Mini-Review

iMedPub Journals http://www.imedpub.com

Archives of Clinical Microbiology ISSN 1989-8436 2021

Vol. 12 No. 6:180

Natural Emergence and Severity of Leaf and Glume Blotch Diseases of Winter Wheat and Winter Triticale Incited by Complex of Fungi *Parastagonospora* spp./*Zymoseptoria tritici* under Climatic Conditions of Poland

Abstract

The overall purpose of the presented minireview is to show natural emergence of very destructive necrotrophic fungi from genera of Parastagonospora spp. and Zymoseptoria tritici known to infect grasses and cereal crops including winter wheat and winter triticale worldwide. The study of natural emergence of the fungal species was conducted in 8 different geographic locations of Poland. The experiments were done with 10 winter wheat and 10 winter triticale varieties planted to field plots of 10 m² in a randomised block design. In spring and early summer plants on the plots were assessed visually om emergence of diseases caused by the pathogens and diseased plant parts from each plot were collected for microscope inspection. The reason of the inspection was to determine by which pathogen plants were affected and how the dieseases progressed upwards on the cereal plants. Significant differences among the eight locations spread around the country were found. Zymoseptoria tritici was most frequently identified on the studied genotypes of winter wheat. On the other hand the most frequently isolated pathogen from different heights of winter triticale plants was *P. nodorum*. The incidence of *P. avenae* f. sp. triticea on both crops, wheat and triticale appeared to be much less frequently isolated from infected plant parts at different heights than that of Z. tritici and P. nodorum. The present research also aimed to prove statistically the impact of climatic factors on the natural emergence of the pathogens affecting green plant parts at various plant heights of winter wheat and winter triticale in Poland. However, the plant-pathogen-climate interactions were not proven statistically. The bunch of correlation coefficients, sometimes positive, sometimes negative were very low, thus making inferences about disease emergence at different plant heights appeared to be quite risky.

Keywords: Winter wheat; Winter triticale; Natural leaf and glume blotch incidence; *Parastagonospora* spp., *Zymoseptoria tritici*

Edward Arseniuk*

Department of Plant Breeding and Acclimatization, Institute-National Research Institute, Blonie, Poland

* Corresponding author: Arseniuk E,

Plant Breeding and Acclimatization, Institute-National Research Institute, Blonie, Poland, India

e.arseniuk@ihar.edu.pl

Citation: Arseniuk E (2021) Natural Emergence and Severity of Leaf and Glume Blotch Diseases of Winter Wheat and Winter Triticale Incited by Complex of Fungi *Parastagonospora* spp./ *Zymoseptoria tritici* under climatic conditions of Poland. Arch Clin Microbiol Vol.12 No.S5: 180.

Introduction

Complex of necrotrophic fungi from genera of *Parastagonospora* spp./*Zymoseptoria tritici* has been observed to occur naturally on cereal and grass species for quite a number of years. The occurrence of *P. nodorum, Z. tritici,* and *P. avenae* f.sp. *triticea* depends not only on cereal species but also on multiple climatic factors [1-14]. Symptoms and severity of diseases evoked by the above pathogens appear to be of different magnitude depending upon course of weather conditions to which belong: frequent rainfalls, high relative air humidity and air temperatures about 20-

25°C. The latter conditions are very conducive for development of blotches incited by *Parastagonospora* spp. and especially by *P. nodorum, Zymoseptoria tritici* and *P. avenaria* f.sp. *triticea*.

The above pathogens are considered the most devastating for the small-grain cereals and especially for wheat (*Triticum aestivum* L.) and triticale (× *Triticosecale* Wittm.) [3, 15-20]. Diseases incited by *Parastagonospora* spp. and *Zymoseptoria tritici* cause significant quantitative and qualitative grain yield losses and hamper grain production of the above indicated small grain cereal species.

