts of the world, and especially under drought stress, they have caused yield loss exceeding 50% (35,37,41). Crown rot caused by *Fusarium* spp. is of economic importance in Australia, Europe, North America, South America, West Asia, North Africa, and South Africa (14). Recently, losses of up to 43% on common bread wheat cultivars have been reported from the Central Anatolian Plateau of Turkey (29), with additional detrimental effects on grain quality (2,4).

Rhizoctonia spp. are also a commonly reported cause of damaged cereal root and crown tissue. *Rhizoctonia* root rot and bare patch are caused by *Rhizoctonia solani* Kühn AG-8, which causes lesions and pruning of seminal and crown roots, particularly in rained cereal production systems in Australia and the U.S. Pacific Northwest (41). *Rhizoctonia oryzae* Ryker & Gooch also causes root rot in the Pacific Northwest (41). Sharp eyespot, caused by *Rhizoctonia cerealis* E.P. Hoeven (teleomorph *Ceratobasidium cereale* D. Murray & L.L. Burpee), causes lesions on lower tillers resulting in premature death of tillers and whitehead formation. Sharp eyespot is found particularly in temperate regions of the world including The Netherlands, Germany, South Africa, China (28), United Kingdom (15), and New Zealand (20). This disease has caused losses of up to 18% on winter cereals in New Zealand (20) and 26% in England and Wales (15). *R. cerealis* has also been reported to be widespread in Turkey and is a potentially economically important crown pathogen of wheat (4,5).

To date in Turkey three smaller regional surveys (3,4,33) have also implicated *F. culmorum*, *B. sorokiniana*, and *Rhizoctonia* spp. as the main causal agents of dryland root rot on wheat.

The high rainfall root rot complex (HRRRC) is associated with several species of fungi, including *Gaeumannomyces graminis* (Sacc.) Arx & Olivier var. *tritici* Walker, the cause of take-all disease, and various species of *Pythium* which have been reported to cause losses under wetter soil conditions. Take-all is most damaging under wet conditions where it causes patches of stunted, prematurely blighted plants often with black mycelial growth on and within the roots, crown, and tiller bases (41,50). Take-all appears to be under-recognized in low-rainfall dryland environments such as the U.S. Pacific Northwest (41), where the fungus infects individual plants instead of plants in patches, and growth of the fungus from

roots upward usually stops in the crown rather than moving upward onto tiller bases.

Pythium spp. infect germinating seeds and root tips, resulting in the stripping of fine rootlets and root hairs of plants (41). Cook and Veseth (19) have referred to *Pythium* as the "common cold", with its highest frequency and severity being found in high-rainfall areas with cool, wet soils covered with crop debris (17). *Pythium* root rot has been reported as a yield-limiting disease on wheat and barley produced in North America, Australia, and the United Kingdom (9,18,39,41,50).

The survey described in this paper was conducted throughout the main cereal growing regions of Turkey with the objective of determining the distribution frequency of root and crown rotting fungi. The distribution of these pathogens was assessed in relation to agroecological factors associated with wheat production in different parts of the country.

MATERIALS AND METHODS

Ecological and regional zones for survey areas and sampling. The study was carried out in six representative wheat growing regions of Turkey including the Central Anatolia Plateau, Marmara, Aegean, Black Sea, Mediterranean, and South East Anatolia agroecological wheat zones defined by Guler et al. (27; Fig. 1 and Table 1). Bread and durum wheat samples were collected near plant maturity (growth stage 92 of the Zadoks scale) during June and July in 2000 and 2001 (53). Fields were selected at random based on an approximate separation distance of 20 km, field size of approximately 10 ha, the maturity of wheat plants, and field accessibility. At each site, five sampling points were made in a zig zag pattern (36), with each point being 20 m apart. At each of the five points, approximately 20 plants were collected, and on return to the laboratory 12 main stems with intact crown and sub-crown tissue were randomly selected from each transect point, making a total of 60 main stems per site. The plants and roots collected at each site were stored in paper bags at 4°C until fungal isolation was performed.

A differing geographical focus of sampling occurred during the two survey years. In 2000, the lower elevation and wetter environments of the Aegean and Marmara regions were sampled, and all other regions were sampled in 2001.

