

GENETIC VARIABILITY OF MAIZE PATHOGENS IN SERBIA

Slavica STANKOVIĆ, Jelena LEVIĆ and Dragica IVANOVIĆ

Maize Research Institute „Zemun Polje“, Belgrade-Zemun, Serbia

Stanković S. J. Lević and D. Ivanović (2007): *Genetic variability of maize pathogens in Serbia*. – Genetika, Vol.39, No. 2, 227-240.

Variability of some maize pathogens was identified in the last 50 years of research in Serbia, mostly by their cultural characteristics and susceptibility of test genotypes and only in some cases by determination of mating types, vegetative compatibility or biochemical methods. Although more advanced methods that can determine within population variability at the molecular level were developed, they are still not applied in research in Serbia. The highest variability was determined for maize leaf pathogens - *Exserohilum turcicum* (Pass.) Leonard & Suggs (2 races) and *Bipolaris zeicola* (Stout) Shoemaker (2 races), although this variability is significantly lower than the variability of the same pathogens found in the world. Researches conducted with the aim to determine mating types and vegetative compatibility of the *Fusarium* species, a maize root and stalk pathogen, indicated their high variability in Serbia. Considering the pathogen ability to adapt easily and quickly to new genotypes, agro-ecological conditions and crop practice, a constant

Corresponding author: Slavica Stanković, Maize Research Institute „Zemun Polje“, S. Bajića 1, 11185 Belgrade, Serbia, Tel: 011/3756704, Fax: 011/3756707
e-mail: sstojkov@mrizp.co.yu

surveillance of parasite divergence and epidemiology is necessary in order to avoid detrimental consequences on maize yield and quality.

Key words: maize, variability, VCG, mating population, races, pathogen

INTRODUCTION

Development of maize genotypes resistant or tolerant to pathogen microorganisms is the cheapest, the simplest and the most effective way of suppressing their impact on yield losses and kernel quality. Maize genotypes resistant towards a specific pathogen race display this resistance no matter in which region of the world they are grown. For example, genotypes resistant towards *Exserohilum turcicum* (Pass.) Leonard & Suggs (syn. *Helminthosporium turcicum* Pass.), *Bipolaris zeicola* (Stout) Shoemaker (syn. *Helminthosporium carbonum* Ull.) that were developed in USA were also resistant to the same pathogens when grown in Serbia (PENČIĆ *et al.*, 1981). However, development of these genotypes is characterized by a constant quest for new sources of resistance that have to be different in target genes, because of the possibility of pathogen new race or patotype occurrence within one species or occurrence of new species, mainly by introduction. Pathogen ability, especially fungi, is to adapt easily to new maize genotypes (mainly to those which are grown on large areas), as well as to agro-ecological conditions and crop practices. Hitherto results indicate that the largest number of races was found within fungi which cause maize leaf diseases.

Different types of variability within a population deriving from one species are possible, such as cultural (colony appearance and substrate pigmentation), morphological (conidia and other structure shape and size), physiological (races) and toxicological (potential for mycotoxins biosynthesis). Cultural characteristics are the usual approach for variability determination, which is distinctive for isolates with different origin of species and very rare for races of species. Interaction between a pathogen and maize genotype is very important for determination and definition of a race. However, one isolate can cause different symptoms on different genotypes. In PENČIĆ and LEVIĆ (1979) it was shown that 30 maize genotypes inoculated with one *Kabatiella zae* Narita et Hiratsukja isolate showed typical eyespot symptoms and also six different symptoms that were connected with the level of genotype resistance. The list of pathogen microorganisms is constantly increasing due to novel information on relationships between host plants and parasitic, saprophytic or endophytic pathogens. Pathogen and non-pathogen isolates of *Fusarium oxysporum* Schlecht, the causer of maize stalk and root rot, can be distinguished solely on the results of pathogenicity test (BURGESS *et al.*, 1994). *F. verticillioides* (Sacc.) Nirenberg (syn. *F. moniliforme* Sheld.) which also causes root, stalk and ear rot can be isolated from tissues with no disease symptoms (MUNKVOLD and DESJARDINS, 1997), in which the pathogen stays inactive as long as the tissue is physiologically active.

Phaeocystroma ambiguum (Mont.) Petrak, the causer of root and stalk rot produces α and β conidia. The first are ellipsoid, egg-shaped and dark, while β conidia are thready and colorless, sometimes intermediate (LEVIĆ and PETROVIĆ, 1998). Races of *K. zea*, the causer of maize eyespot, are not known, but differences in carbon and nitrogen utilization between the isolates were determined (LEVIĆ and PENČIĆ, 1990). Different cultural, but similar morphological characteristics were found for races 2 and 4 of *B. zeicola* (LEVIĆ and PENČIĆ, 1993, navesti i druge autore).

In this paper an overview of population variability of the pathogens which are the main causers of maize diseases in Serbia (and worldwide) is given. In Serbia maize pathogen variability was identified mostly by their cultural characteristics and susceptibility of the test genotypes. In some cases determination of mating types, vegetative compatibility or biochemical markers were used. On the other hand, within population variability determination on molecular (DNA) level is used worldwide.

Races

Race is a sub-compartment or a group of a species similar in morphological, but different in physiological, biochemical, pathological or other characteristics. In this context, pathogenic races, physiological races or pathotypes of species are mentioned. Pathogenic race is species specific, even for a distinct genotype of species. For example, southern leaf blight epidemic that produced severe losses in maize production in USA in 1970. was explained by occurrence of T race of *Bipolaris maydis* (syn. *Helminthosporium maydis* (Nisikado) Sub. & Jain), to which maize genotypes with cms-T cytoplasm that were grown on large areas were extremely susceptible (OLDFIELD, 1989).

