



Communication

# Sources of Resistance to Powdery Mildew in Barley Landraces from Turkey

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Abstract: Powdery mildew on barley, caused by the pathogen Blumeria graminis f. sp. hordei, occurs worldwide and can result in severe yield loss. Germplasm of barley, including landraces, commercial cultivars, wild relatives and breeding lines are stored in more than 200 institutions. There is a need for characterization of this germplasm in terms of resistance to biotic and abiotic stresses. This is necessary in order to use specific accessions in breeding programs. In the present study, 129 barley landraces originated from Turkey and provided by the ICARDA genebank were tested for resistance to powdery mildew. Seedling resistance tests after inoculation with 19 differentiated isolates of B. graminis f. sp. hordei were used to postulate the presence of resistance genes. From the 129 landraces studied, plants of 19 (14.7%) of them showed resistance to infection with powdery mildew. Based on preliminary tests from these 19 landraces, 25 resistant single plant lines were selected for testing with differential powdery mildew isolates. Seven lines were resistant to all 19 isolates used. However, only one line (5583-1-4) showed resistance scores of zero against all isolates used. It is likely that this line possesses unknown, but highly effective genes for resistance. In five resistant lines it was not possible to postulate the presence of specific resistance genes. In 19 lines the presence of the genes Mlp, Mlk, Mlh, Mlg, Ml(CP), Mlat, Mla3, Mla6, Mla7 and Mla22 were postulated. These new sources of highly effective powdery mildew resistance in barley landraces from Turkey could be successfully used in breeding programs.

**Keywords:** *Hordeum vulgare*; barley; *Blumeria graminis*; powdery mildew; landraces; germplasm; resistance genes; genebank



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# 1. Introduction

Barley (*Hordeum vulgare* L.) is an economically important cereal crop which is known to be drought, cold, and salt tolerant and well-adapted to low-input environmental conditions [1,2]. It is cultivated at high altitudes and commonly under rain-fed conditions. It is often grown in marginal agricultural areas with low annual precipitation, often less than 220 mm [3]. Barley ranks as the fourth crop in the world, after wheat, maize and rice, in terms of the area of cultivation. Almost half of the world's barley area is in Europe, where barley is second crop after wheat in cultivated area [4].

Germplasm of barley, including landraces, commercial cultivars, wild relatives and breeding lines is very diverse and is stored in more than 200 institutions [5,6]. Barley landraces are an important source of genetic variation and resistance to biotic stresses including powdery mildew [7–9]. Turkey is characterized by the presence of diverse agroecological zones and a long history of agriculture. It is known to be a rich source of barley landraces. They are still planted in this country, and they are characterized by a high level of resistance to biotic and abiotic stresses. There is a need for characterization of this germplasm in terms of resistance to biotic and abiotic stresses. This knowledge is necessary in order to use specific accessions in breeding programs [10–12].

Barley is often infected by barley powdery mildew fungus (*Blumeria graminis* DC. Golovin ex Speer f. sp. *hordei*). Loss of yield caused by this disease can reach up to

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30%, with averages of 5–10% [13–16]. Powdery mildew on barley is considered one of the most well-characterized host–pathogen genetic interaction systems. Barley cultivars with effective genes for resistance to powdery mildew have been an efficient means for controlling this disease [17–21]. Barley breeders have used major resistance genes: *Mla6*, *Mla7*, *Mla9*, *Mla12* and *Mla13*, *Mlk*, *Mlg*, *MlLa*, *Mlh* and *Mlra*, which originated from landraces as well as from the subspecies *H. vulgare* ssp. *spontaneum*. Durable mlo-resistance (gene *mlo*) has been identified in landraces. Since 1984, it has been deployed in many barley cultivars throughout Europe [20,22–24].

Effective controlling of barley powdery mildew is possible by growing genetically resistant barley cultivars. This method of crop protection is relatively inexpensive and it is environmentally friendly. These cultivars started being used from the beginning of the application of modern, intensive methods in barley production because these production methods created favorable conditions for development of this disease [25–30]. Currently, powdery mildew of barley is one of the most common and most widespread disease of barley in Europe and another barley regions of the world, causing significant yield losses [20,21,31].