Isolation and identification of fungi. The 60 selected stems from each field were washed thoroughly under running water. Two diseased 2-cm crown sections and one diseased 2-cm root section were taken from each stem (180 sections per survey site). These sections were surface-sterilized with 1% w/vol sodium hypochlorite for 3 min, rinsed three times in sterile distilled water, and dried on sterile

filter paper. Sections were plated onto full-strength potato dextrose agar (PDA) medium. The plates were incubated at $24 \pm 1^\circ\text{C}$ for 4 to 5 days under a 12-h photoperiod. Isolates were then transferred to full-strength PDA tubes for further identification.

The isolates resembling *Fusarium* species were subcultured onto synthetic nutrient agar (SNA) and carnation leaf agar (CLA) for sporulation and identification. All plates were incubated under a combination of white fluorescent and black lights with a 12-h photoperiod at $23 \pm 1^\circ\text{C}$ for at least 7 days. Identification of *Fusarium* species was based on macro and micro conidial morphology, phialid structure, and hyphal characteristics observed under a compound microscope at $\times 200$ to $\times 400$ magnification (9,13,23,26,44). Other fungi were identified using keys based on hyphal and conidial morphologies (7,24).

Data were recorded based on the presence or absence of different fungal species in a field. If a particular fungus was isolated from one or more of the 180 plated plant sections from a given field, then the fungus was considered present. These were then collated for a particular agroecological region or geographic zone (Fig. 1) and expressed as percent frequency based on the number of fields sampled in that region or zone.

Environmental characterization. All fields were geo-referenced and incorporated into a geographical information systems (GIS) framework (Environmental Systems Research Institute, Inc. (ESRI), Redlands, CA; ArcGIS v 9.1). In cases where no precise GPS location data existed, geographical place names recorded in the survey were matched to existing spatial databases of populated places in Turkey. For each survey location, climate (long-term normal precipitation, i.e., monthly averages for the period 1950 to 2000) and elevation data were extracted from the WorldCLIM database (30) using standard point intersect tools within the GIS. The spatial resolution of the WorldCLIM data was 30 arc seconds (approximately 1 km² grid cell sizes). All environmental data were analyzed on either an individual species basis or as a group of species according to their environmental association including DLRRRC and HRRRC.

Data analysis. For the extracted environmental variables, standard parametric statistical tests (chi-square, *t* test, and ANOVA) were used to determine if significant differences existed between means for individual species or groups of species. Proportions of species associated with specific regions or agro-zones were analyzed in the same manner.

In order to determine any significant spatial distribution patterns for the different fungal species, geospatial statistics were used. All fields (with or without

fungi) were included in the analysis, and tests for spatial autocorrelation were undertaken using Moran's I statistic (49). This statistic provides an evaluation of whether the observed distribution pattern for an exhibited fungal species is clustered, dispersed, or random. All tests were implemented within the spatial statistics component of ArcGIS (ESRI). In this analysis, inverse distance was used as the conceptualization of spatial relationships and Euclidean distances with no threshold limit.

For analytical purposes, the root and crown rotting fungi were divided into two groups based on the literature describing the main cereal pathogens and their damage associated with the low or high-rainfall environments. The groupings considered were the DLRRRC, including the most reported causal agents *B. sorokiniana*, *F. culmorum*, and *F. pseudograminearum*, and the HRRRC, including *G. graminis* var. *tritici* and *Pythium* spp. All other species of root and crown rotting fungi were placed in the category of other root and crown fungi.

RESULTS

DLRRRC. More than 26% of fields had one or more of the common DLRRRC species isolated from crown and root tissue of wheat. Considering individual species, 14% of fields had *F. culmorum*, 10% *B. sorokiniana*, and 2% *F. pseudograminearum* (Table 2). A widespread distribution of DLRRRC species was documented throughout Turkey, with at least one of the three key DLRRRC species being found in each of the six geographical regions included in the survey. Similarly, at least one DLRRRC

species was detected in all of the wheat agroecological zones (Table 2). No significant associations were found in the proportion of DLRRRC (all three species combined) by geographical region ($\chi^2 = 0.004$, df 5, NS) or agroecological zone ($\chi^2 = 0.036$, df 10, NS). Despite no association with any particular region or zone, point pattern analysis of the spatial distributions of the three DLRRRC species revealed highly significant spatial autocorrelation (clustering) patterns for each individual DLRRRC species. Using Moran's I statistic, the *z* scores and significance levels for the three DLRRRC species were all highly significant (*F. culmorum* *z* = -9.8, *P* < 0.001; *B. sorokiniana* *z* = -8.6, *P* < 0.001; *F. pseudograminearum* *z* = -3.1, *P* < 0.001). Hence, the spatial distribution of each DLRRRC species exhibited a highly clustered, nonrandom pattern.