Research has shown that races develop due to growing maize genotypes with monogenic resistance on large areas and for long periods, thus enabling the increase in the number of individuals within the parasite population adapted to a given genotype. At one point avirulent population becomes virulent to the cultivar and a new race occurs. A genotype that was resistant towards one race becomes susceptible to another race. This phenomenon is known as genetic vulnerability and its biochemical and physiological base is the consequence of the changes that occur in genes and enzymes (VAN DER PLANK, 1975). In these cases, genotype-pathogen interaction functions on the gene-for-gene concept (FLOR, 1942), meaning that each resistance gene in the host corresponds to the pathogen avirulence gene or that each susceptibility gene in the host corresponds to the pathogen virulence gene.

In Serbia cases of genetic vulnerability towards *B. zeicola* race 4 (LEVIĆ *et al.*, 1994) and BYDV (IVANOVIĆ *et al.*, 1990, 1992) were recorded. Their occurrence was most intensive and extensive when maize production was based mostly on genotypes of the same BSS and Lancaster origin, which were extremely susceptible towards *B. zeicola* race 4 and BYDV, respectively. Literature data

show that maize pathogen variability is much lower in Serbia than in the world, when compared with the variability of the same species.

Exserohilum turcicum (Pass.) Leonard & Suggs (syn. *Helminthosporium turcicum* Pass.). The initial designation of races of *E. turcicum* was done by BERQUIST and MASIAS (1974) after they characterized an isolate from Hawaii, which was virulent to *Ht1* as race 2. Since then 11 races have been identified (R0, R1, R2, R3, RN, R12, R1N, R12N, R123N, R23, R23N), with race R0 being economically the most important, while race R1 shows spreading tendency (BERQUIST and MASIAS, 1974). LEONARD *et al.* (1989) consider that the appearance of much higher number of pathogenic races is possible than it was identified previously.

Following a classification system proposed by LEONARD *et al.* (1989), *E. turcicum* races are defined based on their phenotypic reactions when inoculated onto a set of differential maize lines. In this system, *E. turcicum* race designations are assigned according to the maize resistance genes that their virulence matches, e.g. *E. turcicum* race 0 is ineffective (avirulent) against all *Ht* genes described above whereas *E. turcicum* race 1 is only effective (virulent) against *Ht1*.

Both races are effective against maize genotypes lacking all resistance genes. Race 0 has the resistance formula, *Ht1, Ht2, Ht3, HtN*, race 1, *Ht2, Ht3, HtN / Ht1*; race 2, *Ht1, Ht3, HtN/Ht2*, race 3, *Ht1 / Ht2 Ht3*, race 12, *Ht3, HtN / Ht1, Ht2*, race 23, *Ht2, Ht3 / Ht1, HtN* and race 23N, *Ht2, Ht3, HtN / Ht1*. This classification left room for the accommodation of new races that may be encountered in future studies. The *Ht1, Ht2* and *Ht3* resistant genes occur as chlorotic lesions with minimum sporulation, while the *HtN* induced resistance is expressed as a delay in disease development until after pollination (LEONARD *et al.* 1989).

In North America *E. turcicum* races R0 and R1 predominate whereas R2N and R23N rarely occur (BERQUIST and MASIAS, 1974; FALLAH MOGHADDAM and PATAKY, 1994; JORDAN *et al.*, 1983; LIPPS and HITE, 1982; PIECZARKA, 1980; SMITH AND KINSEY, 1980; THAKUR *et al.*, 1989; WINDES and PEDERSEN, 1991). TURNER and JOHNSON (1980) reported the presence of race 1 in Indiana that was virulent on *Ht1* but not *Ht2*. LIPPS and HITE (1982) reported the presence of race R1 in Ohio that was found also virulent on *Ht1* but avirulent on *Ht2*. SMITH and KINSEY (1980) reported a new race designated race 3 with a resistance formula *Ht1/Ht2, Ht3*. Later in 1983, JORDAN *et al.* (1983) reported the occurrence of race R1 and race R2 from seven states in the central and eastern USA where race R1 was virulent on B37 only and race R2 virulent on B37Ht1. No isolate was found virulent on B37Ht2 or Oh43Ht3.

WELZ *et al.* 1993 indicated the presence of races R0 and R1 in China, R23N, R23, and R2N in Mexico, R23, R23N, and R0 in Zambia, and R0, R2, RN, and R23N in Uganda. ADIPALA *et al.* (1993a) found that all the isolates tested were virulent on A619 and avirulent on A619Ht1, A619Ht2, A619Ht3 and A619HtN, hence they were classified as race R0. In Brazil *E. turcicum* populations seem to be more diverse in terms of race composition. GIANASI *et al.* (1996), for instance,

observed a predominance of R0 but also detected races capable of overcoming resistance conferred by *Ht1* (R1N, R12N, and R123N).