When a cultivar containing one dominant resistance gene is grown on a large acreage, new virulent *B. graminis* races may occur within 4–5 years. Exceptions are recessive genes for resistance such as *mlf* and *mlo*. However, many factors, e.g., temperature, water stress or light intensity, may affect the use of these genes in breeding programmes [20,22,28]. At least 38 different genes/alleles have been used in varieties grown in Europe [32]. Nevertheless, barley breeders, geneticists and plant pathologists are constantly looking for new, efficient sources of powdery mildew resistance, in order to combine them with those already used in modern cultivars, and to increase their resistance durability [31,33,34].

Most of the original sources of powdery mildew resistance genes came from domesticated cultivars in Europe [25,26,35]. These sources of resistance were easy to be used in breeding but the number of resistance genes was limited. Breeders and geneticists have been looking for new sources of resistance in non-European germplasm. Most of these studies were conducted using collections of landraces and differential sets of powdery mildew isolates [36,37]. Previous studies showed that barley landraces from Turkey are rich sources of genetic diversity for plant breeding, including resistance to pathogens [10–12,38,39].

Identification of powdery mildew resistance genes based on tests performed on seedlings using differential sets of pathogens is effective and sufficient for breeders' and pathologists' needs [25,30,40,41]. This study aimed at detecting sources of powdery mildew resistance in barley landraces from Turkey.

#### 2. Materials and Method

### 2.1. Plant Material

Seed samples of 129 *H. vulgare* L. landraces from Turkey were provided by Dr. J. Valkoun, J. Konopka and Prof. S. Ceccarelli (International Center for Agricultural Research in the Dry Areas—ICARDA, Aleppo, Syria) (Table 1). For 53 landraces, details about the places of collection were known. These landraces originated from 26 provinces: 11 landraces were from Ismir province, 4—Kars, 3—Eskisehir, 3—Agri, 3—Erzincan, 3—Sivas, 3—Mugla, 3—Bilecik, 3—Kayseri, 2—Kutahya and 1 landrace originated from each of the following provinces—Bayburt, Denizli, Sanli Urfa, Manisa, Van, Mus, Hakkari, Tokat, Icel, Antakya, Gaziantep, Isparta, Adana, Afyon, Usak, and Bursa. They were collected at altitudes from 15 m above sea level in Izmir province, to 1900 m above sea level in Kars province, and 2250 m above sea level in Bayburt province.

## 2.2. Pathogen

Nineteen differential *Bgh* (*B. graminis* f. sp. *hordei* Em Marschal) isolates with virulence genes corresponding to known resistance genes were used (Table 2). Isolates originated from the collections in Risø National Laboratory, Roskilde, Denmark; Danish Institute for Plant and Soil Science, Lyngby, Denmark; Edigenossische Technische Hochschule—ETH,

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Zurich, Switzerland provided kindly by Dr. H. J. Schaerer (ETH, Zurich, Switzerland) and the Plant Breeding and Acclimatization Institute—National Research Institute (PBAINRI) IHAR, Radzikow, Poland. A set of isolines of barley cultivar Pallas with different resistance genes was used [42], provided by Dr. L. Munk (Royal Agricultural and Veterinary University, Copenhagen, Denmark) and on 8 additional cultivars.

Isolate  $Bgh\ 33$  was the most avirulent isolate. It was avirulent to resistance genes, or their combinations, such as: Mla1, Mla3, Mla6+Mla14, Mla7+Mlk+?, Mla7+?, Mla7+MlLG2, Mla9+Mlk, Mla9, Mla12, Mla13+MlRu3, Mla22, Mla23, MlRu2, Mlk, Mlp, Mla1 and to resistance genes present in additional cultivars included in a differential set: Benedicte (Mla9, Ml(Ml)), Lenka (Mla13, Ml(Ab)), Steffi (Ml(St1)), Ml(St2), and Kredit Ml(Kr). Isolates  $Bgh\ 1$ ,  $Bgh\ 29$ , and  $Bgh\ 51$  were the most virulent group. Isolate  $Bgh\ 51$  was virulent to resistance genes or their combinations present in 18 Pallas isolines,  $Bgh\ 29$  was virulent to resistance genes or their combinations present in 17 Pallas isolines and  $Bgh\ 1$  virulent to resistance genes or their combinations present in 16 Pallas isolines. They were purified by single pustule isolation and were maintained and propagated on young seedlings of the powdery mildew-susceptible cultivar Manchuria (CI 2330). Virulence checks were conducted to assure the purity of isolates throughout the experiment.

