

## АГРОТЕХНИЧЕСКИ ИЗСЛЕДВАНИЯ ЗА УСТОЙЧИВО ПРОИЗВОДСТВО НА ПОЛСКИ КУЛТУРИ

### WHEAT AND BARLEY MYCOSIS DISSEMINATED IN THE REPUBLIC OF MACEDONIA

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#### Abstract

Wheat and barley in the Republic of Macedonia are grown on the area of approximately 200 000 ha with the yield of 3 000 kg/ha and 5 000 kg/ha, respectively. The most important regions are Kumanovo, Bitola, Skopje, Probishtip, Kochani, St. Nikole and Stip. The importance of seed without seed health control results with the introduction of many new diseases on barley and wheat in the Republic of Macedonia. Some of them are detected and identified in the Laboratory of plant and environmental protection in the agriculture faculty in Stip. The most important are: *Tapesia yallundae* Wallwork & Spooner with its anamorphic stage *Pseudocercospora herpotrichoides* (Fron) Deighton causer of the disease “eyespot” on barley; *Rhynchosporium secalis* (Oudem.) Davis causer of the disease “leaf scald” of barley; *Cochliobolus sativus* (Ito.&Kurib.) Drechler ex Dastur causer of the disease “spot blotch and common root rot” of barley; *Septoria tritici* Rob ex Desm and its teliomorphic stage *Mycosphaerella graminicola* (Fuckel.) Schroter causer of “septoria leaf blotch” on wheat. The health condition of wheat and barley has been critical mostly in the early stage of the production in last several years especially on the area where crop rotation is not present. The study deals with the health condition on barley and wheat in the most important production area in the republic of Macedonia during the period 2007-2014.

**Key words:** eyespot, leaf scald, spot blotch, septoria leaf blotch.

#### INTRODUCTION

Wheat and barley are the most important small grain crops used in the daily diet of people and animals with high business, agro-technical and economic importance for the Republic of Macedonia. These small grain crops occupy an area of approximately 200 000 ha from which around 140 000 ha are sown with wheat and 60 000 ha with barley. The most common wheat varieties are Mila, Pobeda, Orovchanka, Radika, Novosadska rana 5, Lepoklasa, Zvezdana, Simonida and Milenka with the average yield of around 3 000 kg/ha. The most common barley varieties are: Barun, Hit, Fantasy, Izvor, Egej, Rex, NS 525, NS 565 and Zlatko, with the average yield estimate at 5 000 kg/ha. The production in Macedonia is intended only for domestic purposes. This quantity doesn't satisfy the real needs of the consumers therefore each year the country imports near 50% of the needs in primary seed material and mercantile. A constant monitoring of health condition is performed on the area under cultivation during the vegetation period. Field control is performed mostly in the area of Kumanovo, Bitola, Skopje, Probishtip,

Kochani, St. Nikole and Stip. Many new diseases on wheat and barley are observed during this field approbation causing significant economic damage to the producers (Karov et al., 2009<sup>a</sup>). The consumption of pesticides increase in order to reduce the yield decrease. Facing the actual need the Laboratory for Plant and Environmental Protection in the frame of UGD – Stip in 2010 begun to perform a seed health control of wheat and barley seed material. The health condition of barley show that 5 to 30% of entire yield lost is caused by several types of phytopathogenic fungi in Macedonia. The presence of *Bipolaris sorokiniana* (Sacc.) Shoemaker, *Rhynchosporium secalis* (Oudem.) Davis, *Pseudocercospora herpotrichoides* (Fron) Deighton teleomorph *Tapesia yallundae* Wallwork & Spooner (Rostr.) on barley and the presence of the anamorphic stage *Septoria tritici* Rob ex Desm. and telomorphic stage *Mycosphaerella graminicola* (Fuckel.) Schroter on wheat are confirmed for the first time in the Republic of Macedonia (Karov et al., 2008<sup>a,b</sup>; Karov et al., 2009<sup>a,b,c</sup>). The aim of this article is to present symptoms, morphology and protective measures of some new fungal pathogens on wheat and barley introduced with the seed import in the Republic of Macedonia.