According to a number of reports the pathogens from the *Parastagonospora* spp/*Zymoseptoria tritici* complex afflict cereal species with substantial grain yield losses in quantity and quality. Parastagonospora nodorum blotch (syn. Stagonospora nodorum blotch, SNB) may cause grain yield losses in wheat of up to 31% [15]. On the other hand Zymoseptoria tritici blotch (syn. Septoria tritici blotch, STB) may reduce grain yield losses in wheat as much as 50% [20]. It should be mentioned, that Zymoseptoria tritici blotch did not occur on triticale at all, however, in recent years appeared reports claiming that the latter disease occurs also on triticale, which hectarage in Poland reached 1,3 millions ha. Along with increased triticale popularity, the crop is being affected by a number of new, mainly of wheat origin diseases.

It is to underline that the above reported losses afflicted by necrotrophic pathogens *Parastagonospora* spp./*Zymoseptoria tritici* adversely affect primarily wheat production in Poland and other European countries [21,22]. On the other hand some time ago Arseniuk et al. [15] reported a negative impact of *Parastagonospora* spp./*Z. tritici* complex on the quality and quantity of wheat and triticale grain, which is shriveled and of reduced size. Since then the situation got aggravated. New pathogens and diseases appeared on triticale in the last decades.

It also should be indicated that another fungal pathogen from Parastagonospora/Zymoseptoria complex, namely P. avenae f. sp. triticea, is considered as a minor pathogen of wheat [10,23,24] and triticale [1]. Two special forms of P. avenae were identified. From our study results, that the first one, the *P. avenae* f. sp. triticea (NCBI:txid54790: Parastagonospora avenae f. sp. tritici) may severely affect wheat and triticale. The second one, the P. avenae f. sp. avenaria (NCBI:txid215456: Parastagonospora avenae f. sp. avenae), is destructive to oats [14]. In later studies, McDonald et al. [25], determined genetic differences in groups of isolates of Phaeosphaeria spp., and 5 phylogenetically distinct clades were identified: Pat1, Pat3, Pat4, Pat5, and Pat6. In Poland, P. avenaria f.sp. triticea isolates Pat2 and some homothallic P. avenaria f.sp. triticea isolates Pat1 occurred on foxtail barley (Hordeum jubatum) [5]. Some oat, wheat, rye, and triticale isolates from Poland also appeared to be Pat1 [11]. In McDonald et al. [25] study appeared, that the isolates of the Pat5 clade were avirulent to susceptible wheat cultivars.

The research reported in this paper is based on data collected over 2015 to 2020. Its aim was to provide more information on the natural occurrence, its spatial and temporal distribution of the complex of fungi of *Parastagonospora* spp./*Zymoseptoria tritici,* the common necrotrophic pathogens of wheat and triticale worldwide and especially in Poland.

Field Experiments

The studies were conducted in 8 experimental locations of Poland. These were: Bartążek: 20.482843 E 53.71086 N, Bonin: 16.2477 E 54.153613 N, Borowo: 16.790134 E 52.114888 N, Grodkowice: 20.27488 E 49.970209 N, Małyszyn: 15.174479 E 52.744631 N, Oleśnica Mała: 17.260902 E 50.847896 N, Smolice: 17.183084 E 51.691261 N and Ożańsk: 15.17448 E 52.74463 N) during the years 2015-2020. The experiments consisted, respectively, 10 winter wheat and 10 winter triticale varieties. The varieties varied in their response to *P. nodorum* and to *Z. tritici*. The seeds were planted to field plots of size 1×10 m in a randomised block design. Plants on plots were not chemically protected [26-28]. In the summer plants on the plots were assessed visually and diseased plant parts were collected. The collected diseased plant samples were analysed mycologically to determine by which pathogen these were affected.