A five-point (0 to 4) reaction type (RT) scale was used, as follows: 0, no visible symptoms; 1, minute necrotic flecks, no mycelial growth and no sporulation; 2, frequent chlorosis, reduced mycelial growth and no or very scarce sporulation; 3, moderate mycelial growth, moderate sporulation, and occasional chlorosis; 4, profuse sporulation of well-developed colonies, 0(4) sparse small colonies originating from the stomatal subsidiary cells [26,43].

## 2.3. Landraces and Single Plant-Lines Resistance Tests

First, samples of 30 plants from each of the landraces were tested with the Bgh 33 isolate (the most avirulent one) under controlled chamber conditions with a 16/8 h day/night photoperiod and a 22/16  $^{\circ}$ C temperature regime.

Seedlings with a fully expanded first leaf were inoculated with *Bgh* isolate by shaking conidia from the susceptible cv. Manchuria. After 8–10 days, infection types were scored. Plants with disease scores of 0 to 1 were classified as highly resistant (R), plants that scored 2 as a moderately resistant (M) and rating of 3 or 4 as susceptible and very susceptible (S). Plants with the score 0(4) possess a resistance gene in locus *Mlo*. The cultivar Manchurian CI 3230 was used as a susceptible control.

Based on the results of this preliminary experiment, 25 resistant single plant lines from 19 landraces were selected. A highly resistant reaction type was observed on 13 lines, and a moderately resistant reaction type was observed on 11 lines. In 5 landraces, segregation of RT was observed (Table 3). Next, they were grown in greenhouse conditions to obtain seeds for future evaluations using a set of 19 *Bgh* differential isolates.

Postulation of resistant genes in tested lines was based on a comparison of reaction spectra observed on tested plants and the barley differential set infected with differential *Bgh* isolates (Table 1). This was performed on the basis of the gene-for-gene hypothesis [44].

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**Table 1.** Collected data of 129 landraces from Turkey.

No.	ICARDA IG	IHAR No	Longitude (E)	Latitude (N)	Altitude (ppm)	Province	No.	ICARDA IG	IHAR No	Longitude	Latitude	Altitude	Province
1	18,781	5177	-	-	-	-	66	115,938	5468	E28 22	N37 13	-	Mugla
2	18,848	5178	31 32-E	N39 27	-	Eskisehir	67	115,939	5469	-	-	-	-
3	18,849	5179	E31 32-E	N39 27	-	Eskisehir	68	115,940	5470	-	-	-	
4	18,851	5180	E27 11	N39 07	-	Izmir	69	115,946	5471	E28 53	N40 35	-	Bursa
5	19,056	5181	-	-	-	-	70	115,947	5472	-	-	-	-
6	19,058	5182	-	-	=	-	71	115,948	5473	-	-	-	-
7	19,062	5183	-	-	-	-	72	115,949	5474	-	-	-	-
8	19,068	5184	E27 11	N39 07	-	Izmir	73	115,958	5475	-	-	-	-
9	19,077	5185	-	-	-	-	74	19,711	5511	-	-	-	Usak
10	18,541	5186	E28 38	N38 33	-	Manisa	75	19,742	5512	-	-	-	Afyon
11	19,545	5187	-	-	-	-	76	19,909	5513	-	-	-	Kutahya
12	19,546	5188	-	-	-	-	77	19,920	5514	-	-	-	Kutahya
13	19,547	5189	-	-	-	-	78	19,929	5515	-	-	-	Bilecik
14	19,550	5190	-	-	-	-	79	19,992	5516	-	-	-	Bilecik
15	19,562	5191	-	-	-	-	80	26,310	5523	-	-	-	Adana
16	19,565	5192	-	-	-	-	81	26,758	5524	-	-	-	-
17	19,566	5193	-	-	-	-	82	27,189	5525	E35 29	N38 44	-	Kayseri
18	19,568	5194	-	-	-	-	83	27,246	5526	E30 32	N37 46	-	Isparta
19	19,569	5195	-	-	-	-	84	27,251	5527	E37 21	N37 08	-	Gaziantep
20	19,576	5196	E27 11	N39 07	-	Izmir	85	28,537	5532	E36 34	N36 16	-	Antakya
21	37,224	5340	-	-	-	-	86	28,543	5533	E33 39	N36 21	-	Icel
22	37,225	5341	-	-	-	-	87	28,675	5534	E35 37	N40 17	-	Tokat
23	37,230	5342	-	-	-	-	88	28,676	5535	E43 06	N40 37	-	Kars
24	37,234	5343	-	-	-	-	89	28,704	5536	E35 22	N38 11	-	Kayseri
25	37,235	5344	-	-	-	-	90	28,706	5537	E43 04	N39 44	-	Agri
26	37,237	5345	-	-	-	-	91	28,728	5538	E38 29	N39 16	-	Erzincan
27	37,239	5346	-	-	-	-	92	28,734	5539	E36 36	N39 52	-	Sivas
28	37,240	5347	-	-	-	-	93	28,809	5540	E44 02	N39 34	-	Agri
29	37,241	5348	=	-	-	-	94	28,811	5541	=	-	702	Hakkari