Investigation of environmental variables at survey locations (Table 1) revealed indications of potential niche separation between DLRRRC species. As an example, data from the most common species, *F. culmorum*, provides an illustrative example, as year 2000 sites were significantly lower elevation (*t* = 6.16, df 64, *P* < 0.001) and higher annual precipitation (*t* = 7.1, df 57, *P* < 0.001) than year 2001 sites. However, due to the presence-or-absence nature of the survey results and the extensive multi-environment nature of the surveys, it was decided to combine both survey years for analytical purposes. This decision was further justified as analysis of 2001 data revealed the same significant trends as the combined year data results reported here.

Analysis of elevation values extracted from survey locations revealed significant

differences between DLRRRC species (ANOVA, $F_{2,129} = 6.61$, *P* ≤ 0.002), with *B. sorokiniana* occurring at significantly higher elevations than either *F. culmorum* (*t* = 2.9, df 116, *P* < 0.005) or *F. pseudograminearum* (*t* = 3.2, df 62, *P* ≤ 0.002). No significant difference in elevation was observed between *F. culmorum* and *F. pseudograminearum* (*t* = 1.36, df 77, *P* < 0.09, NS).

Annual precipitation, based on extracted historical long-term averages, was not significantly associated with differences of any DLRRRC species (ANOVA, $F_{2,129} = 0.84$, *P* < 0.43, NS). However, investigation of long-term normal precipitation data for specific 3-month periods revealed that *F. pseudograminearum* occurred in significantly higher long-term normal precipitation sites when considering the driest quarter (3 consecutive driest months) than either *F. culmorum* (*t* = 2.2, df 11, *P* < 0.05) or *B. sorokiniana* (*t* = 2.4, df 11, *P* < 0.04). *F. pseudograminearum* also occurred in significantly wetter environments than *B. sorokiniana* in the warmest quarter (*t* = 21.6, df 11, *P* < 0.05).

The incidence of DLRRRC species, calculated as the percentage of fields (*n*) in each region with one or more of the DLRRRC species present, was highest in the South East Anatolia region (40%, *n* = 60), followed by the Aegean (34%, *n* = 137), Mediterranean (24%, *n* = 17), Central Anatolian Plateau (21%, *n* = 200), Marmara (20%, *n* = 80), and Black Sea (8%, *n* = 24) regions (Table 2). Using the same metric for the agroecological zones, the winter wheat zone 1 covering the far eastern part of Turkey had the highest incidence of DLRRRC (45%).

Table 2. Incidence on a field basis of species of the dryland root rot complex^a from wheat roots and crowns by agroecological zone and geographical region in 2001 and 2002

Agroecological zone ^b	No. of fields	Number of fields with species present				Incidence (%)			
		<i>Fc</i>	<i>Fp</i>	<i>Bs</i>	Total DLRRRC	<i>Fc</i>	<i>Fp</i>	<i>Bs</i>	Total DLRRRC
1	11	1	0	4	5	9	0	36	45
3A	185	26	1	12	39	14	1	6	21
4A1	35	6	2	0	8	17	6	0	23
4A2	49	0	0	15	15	0	0	31	31
4B	16	4	1	0	5	25	6	0	31
5A1	22	2	0	0	2	9	0	0	9
5A2	41	3	4	1	8	7	10	2	20
5B	52	7	2	13	22	13	4	25	42
5C	33	7	1	6	14	21	3	18	42
6	57	10	0	2	12	18	0	4	21
7	17	4	0	0	4	24	0	0	24
Geographical region ^c									
Cent. Anatolian Plateau	200	27	1	13	41	14	1	7	21
Aegean	137	17	2	28	47	12	1	20	34
Marmara	80	8	6	2	16	10	8	3	20
Mediterranean	17	4	0	0	4	24	0	0	24
South East Anatolia	60	12	2	10	24	20	3	17	40
Black Sea	24	2	0	0	2	8	0	0	8
Total	518	70	11	53	134				
Incidence (%)		14	2	10	26				

^a Agroecological zone after Guler et al. (27) given in Figure 1.

^b Dryland root rot complex (DLRRRC), which includes the species *Fusarium culmorum* (*Fc*), *F. pseudograminearum* (*Fp*), and *Bipolaris sorokiniana* (*Bs*).