Lesions on infected plant parts were inspected with a binocular microscope (25 ×). Mature pycnidia were transferred onto an agar solid medium with a glass needle. Pycnidiospores from cirrhi were spread over the surface of the agar medium. Conidia were measured with a use of digital microscope camera. To produce monospore cultures individual spores were isolated on solid media: V8 juice-PDA was used for *Parastagonospora* spp. [29] and YMDA was used to develop *Z. tritici* [30].

The following taxonomic criteria were applied for species identification of developed isolates: visual assessment of disease symptoms, morphology and dimension of conidia, morphology of colony on artificial solid media (in vitro). Dimensions of conidia for individual species were following: $25-45 \times 3-4 \mu m$ for *P. avenae* f. sp. *triticea*, $15-32 \times 2-4 \mu m$ for *P. nodorum* [1], and $35-98 \times 1-3 \mu m$ for *Z. tritici* [28]. The morphology of colony of each isolate was also used as a taxonomic criterium.

The data sets obtained in experimental locations were also statistically analysed with selected meteorological factors such as: absolute maximum temperature (°C), average maximum temperature (°C), absolute minimum temperature (°C), average minimum temperature (°C), monthly average temperature (°C), minimum near-surface temperature (°C), monthly sum of rainfall (mm), maximum daily rainfall (mm), first day of maximum rainfall, number of days with snow cover, number of days with rainfall, and number of days with snowfall. The interpolation of data obtained in each of the experimental location was estimated by software program language using such models as: Scikit-learn, pandas, geopandas, numpy and matplotlib. The training data set was expanded by rotation matrix of two-dimensional geographical coordinates in Euclidean space using the following equation of Harris et al. [31-34].

Description and discussion of results

Occurrence of pathogens on wheat and triticale

It is to state that significant differences among the eight locations spread around the country to study the natural incidence frequency of the pathogens were found. The average incidence of *Z. tritici* was most frequently identified on the studied genotypes of winter wheat. The average number of isolations of *Z. tritici* from the infected plant parts was statistically different from ones of *P. nodorum* (p < 0.001) and *P. avenae* f. sp. *triticea* (p<0.001). The incidenc of *P. avenae* f. sp. *triticea* appeared to be 86 times less frequent than that of *Z. tritici*, while *P. nodorum* was isolated 10 times less often as compared to *Z. tritici*. It was found that the

incidence of *P. avenae* f. sp. *triticea* on winter wheat was 8 times less frequent than that of *P. nodorum*.

The most frequently isolated pathogen from winter triticale was P. nodorum. The difference between the average number of P. nodorum isolations from winter triticale in experimental plots over the study period of 2015-2020 and the average number of isolations of P. avenae f. sp. triticea and Z. tritici was statistically significant. Nevertheless, the difference in the average occurrence of P. avenae f. sp. triticea and Z. tritici was not statistically significant [35]. The mean number of isolations of P. avenae f. sp. triticea was 6.8 and the average number of isolations of Z. tritici was 7.7. According to previous studies, P. nodorum and P. avenae f. sp. triticea were considered as less host preferring pathogens, while the Z. tritici host preference to wheat was more distinct [1,13,36-38]. In the research conducted by Arseniuk et al. [1,39], Tian et al., and Sapkota et al., wheat was determined as the primary host for Z. tritici, while the occurrence of Z. tritici on triticale was not observed at all or it was observed only very rarely [1,6,35,40].

Lesions with mature pycnidia of P. nodorum on glumes of winter wheat and winter triticale were observed very rarely, i.e., in 53 cases on winter wheat and in 28 cases on winter triticale. Surprisingly, in 12 cases, Z. tritici symptoms were identified on winter wheat heads (cvs. Fidelius, Bogatka, Ostroga, Muszelka), while these were observed only one time on winter triticale cv Cyrkon. The latter phenomenon is not common, although it was already reported by Jones and Cooke [37]. There were also symptoms of P. avenae f. sp. triticea on heads of triticale cultivars Baltiko and Atletico (n=5 isolations). The rare incidence of P. nodorum on glumes could be attributed to breeding for resistance [27] and/or climatic changes [6]. In the studies by Arseniuk et al. [38], necrotrophic effector genes were identified in populations of breeding lines and cultivars; 57% of triticale lines and 30% of winter wheat cultivars were susceptible to the SnTox3 necrotrophic effector. The other effectors, namely SnTox1, SnTox5, and SnToxA had a negligible effect on the susceptibility of lines at seedling and adult plant growth stages. In the present study, the average isolation number of P. nodorum from disease plant parts of winter triticale plants amounted to 25 times, while on winter wheat it amounted to 16 times only.