## MATERIALS AND METHODS

Symptomatic plant material is collected during the field approbations of wheat and barley in the regions of Kumanovo, Skopje, Bitola, Tetovo, Prilep, Stip, Kocani, Sveti Nikole and Probistip in the period 2004-2014. Symptoms in the field are photographed and observed under binocular and microscope, mark OLYMPUS, model XS-402. Seed control was maintained according to Mathur & Kongsdal (2003). Pathogens are isolated on nutrient agar PDA (Nelson et al., 1983) and grown on 22-25°C for 7-10 days. For conidia induce, the pathogen was maintain on Czapeck's Solution Agar (Tuite, 1969), 10 days at temperature of 20-25°C. The identity of fungi is confirmed by the morphology of the pathogen and the use of identification key (Agrios, 2007; Bozidar & Milosevic, 1990). Pathogenicity on barley is confirmed infecting health barley plants variety of Barun, spraying the suspension with 10<sup>7</sup> CFU on the leaf surface and setting healthy seeds in infected soil. The some method is used for testing pathogenicity on wheat infecting healthy plants variety of Mila.

## RESULTS

***Bipolaris sorokiniana* (Sacc.) Shoemaker** teleomorph *Cochliobolus sativus* (Ito & Kurib.) Drechsler ex Dastur is a casual agent of common root rot, leaf spot, seedling blight and black point of wheat and barley. Our investigation discovered only the presence of anamorphic stage *B. sorokiniana* in all investigated area. The perfect stage was not found. All barley varieties grown in the Republic of Macedonia are susceptible to black point unlike to wheat varieties which didn't show susceptibility. According to literature data teleomorph stage in nature was found only in Zambia (Raemaekers, 1988). Primary symptoms, occurred in the early stage of barley development due to the presence of conidia inoculum in soil and seed. At the end of the winter and early in the spring a black brown elliptical spots appear on the young leaves (Figura 1). The second infections become from conidia produced in this spots or from diseased grass hosts (Figura 2). Some of the plants recover from the infection (Figura 3) and mature normally, but the infection progress every year

due to the multiplication of the inoculum in the soil in mono cultural production. Large number of conidia having 3-6 septa, dark green to brown colored, 21-47 μm in lenght and 16-18 μm width (Figura 4).



Figura1. Primary infection of *B. sorokiniana*



Figura 2. Secondary infections



Figura 3. Plant recovering from the infection

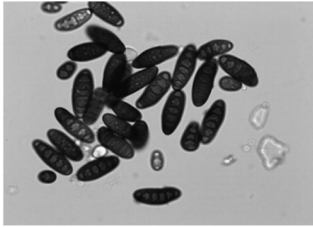


Figura 4. Conidia of *B. sorokiniana*

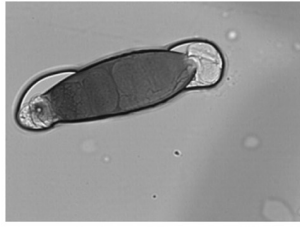


Figura 5. Bipolar germination of conidia

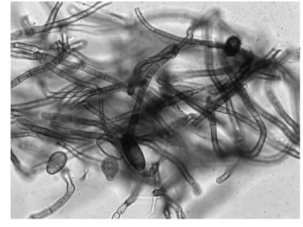


Figura 6. Olivaceous mycelium

***Tapesia yallundae* Wallwork & Spooner** cause disease called “Eyespot”. It is a teleomorphic stage of the fungus which anamorph is *Pseudocercospora herpotrichoides* (Fron) Deighton. Symptoms appeared as elliptical or “eye” shaped lesions at the stem basis (Figura 7 and Figura 8). The eyespot lesions are elliptical, greyish to olivaceous inside, with dark brown margins (Figura 8). Later the inside of the lesions become sunken, and develop fungus-darkened centers. Poor root systems associated with whitehead formation are usually observed. In future

