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SENSITIVITY OF TOKAY VINE VARIETIES TO *PLASMOPARA VITICOLA* (BERK&CURT.) BERK.&DE TONI

CITLIVOSŤ TOKAJSKÝCH ODRÔD VINIČA NA *PLASMOPARA VITICOLA* (BERK&CURT.) BERK.&DE TONI

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For centuries, the vine was produced in the Tokay region. Acreages and vine varieties convenient to growing (cv. Lipovina, cv. Yellow Muscat, cv. Furmint) are strictly concluded by the legislation. Due to the monocultures and low intervarietal diversity, better conditions for more aggressive manifestation of pathogens have been formed. During 2005–2008 there were observed the occurrences of grapevine downy mildew *Plasmopara viticola* (Berk&Curt.) Berk.&DeToni in listed varieties and in selected experimental sites with different altitude. At the same time the influence of microclimatic factors and bush structure on extension and occurrence of grapevine downy mildew was observed. According to our experimental observations and results the most vulnerable time period in connection with grapevine downy mildew infection is the BBCH 68 phenophase. Even though several handbooks describe the etiology of the disease outbreak and course they do not describe some possible changes of pathogenic fungus in relation to host plant or environment. Based on our research results an integrated vine protection was proposed.

Key words: monoculture, pathogen, Grapevine downy mildew, monitoring, integrated pest management

The Tokay region belongs to historically and economically important viticultural areas of Slovakia. Historians assume that the viticulture has existed in this area from the 3rd century A.D. (Žadanský, 2002). Specific feature of closed Tokay area is the soil, its geological basement, soil skeleton content and the soil type (Kolárik, 2004). Soils are formed on the Secondary andesite and rhyolite tuffs mixed up with soil skeleton of volcanic origin generating favourable thermal regime and microclimate for wine grape growing. Distinctive, naturally sweet, high quality wines are produced in this region. The Tokay region (908.77 hectares), area limits and allowable vine varieties (cv. Lipovina, cv. Yellow Muscat, cv. Furmint) are defined and characterised according to the Act No. 182/2005. Populations of Tokay vine varieties are endangered by genetic erosion according to Brindza (2002). Strict conditions of long-term monoculture wine grape growing and low intra-species diversity generates conditions for more aggressive manifestation of pathogens and more frequent occurrence of epidemics (Eftimová, 2006; Eftimová and Bacigálová, 2008). Grapevine downy mildew *Plasmopara viticola* (Berk&Curt.) Berk.&De was spreaded to Europe from the North America in the 19th century and caused catastrophic losses on vine yield. As the history of wine grape growing in Tokay region Žadanský (2002) indicates, the grapevine downy mildew and phylloxera epidemic occurred in the Zemplin region in 1880yet. In that period 9 078 hectares (i.e. 87 %) from totally 10 286 were devastated. Even today this disease belongs to the most dangerous in the years favourable for fungus development. In 2004–2008 within semi-production trials in the Tokay region the variety sensibility to grapevine downy mildew infection depending on different altitudes and plot location were tested and the results were compared to those of Pospišilová (1981). An anatomic/morphologic characteristic of fungus was elaborated. Part of the research was also the evaluation of microclimatic factors and bush structure impact on fungus infection.

Material and method

Stand description – the Tokay wine production area is located on south-east to south-west downhills of Zemplin Highlands in the cadastres of Malá Tŕňa, Veľká Tŕňa, Bara, Černochov, Čerhov, Viničky, and Slovenské Nové Mesto.

Climatic conditions – continental weather conditions are typical for this region. Atmospheric precipitations are distributed unevenly. Dry and warm autumn is of big importance because of cibéby-raisins formation.

Soil conditions – soils are situated on Secondary andesite and rhyolite tuffs. Disintegration of volcanic rocks ends in heavy clay soils rich in potassium. Concerning soil types most typical for this region are brown soils.

Varieties – Furmint 65–75 %, Lipovina 15–20 % and Yellow Muscat up to 10 %.

Agro-technology – Middle Rhein-Hessen training system with 2.20 x 1 m spacing in older vineyards, in younger ones with 2.4 x 0.85 m spacing and horizontal cordon training system. Grass covered interrows.

