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SCREENING FOR LEAF RUST RESISTANCE IN COLLECTIONS OF BARLEY LANDRACES FROM SOUTHERN MEDITERRANEAN REGION

SHORT COMMUNICATION

ABSTRACT

(*Hordeum vulgare* L.) landraces are grown commonly in Southern Mediterranean region. Leaf rust caused by fungus *Puccinia hordei* has economic importance in many barley growing regions. There is need for new sources of effective leaf rust resistance. Landraces were proven to be rich source of resistance genes for resistance to major pathogens of barley. A total of 880 landraces collected in 7 countries (Morocco – 320, Algeria – 67, Tunisia – 104, Libya – 159, Egypt – 137, Jordan – 93 and Lebanon - 15) were used. These landraces were collected in period of 1981-1995 in 23 germplasm collecting expeditions: 4 in Morocco, 3 in Algeria, 2 in Tunisia, 5 in Libya, 2 in Egypt, 5 in Jordan and 2 in Lebanon. Isolate Ph 25 of *P. hordei* was used. Isolate Ph25 represented the most virulent isolate available allowing the expression of highly effective (in breeders point of view) resistance to leaf rust. In samples of 69 landraces were observed resistant plants. These landraces originated from germplasm collection expeditions in 6 countries: Morocco – 23, Tunisia – 28, Libya – 9, Egypt – 1, Lebanon – 4 and Jordan – 4. None of plants sampled from landraces from Algeria showed resistance to leaf rust. The highest frequency for landraces from duraces from Tunisia (27%) and Lebanon (26,7%). The frequency for landraces from duraces from ther countries was: 7,2% for Morocco, 5,7% for Libya, 4,3% for Jordan, and 0,7% for Egypt.

In samples of 16 landraces all plants showed resistance reaction to leaf rust. Majority of these landraces (14) were collected in Tunisia. In 20% of tested landraces all tested plants showed homogenous leaf rust resistance reactions. Five of them (861,871,872, 1027, 1032) showed resistance reaction 2 and other 9 (881, 883, 885, 889, 892, 893, 894, 944, 955) showed resistance reaction 0. Rest of landraces showed mixed infection types after inoculation with leaf rust isolate. Among them plants of 2 landraces (886 and 933) showed only mixed resistant reactions which were 0 and 2. Plants of landrace 218 showed 3 different infection types 0, 1 and 4. The most frequently observed resistant infection type among samples of landraces was 0 (51 samples – 74%). Other resistant infection types (IT) were significantly less frequent: IT 2 in 12 samples, IT 1 in 5 samples and IT 0; in 4 samples. Susceptible plants were observed in 52 (77%) samples of landraces. Single plant lines resistant to leaf rust were selected from tested landraces for further genetic studies and for future incorporation into breeding materials. Different strategies for control of barley leaf rust were described. In addition, the potential "hot spots" in Southern Mediterranean region for sampling of barley landraces for disease resistance were discussed.

Key words: barley landraces, Hordeum vulgare, leaf rust, Puccinia hordei, sources of resistance

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INTRODUCTION

Barley (*Hordeum vulgare* L.) landraces are grown commonly in Southern Mediterranean region (Bothmer *et al.*, 1995; Fischbeck 2003). In countries of this region barley landraces are grown in place where other cereals are not suitable for farming due to harsh environmental conditions such as high altitude, low rainfall (<300 mm annually), or soil salinity (Bothmer *et al.*, 1995; Yahyaoui *et al.* 1996; Ceccarelli *et al.* 1999; ICARDA 2005). Growing of barley landraces is especially important in dry areas of semi-deserts and the high-lands in Morocco, Algeria, Tunisia, Egypt, Jordan and Lebanon. In these countries barley landraces play especially important role because barley grain often is used for human consumption. This consumption is considerably higher during dry years (Damania 1988; Bothmer *et al.* 1995; Ceccareli *et al.* 1999; Grando 2002; Fischbeck 2003).