There is a lack of data for *E. turcicum* races diversity in Europe. To investigate the pathogen's population genetic structure in central Europe, a total of 80 isolates was sampled in Germany, Switzerland, France, Austria, and Hungary and investigated with 52 random amplified polymorphic DNA (RAPD) markers. Among the 73 isolates from maize there were 26 different RAPD haplotypes. All isolates with identical haplotype are considered clonemates. Only a single clone had members in both southeastern Austria and southwestern Switzerland, suggesting that the Alps constitute a major barrier for this pathogen (BERCHARDT *et al.*, 1998). New race of *E. turcicum* was established on the basis of a susceptible response of genotypes with *Ht1* gene in Croatia (PALAVERŠIĆ *et al.*, 1995) and Slovenia (ROZMAN *et al.*, 2003)

Two races of *E. turcicum* were identified in Serbia. Race R0 is present since 1925 (JOSIFOVIĆ, 1925), but it provoked severe losses since the mid 50-ies until mid 70-ies because maize hybrids introduced from USA that were susceptible to this pathogen were grown on large areas. After this period, the majority of the hybrids used to have a polygenic type of resistance. During the last two decades the race specialisation of *E. turcicum* in Serbia was studied on the basis of the response of differential maize inbred lines carrying *Ht* gene. Results showed that until 1999 only race R0 existed. In that year a susceptible response to *E. turcicum* of a genotype carrying the *Ht1* gene was observed, indicating the occurrence of a new race (LEVIĆ and PETROVIĆ, 1999).

Bipolaris zeicola (Stout) Shoemaker (syn. *Helminthosporium carbonum* Ull.). HOOKER and PERKINS (1980) were the first to indicate that the variability of this pathogen is higher in the nature than it was suggested in the literature, what was later proven in DODD, 1993 and TRAUT and WARREN, 1993. The pathogen drew attention of the scientists when race 3 was discovered (CASTOR *et al.*, 1976; HALSETH *et al.*, 1991), because this race was thought to be a new *Bipolaris maydis* Drechslera (syn. *Helminthosporium maydis* Nisikado & Miyake) race which was the cause of great losses in maize (with cms-T cytoplasm) production in that period.

Five races of *Bipolaris zeicola* are described, which are designated as 0, 1, 2, 3 and 4. The rates by which typical spots are formed, as well as their size on susceptible genotypes are useful parameters for race determination (Table 1).

The most dispersed is race 2, while race 3 shows spreading tendency. Incidence of races 1 and 4 is limited, since a small number of maize genotypes are susceptible to these races. *B. Zeicola* patotype that was first described by DODD and HOOKER (1990) is designated as race 4. WELZ and LEONARD (1988) identified race 0 which is avirulent to maize genotypes otherwise susceptible to races 1, 2 and 3. On the basis of the results in their work these authors concluded that race 0 is genetically different from the mentioned races and that its avirulence to maize genotypes indicates that it is spread from some, for now unknown, weed species.

B. zeicola was identified in Serbia for the first time in 1959. in the vicinity of Sivac (JOSIFOVIĆ and MIRIĆ, 1960). The most dispersed is race 2 which can cause huge losses on some genotypes and in some years, due to significant reduction of photosynthetic area (ROSENFELD, 1980). Growing genotypes with narrow genetic basis from BSS populations on huge areas and in long period brought to the occurrence of a new race – race 4 (LEVIĆ and PENČIĆ, 1992). The intensive upgrowth of this pathogen during the 80-ies was not only the consequence of growing susceptible genotypes, but also of climatic conditions, especially favorable rainfall configuration from the end of July to the beginning of August. This pathogen does not occur in Serbia any more, since maize assortment was changed and drought years became more frequent. Although the presence of race 1 in Serbia is mentioned in some literature, our research on *B. zeicola* over 30 years long did not confirm the presence of this race. This can be explained by the fact there are no maize genotypes selected in Serbia that were susceptible to race 1, as it was the case with American inbred lines Pr and K61.

Table 1. Reactions of susceptible and resistant maize genotypes to different races of *Bipolaris zeicola*

Race of <i>B. zeicola</i>	Maize genotype		Authors
	Susceptible ^a	Resistant	
0	Avirulent to maize, genetically different from races 1, 2, 3 and 4		WELZ and LEONARD, 1988, 1993
1	oval to circular spots, with dark center and dark-watery margins, (11.4 x 3.8 mm); spots appear 6 days after inoculation	oval to circular, small, brown spots	WELZ and LEONARD, 1988
2	Necrotic, dark to brown, oval to irregular spots (1.9 x 0.7 mm) appear 6 days after inoculation	Chlorotic or necrotic, circular to oval spots	WELZ and LEONARD, 1988
3	Linear spots length/width- 1/7	Small oval or short linear chlorotic spots	WELZ and LEONARD, 1988
4	Circular to oval spots; 5 to 10 mm in diameter.	Mostly avirulent race, rarely small, irregular spots appears	LEVIĆ and PENČIĆ 1992; DODD and HOOKER, 1990

^aSusceptible genotypes of maize: Pr and K61 inbreds to race 1, W64 inbred to race 2, Pa33 inbred to race 3 and B73 to race 4 by *B. zeicola*.

Pathogen variability determined with biochemical methods

Genetic and molecular techniques as well as classical mycological and plant pathological methods can be used to investigate the variability of pathogens. Analyses of DNA sequences, either directly or as a fragment from a genomic DNA fractionation protocol, are becoming common in fungal determination.

Differences in protein profile within certain isolates of one *B zeicola* pathotype confirmed presence of race 4 of this pathogen in Serbia (IVANOVIĆ *et al*, 1992). Considering morphological, cultural and pathogenic characteristics of 10 investigated isolates, originating from diseased maize in Serbia, three species of the genus *Periconia* were identified: *P. macrospinosa* Lefebvre & A.G. Johnson, *P. circinata* (Managin) Sacc. i *P digitata* (Cook) Sacc. Biochemical changes in the mycelia soluble protein content of these species confirmed their taxonomic differences (STOJKOV *et al.*, 1996).