^c Geographical regions as illustrated in Figure 1.

The most commonly isolated DLRRC pathogen was *F. culmorum*, found in 14% of all fields. The highest frequency of isolation, relative to the total number of fields per region (*n*), was from the Mediterranean region (24%, *n* = 17). Lower incidence occurred in South East Anatolia (20%, *n* = 60), Central Anatolian Plateau (14%, *n* = 200), Aegean (12%, *n* = 137), Marmara (10%, *n* = 80), and Black Sea (8%, *n* = 24) regions. This fungus was most frequently isolated from the facultative and spring eco-regions 4B, 5C, and 7, which represent the South Eastern Anatolia and Mediterranean regions and the winter wheat eco-region 3A on the Central Anatolian Plateau (Table 1 and Fig. 1).

B. sorokiniana was the second most commonly isolated DLRRC pathogen (10% of all fields). The frequency of isolation, relative to the total number of fields per region, was highest in the Aegean (20%) and lower in South East Anatolia (17%), Central Anatolian Plateau (7%), and Marmara (3%) regions. Unlike *F. culmorum*, *B. sorokiniana* tended to be associated more with the winter/facultative wheat environments, with 65% of its isolates found in agroecological zones 1 and 4A2. Compared to other species in the DLRRC, *B. sorokiniana* appeared to occur in the highest elevation environments.

The least commonly isolated of all DLRRC pathogens was *F. pseudograminearum*, found in only 2% of crown

and root samples. The highest incidence of this fungus was in Marmara and South East Anatolia, representing predominately the spring wheat area of agroecological zone 5A2 (Table 1). Compared to other species in the DLRRC, *F. pseudograminearum* appeared to occur in the lowest elevations and correspondingly wetter environments.

Often the various DLRRC pathogens occurred individually in a particular field site; however, in many cases it was common to isolate two or more species from the group of stems. This was particularly the case for *B. sorokiniana* and *Rhizoctonia* spp. in the Aegean region and *F. culmorum* and *Rhizoctonia* spp. on the Central Anatolian Plateau, South East Anatolia, and the Aegean.

HRRRC. The incidence of isolation of *G. graminis* var. *tritici* and various species of *Pythium* was low (2% of fields for *G. graminis* var. *tritici* and 3% of fields for *Pythium* spp.). Unlike the DLRRC species, distribution patterns of *G. graminis* and *Pythium* spp. revealed no significant spatial autocorrelations or clustering (Morans $I_z = 0.5$, $P < 0.35$ for *G. graminis* var. *tritici*, $z = -0.2$, $P < 0.4$ for *Pythium* spp., both NS); hence distributions were considered to be random for both species. Analysis of environmental variables from sites of occurrence revealed no significant differences between the two species for any of the variables analyzed (i.e., elevation, an-

nual precipitation, or precipitation during different quarters of the year).

Of the 12 fields with *G. graminis* var. *tritici*, six were from the Black Sea region, four were from by the Central Anatolian Plateau, and two were from Marmara. Considering the 15 *Pythium* spp. identified in the survey, the South East Anatolia region had the most positive fields (6), followed by the Central Anatolian Plateau (3), Black Sea (4), and Marmara (2).

Analysis of environmental factors comparing combined HRRRC data versus combined DLRRC revealed no significant differences in elevation, annual precipitation, or precipitation in the driest and warmest quarters (approximately summer period in Turkey) between the two groups.

Other species of cereal crown and root rotting fungi. More than 20 species of *Fusarium* were isolated from cereal roots and crowns of wheat (Table 3). Sixty percent of fields contained species of *Fusarium* other than *F. culmorum* and *F. pseudograminearum*. The most commonly isolated species were *F. oxysporum* and *F. chlamydosporum* (each from 11% of fields) and *F. sporotrichioides* (10% of fields), followed by *F. solani* and *F. acuminatum* (8%). *F. solani* was isolated most frequently in Aegean (16%), Central Anatolian Plateau (11%), and Marmara (7%) regions. *F. acuminatum* was most common in Marmara (15%), Aegean, and Central Anatolian Plateau (12%). *F. avenaceum*

Table 3. Incidence on a field basis of isolating *Fusarium* spp. in 2001 and 2002 (excluding *F. culmorum* and *F. pseudograminearum*) from wheat roots and crowns by agroecological zone and geographical region

