# The role of cultivar susceptibility and vineyard age in GTD: examples from the Carpathian Basin

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#### SUMMARY

Grapevine trunk diseases (GTDs) are among the most severe problems in viticulture worldwide. The exact etiology and the role of endophytic microorganisms is not known yet and there is no adequate protection or curative treatment against the disease. Hungarian wine regions are also affected by the disease, and there is restricted information about the rate of infection nation-wide and about the susceptibility of the Hungarian cultivars.

The main objectives of our research are to measure the symptom expression and the damage caused by GTDs, to understand the epidemiology and etiology of the disease to establish a foundation of a proper disease management.

Cultivar susceptibility groups were created with the aim to allocate some Hungarian cultivars and the role of vineyard age was also examined in symptom expression.

Keywords: etiology, Botryosphaeria stevensii, esca, grapevine trunk diseases, symptom expression

#### **INTRODUCTION**

Nowadays, one of the most important problems in viticulture is the grapevine trunk diseases (hereinafter referred as GTDs), as there are no effective methods against them. The disease can cause necrosis either in a segment of shoots and arms or the whole trunk. Although annual percentage of decaying vines is low, economic aging of the vineyard (the point from when the vineyard is not profitable) will accelerate, because of the consecutive plant (Varga, 2009). 1.5 billion USD annual deficit of the sector was estimated by Hofstetter et al. (2012) worldwide, resulting from costs of replacing dead vines. This is a severe problem all across the European Union. GTD disease incidence was reported between 8% to 19% in Italy at 2014, and circa 10% in Spain (Fontaine et al., 2016). In France 13% of the vineyards are unproductive, resulting 1 billion EUR loss. The Hungarian wine regions are similarly affected. The ratio of the symptomatic plants was already over 12% in certain vineyards in 2007 (Dula, 2011). Between 2013 and 2015 disease incidence ranged from 0.17% to 42.11% at different vineyards in the Tokaj Wine Region, in Northeast-Hungary and presumably all Hungarian Wine Regions are affected with GTDs (Kovács et al., 2017).

The most important diseases globally among the GTDs are the 'sensu stricto' esca, eutypa dieback and botryosphaeria dieback, which are complex problems caused by different pathogens and environmental factors as well (Bertsch et al., 2012; Kovács and Sándor, 2016). According to Larignon and Dubos' (1997), the isolated pathogens can be different based on the part of the cross- section of the trunk or the cordon they originate from. *Phaeoacremonium aleophilum* and *Phaemoniella chlamydospora, Phellinus punctatus, Stereum hirsutum* were identified as the main causative fungal pathogens of esca, but *Eutypa lata* was also

found in the brown decaying tissues. In Hungary, the most common GTD pathogen was the *Diplodia seriata* (Csótó et al., 2016; Kovács et al., 2014).

The uncertainty of pathogenicity and the role of several influencing environmental factors makes our image about GTDs even more abstract. Graniti et al. (2000) mentioned, that the pathogens could be present in the vineyards as part of the biocoenosis, without causing any symptoms. Extreme climatic conditions are decreasing the vigor of the vines, therefore the pathogens can proliferate in the woody tissue significantly. The survey of the disease can be difficult, because the infected vines does not show symptoms each year (Lecomte et al., 2011; Kovács et al., 2017).

The threat of early decline of grapevines has been documented since the antique times. The Latin word esca means tinder, comes from the usage of the spongy, dry, decayed wood tissue (Mugnai et al., 1999). Kövics (2009) originates the name from the greek word 'eskhatos', which means the last (stage of disease). At first, an anonymous author (1895) documented the symptom complex called "sunstroke" in California. Ravaz (1898) mentions the symptoms as "folletage" (cited in: Chiarappa, 2000).

Most GTDs can occur both in acute and chronic form. The acute symptoms result the rapid decaying of the arms or another woody parts of the vines. When the infected, decaying tissues are in the lower parts of the trunk, usually the whole plant dies suddenly. This symptom is called "apoplexy". Just before this stage, the shoots show significant alterations in photosynthetic activity (Letousey et al., 2010.) The tissues may show different changes in color and consistence depending on the pathogens present in the dissection of the apoplectic trunk (Larignon and Dubos, 1997). In case of Eutypa or Botryosphaeria dieback, although the woody tissues are brown and necrotic, but they remain intact (Moller et al., 1974; Hluchy et al.,

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2007). Besides them, basidiomycetous fungi of esca complex cause white spongy rot in the inner tissues (Fischer, 2006) (*Figure 1*).

### Figure 1: Necrosis and white rot on the cross-section of a decaying trunk (Photo: András Csótó)



The foliar symptoms are very specific. The toxins produced by the pathogens in the trunk translocate to the leaves. This will cause color gradient interveinal striping, vernacularly the "tiger strip" (Haviland et al., 2019) (*Figure 2*).

