Anadolu Tarım Bilimleri Dergisi,
Anadolu Journal of Agricultural Sciences

e-ISSN: 1308-8769, ANAJAS Haziran 2023, 38 (2): 421-430

Screening of Some Registered Turkish Barley Cultivars Reactions Against *Rhynchosporium Commune*

Bazı Tescilli Çeşitlerin *Rhynchosporium Commune* Hastalığına Karşı Reaksiyonlarının Değerlendirilmesi

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Makale Bilgisi/Article Information

Makale Türü/Article Types: Araştırma Makalesi/Research Article
Geliş Tarihi/Received: 18 Mayıs/May 2022
Kabul Tarihi/Accepted: 5 Haziran/June 2023
Yıl/Year: 2023 | **Cilt-Volume:** 38 | **Sayı-Issue:** 2 | **Sayfa/Pages:** 421-430

Atıf/Cite as: Ertas Oz, M. N., Turgay, E. B., Bulbul, S., Ergun, N. "Screening of Some Registered Turkish Barley Cultivars Reactions Against *Rhynchosporium Commune*" Anadolu Journal of Agricultural Sciences, 38(2), Haziran 2023: 421-430.

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https://doi.org/10.7161/omuanajas.1115743

SCREENING OF SOME REGISTERED TURKISH BARLEY CULTIVARS REACTIONS AGAINST *RHYNCHOSPORIUM COMMUNE*

ABSTRACT

Barley is one of the most significant cereals used as feed and malt both in the world and in Turkey. There are several abiotic and biotic factors affecting the production and quality of barley. Of these factors, fungal diseases play a vital role. Especially, barley scald (*Rhynchosporium commune*), a fungal pathogen, is threatening the barley grown areas in terms of quality and production. One of the most common methods of controlling of this fungal disease is to develop resistant barley cultivars. The study was carried out in the laboratories and greenhouses of Field Crop Central Research Institute, Yenimahalle, Ankara in 2020-2021. The aim of this study was to investigate the reaction of 55 registered barley cultivars against barley scald. The results show that Erginel-90 is immun, as well Avcı-2002, Ocak, Kendal, Hevsel, Yükse, Şahin-91 and Akhisar-98 cultivars are resistant. On the other hand, Yesevi-93, Tarm-92, Orza-96, Larende, Bülbül-89, Karatay-94 and Ayrancı are detected as susceptible to this disease.

Keywords: Barley Scald, *Rhyncosporium Commune*, Barley, Cultivars, Reaction.

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BAZI TESCİLLİ ÇEŞİTLERİN *RHYNCHOSPORIUM COMMUNE* HASTALIĞINA KARŞI REAKSİYONLARININ DEĞERLENDİRİLMESİ

ÖZ

Arpa hem dünyada hem ülkemizde hayvan beslenmesi ve malt yapımında kullanılan önemli tahıl grubundan biridir. Arpa alanlarında üretimi ve kaliteyi etkileyen çeşitli faktörler mevcuttur. Bu abiyotik ve biyotik faktörler içerisinde fungal hastalıklar önemli yer tutmaktadırlar. Özellikle arpa yaprak lekesi (*Rhynchosporium commune*) arpa üretim alanlarında sıklıkla görülen ve verim ve kaliteyi etkileyen önemli bir fungal etmendir. Bu fungal etmen ile savaşımda ise en yaygın kullanılan yöntemlerden birisi de dayanıklı çeşit geliştirmektir. Çalışma 2020-2021 yılları arasında Tarla Bitkileri Merkez Araştırma Enstitüsü sera ve laboratuvarlarında yürütülmüştür. Çalışmanın amacı Türkiye'de tescil edilmiş 55 tescilli arpa çeşitlerinin bu hastalığa karşı reaksiyonları araştırılmasıdır. Buna göre Erginel-90 immun, Avcı-2002, Ocak, Kendal, Hevsel, Yükse, Şahin-91 ve Akhisar 98 tescili arpa çeşitleri dayanıklı olarak tespit edilmiştir. Yesevi-93, Tarm-92, Orza-96, Larende, Bülbül-89, Karatay-94, Ayrancı hassas çeşitler olarak tespit edilmiştir.

Anahtar Kelimeler: Arpa Yaprak Lekesi, *Rhyncosporium Commune*, Arpa, Tescilli Çeşit, Reaksiyon.