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 Table 1. Cont.

No.	ICARDA IG	IHAR No	Longitude (E)	Latitude (N)	Altitude (ppm)	Province	No.	ICARDA IG	IHAR No	Longitude	Latitude	Altitude	Province
30	37,242	5349	-	-	-	-	95	28,817	5542	-	-	-	Mus
31	37,243	5350	-	-	-	-	96	28,859	5543	E29 59	N40 10	-	Bilecik
32	37,244	5351	-	-	-	-	97	28,860	5544	-	-	-	Van
33	37,246	5352	-	-	-	-	98	32,679	5546	E43 02	N41 01	1900	Kars
34	37,249	5353	-	-	-	-	99	32,683	5547	E36 55	N39 48	1310	Sivas
35	37,250	5354	-	-	-	-	100	32,684	5548	E27 03	N38 34	15	Izmir
36	37,251	5355	-	-	-	-	101	35,528	5553	E43 56	N39 33	1820	Agri
37	37,252	5356	-	-	-	-	102	35,531	5554	E43 02	N41 01	1900	Kars
38	37,255	5357	-	-	-	-	103	35,532	5555	E40 00	N40 01	2250	Bayburt
39	37,257	5358	-	-	-	-	104	35,533	5556	E40 23	N39 47	1420	Erzincan
40	37,258	5359	-	-	-	-	105	35,534	5557	E40 23	N39 47	1420	Erzincan
41	37,260	5360	-	-	-	-	106	35,535	5558	E36 55	N39 48	1310	Sivas
42	37,262	5361	-	-	-	-	107	35,536	5559	E27 03	N38 34	15	Izmir
43	37,266	5362	-	-	-	-	108	35,867	5563	E28 22	N37 13	-	Mugla
44	37,268	5363	-	-	-	-	109	35,868	5564	E27 10	N38 25	200	Izmir
45	37,269	5364	-	-	-	-	110	37,226	5569	-	-	-	-
46	37,271	5365	-	-	-	-	111	37,227	5570	-	-	-	-
47	37,273	5366	-	-	-	-	112	37,228	5571	-	-	-	-
48	37,275	5367	-	-	-	-	113	37,229	5572	-	-	-	
49	37,285	5368	-	-	-	-	114	37,223	5573	-	-	-	-
50	112,919	5430	E27 32	N38 01	-	Izmir	115	37,236	5574	-	-	-	-
51	112,942	5431	-	-	-	-	116	37,238	5575	-	-	-	
52	112,995	5432	-	-	-	-	117	37,245	5576	-	-	-	
53	113,006	5433	-	-	-	-	118	37,248	5577	-	-	-	-
54	113,008	5434	-	-	-	-	119	37,253	5578	-	-	-	-
55	113,009	5435	E27 32	N38 01	-	Izmir	120	37,256	5579		-	-	-
56	113,010	5436	E27 32	N38 01	-	Izmir	121	37,259	5580	-	-	-	-
57	113,011	5437	E31 32	N39 27	-	Eskisehir	122	37,261	5581	-	-	-	-

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 Table 1. Cont.