The symptoms mentioned above mostly occur in older vineyards (Gubler et al., 2005). Grapevine trunk diseases can potentially affect young vines and freshly planted vineyards too, when it is called "black foot" or Petri-disease. Typical symptoms are stunted growth, weak sprouting or no sprouting at all, leaf chlorosis and marginal necrosis, dieback and decaying of the whole plant. These symptoms are commonly tied to the *Phaeomoniella* and *Phaeoacremonium* genera, likewise the esca complex (Gramaje and Armengol, 2011; Sidoti et al., 2000).

Figure 2: Foliar symptoms on red grape cultivar (Photo: András Csótó)



In the summer of 2019 a survey was carried out in the Carpathian-basin (*Figure 3*), in 7 wine regions involving nearly 20000 vines. Our aims were to get data about the susceptibility of cultivars and about the occurrence of the symptoms in the vineyards with different age.



Figure 3: Locations of the surveyed wine regions (Google Earth 2019)



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#### MATERIALS AND METHODS

The symptom expression of grapevine trunk diseases was measured in Hungary and certain bordering areas in the summer and early autumn of 2019. The survey was carried out in eight vineyards of five wine regions involving seventeen grapevine cultivars, of which ten were white and seven were red ones (*Table 1*).

Table 1

The surveyed vineyards and their cultivars. Cultivars sorted by decreasing number of studied parcels belonging to the given cultivar  $\mathbf{R} = \mathbf{red}$  cultivars,  $\mathbf{W} = \mathbf{white}$  cultivars

	Wine region Vineyard/ parcel	Érmellék (Romania)		Tokaj	Badacsony	Szekszárd	Villány		
		Heit Birtok	Heit Papok földje	Szarvas	VWRI*	Lajvér Borbirtok	Bocor	Göntér	Zuhánya
	Cabernet Franc <sup>R</sup>				+	+	+		+
	Pinot Noir <sup>R</sup>				+	+	+	+	
	Cabernet Sauvignon <sup>R</sup>				+	+	+		
	Olaszrizling <sup>w</sup>				+	+	+		
	Kékfrankos <sup>R</sup>					+	+		+
	Királyleányka <sup>w</sup>	+			+				
	Bakator <sup>w</sup>	+	+						
Cultivar	Sauvignon Blanc <sup>W</sup>					+		+	
Cult	Merlot <sup>R</sup>					+		+	
Ŭ	Chardonnay <sup>w</sup>					+			
	Cserszegi fűszeres <sup>w</sup>					+			
	Kadarka <sup>R</sup>		+						
	Portugieser R							+	
	Juhfark <sup>W</sup>				+				
	Pinot Gris <sup>w</sup>				+				
	Kéknyelű <sup>w</sup>				+				
	Hárslevelű <sup>w</sup>			+					

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The surveyed areas were chosen to be representative for the climate and regional characteristics of the country. Tokaj and Érmellék Wine Regions belong to the continental precipitation regime, while the precipitation regime of Badacsony, Szekszárd and Villány Wine Regions are usually similar to the Mediterranean type (Borhidi, 1981). Despite these differences, the COUNCIL REGULATION (EC) No 479/2008 handles the whole area of Hungary in the C1 wine-growing zone. The studied vineyards were trained differently: mid-high single cordon was the most widespread, but the double Guyot and head training methods were represented too. The directional exposures, row directions of the vineyards were different as well as the inclination of the slopes.

In order to characterize the GTD infection in different vineyards (i) the relative frequency of the foliar symptom expression (hereinafter referred as  $RF_{symptoms}$  or symptom expression) was measured in 2019. Beyond that (ii) the relative frequency of the previously dead vines or hiatuses presumably caused by GTDs and the replacements were also studied and calculated. The sum of all values above (hereinafter referred as  $RF_{damage}$ ) reflected the damage caused by GTDs since the establishment of the vineyard.

Considering the effect of the age of the vineyard on the symptom expression, two age categories were used: young (<9 year old) and mature (at least 9 year old) vineyards.

For statistical analysis IBM SPSS 23 software package was used. Comparisons were made with oneway ANOVA if data fits the assumptions of parametric tests. In this case Bonferroni post-hoc test was used. In other cases Kruskall-Wallis nonparametric test was carried out with Mann-Whitney U-test for post-hoc analysis (Ketskeméti et al., 2011).