It was found that *Z. tritici* occurs on wheat glumes what is surprising although in other studies employing more sensitive monitoring methods demonstrated that the pathogen was detected even without clearly visible lesions on spikes [13]. In studies of Tian et al. [13] the antigen amount of *Z. tritici* was measured using BA-ELISA test, which revealed that there was a greater quantity of antigen in F-1 leaves than in the flag ones. Minimal amounts of antigen were detected in glumes, and none in grains, what is in accordance with data of Shaw et. al. [26] employing real-time PCR method on a large number of samples of *M. graminicola*. DNA was detected in grain samples of wheat. However, the formation of mature pycnidia of *Z. tritici* on spikes under natural conditions was never observed. Nevertheless, it is worth to note that the in present research this phenomenon manifests on very susceptible wheat varieties such as Fidelius, Bogatka, Ostroga, Muszelka. It is suspected that this is due to the fact that the disease progresses upwards on leaves very quickly, and reaches spikes when glumes are still green and quite soft, what speeds up to develop symptoms on glumes. Under favourable conditions *P. nodorum* can easily develop lesions on both, glumes and leaves. In research reported by Shaw et. al. [26], a strong correlation between *Phaeosphaeria nodorum* blotch abundance on wheat grains and leaves was found (r = 0.7). The pathogen was more detrimental to ears as compared to leaves. Pycnidiospores much easier are transferred with seed and no airborne spores would be necessary to begin the disease outbreak in consecutive years [26]. There is also always a high risk of grain quality deterioration in the following years.

Correlation between meteorological factors and occurrence of pathogens

Pearson correlation coefficients were calculated between meteorological factors and the occurrence of the studied pathogens. In total, 137 meteorological factors were considered but because of very low values of correlation coefficients their analysis is omitted. Using large numbers degrees of freedom it is statistically possible to prove significant of anything. Methods of statistics should not serves these purposes, despite the facts that some stubborn colleague researchers (28) try to use statistics to prove and to make convincing their, sometimes horrible, ideas, e.g. that the snow cover and freezing temperatures stimulate development of *Parastagonospora/Septoria tritici* blotches. However, I personally disagree with such conclusions as the above.

Conclusion

Climate changes are known to occur depending upon regions and seasons and affect natural incidences of pets and pathogens, and agricultural production as well. All these factors affect agricultural production and this way also food security and safety, since the yields are reduced in quantity and poorer in quality. To deterioration of yields contribute not only climate changes but also changes in populations of pests and pathogens stimulated by climatic conditions. It is not a mystery, that actual yields are being affected to a large extent by climate-dependent pathogens, to which belongs the complex of necrotrophic complex of fungi Parastagonospora spp./Zymoseptoria tritici. The present research aimed to statistically prove the impact of climatic factors on the on the natural incidence of the pathogens affecting green plant parts of winter wheat and winter triticale in Poland, however, the plantpathogen-climate interactions were not proven statistically. The bunch of correlation coefficients, sometimes positive, sometimes negative was very low, thus making inferences very risky. It is generally known that the studied pathogens flourish under humid conditions and moderate temperatures. Zymoseptoria tritici prefers climate of north-west Europe and P. nodorum strives well in continental, mid-eastern and Eastern Europe. Regarding affected crops it was proven that wheat under natural conditions is largely affected by Z. tritici and triticale by P. nodorum.

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