STOJKOV and IGNJATOVIĆ (1998) reported that the biochemical characterisation revealed that four of five analyzed isolates had qualitatively the same protein pattern of *Microdochium bolleyi*, (Spregue) De Hoog & Hermandies-Nijhof, maize root and stalk pathogen. A different set of bands for the fifth isolate points the possibility of the existence of the different pathotype in Serbia.

Variation among *Gibberella fujikuroi* species complex

G. fujikuroi species complex includes nine mating populations and all of them correspond to particular anamorphic species in the *Liseola* section of *Fusarium*. Significant differences in morphology, genetics and mycotoxins production exist. between the populations Populations A, D and E are widely distributed and are the most frequent fungi isolated from diseased maize plants (with or without symptoms) in Serbia (LEVIĆ *et al.*, 1997).

Results of research conducted to determine mating types for three species: *F. verticillioides*, *F. proliferatum* (Wollenw. & Reink.) Nelson et al. i *F. subglutinans* (Matsushima) Nirenberg, originating from diseased maize in different agroecological conditions, showed that mating populations A1 (*F. verticillioides*), D2 (*F. proliferatum*) and E1 (*F. subglutinans*) are prevalent in Serbia.

Results on the effective population size based on mating type of three biological species belonging to the *Gibberella fujikuroi* species complex are given in Table 2. (LEVIĆ, J., S. STANKOVIĆ and T. KOVAČEVIĆ, nepublikovani podaci). The effective population number for one mating type [$N_{e(mt)}$] was evaluated following Leslie and Klein (1996) by the equation: [$N_{e(mt)}$] = $(4 N_+ N_-)/(N_+ + N_-)$, where N_+ , i.e. N_- is the number of isolates with the mating type *MATD-1*, i.e. the mating type *MATD-2*, respectively. Number of inbreed effective population [$N_{e(f)}$] was evaluated following by the equation: [$N_{e(f)}$] = $(4N^2 \times N_h)/(N + N_h)^2$, where N is the total number of isolates and N_h is the number of hermaphrodites. Based on the proportion between the mating population and effective population, the highest variability was indicated in *F. proliferatum* (Table 2).

Stanković *et al.* (2004) showed that isolates of *F. proliferatum* originating from different localities in Serbia had the highest potential for the biosynthesis of fumonisin B1 and bovericin. Strains of *F. verticillioides* possessed the highest potential for the synthesis of fusaproliferin, while isolates of *F. proliferatum* and *F. subglutinans* displayed approximately the same potential for the synthesis of beauvericin. Similar results about the mycotoxins production ability of some

Fusarium species from USA were obtained by MUNKVOLD and DESJARDINS (1997)

Table 2. - Experimental data and effective population size parameters for three biological species in the *Gibberella fujikuroi* species complex origin from maize in Serbia

Biological species	Mating type ^a	$N_{fs} : N_h$	N_e		
			M.T. ^b	N_{ef} ^c	$N_{e(v)}$ ^d
A	6 : 5	14 : 1	99.1	16.7	93.7
D	8 : 13	20 : 4	94.0	66.7	85.7
E	5 : 6	13 : 0	99.0	0.0	100.0

^aMating type: *MAT-1:MAT-2*; ^bEffective population number based on mating type and expressed as percent of actual count; ^cInbreeding effective number based on numbers of males and hermaphrodites and expressed as a percentage of the actual count; ^dVariance effective number based on numbers of males and hermaphrodites and expressed as a percentage of the actual count; N_{fs} – number of female-sterile male-fertile strains; N_h – number of hermaphroditic strains; N_e – effective population number.

Variation based on vegetative compatibility

Vegetative compatibility also known as heterocaryon compatibility has been documented in numerous ascomycetes and it means that two hyphae can anastomose and fuse to form a stable heterocarion. The vegetative compatibility phenotype has a multigenic basis, and can be used as a means of identifying a set of isolates that all share common alleles at those loci. As the number of loci governing the trait is large, the number of possible VCGs is also very large.

In Serbia, *F. verticillioides*, as a pathogen of grain, was identified on maize, wheat and sorghum up to 77.8% (LEVIĆ *et al.*, 1997, 2003), 10% (DOPUĐA and LEVIĆ, 2004) and 7.5% (LEVIĆ *et al.*, 2006), respectively. The characterisation of *F. verticillioides* isolates can be done on the basis of the seedling pathogenicity test or vegetative compatibility, since it was determined that isolates of the similar pathogenicity belonged to the same vegetative compatibility group (VCG) (KLEIN and CORRELL, 2001).

Vegetative compatibility of *F. verticillioides* isolates, originating from maize and wheat grown on different locations in Serbia, and their pathogenicity to maize seedlings were studied by KRNJAJA *et al.* (2006). According to these authors a total of 10 VCGs of *F. verticillioides* were determined in the complementation tests. The number of vegetative compatible groups indicates a high genetic diversity of observed *F. verticillioides* populations in Serbia.

Variation between virus pathogens

Maize dwarf mosaic virus. Two viruses belonging to the *Potyviridae* (PVY) family are currently recognized in Serbia - Maize Dwarf Mosaic Virus (MDMV) and Sugarcane Mosaic Virus (SCMV). The main symptoms on maize plants infected by these viruses are mosaic patterns of light and darker green

streaks and reduction in plant height. Based on the symptoms it is not possible to distinguish which of the viruses infected the plants. Infection with both MDMV and SCMV causes more severe symptoms, such as mosaic with chlorosis, i.e. mosaic with whitening of the leaves, thus even more complicating pathogen identification (IVANOVIĆ and IVANOVIĆ, 1994).