No.	ICARDA IG	IHAR No	Longitude (E)	Latitude (N)	Altitude (ppm)	Province	No.	ICARDA IG	IHAR No	Longitude	Latitude	Altitude	Province
58	113,019	5438	-	-	-	-	123	37,264	5582	-	-	-	-
59	113,020	5439	-	-	-	-	124	37,267	5583	-	-	-	-
60	113,022	5440	E37 56	N37 04	-	Sanli Urfa	125	37,270	5584	-	-	-	-
61	113,024	5441	-	-	-	-	126	37,274	5585	-	-	-	-
62	113,028	5442	-	-	-	-	127	37,276	5586	-	-	-	-
63	113,029	5443	E27 10	N38 26		Izmir	128	37,289	5587	E43 05	N40 51	1900	Kars
64	113,053	5444	E28 50	N37 27		Denizli	129	37,295	5588	E40 00	N40 01	2250	Bayburt
65	115,937	5467	E27 10	N38 26	-	Izmir							

**Table 2.** *B. graminis* f. sp. *hordei* isolates used for artificial inoculation and their virulence spectra against resistance genes on differential set of Pallas near-isogenic lines and 8 cultivars.

	Pallas											Bgh Isola	ates								
No.	Isolines and Cultivars	Virulence	Bgh 1	Bgh 2	Bgh 4	Bgh 8	Bgh 9	Bgh 11	Bgh 13	Bgh 14	Bgh 24	Bgh 28	Bgh 29	Bgh 31	Bgh 33	Bgh 36	Bgh 40	Bgh 48	Bgh 51	Bgh 57	Bgh 63
1	P1	Mla1	0	0	4	4	0	0	0	0	0	0	4	0	0	4	0	0	4	0	0
2	P2	Mla3	1	0	0	0	0	0	0	0	0	0	4	0	0	4	4	0	0	0	0
3	P3	Mla6, Mla14	0	0	0	0	0	4	0	4	0	4	0	0	0	4	4	4	4	4	4
4	P4A	Mla7, Mlk, +?	2	2	2	2	2	2	2	2	2	4	2	4	0	2	2	2	4	4	2
5	P4B	Mla7, +?	4	4	1	0	2	2	4	4	0	2	4	4	1	4	4	1	4	4	4
6	P6	Mla7, MlLG2	4	4	0	0	2	1	2	4	0	2	2	4	0	4	2	0	4	4	4
7	P7	Mla9, Mlk	4	0	0	0	0	0	4	0	0	0	4	0	0	0	0	0	0	4	0
8	P8A	Mla9, Mlk	4	0	0	0	0	0	4	0	0	0	4	0	0	0	0	0	0	4	0
9	P8B	Mla9	4	0	0	0	0	0	4	0	4	0	4	0	0	0	0	0	0	4	0
10	Р9	Mla10, MlDu2	4	4	0	1	4	0	4	0	2	0	4	4	4	4	0	0	4	4	4
11	P10	Mla12	0	0	0	0	4	0	0	4	0	0	4	4	0	4	4	0	4	0	4
12	P11	Mla13, MlRu3	4	0	0	0	0	0	0	4	4	0	0	4	0	0	0	0	4	0	4
13	P12	Mla22	4	4	4	4	0	4	0	4	4	4	4	4	0	0	4	4	4	0	0
14	P13	Mla23	1	1	1	2	1	2	1	1	1	1	2	1	1	1	1	1	1	1	1
15	P14	Mlra	4	4	0	4	4	4	4	0	4	4	4	4	4	4	4	4	4	4	4
16	P15	Ml(Ru2)	4	4	4	3	4	2	4	4	2	0	4	4	2	4	4	4	4	4	4
17	P17	Mlk	4	4	2	2	2	2	4	2	2	0	4	4	2	4	2	2	4	4	4
18	P18	Mlnn	4	4	4	4	2	4	4	4	2	2	4	4	4	4	4	4	4	2	2

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Table 2. Cont.