Agroecological zone ^b	Total fields	Number of fields with specific <i>Fusarium</i> species ^a																				% ^c
		<i>a</i>	<i>av</i>	<i>ch</i>	<i>co</i>	<i>de</i>	<i>eq</i>	<i>fl</i>	<i>he</i>	<i>in</i>	<i>ja</i>	<i>la</i>	<i>ox</i>	<i>po</i>	<i>pr</i>	<i>se</i>	<i>so</i>	<i>sp</i>	<i>tr</i>	<i>ve</i>	spp	
1	11	0	3	0	0	0	0	0	0	0	0	0	1	0	0	0	2	0	0	0	0	54
3A	166	12	6	5	2	0	4	1	3	3	3	1	5	0	0	5	11	5	1	8	2	45
4A1	42	5	3	2	0	2	1	0	2	0	0	1	2	0	0	0	0	3	0	1	2	57
4A2	103	16	11	22	4	0	3	3	4	5	0	0	22	0	8	0	8	30	0	0	1	85
4B	13	1	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	0	0	31
5A1	23	3	5	0	1	0	0	0	1	0	1	0	0	1	1	0	1	0	0	0	2	70
5A2	37	2	2	1	1	1	0	0	2	0	0	0	3	0	0	0	6	1	1	1	1	57
5C	37	0	1	4	0	0	0	0	2	0	0	0	1	0	0	0	4	5	0	0	0	46
6	69	4	1	19	0	0	3	3	16	0	0	0	18	2	6	0	9	8	0	0	0	72
7	17	1	0	1	0	0	0	0	0	0	0	0	4	0	0	0	3	1	0	1	0	65
Total	518	44	32	56	8	3	11	7	30	8	4	2	56	3	15	5	44	54	2	11	8	
Geographical region ^d																						
Cent. Anatolian Plateau	166	12	6	5	2	0	4	1	3	3	3	1	5	0	0	5	11	5	1	8	2	46
Aegean	137	12	3	40	1	0	5	6	19	5	0	0	39	2	14	0	16	38	0	0	0	81
Marmara	114	15	14	4	4	3	2	0	5	0	0	1	6	0	0	0	7	4	1	2	4	61
Mediterranean	17	1	0	1	0	0	0	0	0	0	0	0	4	0	0	0	3	1	0	1	0	65
South East Anatolia	50	1	1	6	0	0	0	0	2	0	0	0	1	0	0	0	4	6	0	0	0	42
Black Sea	34	3	8	0	1	0	0	0	1	0	1	0	1	1	1	0	3	0	0	0	2	65
Total	518	44	32	56	8	3	11	7	30	8	4	2	56	3	15	5	44	54	2	11	8	311
Incidence of isolation (%)	8	6	11	2	1	2	1	6	2	1	0	11	1	3	1	8	10	0	2	2	60	

^a *F. acuminatum* (*a*), *F. avenaceum* (*av*), *F. chlamydosporum* (*ch*), *F. compactum* (*co*), *F. detonianum* (*de*), *F. equiseti* (*eq*), *F. lateritium* (*fl*), *F. heterosporum* (*he*), *F. inflexum* (*in*), *F. javanicum* (*ja*), *F. lateritium* (*la*), *F. oxysporum* (*ox*), *F. poae* (*po*), *F. proliferatum* (*pr*), *F. semitectum* (*se*), *F. solani* (*so*), *F. sporotrichioides* (*sp*), *F. tricinum* (*tr*), *F. verticillium* (*ve*), other *Fusarium* species unidentified (spp).

^b Agroecological zone after Güler et al. (27) given in Figure 1.

^c % samples with *Fusarium* species (excluding *F. culmorum* and *F. pseudograminearum*).

^d Geographical regions in Turkey as illustrated in Figure 1.

and *F. heterosporum* were found in 6% of all fields, and all other species were found in frequencies less than 3%.

The Aegean region had the highest percentage of fields with *Fusarium* spp. (81%). Lower percentages occurred in the Mediterranean and Black Sea (each with 65%), Marmara (61%), Central Anatolian Plateau (46%), and South East Anatolia (42%) regions. The highest *Fusarium* diversity in a given sample (data not shown) occurred in agroecological zone 4A2, including much of the transitional zone of the Central Anatolian Plateau, Aegean, and parts of Marmara and zone 6, representing predominately the Aegean. At least one species of *Fusarium* was found in 85 and 72% of samples from zones 4A2 and 6, respectively, and 34 and 40% of samples, respectively, had two or more species.