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INTRODUCTION

Barley, the most important cereal that grows in temperate regions, is used in animal feeding as well as malt production and human nutrition (Ullrich, 2010). For Turkey, a rise in both barley grown areas and production was reported in 2020, however, a decrease in cultivation area (3,2 million ha) and an increase (5.8 million metric tonnes) in yield were observed in 2021 (Anonymous, 2021). In addition to this, the world's yield of barley is 147 million metric tonnes (Anonymous, 2022).

There are various factors affecting the production and quality of barley negatively which are abiotic (heat, drought and cold stresses) and biotic (diseases, pests and weeds) factors. Barley scald, a biotic factor, is an important pathogen and brings about approximately 20% loss in barley areas (Aktaş, 2001). According to a study conducted in the UK, this causes a 100 £ financial loss per tonne (HGCA, 2011). This disease, reported nearly all over the world (Avrova and Knogge, 2012), might cause lesions in different parts of the plant (Anonymous, 2008). Those lesions have a green-gray colour and oily texture in the beginning, after that they become lesions gray-white in the center and brownish around them (Anonymous, 2008). The life cycle of the pathogen is not fully understood yet, however, it is known that the disease spreads through beak shaped conidia splashing from rain or irrigation water. Additionally, pathogens can infect the plant when the temperature is 4-25 °C and the humidity is high (Anonymous, 2008; Avrova and Knogge, 2012).

The most significant control method for this disease is the development of resistant cultivars since the pathogen might cause up to 40% loss in susceptible varities (Paulitz and Steffenson 2010). The pathogen is polycyclic, therefore the chance of seeing recombinations/mutations among the individuals rises, which means new cultivars might emerge in a season (Zhang et al. 2020). Therefore, the diversity of this pathogen is high both phenotypically and genotypically (Stefansson et al. 2014). As the pathogen exhibits highly virulent patterns even in small-scale geographic locations (Zaffarano et al., 2006; Linde et al., 2009; McDonald, 2015,)366 Rhynchosporium secalis isolates causing scald on barley, rye, and wild barley (Hordeum spontaneum, the pathogen may easily overcome resistance genes deployed in barley cultivars (Zhang et al., 2020). In 2018, thirthy pathohtypes has been reported in Turkey (Azamparsa et al 2019). Additionally, a variety of reaction studies were conducted in Turkey. Azamparsa et al. 2018 tested 198 landraces and 106 wild relatives against six different *R.commune* isolates and found that one landrace and 27 wild relatives were as immune to all isolates Exploring the new durable genes in barley germplasm is an important step in terms of controlling the pathogen. So far, 150 resistant loci have been identified from different barley materials, which wild relatives and landraces are the most significant sources of resistance genes.

Recently, a study done on wild relative and landrace populations of barley collected from Turkey revealed 21 QTL, of which 13 were reported as new. (Clare et al 2023)

This study aims to identify the registered barley cultivars' phenotypical reactions against to barley scald disease. For this reason, this study is a guide for producers and farmers in terms of controlling the disease, and it is also thought that this will be helpful for breeders in their breeding programmes.

MATERIALS AND METHOD

The material for this study consists of fifty-five registered barley cultivars developed by General Directorate of Agricultural Research and Policies (TAGEM) research institutes in Turkey, a susceptible cultivar (Bülbül-89) to barley scald and the most virulent *R. commune* isolate, 75, isolated from a single spore and determined its virulence pattern kept at the fungal collection of Field Crops Central Research Institute (FCCRI).The study was carried out in the laboratories and greenhouses of FCCRI, Yenimahalle, Ankara.

Each genotype was planted in 7x7x9 cm pots with a mixture of soil: sand: fertilizer (60: 20: 20) with 5-7 seeds for each genotype under greenhouse conditions at 22-25°C. Plants were developed up to the 1.5-leaf stage (Zadoks scale 11-12) (Zadoks et al., 1974). Isolate 75 was transferred to petri dishes in Lima Bean Agar (LBA) medium and incubated for 14 days in a 17±2°C incubator for the inoculation. The new isolates developed were removed from the petri dishes with the help of sterile slides by adding sterile distilled water. For inoculation, the obtained suspension was filtered with the help of a cheesecloth and large colony residues were removed and adjusted to $1x10^6$ spore concentration using a thoma slide and Tween 20 (1000µl/l) was added to it. After that the suspension was sprayed on the plants in the 1.5-leaf period with the help of a hand pulverizer. After inoculation, the plants were incubated for 48 hours at 100% humidity and 16-17°C and then transferred to the greenhouse at 22-25°C (Mert and Karakaya, 2004). Plants were watered as needed. After 14 days of inoculation, the plants were evaluated according to the 0-4 scale (El-Ahmed, 1981). This scale indicates 0 as immune (I), 0.1-1.0 as resistant (R), 1.1-2.0 as moderately resistant (MR) 2.1-3.0 as susceptible (S) and 3.1-4.0 as highly susceptible (S). Experiments were carried out in a randomized block design with three replications. Statistical analysis of variance was made with Duncan test in R Environment (version 4.3.0) using *agricolae* library. Briefly, a 0-to-5 scale was used, where 0 = symptom free, 1 = traces or small necrotic flecks, 2 = some chlorosis or necrosis along margins, 3 = necroses but less than 40% affected tissue, 4 = necroses on 40 to 80% of the lamina, and 5 = more than 80% and up to a fully wilted leaf (Abang et al., 2006).