Experiments on distinguishing MDMV and SCMV have been conducted on Johnson grass for a long time. It was at the beginning of the nineties of the last century that the virus-specific antibodies for these two viruses were developed (TOŠIĆ *et al.*, 1990, IVANOVIĆ *et al.*, 1995a). Based on the antigen specificity of the virus proteins that was revealed in a study on about 20 virus strains causing the same mosaic symptoms on maize and sugar cane worldwide, these varieties were aligned as four discrete viruses of the *Potyviridae* family: MDMV, SCMV, JgMV (Johnson grass mosaic virus) and SMV (Sorghum mosaic virus).

One of the specificity of Potyvirus group is forming cylindrical or pinwheel like inclusion bodies in the host cell cytoplasm. Serological studies of the inclusion bodies formed by 18 *Potyvirus* strains suggested MDMV specificity, i.e. MDMV was found to be related only to JgMV. Virus specific epitopes were found on C-terminal domains, while epitopes common for most *Potyvirus* were found on N-terminal domains, thus indicating their preservation (HAMMOND, 1998).

Barley yellow dwarf virus. Barley yellow dwarf virus – BYDV (*Luteoviridae* family) was observed on barley, wheat and maize for the first time in Serbia in 1990. (BALAŽ, 1990; IVANOVIĆ *et al.*, 1990). This virus infects over 150 hosts from *Poaceae* family, residing in the phloem tissues. Aphids, vectors for BYDV, become infective 24-28 hours after adopting the virus from phloem tissue (persistent transmission) and stay infective for several weeks or throughout the life cycle, depending on the aphid specie and virus strain. Different studies revealed high genetic variability of the virus, as well as the possibility of new strains development through genotypic and phenotypic mixation, thus complicating development of resistant plant (such as wheat, rye, barley, maize) varieties. Infection with different BYDV strains does not provide cross protection, but has additive effect on severe symptom manifestation.

Different strains have been described as BYDV. Taxonomy of these viruses is based on genome organization, serological characteristics and vector type. Five strains of BYDV were recognized based on the vector specificity (ROCHOW, 1979; DUFFUS *et al.*, 1990). Each of the strain was designated by the first letters in the names of the vector. For example, *Rhopalosiphum padi* is the vector of the RPV strain; *R. padi* and *Sitobion avenae* are the vectors of SGV strain. The strain is described as specific if it is transmitted by a single vector (RPV) and as non-specific if it is transmitted by several vectors (PAV, MAV). PAV strain originating from maize is designated as M-PAV (M-maize).

Studies on cytopathological changes in infected rye plants with monoclonal antibodies indicate existence of two sub-groups within BYDV strains (HSU *et al.*, 1984). Strains whose progeny develop in cytoplasm and form one membrane vesicles (PAV, MAV, and SGV) belong to sub-group I. RPV and RMV strains belong to sub-

group II and the progeny of these strains develop in the nucleus of the infected cells and form two membrane vesicles.

In an experiment with DNA probes WATERHOUSE *et al.* (1986) did not find genome homology between RPV and PAV strains. On the other hand they found homology between these strains and Potato leafroll virus (PLRV) and Western yellows virus (BWYV). These results indicate that, although intensive research has been conducted on relationships between BYDV strains, their classification is not definitive and clear and that revision is necessary. Virus classification based on vector specificity has been replaced with a new concept – serological specificity of the strains. Using this concept two genus, within *Luteoviridae* family, has been separated: Luteovirus and Polerovirus. BYDV-MAV, BYDV-PAV Cereal Yellow Dwarf Virus (CYDV, with the strain CYDV-RPV) are the most common, while RMV, SGV and Chinese strain GPV are still indisposed (ZITTER, 2001; D'ARCY and DOMIER, 2005.).

According to OSLER *et al.* (1985) and IVANOVIĆ *et al.* (1992) BYDV is a solemn maize pathogen in south Europe regions, where maize is also the main summer host for BYDV. M-PAV is the dominant strain in this part of Europe, while MAV strain is less frequent (IVANOVIĆ *et al.*, 1995b). BYDV and CYDV are important sweet corn pathogens in New York State (ZITTER, 2001). A huge incidence of BYDV and MDMV attacks on sweet corn was observed in Serbia in 2007, which led to severe yield losses (Ivanović M., personal communication).

Received July 22nd, 2007

Accepted August 17th, 2007

REFERENCES

- ADIPALA, E., P.E. LIPPS and L.V. MADDEN (1993): Reaction of maize cultivars from Uganda to *Exserohilum turcicum*. *Phytopathology* 83: 217–223.
- BALAŽ, F. (1990): Pojava virusa žute patuljivosti ječma (Barley Yellow Dwarf Virus) u Vojvodini. *Zaštita bilja* 193: 241–210.
- BERQUIST, R.R. and O.R. MASIAS (1974): Physiologic specialization in *Trichometasphaeria turcica* f. sp. *zeae* and *T. turcica* f. sp. *sorghii* in Hawaii. *Phytopathology* 64: 645–649.
- BORCHARDT, D.S., H. G. WELZ and H.H. GEIGER (1998): Molecular marker analysis of European *Setosphaeria turcica* populations. *European Journal Of Plant athology* 104: 611–617.
- BURGESS, L.W., B.A. SUMMERELL, S. BULLOCK, K.P. GOTT and D. BACKHOUSE (1994): Laboratory Manual for *Fusarium* Research. Third Edition. *Fusarium Research Laboratory, Department of Crop Sciences, University of Sydney and Royal Botanic Gardens, Sydney*, 133.
- CARSON, M.L. (1995): A new gene in maize conferring the chlorotic halo reaction to infection by *Exserohilum turcicum*. *Plant Dis.* 79: 717–720.
- CARSON, M.L. and Z.W. WICKS (1993): Response of a maize synthetic to S1 recurrent selection for grain yield in a disease-stress environment. *Maydica* 38:193–199.
- CASTOR, L.L., J.L. AYERS, R.R. NELSON and A.J. SCHREIBER (1976): The reaction of several corn inbreds to race 3 of *Helminthoaporium carbonum*. *Plant Dis. Rep.* 60: 827–830.