Monitoring and diagnosis of grapevine downy mildew:

We have monitored the occurrence of this disease during the vegetation period in localities of Malá Tŕňa, Čelejka – Káty, Viničky – škola, and Makovisko – Ostrožovič. In order to make the diagnosis, 100 leaves and 50 bunches of grapes were taken from each locality on 12. 9. Microscopic preparations were prepared from each sample in order to make the microphotography and determine the kind of fungus, and biometrical data. 50 % milk acid or lactophenol with aniline blue was used to prepare them. Microscopic observations were made using the Zeiss Amplival optical microscope coupled with microphotography equipment. The kind of single fungus was determined according to Fungi

Identification Handbooks, Guides and monographs. Herbal material is stored in the Mycological Herbarium of the Institute of Botany, Slovak Academy of Sciences (SAS). The infestation degree in leaves and bunch of grapes were evaluated using the Townsend and Heuberger formula (1943).

Results and discussion

The grapevine downy mildew – *Plasmopara viticola* (Berk. et Curt.) Berk. et de Toni belongs to the most serious diseases in the closed Tokay wine production region. Under favourable climatic conditions for pathogen (temperature, humidity) and under deficient vine protection conditions the fungus infect young shoots, leaves, and grape berries and cause huge damages on yield amount and affects the quality of Tokay wine significantly. The harmfulness of this disease lies in the reduction of assimilation area and vegetation potential of shrubs.

Grapevine downy mildew – *Plasmopara viticola* (Berk. et Curt.) Berk. et de Toni – *Plasmopara viticola,* Syn. *Botrytis viticola* Berk. et Curt.; *Grapevine downy mildew* (Berk.et Curt.) Casp. Oomycota

Plasmopara viticola belongs to cosmopolitan species and occurs in representatives of *Vitis* L. and *Partenocissus* Planch. genus. It outlast the winter period in the form of oospora spores in infected fall off leaves. From graafian oospores germinates the carrier of sporangia during the spring period, and macrosporangium with differenced zoospores is formatted. The process of oospores germination starts when there are 10 mm precipitations within 24 hours, the average temperature reaches 12 °C, and the minimal one does not drop under 8 °C. Primary infection arise out of vine leaves by zoospores penetration through pores when leaves are wet and the temperature range from 22 to 25 °C. Pathogen creates an intercellular mycelium which produces sporangiophores on bottom of leaves or on the top of bunch of grapes. Zoospores release from zoosporangium and cause the secondary infection. The incubation period of fungus depends on the temperature. The darkness is essential for fungus sporulation. The grapevine downy mildew lifespan repeats 8–10 times during the summer period depending on favourable conditions (temperature and moisture). Blossoms and young berries are most predisposed for infection.

In 2004–2008 the pathogenity differences in *Plasmopara viticola* (Berk & Curt.) Berk. & De Toni. depending on variety, locality, and stand architecture were studied in the Tokay region. Within the experimental period the influence of weather on BBCH phenophases in vine (Lorenz et al., 1994) were studied and compared to the results of Pospíšilová (Pospišilová, 1981) (Tab.1). We find out that the vine starts phenophases with only slight differences except for the 2007 growing season, when the vegetation period shifted about 14 days due to the warmer weather conditions.

In order to diagnose the pathogen we sampled leaves and berries from the control variant and treated varieties cv. Furmint, cv. Yellow Muscat and cv. Lipovina in locality of Malá Tŕna – Pahorok, Čelejka – Káty, Viničky – škola and Ostrožovič – Makovisko during and at the end of the vegetation period. The occurrence of different size yellow and brown oil spots underneath with white floccose fur of fungus was determined on vine leaves (Fig. 1).

The fungus mycelium expanded to interstitium of leaves tissue and formatted spherical haustoria which permeate into living cells of leaves tissue. Sporangiophores growing out in bunches through stomata on thelower side of leaves are differenced on mycelium (Fig. 1B, C). Each sporangiophora furcates 2–6 times primary and 2–3 times secondarily. Side springs angle from sporangiophorum on the square and on short secondary shoots egg-shaped or elliptic sporangiophora of 14–34 x 9–19 µm size are formatted (Fig. 1C).