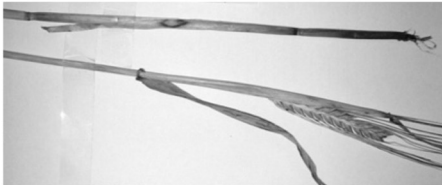


Figura 7. Eyespot symptoms

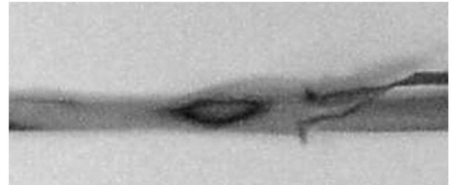


Figura 8. Eyespot lesion on the steam base

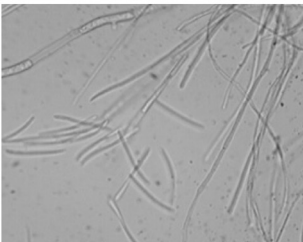


Figura 9. Conidia of *P. herpotrichoides*

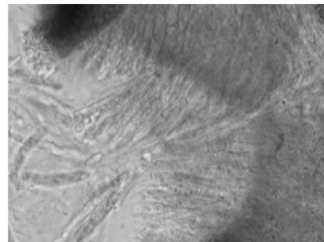


Figura 10. Apothecia and asci of *T. yallundae*

development of the disease, basal lesions cause stem breakage. In the beginning, mycelium is white to gray, latter becoming light brown to olivaceous. It produces conidiophores and long, tight conidia (Figura 9), which cause the secondary infections in spring. Primary infections due to the presence of ascospores. These infections cause the eyespot symptoms on the base of the steam. Our observations discovered the presence of apothecia during the vegetation period in May (Karov et al., 2009<sup>c</sup>). Apothecia carry a great number of asci (Figura 10). Each ascus bear eight ascospores responsible for primary infections (Lucas et al., 2000).

***Rhynchosporium secalis* (Oudem.) Davis** cause the disease called leaf scald on barley. The symptoms appear on leaves and steam in form of oval shaped and elongate lesions up to 1.5 cm surrounded with dark brown borders (Figura 11). Microscopic observation show the presence of two celled conidia with dimensions from 10-20 x 2-4  $\mu\text{m}$ . Conidia are hyaline and the terminal cell hooks in to a beak (Figura 12). Teleomorphic stage is not observed. The fungus overwinter in plant debris and in seed in form of mycelium and conidia. Conidia cause primary infections early in the spring and spread the disease with the secondary infections during the vegetation period. The climate in Macedonia is favorable for development of the disease. According to Nazarova et al., (1998) the disease can develop in around two weeks if the climate conditions are favorable. The Macedonian variety of barley called Barun was very susceptible to the attack of this pathogen. The yield loss in this variety was estimate up to 30% according to the weather condition in spring and summer (Karov, 2008<sup>a</sup>).



Figura 11. Symptoms of “leaf scald” on barley



Figura 12. Conidia of *R. secalis*

***Mycosphaerella graminicola* (Fuckel.) Schroter** or “Septoria leaf blotch”. First symptoms are observed during the period of June 2007 like small yellow areas in the field of barley area in Bitola and Prilep. First symptoms appear on lower leaves as chlorotic spots. The infection progress on upper leaves, while lower leaves become chlorotic with the presence of black pycnidia (Figura 13) representing the anamorphic stadium of the fungus called *S. tritici* Rob ex Desm. Pycnidia are oval and black with diameter of 71-96  $\mu\text{m}$ . Pycnidium cracks and liberates a great number of micro (8-9,5  $\mu\text{m}$ ) and macro-pycnosporos (Figura 13 and Figura 14) which cause the infection. Teleomorphic stage was observed for the first time in May in 2008 in the area of Probishtip in form of perithecia on

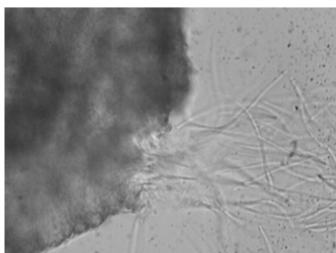


Figura 13. Pycnidia of *S. tritici* liberating pycnospores

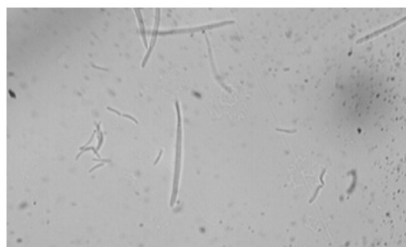


Figura 14. Macro and micro-pycnospores of *S. tritici*

the lower part of the stem (Karov et al., 2008<sup>b</sup>). Perithecia were dark brown to black in color with diameter of 72-95  $\mu\text{m}$  caring a lot of asci (Figura 15). Asci are limpid with two layers bearing eight ascospores. Ascospores are hyaline, elliptical and two – celled (Figura 16). Field investigations showed that all wheat varieties are susceptible to the disease.

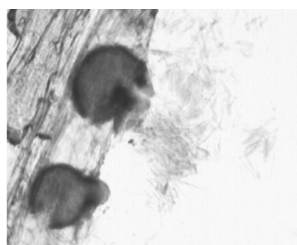


Figura 15. Perithecia of *M. graminicola* on straw and liberation of asci

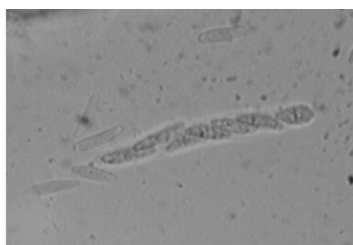


Figura 16. Ascus and ascospores of *M. graminicola*



Figura 17. Testing the effectiveness of the fungicide Duett Ultra against septoriosis

The pathogen overwinters as perithecia in diseased plant refuse, left in the field and in infected seed. Seedling infections result in damping off, and provide inoculum for subsequent infections (Halama, 1996).