Leaf rust caused by fungus *Puccinia hordei* has great economic importance in many barley growing regions in Europe, North America, Australia and West Asia and North Africa (WANA) (Parlevliet *et al.* 1981; Reinhold and Sharp 1982; Lim and Gaunt 1986; Yahyaoui and Sharp 1987; Park *et al.* 1992; Park 2003; Woldeab *et al.* 2006). In last 70 years the use of disease-resistant barley cultivars has been an efficient means for controlling major diseases and preventing yield losses (Brooks *et al.* 2000; Finckh *et al.* 2000; Fischbeck 2003; Weibull *et al.* 2003; Czembor 2005). Based on many studies on the genetics of barley-leaf rust host-pathogen system 19 loci with major genes for resistance were identified: *Rph1*, *Rph2bj*, *Rph2k*, *Rph2l*, *Rph2m*, *Rph2n*, *Rph2q*, *Rph2r*, *Rph2s*, *Rph2t*, *Rph2u*, *Rph2y*, *Rph3c*, *Rph3w*, *Rph3aa*, *Rph4*, *Rph14*, *Rph15*, *Rph16*, *Rph17*, *Rph18*, *Rph19*) (Franckowiak *et al.* 1997; Park and Karakousis 2002; Steffenson 2002; Park *et al.* 2003; Weerasena *et al.* 2004).

Recently it is observed increase of incidences of leaf rust on barley plantations in Europe (Mazaraki and Grabowska, 1998; Niks *et al.* 2000; Czembor *et al.* 2006; Czembor and Czembor 2007a, 2007b). Most probably it is caused by observations of increases in fitness of leaf rust populations to many currently grown barley cultivars and to cultivar Vada. Cultivar Vada is well known for possessing high level of partial resistance. It was used commonly as parent in major European barley breeding programs (Czembor and Czembor 2007a, 2007b). Taking this fact into account, barley breeders, geneticists and plant pathologists are constantly looking for new efficient sources of resistance to leaf rust to combine them with already used in modern cultivars in order to increase the resistance durability (Levine and Cherevick 1952; Nover and Mansfeld 1959; Nover and Lehmann 1968, 1974; Tan 1977; Walther and Lehmann 1980; Sharp and Reinhold 1982; Yahyaoui *et al.* 1988; Alemayehu 1995; Brian *et al.* 1995; Alemayehu and Parlevliet 1996; Chicaiza *et al.* 1996; Backes *et al.* 2003; Bonman *et al.* 2005).

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The original area of cultivation of *H. vulgare* L. was most probably the area of the Fertile Crescent. In this area barley was derived from its wild ancestor *Hordeum spontaneum* C. Koch. when Neolithic men selected spikes with tough rachis. *H. spontaneum* is common in West Asia often growing on the same fields as cultivated barley and it occurs also in Egypt and Libya (Bothmer *et al.* 1995; Hawkes 1995; Badr *et al.* 2000). In addition, the discovery of stands of wild barley in southern Morocco has been reported (Molina-Cano and Conde 1980; Molina-Cano *et al.* 1982). This report suggests that the area of North Africa may be considered as possible additional center of origin for cultivated barley (Molina-Cano *et al.* 1987, 1992, 2002, 2005). Considering this fact, barley landraces collected in Mediterranean region including Sardinia may be rich source of new genes for resistance to leaf rust due to their high degree of diversification resulting from the long co-evolution with populations of pathogen (Wolfe, 1988).

It was shown that barley landraces, especially those collected in Southern Mediterranean region, can be valuable source of many breeding characteristics including resistance to major pathogens (Caddel 1976; Sharp and Reinhold 1982; Yahyaoui *et al.* 1988; Leur *et al.* 1989; Czembor 1996, 2001, 2005; Czembor and Czembor 2001; Martinez *et al.* 2001; Shtaya *et al.* 2006a). The objective of this study was to study leaf rust resistance in barley landraces collected in germplasm collection missions in Southern Mediterranean region.