	Pallas	37' 1										Bgh Isola	ites								
No.	Isolines and Cultivars	Virulence	Bgh 1	Bgh 2	Bgh 4	Bgh 8	Bgh 9	Bgh 11	Bgh 13	Bgh 14	Bgh 24	Bgh 28	Bgh 29	Bgh 31	Bgh 33	Bgh 36	Bgh 40	Bgh 48	Bgh 51	Bgh 57	Bgh 63
19	P19	Mlp	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2
20	P20	Mlat	2	2	4	2	2	2	4	2	2	2	2	2	2	2	4	2	4	2	2
21	P22	mlo5	0(4)	0(4)	0(4)	0(4)	0(4)	3	0(4)	0(4)	0(4)	0(4)	0(4)	0(4)	0(4)	0(4)	0(4)	0(4)	0(4)	0(4)	0(4)
22	P23	Ml(La)	4	4	4	4	2	4	4	4	4	4	4	4	4	4	4	4	4	4	4
23	P24	Mlh	4	4	0	4	4	4	4	4	4	4	4	4	4	0	4	4	4	4	4
24	P21	Mlg, Ml(CP)	4	4	0	0	0	4	0	4	0	4	4	4	4	4	4	0	4	0	4
25	Pallas	Mla8	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4
26	Benedicte	Mla9,Ml(IM9)	0	0	0	0	0	0	0	4	0	0	4	4	0	4	4	0	4	0	4
27	Borwina	Ml(Bw)	4	3	0	4	0	4	4	4	2	2	3	4	4	4	3	4	4	2	2
28	Gunnar	Mla3, Ml(Tu2)	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
29	Jarek	Ml(Kr), +?	4	4	4	4	2	4	4	4	4	4	4	4	4	4	2	4	4	2	4
30	Kredit	Ml(Kr)	4	2	0	2	0	0	2	4	4	4	2	4	0	4	2	2	4	4	4
31	Lenka	Mla13,Ml(Ab)	4	0	0	0	0	0	0	4	0	0	0	4	0	0	0	0	4	0	4
32	Steffi	Ml(St1), Ml(St2)	2	2	0	0	0	0	0	4	0	0	2	3	0	4	2	0	2	0	4
33	Trumph	Mla7, Ml(Ab)	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4
34	Manchuria	-	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4	4

A five-point (0 to 4) reaction type (RT) scale was as follows: 0, no visible symptoms; 1, minute necrotic flecks, no mycelial growth and no sporulation; 2, frequent chlorosis, reduced mycelial growth and no or very scarce sporulation; 3, moderate mycelial growth, moderate sporulation, and occasional chlorosis; 4, profuse sporulation of well-developed colonies, 0(4) sparse small colonies originating from the stomatal subsidiary cells.

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**Table 3.** Resistance of barley (*H. vulgare* L.) lines selected from landraces originating from Turkey to *B. graminis* f. sp. hordei isolates after inoculation at the seedling stage.