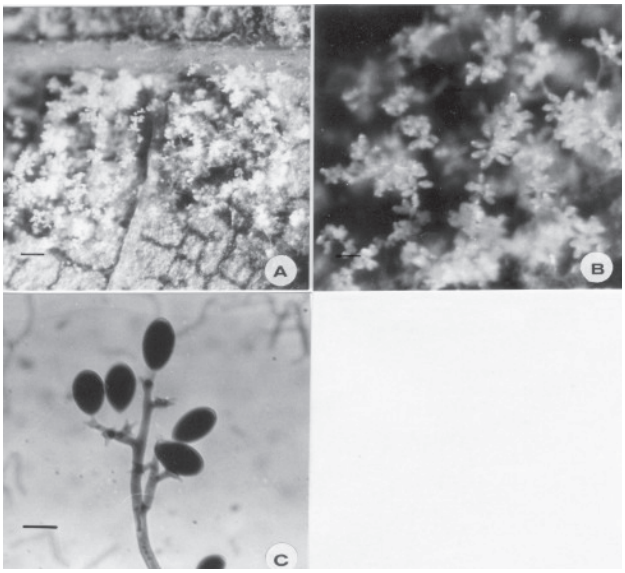

Figure 1 Microphotography of *Plasmopara viticola* (Berk&Curt.) Berk.&De Toni
Obrázok 1 Mikrofotografické zábery *Plasmopara viticola* (Berk&Curt.) Berk.&De Toni

Our evaluation of variety sensitivity to the infection of grapevine downy mildew issued from the last research made by Pospíšilová (1981). This author characterized the cv. Furmit as slightly resistant, cv. Lipovina as very sensitive and the Yellow Muscat as medium resistant to the grapevine downy mildew. The sensitivity of varieties against the disease infection was compared to the results published by Pospišílová (Pospíšilová, 1981). The infection intensity was evaluated using the Townsend and Heuberger formula (1943) and published in the Table 2.

Resulting from the Table (2) in 2005 strong infection by grapevine downy mildew (92–97 %) in all control variants and in all varieties was stated. In 2006 this was 80–87 %. The weakest infection (61–67 %) in the control variant was stated in 2007, when the weather conditions were not convenient for disease development. On the contrary – in 2008 – the varieties were infected only to 76–92 %. On June 26, 2008 strong hail damage in BBCH 69 growing phase was recorded, this was the cause of convenient conditions not only for grapevine downy mildew infection but also for another considerable vine disease – botrytis bunch rot. In some closed hermetic localities of Černochov the wine grape was not even harvested because of its poor quality. According to the Table (2) the most sensitive reaction to grapevine downy mildew infection was observed in cv. Lipovina, which corresponds to the conclusions of Pospíšilová (1981). The leaves of cv. Lipovina compared to those of cv. Furmint and Yellow Muscat are subtle and thin, which makes it easier for initial fungus fibre to penetrate through the leaf surface. Medium sensitive reaction to grapevine downy mildew infection was observed in cv. Furmint and the lowest one in cv. Yellow Muscat. According to our observations the most vulnerable period regarding grapevine downy mildew infection is the BBCH 68, when 80 % of blossoms are passed.

Table 1 Grapevine BBCH phenological scale – Furmint, Lipovina, Yellow Muscat varieties, Pahorok Malá Tŕňa, 2005–2008

BBCH (1)	Start (3) BBCH 2005	Start (3) BBCH 2006	Start (3) BBCH 2007	Start (3) BBCH 2008	Start (3) BBCH Pospíšilová	Varieties (4)
01	23. 3.05	10. 4.06	11. 3. 07	31. 3. 08		
08	18. 4. 05	20. 4. 06	5. 4. 07	15. 4. 08		
13–15	30.5. 05	20. 5. 06	4. 5. 07	12. 5. 08		
60	15. 6. 05 17. 6. 05 17. 6. 05	17. 6. 06 18. 6. 06 18. 6. 06	28. 5. 07 30. 5. 07 30. 5. 07	9. 6. 08 11. 6. 08 11. 6. 08		Furmint Lipovina, Yellow Muscat
65	14. 6. 05 20. 6. 05 22. 6. 05 22. 6. 06 30. 6. 05	14. 6. 06 21. 6. 06 22. 6. 06 22. 6. 06	30. 5. 07 2. 6. 07 3. 6. 07 3. 6. 07	6. 6. 08 11. 6. 08 13. 6. 08 13. 6. 08	 27. 5–10. 6 15. 6. 9. 6.	Rulanské biele Furmint Lipovina Yellow Muscat Lipovina – Čelejka
69	25. 6. 05	25. 6. 06	6. 6. 07	16. 6. 08		
75	11. 7. 05	10. 7. 06	25. 6. 07	16. 7. 08		
79	21. 7. 05 23. 7. 05 23. 7. 05	25. 7. 06 27. 7. 06 27. 7. 06	9. 7. 07	2. 8. 08 5. 8. 08 7. 8. 08		Yellow Muscat Furmint, Lipovina
85	23. 8. 05 25. 8. 05 25. 8. 05	15. 8. 06 17. 8. 06 17. 8. 06	20. 8. 07	15. 8. 08 18. 8. 08 20. 8. 08		Yellow Muscat Furmint, Lipovina
89 Harvest of raisins (2)	17. 10. 05 24. 10. 05	10. 10. 06 16. 10. 06 23. 10. 06	1. 10. 07 16. 10. 07	13. 10. 08 –		Yellow Muscat Furmint, Lipovina