MATERIALS AND METHODS

Plant material

A total of 880 landraces collected in 7 countries (Morocco – 320, Algeria – 67, Tunisia – 104, Libya – 159, Egypt – 137, Jordan – 93 and Lebanon - 15) were used. These landraces were collected in period of 1981-1995 in 23 germplasm collecting expeditions: 4 in Morocco (Mar84, Mar85, Mar87-1, Mar90), 3 in Algeria (Dza89A, Dza89B, Dza90), 2 in Tunisia (Tun90, Tun90-2), 5 in Libya (Lby81, Lby82, Lby83, Lby90, Lby91), 2 in Egypt (Egy87, Egy89), 5 in Jordan (Jor81-2, Jor81-3, Jor85, Jor91, Jor95) and 2 in Lebanon (Lbn93, Lbn94-2) (Table 1).

Pathogen

Isolate Ph-25 of *P. hordei* was used (Table 2). Isolate Ph-25 represented the most virulent isolate available allowing the expression of highly effective (in breeders point of view) resistance to leaf rust. This isolate originated from IHAR Radzikow collection and was chosen according to differences in virulence spectra observed on 12 differential cultivars.

| Expedition code | Country | Year | Number of accession |
|-----------------|---------|------|---------------------|
| Mar84 | Morocco | 1984 | 90 |
| Mar85 | Morocco | 1985 | 168 |
| Mar87-1 | Morocco | 1987 | 59 |
| Mar90 | Morocco | 1990 | 3 |
| Dza89A | Algeria | 1989 | 17 |
| Dza89B | Algeria | 1989 | 25 |
| Dza90 | Algeria | 1990 | 25 |
| Tun90 | Tunisia | 1990 | 2 |
| Tun90-2 | Tunisia | 1990 | 102 |
| Lby81 | Libya | 1981 | 49 |
| Lby82 | Libya | 1982 | 39 |
| Lby83 | Libya | 1983 | 66 |
| Lby90 | Libya | 1990 | 2 |
| Lby91 | Libya | 1991 | 3 |
| Egy87 | Egypt | 1987 | 125 |
| Egy89 | Egypt | 1989 | 12 |
| Jor81-2 | Jordan | 1981 | 4 |
| Jor81-3 | Jordan | 1981 | 10 |
| Jor85 | Jordan | 1985 | 28 |
| Jor91 | Jordan | 1991 | 45 |
| Jor95 | Jordan | 1995 | 6 |
| Lbn93 | Lebanon | 1993 | 3 |
| Lbn94-2 | Lebanon | 1994 | 12 |
| Total | | | 880 |

Infection types of isolate Ph-25 on differential set.

Table 2

Table 1

| Accession name | Accession number | Gene | Isolate Ph-25 |
|----------------|------------------|-----------|---------------|
| Sudan | CIho 6489 | Rph1 | 4 |
| Peruwian | CI 935 | Rph2 | 4 |
| Estate | CI 3410 | Rph3 | 4 |
| Gold | CI 1145 | Rph4 | 4 |
| Magnif | CI 13860 | Rph2+Rph5 | 4 |
| Bolivia | CI 1257 | Rph2+Rph6 | 4 |
| Cebada Capa | CI 6193 | Rph7 | 0; |
| Egypt 4 | CI 6481 | Rph8 | 4 |
| HOR 2596 | CI 1243 | Rph9 | 4 |
| Cliper C8 | None | Rph10 | 4 |
| Cliper C67 | None | Rph11 | 4 |
| Triumph | PI 290195 | Rph12 | 4 |

Resistance tests

In preliminary test about 30 plants per line were evaluated in a greenhouse with the Ph-25 isolate of *P. hordei*. This study was carried out in the IHAR Radzikow greenhouse. Cultivar L94, which does not carry any known genes for resistance to *P. hordei*, was used as a susceptible control. The plants were grown with 16 h light and temperature range of 18-22°C. Urediniospores of *P. hordei* were suspended in deionized water with couple drops of "Tween 20" and inoculated onto one-week old seedling plants (primary leaf fully expanded) using a rate 3 mg urediniospores and 10 ml of water⁻ for 100 plants. Inoculated plants were incubated for 24 hours in a chamber in which the humidity was maintained near saturation by mist from ultrasonic humidifiers n complete darkness and with a temperature range of 12-15°C. Then plants were transferred to a greenhouse bench.