Cereal pathogens other than *Fusarium* and *B. sorokiniana* are shown in Table 4 and include *Microdochium nivale*, *Phoma* spp., *Pseudocercospora herpotrichoides*, *Rhizoctonia solani*, and other *Rhizoctonia* spp. Most striking is the high incidence of *Rhizoctonia* spp., with 22% of the fields harboring species other than *R. solani*, which was detected at a low incidence of 2%. *Rhizoctonia* spp. were highly prevalent in this study, with the majority of isolates from the Aegean (39%), Marmara (37%), and Central Anatolian Plateau (31%) regions. This field site incidence was higher than that of the two most commonly documented root rot pathogens *F. culmorum* and *B. sorokiniana* for the Aegean (17 and 28%), Marmara (8 and 2%), and Central Anatolian Plateau (27 and 13%). The main *Rhizoctonia* species ap-

peared to be *R. cerealis*; however, further confirmation studies are necessary. As with *Fusarium* spp. other than *F. culmorum* and *F. pseudograminearum*, the highest incidence was found again in agroecological zones 4A2 and 6, representing Marmara, Aegean, and Central Anatolian Plateau regions.

Microdochium nivale was found at a low incidence of 3% from the Marmara, Central Anatolian Plateau, and South East Anatolian regions. *Phoma* spp. was found in 11% of all fields, most commonly from the Aegean region and to a limited extent the Central Anatolian Plateau. *P. herpotrichoides* was found at a low incidence of 1% and only from Marmara, which could be expected for a basal stem pathogen that is unlikely to be detected by isolations from root and crown tissues performed in this survey.

DISCUSSION

This is the first report of a nationwide survey of wheat crown and root rots in Turkey. The findings presented here are in general agreement with other regional studies conducted across cereal growing regions of Turkey (3,4,33), each of which reported that *F. culmorum*, *B. sorokiniana*, and *Rhizoctonia* spp. were the most common causal agents of dryland root rot on wheat. Bentley et al. (8) isolated 16 *Fusarium* species from wheat stem bases across 25 sites in parts of the Aegean, Marmara, and Black Sea regions, with *F. oxysporum*, *F. equiseti*, *F. acuminatum*, *F. sambucinum*, *F. culmorum*, and *F. armeniacum* being the most frequent. *F. solani*, *F. pseudograminearum*, and other species were

found at low frequencies. The largest numbers of *Fusarium* species in Bentley's study (8) were isolated from the Marmara coastal region. In contrast, comparing the same three regions in this study, the Aegean had both a higher diversity and frequency of *Fusarium* spp. The low frequency of isolation of *F. pseudograminearum* was observed in both surveys. As with this report, Demirci and Dane's (22) study in the eastern part of the Anatolia region indicated isolation of both *F. acuminatum* and *F. oxysporum*. However, they did not report *F. avenaceum*, unlike our study, and we did not detect *F. equiseti* and *M. nivale*, as was indicated in their study. These differences in part may be explained due to different sampling and isolation methods or possibly the more limited number of samples taken in these smaller regional surveys.

It is accepted that *F. culmorum*, *F. pseudograminearum*, and *B. sorokiniana* are pathogenic on wheat plants (12,41,46). Aktaş (2) reported that *B. sorokiniana* is the major pathogen causing severe root rot disease and important yield losses of barley in the Central Anatolia region. *F. pseudograminearum* was the dominant *Fusarium* species causing crown and root rot in the eastern wheat belt of Australia (11,15); however, other *Fusarium* spp. were important pathogens in specific eco-climatic regions. For example, *F. culmorum* accounted for more than 70% of the isolates obtained from the high rainfall and colder cereal region of Victoria and southeast South Australia (52). Other *Fusarium* spp., including *F. acuminatum*, *F. avenaceum*, *F. culmorum*, *F. crookwellense*, and *F. equi-*

Table 4. Incidence on a field basis of other fungi (excluding *Fusarium* spp. and *Bipolaris sorokiniana*) isolated from wheat roots and crowns by agroecological zone and geographical region in 2001 and 2002

