

# INTRODUCTION OF NEW PATHOGENS ON BARLEY IN REPUBLIC OF MACEDONIA

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## ABSTRACT

Barley in the Republic of Macedonia is grown on the area of approximately 50000 ha with the yield of 3 500 kg/ha. Although this area is very small, barley growers constantly, have problems with the health condition of the crop due to the seed import. In the period of 2009 – 2010, introduction of several new diseases are observed on the territory of the Republic of Macedonia: “leaf scald”, “food and root rot” and “septoria leaf blotch”. Diseased material is investigated in the laboratory and the pathogens are isolated and identified as *Rhynchosporium secalis* (Oudem.) Davis, *Cochliobolus sativus* (Ito.&Kurib.) Drechler ex Dastur, and *Septoria tritici* Rob ex Desm. Preventive measures are conducted pre sowing, and several fungicides are investigated for their efficiency in *in vitro* conditions.

**Key words:** leaf scald, food and root rot, septoria leaf blotch

## INTRODUCTION

Mycoses are the most economically important diseases at barley in the world and the Republic of Macedonia, too. The most common diseases are powdery mildew (*Blumeria graminis*), rust (*Puccinia graminis* f.sp. *hordei*), head blight (*Fusarium graminearum*), septoria leaf blotch (*Septoria tritici*) and loose smut (*Ustilago nuda*) (Pejcinovski F. & Mitrev S., 2009; Ivanovic M., 1992). These diseases didn't cause much losses because of the effective treatment they have. But three years ago new symptoms were observed in the barley fields in Macedonia. Laboratory investigations show the presence of three new fungal diseases: leaf scald, food and root rot (Karov et al., 2007) and septoria leaf blotch (Karov et al. 2008). These diseases have become the serious threat of the barley production in Republic of Macedonia.

The pathogen causing leaf scald is fungus *Rhynchosporium secalis*. This pathogen has become the most economically important last two years, occurring in every region where barley is grown with the intensity of approximately 30%.

Food and root rot is caused by the fungus *Bipolaris sorokiniana*. The fungus has teleomorph (sexual stage) *Cochliobolus sativus*. It is the causal agent of a wide variety of cereal diseases. The pathogen can infect and cause disease on the root (where it is known as common root rot), leaf and stem, and head tissue. *C.sativus* is extremely rare in nature, thus its asexual or anamorphic stage *Bipolaris sorokiniana* causes infections. The two most common diseases caused by *B.sorokiniana* are spot blotch and common root rot.

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*Septoria tritici* is fungal that cause the disease septoria leaf blotch. Three different fungi cause blotch diseases: *Stagonospora nodorum* (*Phaeosphaeria nodorum*), *Septoria tritici* (perfect state *Mycosphaerella graminicola*) and *Stagonospora avenae* f. sp. *triticea* (*Phaeosphaeria avenaria* f. sp. *triticea*). *Stagonospora nodorum* causes disease on leaves and glumes of the head, whereas *Septoria tritici* attacks leaves only (Pejcinovski F. & Mitrev S., 2009; Ivanovic M., 1992).

## MATERIALS AND METHODS

Plant material is collected during the field approbations of *Hordeum* L. in the regions of: Kumanovo, Skopje, Bitola, Tetovo, Prilep, Stip, Kocani, Sveti Nikole and Probistip in 2008, 2009 and 2010. Symptoms in the field are photographed and observed under binocular and microscope, mark OLYMPUS, model XS-402. Symptomatic plants are investigated in *in vitro* conditions. Facultative fungal parasites are isolate on nutrient agar PDA, and grown on 25 °C, 7-10 days. For conidia induce, some pathogens are maintain on Czapeck's Solution Agar (Tuite, 1969), for 7 - 10 days at temperature of 20-25 °C.

The identity of fungi is confirmed by the morphology of the pathogens and the use of identification key (Agrios, 2005; Halama P. 1996; Pejcinovski F. & Mitrev S., 2009; Wiese M.V., 1977).

Pathogenicity is confirmed infecting health barley plants cv. "Barun", spraying the suspension with  $10^7$  CFU on the leaf surface and setting healthy seeds in infected soil, setting infected seed in no infected soil and for control setting healthy seeds in no infected soil was conduct.

## RESULTS

The investigation of barley health condition in the field, show the presence of common fungal diseases and some new symptoms that where not present in the field before. According to the symptoms we notice, we assumed to the presence of new pathogens causing "leaf scald", "food and root rot" and "septoria leaf blotch". *In vitro* investigation confirmed the presence of the pathogens that cause these diseases.