BBCH (1) Vegetative stage, 01 – Beginning of bud swelling: buds begin to expand inside the bud scales, 08 – Bud burst: green shoot tips clearly visible, 13–15 – Shouts 0.4–0.5 m, 60 – Before flowering, 65 – Flowering, 69 – At the end of flowering, 75 – Grapes of peas size, 79 – Majority of berries touching, 85 – Softening of berries, 89 – Berries ripe for harvest.

Tabuľka 1 Fenofázy viniča BBCH – Furmint, Lipovina, Yellow Muscat, Pahorok Malá Tŕňa, 2005–2008
(1) vegetačné štádium, (2) zber cibéb (3) nástup BBCH, (4) odrody

In the course of our trials a strong occurrence of grapevine downy mildew was registered in 2005, 2006, and 2008 (Fig. 2.) Our results were confirmed by observations of Ackrmann (2000) that the intensity of grapevine downy mildew infection depends on the varietal predisposition to the disease, the quantity of infectious resource, and on favourable meteorological conditions. The meteorological characteristics of the Tokay area during 2005–2008 was documented in pictures (temperature and precipitation, temperature and leaves wetness) and in the Walter's climatographs 1, 2, 3 and 4.

As for the grapevine downy mildew epidemiology, the infection period is very important, which means appropriate conditions for sporulation, germination and infection. The vineyard location and the stand density are a crucial precondition for the disease development according to our results from 2005–2008. On localities with insufficient "green works" the infestation degree was higher. Bottom leaves (close to the soil surface) were infected at first. Wet leaves maintain longer in the stand, which creates suitable conditions for pathogen development.

Furthermore, we found out and confirmed the conclusions of Dula and Szendrey (2002) that the temperature exerts influence up the time period when the first symptoms of infection occur on leaves and bunch of grapes (Tab. 3). We can confirm that temperatures higher than 16 °C differently influence the first occurrence of symptoms. On the leaves they occur in one week, but on the bunch of grapes 7–10 days later.

This fact is very important for the decisions of further chemical vine protection. False satisfaction that there are not any infections on the bunch of grapes can lead to yield devaluation.

In 2005 the first symptoms after infection by grapevine downy mildew were detected on the leaves of cv. Lipovina and Furmint (Pahorok, control variant) in June, 21. In 2006 the first symptoms of the grapevine downy mildew infection on leaves were detected in cv. Lipovina and Furmint in June, 12. In 2007 the first disease symptoms on leaves were detected in cv. Lipovina on 8th June, which confirms again that cv. Lipovina is very sensitive. In 2008 the first disease symptoms (June, 19) were detected in cv. Lipovina, again. During 2005, 2006, and 2008 we detected infected cv. Lipovina and cv. Yellow Muscat on protected localities in the same date as on the hermetically

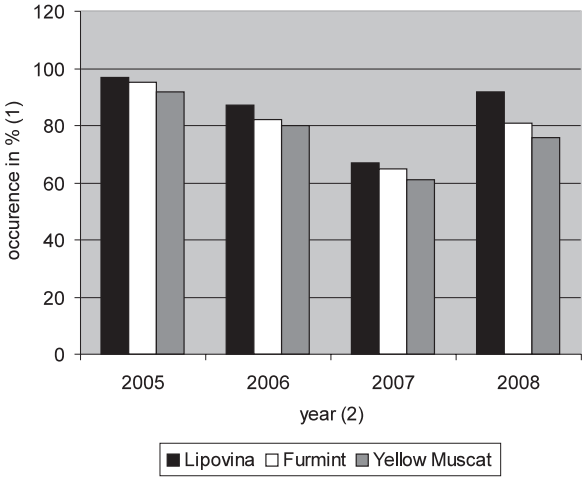

Figure 2 The percentage of *Plasmopara viticola* infection in vine leaves
Obrázok 2 Percento napadnutia listov viniča *Plasmopara viticola*
(1) % napadnutia, (2) rok