One to five seedlings from each landrace showing resistance were transferred to greenhouse in order to obtain their seed. In this manner single plant lines of tested landraces were created for further genetic studies and breeders needs.

Disease assessment

Reactions of each accession were evaluated after an incubation period of 12-14 days in a greenhouse at 20-24°C. Disease symptoms were assessed on the primary leaf of the seedlings according to 0-4 scale adapted from Levine & Cherewick (1952) (Table 3). Infection types 0, 0;, 1 and 2 were considered indicative of incompatibility whereas infection types 3 and 4 of compatibility.

Table 3 Description of infection types and codes used (adapted from Levine and Cherewick 1952).

| Host Response | Symptoms |
|------------------------|--|
| Immune | No vivible uredia |
| Very resistant | Hypersensitive flecks |
| Resistant | Small uredia with necrosis |
| Moderately resistant | Small to medium sized uredia with green islands and surrounded by necrosis or chlorosis |
| Moderately susceptible | Medium sized uredia with or without chlorosis |
| Susceptible | Large uredia without chlorosis |
| | Immune Very resistant Resistant Moderately resistant Moderately susceptible |

RESULTS

After screening for leaf rust resistance in samples of 69 (7,8%) landraces were observed resistant plants (Table 4). These landraces originated from germplasm collection expeditions in 6 countries: Morocco – 23, Tunisia – 28, Libya – 9, Egypt – 1, Lebanon – 4 and Jordan – 4 (Table 5). None of plants sampled from landraces from Algeria showed resistance to leaf rust. The highest frequency of landraces showing resistance was among landraces from Tunisia (27%) and Lebanon (26,7%). This frequency for landraces from other countries was: 7,2% for Morocco, 5,7% for Libya, 4,3% for Jordan, and 0,7% for Egypt.

| Table | ate Ph-25 | fter inoculation with isola | ts of landraces a | types observed on plant | Infection |
|-------|-----------|-----------------------------|-------------------|-------------------------|-----------|
| | | Infection type | | Landrace | No. |
| | | 4 | 0 | 515 | 1 |
| | | 4 | 0 | 516 | 2 |
| | | 4 | 0 | 517 | 3 |
| | | 4 | 0 | 532 | 4 |
| | | 4 | 0 | 567 | 5 |
| | | 4 | 0 | 569 | 6 |
| | | 4 | 0 | 573 | 7 |
| | | 4 | 0 | 580 | 8 |
| | | 4 | 0 | 584 | 9 |
| | | 4 | 0 | 586 | 10 |
| | | 4 | 0 | 602 | 11 |
| | | 4 | 0; | 186 | 12 |
| | | 4 | 0; | 189 | 13 |
| | | 4 | 0 | 193 | 14 |
| | 4 | 1 | 0 | 218 | 15 |
| | | 4 | 0 | 222 | 16 |
| | | 4 | 0; | 244 | 17 |
| | | 4 | 1 | 253 | 18 |
| | | 4 | 0 | 292 | 19 |
| | | 4 | 0 | 296 | 20 |
| | | 4 | 0 | 347 | 21 |
| | | 4 | 0 | 348 | 22 |
| | | 4 | 0 | 383 | 23 |
| | | 4 | 0 | 689 | 24 |
| | | 4 | 0 | 856 | 25 |
| | | 4 | 0 | 858 | 26 |
| | | 4 | 0 | 859 | 27 |
| | | 4 | 0 | 860 | 28 |
| | | | 2 | 861 | 29 |
| | | | 2 | 871 | 30 |
| | | | 2 | 872 | 31 |
| | | 4 | 0 | 873 | 32 |
| | | 4 | 2 | 874 | 33 |
| | | 4 | 0 | 879 | 34 |