- D'ARCY, C.J., and L.L. DOMIER (2005): Barley yellow dwarf. www.APSnet Education Center – Plant Disease Lessons – Barley yellow dwarf.
- DODD, J.L. and A.L. HOOKER (1980): Previously undescribed pathotype of *Bipolaris zeicola* on Corn. Plant Dis. (Disease Notes) 74: 530.
- DOPUDA, M. AND J. LEVIĆ (2004): Sastav mikrobiote (Fusaria) semena pšenice na području Srema, Zb. radova str. 112, 5. Kong. z. bilja, Zlatibor, 22–26. novembar 2004.
- DUFFUS, J.E., B.W. FALK and G.R. JOHNSTONE (1990): In: P.A. Bumett (Ed.) World Perspectives on Barley Yellow Dwarf. CIMMYT, Mexico, 86–104.
- ESTEVEZ, M.C.F. (1989): Reações a *Exserohilum turcicum* (Pass.) Leonard&Suggs em milho (*Zea mays* L.) e variabilidade do patógeno. Master's Thesis, Universidade de São Paulo, Escola Superior de Agricultura "Luiz de Queiróz", Piracicaba.
- FALLAH MOGHADDAM, P. and J.K. PATAKY (1994): Reactions for isolates from mating of races 1 and 23N of *Exserohilum turcicum*. Plant Dis. 78: 767–771.
- FLOR, H.H. (1942): Inheritance of pathogenicity in *Melampsora lini*. Phytopathology 32: 653-669.
- GEVERS, H.O. (1975): A new major gene for resistance to *Helminthosporium turcicum* leaf blight of maize. Plant. Dis. Rep. 59: 296–300.
- GIANASI, L., H.A. DE CASTRO and H.P. DE SILVA (1996): Raças fisiológicas de *Exserohilum turcicum* identificadas em regiões produtoras de milho no Brasil, Safra 93/94. Summa Phytopathol. 22: 214–217.
- HALSETH, D.E., W.D. PARDEE and D.R. VIANDS (1991): Inheritance of resistance to *Helminthosporium carbonum* race 3 in maize. Crop Sci. 31: 612–617.
- HAMMOND, J. (1998): Serological Relationships Between the Cylindrical Inclusion Proteins of Potyviruses. Phytopathology 88: 965–971.
- HOOKE, A.L. (1961): A new type of resistance in corn to *Helminthosporium turcicum* in seedling corn. Plant Dis Rep. 45: 780–781.
- HOOKE, A.L. (1963a): Inheritance of chlorotic-lesion resistance to *Helminthosporium turcicum* in seedling corn. Phytopathology 53: 660–662.
- HOOKE, A.L. (1963b): Monogenic resistance of *Zea mays* L. to *Helminthosporium turcicum*. Crop Sci. 3: 381–383.
- HOOKE, A.L. (1975): *Helminthosporium turcicum* as a pathogen of corn. Reporter Tottori Mycology Institute 12: 115–125.
- HOOKE, A.L. (1977): A second major gene locus in corn for chlorotic-lesion resistance to *Helminthosporium turcicum*. Crop Sci. 17: 132–135.
- HOOKE, A.L. (1978): Additional sources of monogenic resistance in corn to *Helminthosporium turcicum*. Crop Sci. 17: 132–135.
- HOOKE, A.L. (1981): Resistance to *Helminthosporium turcicum* from *Tripsacum floridanum* incorporated into corn. Maize Genet Cooperation Newslett 55: 87–88.
- HOOKE, A.L., R.R. NELSON and H.M. HILU (1965): Avirulence of *Helminthosporium turcicum* on monogenic resistant corn. Phytopathology 55: 462–463.
- HOOKE, A.L. and J.M. PERKINS (1980): *Helminthosporium* leaf blight of corn - the state of the art. Annu. Corn. Sorghum Res. Conf. 34: 68–87.
- HSU, H.T., J. AEBIG, and W.F. ROCHOW (1984): Differences among monoclonal antibodies to barley yellow dwarf viruses. Phytopathology 74: 600–605.