## DISCUSSION AND CONCLUSIONS

Sowing the imported seed without seed health control results in the introduction of many new diseases of barley and wheat before a decade ago in the Republic of Macedonia. Some of them are observed for the first time in 2007 like eyespot, leaf scald, spot blotch and septoria leaf blotch. Symptoms of these diseases are observed each year with higher or lower intensity depending on the weather conditions early in spring and during the summer. Each of them can be transmitted with the seed and overwinter in plant debris and weeds. The most spread is spot blotch caused by the pathogen *B. sorokiniana*. According to Bakonyi et al. (1997) primary monocotyledon hosts are *Triticum aestivum*, *Secale cereale*, *Hordeum sativum*, *Avena sativa* and grasses *Agropyron repens*, *Alopecurus pratensis*, *Bromus erectus*, *Dactylis glomerata*, *Festuca ovina*, *Lolium perenne*, *Poa pratensis* and *Setaria viridis*. Symptoms are observed only on barley. The most susceptible varieties are Rex and Barun. The infection in the diseased area in Macedonia is spread from the diseased plant straw left in the field. Climate conditions in Macedonia are favorable for anamorphic stage to overwinter mostly on the grass flora and make the primary infections early in spring. Septoria leaf

blotch is caused by *S. tritici* teleomorph *M. graminicola* is currently the most important foliar disease on wheat in Europe and US (Ponomarenko, 2011). Both, anamorphic and teleomorphic stages are involved in the disease cycle in the Republic of Macedonia. Teleomorphic stage is observed in May on lower part of the stem. It can survive in seed, straw and stubble. The main host is wheat and occasionally rye, triticale and some grass species. Leaf scald is wide distributed disease on barley. It is caused by the pathogen *R. secalis*. It attack barley, rye, triticale and a number of grasses (Mathre, 1997). Wheat is not a host of leaf scald. The fungus overwinters on the crop debris and stubble. It produces abundant conidia on wet lesions during cool, damp weather after the leaf tissue has become necrotic. Conidia, spread by wind and splashing rain, infect young leaves of spring- planted grain. Optimum temperatures for sporulation and infection range from 10-18 C. Hot and dry weather reduces the rate of disease development. The disease called eyespot is caused by the fungus *P. herpotrichoides* and the teleomorphic stage *T. yallundae* (Wallwork & Spooner, 1988). Both stages were found in the climate conditions in Macedonia. The teleomorphic stage was rarely found in the field. It is responsible for genetic recombination and therefore it would influence the fungicide sensitivity (Nicholson et al., 1991). This means that if frequent production on straw in the field is present the development of fungicide resistance will be often present and should be investigated each year. The experience in the field for suppression of leaf diseases in barley and wheat shows that the application of Tilt 250 EC (Propiconazole 250 g/l) for the first treatment at barley and wheat in the early stage of symptom development when the infection had reached 5% of the first or second leaf, show a significant results. The fungicide Propiconazole is an ergosterol-biosynthesis inhibitor and controls a broad-spectrum of diseases at small grains like septoriosis (*Septoria* spp.), eye spot (*P. herpotrichoides*), leaf scald (*R. secalis*), black spot (*B. sorokiniana*), rust (*Puccinia* spp.), powdery mildew (*Blumeria graminis*) and net blotch disease (*Pyrenophora teres*) (Thomson, 1997). The application of Duett Ultra (187 g/l Epoksikonazol and 310 g/l Tiofant-metil) in the second and eventually third treatment was highly effective in the disease control (Fig. 17). The combination of triazole and carbamate in the formulation of Duett Ultra decrease the possibility for occurring the resistant pathogen varieties. Considering the fact that all of these pathogens are introduced in the field with the diseased seed and maintained on plant debris and weed flora a good soil and weed management is essential for the inoculum decrease. The best results are achieved when Propiconazole is used for the first treatment and Dual gold for the second and third application in a dose of 1 and 0,5 l/ha, respectively. In 2014 the weather conditions were suitable for disease development. Plots where no chemical measures were conducted were completely destroyed from diseases mostly the rust such as in St. Nikole, Kumanovo and Kochani. Plots where three fungicide applications were conducted have above 6 t of yield per hectare.

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