Agroecological zone ^b	No. of fields	Number of fields with particular species ^a						
		<i>Ggt</i>	<i>Mn</i>	<i>Ph</i>	<i>Ps</i>	<i>Py</i>	<i>Rs</i>	<i>R. spp</i>
1	11	0	0	0	0	0	0	0
3A	166	6	6	5	0	7	0	25
4A1	42	1	7	0	0	0	0	3
4A2	103	1	0	32	2	3	5	28
4B	13	1	1	0	0	2	0	2
5A1	23	2	0	1	0	1	0	1
5A2	37	0	2	1	1	0	1	2
5C	37	0	1	1	0	1	0	1
6	69	0	0	17	0	0	5	28
7	17	0	0	0	0	1	0	1
Total	518	11	17	57	3	15	11	91
Geographical region ^c								
Cent. Anatolian Plateau	166	6	6	5	0	7	0	31
Aegean	137	0	0	49	0	1	9	39
Marmara	114	2	9	1	3	2	2	37
Mediterranean	17	0	0	0	0	1	0	1
South East Anatolia	50	1	2	1	0	3	0	3
Black Sea	34	2	0	1	0	1	0	1
Total	518	11	17	57	3	15	11	91
Incidence of isolation (%)		2	3	11	1	3	2	22

^a *Gaeumannomyces graminis* var. *tritici* (*Ggt*), *Microdochium nivale* (*Mn*), *Phoma* spp. (*Ph*), *Pseudocercospora herpotrichoides* (*Ps*), *Pythium* spp. (*Py*), *Rhizoctonia solani* (*Rs*), other *Rhizoctonia* spp. (*R. spp*).

^b Agroecological zone after Guler et al. (27) given in Figure 1.

^c Geographical region as illustrated in Figure 1.

seti, are also regularly isolated from crown and stem tissue of wheat in eastern Australia but are regarded as saprophytes acting as secondary colonizers rather than pathogens (11,15). Smiley and Patterson (47) reported isolates of *F. culmorum*, *F. pseudograminearum*, and *B. sorokiniana* as being highly pathogenic to winter wheat, causing damping-off and/or rotting of stem bases leading to seedling death. Isolates of *F. avenaceum* and *M. nivale* were also considered pathogenic. Similarly in Australia, 10 *Fusarium* species (*F. acuminatum*, *F. avenaceum*, *F. baninda*, *F. graminearum*, *F. crookwellense*, *F. proliferatum*, *F. pseudograminearum*, *F. subglutinans*, *F. torulosum*, and *F. tricinctum*) isolated from the crown and spike of wheat were found capable of producing crown rot under laboratory conditions (1). In Canada, Fernandez and Chen (25) also found that *F. culmorum* caused greater discoloration of the subcrown internodes and affected plant emergence to a greater extent than *F. graminearum*, *F. avenaceum*, *F. equiseti*, or *F. poae*. It is plausible to expect that these species could also be pathogenic under conditions in Turkey, and several pathogenicity studies have been reported. Aktaş et al. (5) revealed that *F. verticillioides* was one of the most virulent pathogens on crowns of wheat and that *B. sorokiniana*, *G. graminis* var. *tritici*, and *F. culmorum* also had highly pathogenic reactions, while *P. graminicola* had moderate pathogenicity on winter wheat variety Katea A-1 under greenhouse conditions. Recent work by Tunalı et al. (48) has clearly demonstrated under greenhouse conditions that a range of *F. culmorum*, *F. pseudograminearum*, and *F. graminearum* isolates are all highly pathogenic on crowns of winter wheat Pehlivan, whereas other species such as *F. subglutinans*, *F. oxysporum*, *F. acuminatum*, *F. solani*, and *F. verticillioides* were weak pathogens. This agrees with Aktaş (5) with the exception of *F. verticillioides*. Demirci and Dane (22) examined the cause of crown and root rots on winter wheat Kirik under greenhouse conditions and found that *M. nivale* was the most virulent pathogen, while *F. acuminatum*, *F. equiseti*, *F. oxysporum*, and *F. solani* were slightly virulent. Under field conditions, the dominant species in the DLRRRC varied in individual fields from year to year and among regions in the U.S. Pacific Northwest, indicating a high level of adaptation as members of the pathogen complex respond to changes in temperature, seasonal distribution, and amount of moisture, and other edaphic factors.