- IVANOVIĆ, D., J. LEVIĆ, M. IVANOVIĆ and V. PENČIĆ (1990): Uloga virusa u etiologiji nastanka crvenoljubičaste pigmentacije biljaka kukuruza. VIII jugoslovenski simpozium o zaštiti bilja - za više hrane i bolju životnu sredinu. 3-6. dec., Opatija. Glasnik zaštite bilja 9-10: 310.
- IVANOVIĆ, D., J. LEVIĆ, PENČIĆ, V., and S. STOJKOV (1992): Proteinski kompleks novog patotipa *Bipolaris zeicola*. IX jugoslovenski simpozijum o zaštiti bilja, 1-5 decembra, Vrnjačka Banja.
- IVANOVIĆ, D., J. LEVIĆ, M. IVANOVIĆ and V. PENČIĆ (1992): Barley yellow dwarf virus in maize in Yugoslavia. *Maydica* 37: 287-292.
- IVANOVIĆ D. and M. IVANOVIĆ (1994): Disease response of maize to barley yellow dwarf and maize dwarf mosaic infection. CIMMYT, Barley Yellow Dwarf Newsletter, No. 5. Mexico, D. F.: CIMMYT, 21-22.
- IVANOVIĆ, D., R. OSLER, D. IGNJATOVIĆ and M. IVANOVIĆ (1995a): Polyclonal antibodies in detection of Potyviruses as maize pathogens. *Maydica* 40: 173-177.
- IVANOVIĆ, D., R. OSLER, N. KATIS, M. IVANOVIĆ and D. IGNJATOVIĆ (1995b): Principal maize viruses in Mediterranean countries. *Agronomie* 15: 443-446.
- JORDAN, E.G., J.M. PERKINS, R.A. SCHALL and W.L. PEDERSEN (1983): Occurrence of race 2 of *E. turcicum* on corn in the Central and Eastern United States. *Plant Dis.* 67: 1163-1165.
- JOSIFOVIĆ, M. (1926): Bolesti gajenih biljaka u Vojvodini i severnom delu Srbije u 1925. g. Glasnik Ministarstva Poljoprivrede i Voda 13: 72-89.
- JOSIFOVIĆ, M. and MIRIĆ (1960): *Helminthosporium carbonum* Ullstr. nov parazit kukuruza u Jugoslaviji. *Zaštita bilja* 59: 3-9.
- KLEIN, K.K. and J.C. CORRELL (2001): Vegetative compatibility group diversity in *Fusarium*, Chapter 6, 83-96, in: Summerell, B.A., Leslie, J.F., Backhouse, D., Bryden, W.L., Burgess, L.W. (ed.), *Fusarium* - Paul E. Nelson Memorial Symposium, AS Press, The American Phytopathological Society, St. Paul, Minnesota, 392.
- KRNJAJA, V., J. LEVIĆ, S. STANKOVIĆ and Z. TOMIĆ (2006): Pathogenicity and vegetative compatibility groups of *Fusarium verticillioides*. Pp. 70. Book of Abstracts, European Fusarium Seminar, Wageningen, The Netherlands, 19-22 September, 2006.
- LEONARD, K.J., Y. LEVY and D.R. SMITH (1989): Proposed nomenclature for pathogenic races of *Exserohilum turcicum* on corn. *Plant Dis.* 73: 776-777.
- LESLIE, J.F. and K.K. KLEIN (1996): Female fertility and mating type effects on effective population size and evolution in filamentous fungi. *Genetics* 144: 557-567.
- LEVIĆ, J., D. IVANOVIĆ and S. STANKOVIĆ (2003): Paraziti kukuruza, sirka i prosa koji se prenose semenom. *B. lekar* 6: 570-577.
- LEVIĆ, J. and V. PENČIĆ (1990): Utilization of carbon, nitrogen and sulfur compounds by *Kabatiella zae* Narita et Hiratsuka. *J. Phytopathology* 44 (128): 321-332.
- LEVIĆ, J. and V. PENČIĆ (1992): Response of maize inbred B73 to a new pathotype of *Bipolaris zeicola* (Stout) Shoemaker. *Maydica* 37: 355-361.
- LEVIĆ, J. and V. PENČIĆ (1993): Morphology of a new pathotype of *Bipolaris zeicola* (Stout) Shoemaker. *J. Phytopathology* 139: 339-346.
- LEVIĆ, J., L. TAMBURIĆ-ILINČIĆ and T. PETROVIĆ (1997): Maize kernel infection by *Fusarium* species in the period 1994-1996. *Cereal Res. Commun.* 25 (3): 773-775.
- LEVIĆ, J. and T. PETROVIĆ (1998): Formation of α - and β -conidia by *Phaeocystostroma ambiguum*. *Mycopathologia* 140: 149-155.