Table 4

| No. | Landrace | | Infection type | |
|----------|------------|--------|----------------|--|
| 35 | 881 | 0 | • • | |
| 36 | 883 | 0 | | |
| 37 | 885 | 0 | | |
| 38 | 886 | 0 | 2 | |
| 39 | 889 | 0 | | |
| 40 | 891 | 0 | 4 | |
| 41 | 892 | 0 | | |
| 42 | 893 | 0 | | |
| 43 | 894 | 0 | | |
| 44 | 915 | 0 | 4 | |
| 45 | 925 | 0 | 4 | |
| 46 | 933 | 0 | 2 | |
| 47 | 943 | 0 | 4 | |
| 48 | 944 | 0 | | |
| 49 | 953 | 1 | 4 | |
| 50 | 954 | 0 | 4 | |
| 51 | 955 | 0 | | |
| 52 | 784 | 0 | 4 | |
| 53 | 787 | 0; | 4 | |
| 54 | 791 | 0 | 4 | |
| 55 | 1032 | 2 | | |
| 56 | 1037 | 2 | 4 | |
| 57 | 1004 | 1 | 4 | |
| 58 | 1026 | 2 | 4 | |
| 59 | 1027 | 2 | | |
| 60 | 824 | 0 | 4 | |
| 61 | 481 | 0 | 4 | |
| 62 | 132 | 1 | 4 | |
| 63 | 133 | 2 | 4 | |
| 64 | 134 | 2 | 4 | |
| 65 | 162 | 0 | 4 | |
| 66 | 980 | 0 | 4 | |
| 67 | 981 | 0 | 4 | |
| 68 | 986 | 0 | 4 | |
| 68 69 | 986 988 | 0 0 | 4 | |

 Table 4

 Infection types observed on plants of landraces after inoculation with isolate Ph-25 (continued)

| I androad | Constant | Expedition | Date of | I and to I added | | Altitude | Descrines | Cito. |
|-----------|----------|------------|------------|-------------------|-----------|------------|------------|--|
| B | | code | collection | rouginac | | [m.a.s.l.] | LIUVIIIUC | 2110 |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar84 | 1984.05.06 | | | | | |
| | Mar | Mar85 | 1985.05.06 | W02 °53' | N34 °25' | 1400 | Ojda | Taourirt between Bleida and Zagora |
| | Mar | Mar85 | 1985.05.07 | W02° 53' | N34 °25' | 600 | Ouarzazate | Tamgrout, Oued Drea |
| | Mar | Mar85 | 1985.05.07 | W02 °53' | N34 °25' | 700 | Ouarzazate | Amzrou |
| | Mar | Mar85 | 1985.05.12 | | | 200 | | Ouled Saiid |
| | Mar | Mar85 | 1985.05.14 | W08 °50' | N31° 30' | 350 | Marrakech | 15 km W of Chichaoua |
| | Mar | Mar85 | 1985.07.05 | W09 °32' | N30 °36' | 100 | Tiznit | 15 km SE of Agadir |
| | Mar | Mar85 | 1985.07.06 | | | 1700 | Tiznit | Ait Mouia between Irherm and Taliouine |
| | Mar | Mar85 | 1985.07.10 | | | 1400 | | near Arbhaloa |
| | Mar | Mar85 | 1985.07.13 | | | 800 | | Sidi Abbou |
| | Mar | Mar87-1 | 1987.06.17 | W04 °30' N34° 09' | N34° 09' | 006 | Taza | Wad Amlil, 6 km from Matmata to Taza |
| | Mar | Mar87-1 | 1987.06.17 | W04 °30' | N34 °09' | 006 | Taza | Wad Amlil, 6 km from Matmata to Taza |
| | Mar | Mar.07 1 | 1007 06 17 | 10.30 COLIN | 120 01014 | | | |