Table 2 The percentage of *Plasmopara viticola* infection on vine leaves in Pahorok, Malá Tŕňa

Phytopathogenic fungus (1)	Grapevine cultivars (2)	Occurrence in % (3)			
		2005	2006	2007	2008
Plasmopara viticola	Lipovina	97	87	67	92
	Furmint	95	82	65	81
	Yellow Muscat	92	80	61	76

Tabuľka 2 Percento napadnutia *Plasmopara viticola* na listoch viniča, Pahorok Malá Tŕňa
(1) hubové ochorenie,(2) odrody, (3) % napadnutia

Table 3 The impact of temperature on the occurrence of first symptoms in leaves and bunch of grapes

Average temperature in °C (1)	Incubation period in days) on leaves (2)	Incubation period in days on bunches of grapes (3)
10–13	15–18	15–18
14	12–15	12–15
15	11–13	11–13
16	9–11	9–11
17	6–7	11–13
18–19	5–6	13–15
20–25	4–5	14–18

Tabuľka 3 Vplyv teploty na objavenie sa prvých príznakov na listoch a strapcoch
(1) priemerná teplota, (2) dĺžka inkubácie (dni) na listoch,(3) dĺžka inkubácie (dni) na strapcoch

closed localities of the control variants in Čelejka – Káty, Viničky – škola, and Ostrožovič – Makovisko. On the contrary, in cv. Furmint on the open locality of Čelejka – Káty there was not find any grapevine downy mildew infection. Our observations confirm the fact that the locality location participate significantly on the fungal diseases propagation.

According to the results of our semi-production trials in 2005–2008 we came to conclusions that essential for successful protection measures against grapevine downy mildew is an in-depth knowledge of pathogen bionomics, observation of its occurrence, and propagation and consideration about varietal predispositions. Without this knowledge it is impossible to determinate reliably the date when the infection starts and this of antifungal measurements. As to the prevention measurements which have an influence on the infection dissemination there are: proper cut, green works, interrow maintenance, and reduction of infectious resources. Within the evaluated period we proposed an integrated vine protection system, which contributes to the protection of natural and environmental conditions of the Tokay region. Besides prevention measures, a part of the integrated protection system is a direct protection based on short-term prognosis and knowledge on the pesticide impact mechanism, which can protect the yield of damages. In 2005–2008 we applied the short-term prognosis method according to Istvánffi – Pálinkás (1913) and Lehoczky and Reichart, 1968 based on the weather course and the fungus biology. Depending on active substances in chemicals the date of treatment was stated so that the protection was finished till the end of the incubation period.

Súhrn

V Tokajskej oblasti sa vinič pestuje už niekoľko storočí. Legislatívou sú presne vymedzené hony a determinované odrody viniča (cv. Lipovina, cv. Muškát žltý, cv. Furmint), ktoré sa môžu pestovať. V dôsledku monokultúrneho pestovania a malej vnútrodruhovej diverzity, vznikajú väčšie predpoklady pre agresívnejšie správanie sa patogénov. V rokoch 2005–2008 sme sledovali výskyt peronospóry viničovej – *Plasmopara viticola* (Berk&Curt.) Berk.&De Toni, pri uvedených odrodách, vybraných experimentálnych lokalitách s rôznou nadmorskou výškou. Súčasne sme sledovali vplyv mikroklimatických faktorov a štruktúry krov na šírenie a výskyt plesne viničovej. Z našich experimentálnych pozorovaní a výsledkov vyplynulo, že najzraniteľnejším obdobím viniča pre napadnutie peronospórou viničovou je obdobie fenofázy BBCH 68. Aj napriek tomu, že viaceré príručky opisujú etiológiu vzniku a priebeh choroby nezachytávajú niektoré možné zmeny patogénnej huby vo vzťahu k hostiteľovi alebo k prostrediu. Na základe nášho výskumu sme navrhli integrovanú ochranu viniča.

Kľučové slová: monokultúra, patogén, perenospóra viničová, monitoring, integrovaná ochrana

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