GIS tools utilized in this study revealed that the three species of the DLRRRC (*F. culmorum*, *F. pseudograminearum*, and *B. sorokiniana*) exhibited a highly significant nonrandom, clustered spatial distribution of occurrence. There was no significant association with any particular region or agroecological zone for any species. There

were strong indications of niche separation with *B. sorokiniana* occurring at higher elevations and *F. pseudograminearum* occurring at lower elevations and wetter winter environments. As previously reported by several authors, the effects of dryland root rot involving *F. pseudograminearum*, *F. culmorum*, and *B. sorokiniana* were more pronounced under stress conditions during the growing season as is commonly experienced under limited rain conditions found in Australia, the U.S. Pacific Northwest, and Canada (6,7,35,41,42,45). This also has important implications in Turkey, as much of the 5 Mha of winter wheat grown on the Central Anatolia plateau located at average elevation of 900 m in the inner part of Turkey (mostly zone 3A, part 2B, 3B, and 4A2; Fig. 1) is under rain conditions associated with drought stress at the end of the season. Supplementary irrigation is available only in limited cases. In this survey, only key macro-environment variables such as rainfall and elevation were analyzed and not micro-variables such as soil type and moisture. If these were available, they might have explained some of the cluster distributions for specific species.

The 2-year extensive study conducted by Smiley and Patterson (47) with 288 fields in the Pacific Northwest also revealed similar isolation frequencies of the major causal agents of root rots. However in their case, *F. pseudograminearum* was more common, with field isolation frequencies of 27 and 16% in years 1 and 2 compared to 14 and 7% for *F. culmorum*. In our study, *F. culmorum* was more common (14%) than *F. pseudograminearum* (2%). Similar findings were also made by the limited survey in the Black Sea and Aegean regions by Bentley et al. (8). The reasons for these differences are not clear given that the environmental conditions in both the Pacific Northwest and Turkey are somewhat comparable in the winter wheat growing areas. Considering Turkey is one of the regions associated with the evolution of wheat, it may also be a center of diversity for *F. culmorum*. Another possibility is differences in crop rotational patterns and the differential hosting abilities for *Fusarium* species, although much of the Pacific Northwest is similarly a wheat-fallow cropping system.

One of the major findings in this study was the high field incidence (22%) of *Rhizoctonia* spp., which were mostly species other than *R. solani* (2%). It was likely that the majority of the *Rhizoctonia* isolates were *R. cerealis*, which differs from *R. solani* by having binucleate hyphal cells and a relatively slow growth rate (32). These species are similar in other morphological features, making species identification in this type of study difficult. The importance of *R. cerealis* in Turkey has been highlighted in several publications

(4,5), and it has also been reported to be widespread and an economically important crown pathogen of winter wheat in both China and New Zealand (20,27,31). This fungus is also reported from several other temperate countries including the Netherlands, Germany, South Africa, and the United Kingdom (15,32). Sowing date, soil moisture and temperature can affect sharp-eye spot severity (51), with disease incidence usually greatest in early autumn-sown crops (21). Mathre (34) also reported that acid, sandy, and dry (less than 20% moisture holding capacity) soils increased disease risk of *R. cerealis*, as did cool spring temperatures. This is likely an important crown pathogen in Turkey and warrants further investigation.

The isolation of the HRRRC root and crown fungi was very low, which is expected given Turkey's cereal production environment. The various species associated with the HRRRC were found in both the high and low rainfall sites. The highest frequency of isolation of these fungi in the Black Sea region represents the highest rainfall in Turkey (agroecological zone 5A1, 600 mm).

In this survey, the cereal growing regions of South East Anatolia, Central Anatolian Plateau, Marmara, and Black Sea regions had the highest frequency of isolation of DLRRRC species, including *Rhizoctonia* spp. More than two-thirds of Turkey's 10 Mha of wheat is in this area, with 60% being rainfed or under suboptimal irrigation that would allow late season plant stress in most years. Hekimhan et al. (29) demonstrated that the root rot complex (*F. culmorum*, *F. pseudograminearum*, and *B. sorokiniana*) caused 24% yield loss in common bread wheats with two supplementary irrigations over a 3-year study on the Central Anatolian Plateau in Konya province. On this basis, if 60% of this 6.77 Mha of both winter and spring wheat is grown under suboptimal moisture conditions (rainfed with or without suboptimal irrigation), and this region suffered an average conservative estimate of 10% yield loss due to dryland root rots per year, the total loss to production would be 1.33 Mt per year or approximately 7% of the annual production of approximately 20 Mt. This loss is significant and warrants active wheat screening and breeding programs for dryland root rot resistance and tolerance. These improvement programs have been actively initiated in joint Turkey and International (CIMMYT) efforts (37,38).

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