Evaluations were made based on a per/pot basis in which: Highly resistant (0), no visible symptoms; resistant (1), small lesions on the tips or on the base of the leaf; intermediate (2), one to two small lesions on the blade and/or a narrow band of lesions extending at the margin of the leaf; susceptible (3),well-developed lesions on the blade, but without collapse; highly susceptible (4), leaves collapsed (El-Ahmed, 1981)

RESULTS AND DISCUSSION

The resilience of fifty-five cultivars against a virulent isolate was assessed according to 0-5 scale (Abang et al., 2006). In accordance with the scores of the assessments, Erginel-90 was immune, seven cultivars (Avcı-2002, Ocak, Kendal, Hevsel, Yüksel, Şahin-91 and Akhisar-98) showed resistance and twenty-nine cultivars exhibited susceptible reactions (Table 1). In other words, 53.7%, 12.9%, 1.8% of the cultivars were susceptible, resistant and immune, respectively. The differences among the mean values are statistically significant about 5% according to Duncan test (p=0.05) Bülbül-89, among the susceptible cultivars, was used as a control in many studies (Azamparsa et al., 2019, Azamparsa et al., 2020, Azamparsa et al., 2015a, Azamparsa et al., 2015b).

Table 1 The assessments of 55 barley cultivars' reactions to *R. commune* according to El-Ahmad (1981) scale

No	Cultivar	2/6 Rowed	Mean	Reaction
1	Akar	2	3fgh*	S
2	Akhisar 98	6	1bc	R
3	Altıkat	6	2de	MR
4	Anka 06	2	3efg	S
5	Asil	2	4ghi	HS
6	Avcı-2002	6	1bc	R
7	Ay	6	2cd	MR
8	Aydanhanım	2	2de	MR
9	Ayrancı	2	3def	S
10	Barış	2	3efg	S
11	Bilgi-91	2	3efg	S
12	Bolayır	2	3efg	S
13	Bozlak	2	3efg	S
14	Burakbey	2	2de	MR
15	Bülbül 89	2	4hi	HS
16	Cacabey	2	4hi	HS
17	Cumhuriyet 50	2	3efg	S

18	Çetin 2000	6	2de	MR
19	Çıldır 02	2	2de	MR
20	Dara	6	3efg	S
21	Erginel 90	6	0a	I
22	Güldeste	6	3def	S
23	Hamidiye	2	3efg	S
24	Hasat	2	2de	MR
25	Helke	6	3efg	S
26	Hevsel	2	1bc	R
27	İnce-04	2	3cd	S
28	Karatay 94	2	3fgh	S
29	Kendal	6	1bc	R
30	Keser	2	3efg	S
31	Keykubad	2	3efg	S
32	Kıral-97	6	2cd	MR
33	Larende	2	3fgh	S
34	Martı	6	4hi	HS
35	Misket	2	3efg	S
36	Ocak	2	1bc	I
37	Olgun	6	2de	MR
38	Orza 96	2	4fgh	HS
39	Özen	2	3efg	S
40	Sabribey	2	2de	MR
41	Samyeli	2	4i	HS
42	Sladoran	2	2cd	MR
43	Sur-93	2	4ghi	HS
44	Şahin-91	2	1bc	R
45	Tarm-92	2	3fgh	S
46	Tokak 157/37	2	4ghi	HS
47	Tosunpaşa	2	2de	MR
48	Ünver	2	2de	MR
49	Yalın	2	2de	MR
50	Yaprak	6	2cd	MR
51	Yerçil-147(Franken gerste)	2	2de	MR
52	Yesevi 93	2	3efg	S
53	Yeşilköy 387	6	2de	MR
54	Yüksel	2	1ab	R
55	Zeynel Ağa	2	3	S