- LEVIĆ, J. and T. PETROVIĆ (1999): Pojava rase R1 *Exserohilum turcicum* (Pass) Leonard & Suggs patogena kukuruza. Program-Izvodi-Spisak učesnika Drugog kongresa genetičara Srbije, Soko Banja, 10–13. novembar, str. 224.
- LEVIĆ, J., T. KOVAČEVIĆ, S. STANKOVIĆ, D. IVANOVIĆ and J. BERENJI (2006): The first report on *Fusarium thapsinum* on sorghum grains in Serbia. 52. Book of Abstracts, European Fusarium Seminar, Wageningen, The Netherlands, 19–22 September, 2006.
- LIPPS, P.E. and R.E. HITE (1982): *Exserohilum turcicum* virulence on corn with the Ht resistance gene in Ohio. *Plant Dis.* 66: 397–398.
- MUNKVOLD, G. P. and DESJARDINS A.E. (1997): Fumonisin in Maize. *Plant Dis* 81: 556–565.
- OLDFIELD, M.L. (1989): The Value of Conserving Genetic Resources. Sinauer Associates, Inc. Publishers Sunderland, Massachusetts, 376.
- OSLER, R., N. LOI, C. LORENZONI, M. SNIDARO and E. REFATTI (1985): Barley yellow dwarf virus infections in maize (*Zea mays* L), inbreds and hybrids in northern Italy. *Maydica* 30: 285–299.
- PALAVERŠIĆ B., J. BREKALO and J. SEVER (1995): Genetic Control of Major Maize Disease. SVIBOR – Collecting Data on Project in Croatia, Project code 4–01–039.
- PENČIĆ, V., A.L. HOOKER and J. LEVIĆ (1981): Reaction of American maize germ plasm and gene stock to leaf disease pathogens in Yugoslavia. *Genetika* 13 (19): 59–68.
- PENČIĆ, V. and J. LEVIĆ (1979): Prilog proučavanju otpornosti kukuruza prema gljivi *Kabatiella zaeae* Narita et Hiratsuka i simptomi oboljenja. *Arh. poljopr. nauke* 118: 115–120.
- PIEZARKA, D.J. (1980): A new race of *Helminthosporium turcicum* and reaction of sweet corn hybrids to the pathogen. *Proc. Florida State Horticult. Soc.* 93: 281–283.
- ROCHOW, W.F. (1979): Field variants of barley yellow dwarf virus: Detection and fluctuation during twenty years. *Phytopathology* 69: 655–660.
- ROZENFELD, J. (1980): Rezultati ispitivanja otpornosti kukuruza prema *Helminthosporium carbonum* Ullstrup. *Zaštita bilja* 31 (4) 154: 347–356.
- ROZMAN L., B. PALVERŠIĆ, N. VALIČ, F. CELAR and L. MILEVOJ (2003): Investigation of different races of *Exserohilum turcicum* on maize in Slovenia. Conference Prigramme, 6th Slovenian Conference on Plant Protection with International Participation, Zreče, Slovenia, March 4–6, 2003.
- SHURTLEFF, M.C. (1980) *Helminthosporium* leaf spot (blight) and Northern leaf spot. In: Compendium of corn diseases, pp. 7–18. APS Press, St Paul, Minnesota, USA.
- SMITH, D.R. and J.G. KINSEY (1980): Further physiological specialization in *Helminthosporium turcicum*. *Plant Dis.* 64: 779–781.
- STOJKOV, S., J. LEVIĆ, D. IGNJATOVIĆ, D. IVANOVIĆ and V. PENČIĆ (1996): Vrste roda *Periconia* paraziti korena i stabla kukuruza. *Zaštita bilja* 47(3), 217: 229–240.
- STOJKOV, S. and D. IGNJATOVIĆ (1998): Variability of fungus *Microdochium bolleyi* isolate - maize root and stalk pathogen. 7th International congress of Plant Pathology, Edinburgh, 9–16. Avgust
- THAKUR, R.P., K.J. LEONARD and R.K. JONES (1989): Characterization of a new race of *Exserohilum turcicum* virulent on corn with resistance gene *HtN*. *Plant Dis.* 73: 151–155.
- TOŠIĆ, M., R.E. FORD, D.D. SHUKLA and J. JILKA (1990): Differentiation of sugarcane, maize dwarf, johnsongrass and sorghum mosaic viruses based on reactions of oat and some sorghum cultivars. *Plant Dis.* 74: 549–552.
- TURNER, M. T. and E. R. JOHNSON (1980): Race of *Helminthosporium* not controlled by *Ht* genetic resistance in corn in the American corn belt. *Plant Dis.* 64: 216–217.

- VAN DER PLANK, J.E., (1975): Principles of plant infection. New York, APS
- WATERHOUSE, P.M., W.R. GERLACH, and W.A. MILLER (1986): Serotype-specific and General Luteovirus Probes from Cloned cDNA Sequences of Barley Yellow Dwarf Virus. J. Gen. Virol. 67: 1273–1281.
- WELZ, G. and K.J. LEONARD (1988): Genetic variation in field population of races 0, 2 and 3 of *Bipolaris zeicola* in 1987. (Abstr.). Phytopathology 78: 1574.
- WELZ H.G., R.G. WAGNER, H.H. GEIGER (1993): Virulence variation in *Setosphaeria turcica* populations collected from maize in China, Mexico, Uganda, and Zambia. Phytopathology 83: 1356.
- WINDES, J.M. and W.L. PEDERSEN (1991): An isolate of *Exserohilum turcicum* virulent on maize inbreds with resistance gene *Htn*. Plant Dis. 75: 430.
- ZITTE, T. (2001): Virus problems of sweet corn. www.vegetablemendonline.ppath.cornell.edu pp. 1–6.

GENETIČKA VARIJABILNOST PATOGENA KUKURUZA U SRBIJI

Slavica STANKOVIĆ, Jelena LEVIĆ i Dragica IVANOVIĆ

Institut za kukuruz „Zemun Polje“, Beograd-Zemun, Srbija

I z v o d

U Srbiji je u poslednjih pedeset godina istraživanja identifikovana varijabilnost nekih patogena kukuruza, i to najčešće na osnovu njihovih odgajivačkih odlika i osetljivosti test genotipova, a samo u nekim slučajevima na osnovu određivanja mating tipova, vegetativne kompatibilnosti ili biohemijskih metoda. Suprotno tome, u svetu se sve više primenjuju metode kojima se utvrđuje varijabilnost unutar jedne populacije na molekularnom nivou. Najveća varijabilnost utvrđena je kod patogena lista kukuruza - *Exserohilum turcicum* (Pass.) Leonard & Suggs (2 rase) i *Bipolaris zeicola* (Stout) Shoemaker (2 rase), mada je ona značajno manja u odnosu na varijabilnost istih patogena u svetu. Rezultati istraživanja u cilju određivanja mating tipova i vegetativne kompatibilnosti vrsta roda *Fusarium*, patogena korena i stabla kukuruza, ukazali su na njihovu veliku varijabilnost u Srbiji. S obzirom na svojstvo patogena da se lako i brzo prilagodjavaju novim genotipovima, agroekološkim uslovima i agrotehničkim merama, neophodno je stalno praćenje divergentnosti i epidemiologije parazita, da bi se izbegle veće posledice po prinos i kvalitet prinosa kukuruza.

Primljeno 22. VII 2007.
Odobreno 17. VIII 2007.