	Landı	race	Line										Isolat	e									D . 1 . 1
No.	ICARDA IG	IHAR No.	IHAR No.	Bgh 1	Bgh 2	Bgh 4	Bgh 8	Bgh 9	Bgh 11	Bgh 13	Bgh 14	Bgh 24	Bgh 28	Bgh 29	Bgh 31	Bgh 33	Bgh 36	Bgh 40	Bgh 48	Bgh 51	Bgh 57	Bgh 63	Postulated Resistance Genes
	18,781	5177	5177 1 1	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	Mlp
2	18,781	5177	5177 1 4	2	2	2	2	2	2	2	2	2	2	2	2	0	2	2	2	2	2	2	Mla7, Mlk, +?
3	18,849	5179	5179 1 1	0	0	0	2	0	1	0	0	0	2	4	1	0	0	4	0	0	0	0	Mla3, Mlh
4	19,077	5185	5185 1 1	0	2	0	2	0	4	0	4	4	4	2	0	2	2	4	4	4	4	4	Mla6, +?
5	19,077	5185	5185 4 3	2	2	0	0	0	2	0	2	0	2	2	0	2	2	2	0	4	0	2	[Mla7, Mlk+?], [Mlg, Ml(CP)]
6	19,541	5186	5186 2 1	2	2	0	0	0	2	0	2	0	2	2	0	2	2	2	0	4	2	2	[Mla7, Mlk+?], [Mlg, Ml(CP)]
7	19,547	5189	5189 2 1	2	2	0	0	0	4	0	4	2	4	4	0	0	4	4	2	4	2	4	?
8	19,547	5189	5189 3 3	2	2	0	0	0	4	0	4	2	4	0	0	2	2	4	2	4	2	4	?
9	19,550	5190	5190 3 1	0	2	0	2	0	4	0	4	4	4	0	0	2	2	4	4	4	4	4	Mla6, +?
10	25,979	5204	5204 3 2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	Mlp
11	37,230	5342	5342 1 1	0	2	0	2	0	0	0	0	0	1	4	0	0	0	4	0	0	1	0	Mla3, Mlh
12	37,234	5343	5343 1 1	0	2	0	2	0	0	0	0	0	0	4	0	0	0	4	0	0	2	0	Mla3, Mlh
13	37,235	5344	5344 1 1	2	2	0	2	2	4	4	4	4	4	2	2	2	4	4	4	4	4	4	?
14	113,011	5437	5437 1 1	0	0	0	2	0	4	0	4	4	4	0	0	0	4	4	4	4	4	4	Mla6, +?
15	113,020	5439	5439 1 1	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	2	Mlp
16	113,020	5439	5439 5 1	2	2	2	2	2	2	2	2	2	2	2	0	0	2	2	2	2	2	2	Mla7, Mlk, +?
17	113,028	5442	5442 2 2	2	2	1	2	2	2	2	2	2	2	2	2	0	2	2	2	2	2	2	Mla7, Mlk, +?
18	113,028	5442	5442 1 3	nd *	nd	nd	2	2	2	nd	2	2	2	2	2	2	2	2	nd	2	2	2	Mlp
19	115,940	5470	5470 3 1	0	0	0	2	0	4	0	4	4	4	0	0	0	2	4	4	4	4	4	Mla6, +?
20	115,947	5472	5472 1 4	0	0	0	2	0	4	0	4	4	4	0	0	0	4	4	4	4	2	4	Mla6, +?
21	115,948	5473	5473 1 5	4	0	4	4	2	4	4	4	4	4	4	4	2	4	4	4	4	4	4	?
22	115,958	5475	5475 1 1	2	2	4	2	2	2	4	2	2	2	2	2	2	2	nd	2	4	2	2	Mlat
23	28,860	5544	5544 1 1	4	4	4	4	0	4	4	4	4	4	4	4	0	0	4	4	4	0	0	Mla22
24	37,267	5583	5583 1 4	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
25	37,289	5587	5587 2 1	4	4	4	4	0	4	4	4	4	4	4	4	0	0	4	4	4	0	0	Mla22

<sup>\*</sup> nd—no data. A five-point (0 to 4) reaction type (RT) scale was used as follows: 0, no visible symptoms; 1, minute necrotic flecks, no mycelial growth and no sporulation; 2, frequent chlorosis, reduced mycelial growth and no or very scarce sporulation; 3, moderate mycelial growth, moderate sporulation, and occasional chlorosis; 4, profuse sporulation of well-developed colonies, 0(4) sparse small colonies originating from the stomatal subsidiary cells.

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### 3. Results

Plants of 19 (14.7%) of the tested landraces were resistant to infection with *Bgh33* isolate in preliminary testing. In five landraces, segregation of RT was observed. Based on preliminary tests from these 19 landraces, 25 resistant, single plant lines were selected for testing with differential isolates. From these lines, seven were resistant to all 19 isolates used. However, only one line (5583-1-4) showed resistance scores of zero against all isolates used (Table 3). In five lines it was not possible to postulate the presence of specific resistance genes. In 19 lines, the presence of the genes *Mlp*, *Mlk*, *Mlh*, *Mlg*, *Ml(CP)*, *Mlat*, *Mla3*, *Mla6*, *Mla7*, *Mla22* and unknown genes (genes not present in differential set) were postulated.

#### 4. Discussion

Barley landraces from Turkey are a rich source of genetic diversity for plant breeding, including resistance to powdery mildew [10–12,38,39]. This was confirmed in the presented study. Single plant lines selected from 19 (14.7%) of the tested landraces were resistant to infection with powdery mildew.