#### **RESULTS AND DISCUSSION**

#### Cultivar susceptibility groups

Symptom expression was calculated from 200–400 studied plants per cultivars in each of the eight vineyards from five different wine regions in Hugary (*Table 1*). Both white and red cultivars were examined in every wine region, except Tokaj, where only indigenous white cultivar (Hárslevelű) was involved in the study. Most of the examined cultivars (Cabernet franc, Pinot noir, Cabernet sauvignon, Olaszrizling, Kékfrankos, Királyleányka, Bakator, Sauvignon blanc and Merlot) were studied in more than one vineyards, mainly at different wine regions.

There was a strong statistically significant correlation between the RF of symptom and the RF of damaged vines based on Pearson-correlation



(r=0.7053; p<0.05) therefore mainly the values of  $RF_{symptoms}$  were used in further analysis.

Mean symptom expression varied between less than 0.5% (Bakator: 0.12% and Chardonay: 0.25%) and more than 10% (Kadarka: 11.43% and Kéknyelű: 19.88%). It must be mentioned that both of the studied Bakator was in the younger age group, and Kadarka was surveyed only in one vineyard belonging to the older age group. Therefore the susceptibility determination of these cultivars need further study. Based on the mean symptom expression (RF<sub>symptoms</sub>) in 2019, susceptibility groups were created to acquire information about the susceptibility of the different cultivars. Three susceptibility groups could be separated, based on larger steps of increasing symptom expression values. There was significant difference between the main symptom expression values of these groups. The symptom expressions of the slightly (RF<sub>symptoms</sub>:  $0.52\% \pm 0.35$ ) and the highly susceptible (RF<sub>symptoms</sub>:  $9.82\% \pm 4.78$ ) groups were differed statistically, although the transitional moderately susceptible (RFsymptoms: 3.42%  $\pm 1.09$ ) group did not show clear separation from them (Table 2). When four groups formed based on their symptom expression (different by quartiles), they could not be separated clearly based on their susceptibility values either. Only the slightly susceptible group ( $RF_{symptoms}$ : 0.52% ±0.35) showed significant differences from the two most sensitive groups (Susceptible with  $RF_{symptoms}$ : 3.09% ±0.92, and Highly susceptible with  $RF_{symptoms}$ : 6.36% ±1.54) (*Table 2*).

The sequence of susceptibility coincide broadly with the previous, international data. The symptom expression was less frequent in Merlot, Pinot noir and Chardonnay than in Sauvignon blanc, Cabernet sauvignon and Cabernet franc plantations (Edwards et al, 2007; Feliciano et al., 2004; Lorraine et al., 2012; Ramírez et al., 2018). Similarly Chardonnay, Merlot and Pinot noir were in the slightly or moderately susceptible groups, while Sauvignon blanc, Cabernet sauvignon and Cabernet franc in the Susceptible and Highly susceptible groups (Table 2). The following Hungarian cultivars were slightly susceptible (similarly to the Chardonnay): Bakator, Cserszegi fűszeres and Kékfrankos. However Juhfark, Hárslevelű, Kadarka and Kéknyelű were highly susceptible in our survey, similarly to Cabernet sauvignon. These susceptibilities are showing good coincidence with earlier surveys (in Hungary) of our working group too (Csótó et al., 2016).

Table 2

Mean symptom expression of cultivars and susceptibility groups (with mean values ±SD) of the different cultivars by major steps of the symptom expression values and quartiles of them. Differences of mean symptom expression (%) of groups were evaluated by Kruskall-Wallis test and Mann-Whitney U-test. Letters sign significant differences based on U-test

Cultivar	Mean symptom- expression (%)	Susceptibility groups by major steps	Susceptibility groups by quartiles		
Bakator	0.12				
Chardonnay	0.25	01' 1 4 4 41 1			
Cserszegi fűszeres	0.50	Slightly susceptible	Slightly susceptible		
Merlot	0.75	0.52 (±0.35) a	0.52 (±0.35) a		
Kékfrankos	0.96				
Sauvignon Blanc	2.00				
Pinot Noir	2.65	NC 1	Moderately susceptible 3.09 (±0.92) ab		
Királyleanyka	3.83	Moderately susceptible			
Pinot Gris	3.86	3.42 (±1.09) ab			
Portugieser	4.75				
Cabernet Franc	5.34		Susceptible		
Olaszrizling	7.60		6.36 (±1.54) b		
Juhfark	7.76	TT: 11			
Cabernet Sauvignon	8.15	Highly susceptible	Highly susceptible 12.01 (±5.44) b		
Hárslevelű	8.59	9.82 (±4.78) b			
Kadarka	11.43				
Kéknyelű	19.88				
	Kruskall-Wallis test	H=18.7281, p=0.0001	H=19.576, p=0.0002		

## Effect of the vineyard age to the relative frequency of symptom expression

Besides the cultivar susceptibility, the vineyard age was an important factor in symptom expression. The relative frequency of foliar symptom expression in case of older vineyards was significantly higher than the younger ones regardless of the effect of the cultivars and other edaphic conditions. The vineyard parcels were separated to two age groups: less, than 9 year old and at least 9 year old (*Table 3*). While the difference was only marginally significant (at p<0.1 level) in the relative frequency of damaged vines between the age groups (*Figure 4*). These outcomes confirmed results of Díaz and Latorre (2013) and Úrbez-Torres et al. (2008) who found that in mature vineyards (older than 7–8 years) the symptoms occur more likely.