### ***Septoria tritici* Rob ex Desm.**

First symptoms are observed during the period of June 2008. Small yellow areas are observed in the field of barley area in Tetovo, Bitola, Prilep and Strumica. 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 (Fig. 1).



Fig.1 Pycnidia in the necrotic leafe tissue

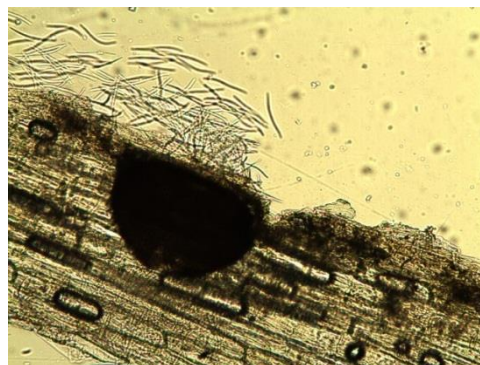


Fig.2 Pycnosporos liberating from pycnidia

Picnidia are oval and black in color with diameter of 71-96  $\mu\text{m}$ . Pycnidium cracks and liberates a great number of micro (8-9,5 $\mu\text{m}$ ) and macropycnosporos (Fig 2) which cause the primer and second infections during the vegetation. The morphological characters and the symptoms allow us to assume the presence of “septoria leaf blotch” caused by the plant pathogen fungus *Septoria tritici* in Macedonia that wasn't the case in the past.

#### ***Cochliobolus sativus* (Ito & Kurib.) Drechsler ex Dastur**

The causal agent of spot blotch and common root rot of barley is discovered almost in every area where the barley is grown: Kumanovo, Skopje, Bitola, Tetovo, Prilep, Stip, Kocani, Sveti Nikole and Strumica. Primary symptoms are observed in the early stage of barley development, on coleoptiles, subcrown, and primary and secondary roots. We assumed that this situation due to the presence of conidia inoculum in the soil and seed infection. At the end of the winter and early in the spring, underground plant parts appeared yellow and not developed enough.



Fig.3 Symptoms of *spot blotch*  
In the field



Fig. 4 Conidia of *Cochliobolus sativus*

The pathogen from soil inoculum destroy the vascular system, thus the plant is unable to supply the nutrients and appear yellow and stunted as compared to healthy plants. The second infections become from conidia produced on the wheat straw or diseased grass hosts. Second infections appear on leaves as black or brown flacks which expand into oval shaped black flack not exceeding 1cm in length (Fig. 3). Some of the plants recover from the infection and mature normally. Our observations in the field show that since 2008 symptoms are present very year with higher intensity, due to the multiplication of the inoculum in the soil.

Laboratory investigation show the presence of large number of conidia having 3-6 septa, dark green to brown colored, 21-47  $\mu\text{m}$  in lenght and 16-18  $\mu\text{m}$  width. In *in vitro* conditions on Chazapeck's medium the presence of olivaceous, septed mycelium with large number of conidiophores and conidia are observed under the microscope. Conidia are olivaceous with three or four septa and germinate after a short period stand in water (Fig. 4).

#### ***Rhynchosporium secalis* (Oudem.) Davis**

The first observation of leaf scald symptoms was in May 2009 in the area of Kochani. After, the symptoms were observed in Shtip, Strumica, Bitola, Prilep and St. Nicole. Symptoms occur mainly on leaves as subcircular, elliptic to oblong lesions. Latter, the lesions become greyish white in the center with dark brown border. Hard diseased plants show symptoms on the steam too. The infection was estimated at 25%.



Fig. 5 Symptoms of leaf scald in the field



Fig.6 Conidia of *Rhynchosporium secalis*

## DISSCUSION

Barley production in the Republic of Macedonia is not intended for export. The biggest production is in the region of Kumanovo with the area of approximately 7700 ha and Bitola with 5 000 ha. Other regions with smaller production are located in Skopje, St. Nicole, Karbinci, Gradsko ect. In our investigation we monitored nine regions independently of the quantity of barley production: Kumanovo, Skopje, Bitola, Tetovo, Prilep, Shtip, Kocani, St. Nikole and Probistip. Barley seed usually is import from the neighboring countries due to the insufficient production, or the small farmers produce barley for their own needs. The total yield production is estimated at approximately 175 000t per year which is insufficient for the needs of the country (Statistical review: Agriculture, 2009). Many plant pathogens are introduced in the country due to the seed import. This results with several problems at farmers and barley producers. In order to overcome these problems producers conduct intensive soil residue management, and laboratory investigations of seed health before sowing.

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