| - | Landrace Country | Expedition code | Date of collection | Longitude | Latitude | Altitude [m.a.s.l.] | Province | Site |
|---|------------------|--------------------|--------------------|-----------|-----------|------------------------|-----------|--------------------------------|
| 1 | Tun | Tun90 | 1990.05.20 | E10° 10' | N36° 00' | 50 | Kairouan | Al Ouled Ameur |
| | Tun | Tun90-2 | 1990.05 | E10° 28' | N36° 19' | 450 | Nabeul | Bou Ficha |
| | Tun | Tun90-2 | 1990.05 | E10° 27' | N36° 11' | | Sousse | Ouled Ameur |
| | Tun | Tun90-2 | 1990.05 | E10° 27' | N36° 11' | | Sousse | Ouled Ameur |
| | Tun | Tun90-2 | 1990.05 | E10° 27' | N36° 11' | | Sousse | Ouled Ameur |
| | Tun | Tun90-2 | 1990.05 | E10° 27' | N36° 11' | | Sousse | Ouled Ameur |
| | Tun | Tun90-2 | 1990.05 | E09° 04' | N35° 36' | | Kasserine | Sbiba, 6 km N of Sbiba |
| | Tun | Tun90-2 | 1990.05 | E09° 02' | N35° 44' | | Siliana | Rouhia, 8 km N of Rouhia |
| | Tun | Tun90-2 | 1990.05 | E09° 02' | N35° 44' | | Siliana | Rouhia, 8 km N of Rouhia |
| | Tun | Tun90-2 | 1990.05 | E09° 02' | N35° 44' | | Siliana | Rouhia, 8 km N of Rouhia |
| | Tun | Tun90-2 | 1990.05 | E08° 40' | N36° 11' | 700 | Le Kef | Le Kef |
| | Tun | Tun90-2 | 1990.05 | E08° 26' | N36° 08' | 006 | Le Kef | 12 km E of Sakiet Sidi Yousef |
| | Tun | Tun90-2 | 1990.05 | E08° 31' | N36° 19' | 820 | Le Kef | Saa Saa, 4 km W of Fernana |
| | Tun | Tun90-2 | 1990.05 | E08° 30' | N34° 52' | 360 | Kasserine | Oueb Gherib, 4 km S of Fernana |
| | Tun | Tun90-2 | 1990.05 | E08° 30' | N34° 52' | 360 | Kasserine | Oueb Gherib, 4 km S of Fernana |
| | Tun | Tun90-2 | 1990.05 | E09° 04' | N36° 29' | | Beja | Nefza |
| | Tun | Tun90-2 | 1990.05 | E 09°19' | N37° 02' | | Bizerte | Sejenane, 10 km E of Sejenane |
| | Tun | Tun90-2 | 1990.05 | E 09°1'9 | N37° 02' | | Bizerte | Sejenane, 10 km E of Sejenane |
| | Tun | Tun90-2 | 1990.05 | E09° 41' | N37° 03' | | Bizerte | Mateur |
| | Tun | Tun90-2 | 1990.05 | E09° 41' | N37° 03' | | Bizerte | Mateur |
| | Tun | Tun90-2 | 1990.05 | E09° 53' | N33° 53' | | Gabes | Gabes, 20 km W of Gabes |
| | Tun | Tun90-2 | 1990.05 | E10° 25' | N34° 30' | | Sfax | Sfax |
| | Tun | Tun00_2 | 1000.06 | E00° 04' | N1250 261 | | Vocconina | Chiho 7 lim N of Chiho |

