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INFLUENCE OF CLIMATIC CONDITIONS ON RIPENING AND THE PHENOLIC CONTENT OF GRAPES FROM CABERNET SAUVIGNON, GAMZA AND RUBIN RED VINE VARIETIES

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Abstract

During the period 2021-2022, the dynamics of ripening and the phenolic content of grapes from the varieties Cabernet Sauvignon, Gamza and Rubin, grown in the region of the town of Pleven, were studied. The changes in the meteorological indicators throughout the vegetation period were monitored. During the winter months, significantly low temperatures for the vine plant growth were not registered in the region. The onset of the vines' vegetative development was in the middle (2021) or the early (2022) April. During the grapes ripening period, the dynamics of sugars and total acids were monitored. The Rubin variety showed the fastest sugar accumulation while maintaining a relatively high acidity rate. The Gamza and Cabernet Sauvignon varieties were later ripening and revealed a gradual increase in sugars and a decrease in total acids. The highest sugar accumulation in the Rubin and Gamza varieties was found in 2021, while for Cabernet Sauvignon - in the grapes from the 2022 harvest. Both the weather conditions during the year and the varietal characteristics, specifics and potential had an impact on the phenolic content in the structural elements of the cluster. The varieties had different reserves of total, flavonoid and non-flavonoid phenolic compounds, which increased in the order of berries < skins < rachis < seeds. The Rubin variety had the highest values of the studied phenolic components, followed by Cabernet Sauvignon, and the lowest were in Gamza. The anthocyanin content of the varieties changed in the same sequence. In all three varieties, the anthocyanin content in the skins was significantly higher compared to the berries.

Keywords: climate, soil, Cabernet Sauvignon, Gamza, Rubin, grapes, ripening, phenolic content

INTRODUCTION

The vine development and the ripening process of grapes strongly depend on the natural conditions in the growing area. The main environmental factors that determine the terroir are climate, soils, geographical location, etc. The quantity and quality of grape harvest largely depend on them. The terroir also includes the varieties grown and the characteristic of the place, as well as some socio-economic factors (Leeuwen and Seguin, 2006; Hatay, 2021; Urvieta et. al., 2021).

Grapevine as a plant has been cultivated in a wide temperature range. The climate in the growing area has the greatest impact on the grapes' composition and quality and, accordingly, on the wine. Due to its influence, components of different composition and quantity accumulate in grapes, thus predetermining the characteristics of wine. The wines obtained from grapes grown in cooler climates differ significantly from those obtained in warmer ones (Jones, 2013; Cogato et. al., 2019). The impact of weather conditions on grapes is crucial during the ripening period (Gonzalez Perez et. al., 2018). The temperature range during the day highly affects the phenolic components accumulation, especially the anthocyanins. The optimal daily temperature for their synthesis is within 25-30°C (Urvieta et. al., 2021).

An essential component of the terroir is the soil with its mineral composition and microorganisms, predetermining the physiological development of vine and the uptake of nutrients. The excess or deficiency of nitrogen in the soil changes plant metabolism along with the biosynthesis of a number of components essential for grapes and wine quality (Leeuwen and Seguin, 2006; Hatay, 2021).

The water status of the soil depends on climate (the precipitation rate), but also on some agricultural factors such as the vine growing practice. With a water deficit, grapes ripen faster, the berry size decreases, as well as the aromatic potential, but the total anthocyanins and phenols ratio increase rapidly (Leeuwen and Seguin, 2006; Gonzalez Perez et. al., 2018).

The stage of ripening of the red varieties grapes begins with the softening and veraison of the berries and ends with the onset of physiological and technological maturity. This period has a different duration for individual varieties. The coloring agents accumulated in the berry skin, the berries and clusters reach specific structure, size, weight and color for the variety (Abrasheva et. al., 2008; Bigard et. al., 2019; Shahood et. al., 2020).

The sugars accumulation processes and the reduction of the organic acids ratio in the berries are most intense at the beginning of the ripening phenophase as thereafter the sugars accumulation continues relatively evenly. The sugar rate reaches its peak as soon as the berries have gained their maximum weight, while the ratio between monosaccharides glucose and fructose (Glu/Fru) decreases. In the case of red varieties, depending on the specifics and characteristics, the sugar content varies from 21 to 24%, and total acids – from 5 to 7 g/dm^3 (Abrasheva et. al., 2008; Chobanova, 2012; Bigard et. al., 2019; Shahood et. al., 2020).

Grapes' ripening is a complex process not only related to the variations of sugars and total acids. During this period, the biosynthesis and accumulation of a number of volatile aromatic components or their non-volatile precursors, phenolic and coloring substances (anthocyanins) take place. That process is most intensive during the last stages of ripening. The berry skins lose their chlorophyll and, with the formation of pigments, acquire the color characteristic of the variety. The pH of the grape juice also goes up (Hellman, 2004; Bigard et. al., 2019).