Resistant lines selected from landraces are a very valuable material for resistance breeding. This kind of germplasm is the simplest source of resistance to use directly in breeding programs. Because of their adaptability to a wide range of conditions, barley landraces are recognized as an important genetic resource for tolerance and resistance to biotic and abiotic stresses. They carry unique traits and are considered a rich resource for resistance breeding and for the expansion of the gene pool [45,46].

Turkey is a rich source of barley genetic diversity because of its geographic location. The south-eastern region of Turkey is at the top of the Fertile Crescent of the Near East, within the centre of origin of cultivated barley [39]. Barley is one of the oldest cultivated plants grown in Anatolia, and it is the second most important cereal crop following wheat. In addition, in Turkey, the ancestor of cultivated barley, *Hordeum spontaneum* C. Koch, grows naturally, and powdery mildew epidemics occur in the western and southern parts of the country [12]. All these factors lead to conclusion that coevolution of barley powdery mildew was occurring in Turkey for very long time, and that barley landraces from Turkey may be a rich source of resistance to powdery mildew. This was confirmed in the presented study, in which many resistance genes were identified in lines selected from Turkish landraces.

The genetic diversity of barley landraces offers many traits for barley breeding, especially concerning resistance to biotic and abiotic stresses [3,33,47–50]. The genetic heterogeneity within the barley landraces is due to a low level of outcrossing occurring in barley, and farmers' management of seed [40,51–53]. This genetic heterogeneity was also observed in the presented study, in which five landraces showed segregation of RT.

Many barley landraces collected in Tunisia [27], Morocco [36,54–59], Australia [60], China [61], Greece [62], Jordan [63,64], Egypt [54], Latvia [65], Libya [66], Yemen [67] and Spain [68–72] have been tested for resistance to powdery mildew, and numerous known and unknown specific resistances have been identified. In addition, collections of landraces from many countries have been studied [35,40,73–76]. Results show that the presence of known and unknown powdery mildew resistance genes have been obtained for barley landraces from Turkey [10,38,39]. The present study confirmed that barley landraces from Turkey have numerous known and unknown specific resistances to powdery mildew. In 24 resistant single-plant lines studied, the presence unknown resistance genes and the genes *Mlp*, *Mlk*, *Mlh*, *Mlg*, *Ml(CP)*, *Mlat*, *Mla3*, *Mla6*, *Mla7* and *Mla22* were postulated.

Seedling resistance tests were used in order to describe infection types expressed by barley lines after inoculation with differentiated isolates of *B. graminis* f. sp. *hordei*. This kind of testing is sufficient for disease-resistance screening. It is used commonly in breeding programmes to postulate the presence of specific resistance genes in modern cultivars and to screen germplasm for new sources of effective resistance [36,40,75,76]. However, these kinds of tests are not very useful for identifying and describing partial resistance. For a description of partial resistance there is a need to conduct measurements of resistance

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characteristics, in addition to the infection type. Furthermore, partial resistance is generally better expressed at the adult plant stage [26,34,77].

Newly identified sources of powdery mildew resistance in 25 single plant lines (originated from 19 landraces) are valuable for barley breeding for resistance. In five lines it was not possible to postulate the presence of specific resistance genes. In 19 lines the presence of the genes Mlp, Mlk, Mlh, Mlg, Ml(CP), Mlat, Mla3, Mla6, Mla7 and Mla22 and unknown genes (not present in differential set) were postulated. Interestingly for barley resistance breeding, seven lines selected from four landraces were resistant to all 19 isolates used in this study. However, the most interesting point from a breeders' point of view was line 5583-1-4, which showed resistance scores of zero for all isolates used. Most probably this line possesses unknown, yet very effective genes for resistance. Future work will include the genetic study of resistance identified in seven single-plant lines by conducting appropriate crosses and the use of molecular markers [58,59,78,79]. Authors intend to introduce these alleles into elite cultivars of barley to create initial materials for European breeding programmes. This is a necessary step between barley genebank collections and the practical use of barley genetic resources in breeding programmes. The new sources of highly effective powdery mildew resistance described in this study could be successfully used in barley breeding programs.

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