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		All surveyed vine	yard parcels			
	Less than 9 years	old	More than 9 years old			
Wine region	Parcel	Cultivar	Wine region	Parcel	Cultivar	
Érmellék	Heit Papok földje	Kadarka	Badacsony	VWRI	Cabernet sauvignon	
Érmellék	Heit Papok földje	Bakator	Badacsony	VWRI	Cabernet franc	
Villány	Zuhánya	Cabernet sauvignon	Villány	Göntér	Merlot	
Villány	Zuhánya	Kékfrankos	Villány	Göntér	Portugieser	
Villány	Zuhánya	Cabernet franc	Villány	Göntér	Pinot noir	
Érmellék	Heit Birtok	Bakator	Villány	Göntér	Sauvignon blanc	
Szekszárd	Lajvér Birtok	Chardonnay	Badacsony	VWRI	Kiralyleányka	
Szekszárd	Lajvér Birtok	Kekfrankos	Tokaj	Szarvas	Harslevelu	
Szekszárd	Lajvér Birtok	Cabernet franc	Badacsony	VWRI	Pinot noir	
Szekszárd	Lajvér Birtok	Cabernet sauvignon	Badacsony	VWRI	Juhfark	
Szekszárd	Lajvér Birtok	Cserszegi fűszeres	Badacsony	VWRI	Juhfark	
Szekszárd	Lajvér Birtok	Pinot noir	Badacsony	VWRI	Juhfark	
Szekszárd	Lajvér Birtok	Sauvignon Blanc	Badacsony	VWRI	Szürkebarát	
Szekszárd	Lajvér Birtok	Olaszrizling	Badacsony	VWRI	Szürkebarát	
Szekszárd	Lajvér Birtok	Merlot	Badacsony	VWRI	Keknyelű	
			Badacsony	VWRI	Keknyelű	
			Badacsony	VWRI	Olaszrizling	
			Villány	Bocor	Kékfrankos	
			Villány	Bocor	Pinot noir	
			Villány	Bocor	Cabernet sauvignon	
			Villány	Bocor	Olaszrizling	
			Villány	Bocor	Cabernet franc	
			Érmellék	Heit Birtok	Kiralyleányka	

#### The surveyed vineyard parcels by age group

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### 20 foliar symptom + 18 damaged vines 16 14 Relative frequency (%) 12 10 8 6 4 2 0 9-<9 Age of vineyard (year)

Figure 4: Symptom expression and ratio of damaged vines by age categories. Signs show significant differences at p<0,1 (+) and p<0,01 (\*\*) levels based on Bonferroni test

The survey will be carried on in these vineyards in the following years, so the relation between symptom expression and its effecting ecological factors may be determined too, like climatic and water stress (Andreini

Table 3

et al., 2014; Songy et al., 2019; Sosnowski et al., 2016; Van Niekerk et al., 2011), rootstock type (Andreini et al., 2014) or the composition of endophytic microorganisms (Gramaje et al., 2015; Del Frari et al., 2019).

#### **CONCLUSIONS**

Strong correlation was found between the relative frequency of retrospectively and currently damaged vines and the relative frequency of symptom expression. Cultivar susceptibility groups we created based on the latter, because of its higher accuracy. The international Merlot, Pinot Noir, Chardonnay, as well as the indigenous Bakator, Cserszegi fűszeres and Kékfrankos cultivars proved to be less susceptible. On the other hand Sauvignon Blanc, Cabernet Sauvignon, Cabernet Franc widespread cultivars, as well as the Juhfark, Hárslevelű, Kadarka and Kéknyelű tipically grown in the Carpathian basin were more sensitive to the grapevine trunk diseases (GTD). These results may help the winegrowers to choose less susceptible cultivars in case other factors (e.g water stress) may prone the GTD symptom expression to reduce economic loss.

Based on previous results, two age groups were created to separate younger (at the eighth year) and older ("mature") cultivars. In accordance with previous statements (Díaz and Latorre, 2013; Kovács et al., 2017; Úrbez-Torres et al., 2008), we also found, that the relative frequency of foliar symptom expression was significantly lower in the younger vineyards. Whether it is due to the lower frequency of infection with GTD pathogen(s), the differences in the endophytic microbial composition and ratio or the difference in the plant resistance, needs further studies.

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