| E09°33' | e | Longitude E09°33' E09°33' |
|--|--|---------------------------------|
| 1990.06 E08°41' 1990.06 E08°41' | | |
| 1990.06 E08°37' 981.06.06 E20°56' | 9 | |
| | 1981.06.07 | |
| 981.06.09 E21°32' 981.06.07 E21°11' | Lby81 1981.06.09 E21°32' Lby81 1981.06.07 E21°11' | |
| 981.06.07 E21°11' N32°26' | | E21°11' |
| 982.05.24 E24°14' N31°50' | | E24°14' |
| 982.05.24 E24°14' N31°50' | | E24°14' |
| 982.05.24 E24°14' N31°50' | | E24°14' |
| 983.04.27 E14°30' | Lby83 1983.04.27 E14°30' | |
| 987.04.24 E28°17' | Egy-82-1 1987.04.24 E28°17' | |
| 1981.05 E36°00' | | |
| 1981.05 E36°00' | | |
| 1981.05 E36°00' | | |
| 985.05.22 E36°00' | Jor85 1985.05.22 E36°00' | |
| 994.05.21 | Lbn94-2 1994.05.21 | |
| 994.05.22 E35°41' | Lbn94-2 1994.05.22 E35°41' | |
| 994.05.22 | Lbn94-2 1994.05.22 | |
| 1994.05.23 E35°40' | | |

In samples of 16 landraces all plants showed resistance reaction to leaf rust. Majority of these landraces (14) were collected in Tunisia (Table 4). In 20% of tested landraces all tested plants showed homogenous leaf rust resistance reactions. Five of them (861,871,872, 1027, 1032) showed resistance reaction 0 and other 9 (881, 883, 885, 889, 892, 893, 894, 944, 955) showed resistance reaction 0. Rest of landraces showed mixed infection types after inoculation with leaf rust isolate. Among them plants of 2 landraces (886 and 933) showed only mixed resistant reactions which were 0 and 2. Plants of landrace 218 showed 3 different infection types 0, 1 and 4).

The most frequently observed resistant infection type among samples of landraces was 0 (51 samples – 74%). Other resistant infection types (IT) were significantly less frequent: IT 2 in 12 samples, IT 1 in 5 samples and IT 0; in 4 samples. Susceptible plants were observed in 52 (77%) samples of landraces.

DISCUSSION

Based on obtained results it may be concluded that barley landraces collected in Southern Mediterranean region posses leaf rust resistance. For total 880 landraces screened for leaf rust resistance this resistance was observed in samples of 69 (7,8%) landraces. These landraces originated from germplasm collection expeditions in 6 countries: Morocco, Tunisia, Libya, Egypt, Lebanon and Jordan. The highest number of landraces with leaf rust resistance was in those originated from Tunisia (28 landraces) and the lowest number of such landraces was in landraces from Egypt (1 landrace). None of plants sampled from landraces from Algeria showed resistance to leaf rust. The highest frequency of landraces showing resistance was among landraces from Tunisia (27%) and Lebanon (26,7%). This frequency for landraces from other countries was: 7,2% for Morocco, 5,7% for Libya, 4,3% for Jordan, and 0,7% for Egypt. These results show that in order to collect or screen landraces of barley aiming at leaf rust resistance researchers should be especially interested in materials collected from Tunisia and Lebanon. Based on detailed data concerning collection of barley landraces with leaf rust resistance it may be proposed so called "hot spots" for future collecting missions. Especially interesting country for such future expedition are Tunisia and Lebanon with highest frequency of resistant landraces in presented study. Collecting missions in Mediterranean region are strongly recommended by many investigators because barley landraces in on this area are subject to rapid genetic erosion (Perrino *et al.*, 1986; Damania, 1988; Podyma, 1989; Tazi et al., 1989; Pistrick et al., 1993; Da'aloul, 1995; Malki et al., 1995; Zine Elabidine et al., 1995; Hammer et al., 1996)

In presented study seedling resistance tests were used in order to describe infection types expressed by barley plants after inoculation with isolate of *P*. *hordei*. This kind of testing as sufficient for screening for disease resistance and it is used commonly in many breeding programs (Parlevliet 1976; Jin *et al.* 1995; Czembor 1996, 2005; Brooks *et al.* 2000; Shtaya 2006b; Czembor and Bladenopoulos 2007; Czembor and Czembor 2007a, 2007b). However, using this kind of tests it is not possible or at least it is difficult to identify and describe partial resistance. For description of this kind of resistance we need conduct additional to infection type measurements characteristic for this kind of resistance. In addition, partial resistance is generally better expressed at the adult plant stage (Parlevliet and van Ommeren 1975; Smit and Parlevliet 1990; Martinez *et al.* 2001; Shtaya *et al.* 2006a; Ochoa and Parlevliet 2007). It will be very interesting if further studies of lines selected from described landraces will also include parameters describing partial resistance and extension of resistance studies to plants at adult stage. Final determination of the number of resistance genes and the type of their action in selected lines may be established by testing with differential leaf rust isolates and by proper crosses among appropriate genotypes (Jin and Steffenson 1994; Alemayehu 1995; Czembor 1996, 2005; Czembor and Czembor 2001; Czembor *et al.* 2006).