*Means with the same letter are not significantly different. (p=0.05)

Mert and Karakaya (2004) conducted a similar study and tested five different isolates on 37 barley cultivars. Findings show that Erginel-90 was detected as resistant against all isolates, which is confirmed by this study. Besides Kıral-97 and Çetin-2000 were assessed as resistant by Mert and Karaya (2004), these cultivars were observed as intermediately resistant in this study. The isolates in the previous study and in this study were obtained from the same province, which may indicate the evolution of the pathogen population. The other studies confirm that the pathogen may adopt rapidly, therefore overcoming the resistance of the plants (Williams, 2003; Abang et al., 2006; Zaffarano et al., 2006)866 isolates recovered from the field experiments showed significant, and sometimes opposite, changes in the frequencies of R. secalis genotypes during the growing season (parasitic phase.)

Additionally, 15 six-rowed and 40 two-rowed barley cultivars were tested. Results show generally six-rowed barley cultivars were resistant while two-rowed barley cultvars were more susceptible (Mert and Karakaya, 2004).

Dusunceli et al. 2008 detected the response of 36 barley cultivars to scald disease both in greenhouse and in the field, resulting reported Çetin 2002, Avcı 2002, Erginel 90, Kıral 97, Kaya 7794, Akhisar 98, Zafer 160 and Yeşilköy 387 cultivars were durable under greenhouse and field conditions. This study similarly found Avcı 2002, Erginel 90 ve Akhisar 98 to be immune/resistant and only Çıldır 02 cv exhibited moderate resistant differently.

Hekimhan et al. 2021 screened sixty-three baley cvs. for five virulent *R.commune* isolates in Eskişehir. In the common cvs, Avcı-2002 was resistant, Aydanhanım, Çetin-2000, Kıral-97 and Olgun were moderate resistant, Bilgi-91, Sladoran and Zeynelağa were susceptible in both studies. The resistance responses of the other cultivars differed from each other.

In another study conducted in 2015, twenty-five advanced barley lines and five cultivars were tested against a virulent scald isolate in the greenhouse. While Bülbül 89 and Karatay 94 were grouped as susceptible and highly susceptible, respectively, Avcı-2002 expressed a resistance (Azamparsa, 2015b). Similarly, this study found Bülbül 89 and Karatay 94 as highly susceptible and susceptible respectively whereas Avcı 2002 was durable.

The dissimilarity in the reactions between this study and the studies above might be due to the virulence differences of the pathogens. Though it was reported that the genetic diversity is low according to a study carried out with sixty isolates collected from various points of Turkey (Celik et al., 2021), thirty pathotypes were detected as a result of the surveying across the country (Azamparsa et al., 2015a)

There are plenty of resistant genes identified in wild relatives and in different barley cultivars where 148 QTLs are summarized by Zhang et al. (2020). It is reported that most of these are located in the 3H chromosome (Bjørnstad et al., 2002), and among them, Rrs 1 is an important locus having different identified resistant genes (Looseley et al., 2020) and recent studies reported various loci with identified with Rrs 1 (Daba et al., 2019)net form (NFNB. A study done recently reported novel durable QTL regions in wild relatives and landraces obtained from Turkey (Clare et al., 2023). It is still complicated to understand the mechanism lying under *R. commune* due to the existence of various genes and the impacts of environmental conditions on these genes, thereby more studies need to be done in this area (Zhang et al., 2020).

CONCLUSION

Breeding is one of the most effective and environmentally-friendly ways to control the plant diseases. In breeding programmes, it is important to identify the resistance of barley cultivars both phenotypically and genotypically. This study determines the phenotypic reactions of barley cultivars against *R.commune* and is the basis for further studies. Not only for breeding studies, this study is also a guide for producers/farmers who struggle with choosing a suitable cultivar.

Conflict of Interest

The authors declare that there is no conflict of interest.

Ethics

This study does not require ethics committee approval.

Author Contribution Rates

Design of Study: MNEO(%40), EBT(%30), SB(%30)

Data Acquisition: MNEO(%50), EBT(%25), SB(%25)

Data Analysis: MNEO(%100)

Writing Up: MNEO(%80), EBT(%15), NE(%5)

Submission and Revision: MNEO(%80), EBT(%10),NE(%10)

Acknowlegment

This study is funded by TAGEM under the TAGEM/TBAD/16/A12/P05/001 project.

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