In many regions of the word many barley varieties had to be discarded because they were the far too disease susceptible to be of any further value. This susceptibility was due to not only by the lost of resistance determined by specific major genes but also by host erosion of partial resistance during breeding of barley for leaf rust race-specific resistance (Parlevliet 1983; Brown and Hovmřller 2002; McDonald and Linde 2002; Ochoa and Parlevliet 2007). Because of all of these facts there is strong need to identify and use new sources of resistance to this pathogen in barley breeding programmes (McDonald and Linde 2002; Backes *et al.* 2003; Czembor and Czembor 2007a, 2007b).

The durability of the resistance genes to leaf rust present in barley cultivars may be increased by using many different strategies for deploying resistance genes. (Parlevliet 1983; Wolfe 1988; Finckh *et al.* 2000; McDonald and Linde 2002). These strategies are: combining partial (minor genes) and race-specific (major) resistance genes, multiline cultivars, partial resistance, combining different race-specific resistance genes into one cultivar and deploying many cultivars with different resistance genes in space (e.g. cultivar mixtures) or time (winter versus spring barley) (Parlevliet 1983; Finckh *et al.* 2000; Brown and Hovmřller 2002; McDonald and Linde 2002). Very important for durability of resistance to leaf rust in agricultural practice is proper use of new sources of resistance to this pathogen (Steffenson *et al.* 1993; Walther 1996; Brown and Hovmřller 2002; McDonald and Linde 2002; Park 2003; Shtaya 2006c).

Studies resulting in description of new sources of resistance are also important because future strategies for the control of barley pathogens will have to focus increasingly on more ecologically acceptable methods. Any usage of chemicals (pesticides, fungicides, herbicides, and mineral fertilizers) in agriculture is increasingly criticized in societies of many countries. Breeding for resistance represent such ecologically safe method. In addition to ecological also economical arguments (use of fuel, labour, special machines) are in favour of breeding for resistance versus chemical control (Nierobca *et al.* 2003; Gullino and Kuijpers 1994).

Barley landraces collected in Mediterranean region are very diverse and often heterogenous concerning many breeding characteristics including disease resistance (Sharp and Reinhold 1982; Yahyaoui *et al.* 1988; Leur *et al.* 1989; Czembor 1996, 2001, 2005; Czembor and Czembor 2001; Martinez *et al.* 2001; Shtaya *et al.* 2006a). This diversity results from big contrasts in this region, both in geographical conditions (e.g. climate, altitude) and in agricultural practices, between the highlands and lowlands (Neffati and Pistrick, 1993; Pistrick *et al.*, 1993; Da'aloul, 1995; Malki *et al.*, 1995; Echikh *et al.*, 1997). This fact was confirmed in presented study because seedlings in 80% of samples expressed mixed infection types.

Results of presented study confirmed findings in other investigations that barley landraces can possess leaf rust resistance different from those in cultivated varieties (Levine and Cherevick 1952; Nover and Mansfeld 1959; Nover and Lehmann 1968, 1974; Tan 1977; Sharp and Reinhold 1982; Yahyaoui *et al.* 1988; Alemayehu 1995; Brian *et al.* 1995; Alemayehu and Parlevliet 1996; Bonman *et al.* 2005). The use of new sources of leaf rust resistance described in this study should result in increasing of leaf rust resistance diversity in barley cultivars.

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