The ripening process of grapes depends on the terroir conditions in the region of cultivation, as well as on the genetic characteristics of the variety. Therefore, the different varieties acquire their specific features and ripen in different periods at the same location. When determining the time of harvest, not only the changes in sugars, total acids and pH are monitored, but also other factors such as the skin elasticity, the easy separation of the berry from the rachis, the homogeneity of the fleshy part and the phenolic maturity concerning the phenols in the solid parts (Hellman, 2004; Mandfield, 2006).

Phenols are an essential group of components in the grapes and wine composition. The content of polyphenols such as anthocyanins, flavanols, flavonols and resveratrol is particularly important for the red varieties. They exhibit a high biological activity and perform a number of protective functions with regard to human health (Xia et. al., 2010; Tomaz et. al., 2016). The amount of phenols in wine depends on the phenol content in the grapes of the respective variety. The structural elements of grapes have different reserves of total, flavonoid and non-flavonoid phenolic compounds. The main groups of phenolic substances are distributed unevenly in the various parts of the cluster. Most of them are found in the rachis and seeds, less are found in the skins and the least - in the fleshy part. About 60-70% of the phenolic components are localized in the seeds, 30-35% in the skins and 5-8% in the fleshy part (Sandhu and Gu, 2010; Godevac et. al., 2010; Hornedo-Ortega et. al., 2020; Rousserie et. al., 2020; Atak et. al., 2021). The flavonoids (catechin, epicatechin, procyanidin polymers) are mainly localized in the rachis and the seeds. The phenolic acids (hydroxycinnamic and hydroxybenzoic derivates) are distributed in the skins and in the fleshy part. The anthocyanins are the main pigment in the red grape skins. Their content is specific and characteristic for the different varieties and depends on the degree of ripeness of the grapes and the region of cultivation (Godevac et. al., 2010; Xia et. al., 2010; Gomez Gallego et. al., 2012; Shi et. al., 2016; Hornedo-Ortega et. al., 2020). In the varieties of *Vitis vinifera* L., the glycosidic forms of the individual representatives of anthocyanins predominate – mainly of malvidin, delphinidin, cyanidin, petunidin, peonidin (Gomez Gallego et. al., 2012; Obreque-Slier et. al., 2013; Shi et. al., 2016; Hornedo-Ortega et. al., 2020). The content of phenolic compounds in grapes is important and decisive for the organoleptic properties of obtained wines – color, astringency and bitterness (Gombau et. al., 2020; Hornedo-Ortega et. al., 2020; Rousserie et. al., 2020).

During the grapes' ripening, the phenolic complex changes significantly, and at a certain moment the amount and composition of the phenolic components are the most favorable for obtaining wines with optimal chemical indicators and organoleptic features. All that necessitated the determination of the phenolic content and the phenolic ripeness of the grapes (Stoyanov et. al., 2004; Stoyanov, 2007; Delcheva et. al., 2013).

The objective of this study was to investigate the dynamics of ripening and the phenolic content of red grapes of the varieties Cabernet Sauvignon, Gamza and Rubin, under the influence of soil and climatic conditions in the region of the town of Pleven.

MATERIALS AND METHODS

The experimental work was carried out in the period 2021-2022 at the Institute of Viticulture and Enology (IVE) – Pleven. The area of the town of Pleven is part of the Northern wine-growing region (the Danube plain), characterized by a typical continental climate, an early spring with frequent late frosts, a hot and relatively dry summer, a long and warm autumn with early frosts, a cold and frosty winter. Soils include all types of black soils – typical, carbonate, leached, highly leached and podzolic, formed on loess (Katerov et. al., 1990).

Trial areas and agricultural practice

The study was focused on grapes from the red varieties Gamza (local), Rubin (intraspecific hybrid with parental forms Nebbiolo x Syrah) and Cabernet Sauvignon (introduced), grown in the Experimental Base of IVE.

The vineyards were created on slightly leached black soil formed on clay loess. This type of soil is weakly strong, moderately eroded, heavy sandy-clay by mechanical composition, which determines the favorable water-physical properties. The content of total nitrogen is low, the supply of phosphorus is relatively weak, and the amount of potassium is not sufficient for the development of vine. The carbonate content is low. The reaction of the soil in the upper horizons is neutral, and in the lower carbonate horizons it is slight to moderate alkaline pH 6.0 - 7.2 (Ivanov, 2016).

The trial vineyards were fruit-bearing and 20 experimental vines were selected from each studied variety. All varieties were grafted on Berlandieri x Riparia SO4 rootstock. The planting distance between the vines of the Gamza and Rubin varieties was 2.20/1.30 m and of the Cabernet Sauvignon variety – 3.00/1.30 m. The Rubin and Cabernet Sauvignon varieties were grown on a medium-stem training system double improved Guyot, while the Gamza variety – on a low-stem system single improved Guyot. The pruning and loading of the vines were carried out depending on the varietal specifics and training: the Gamza variety – 18 winter eyes per vine, the Rubin variety – 28 winter eyes per vine, the Cabernet Sauvignon variety – 40 winter eyes per vine.

Meteorological indicators

During the vines' vegetation period (01.04. – 31.10.), the meteorological indicators were daily monitored every year. Data on temperature (minimum, maximum, average value), relative air humidity, amount of precipitation and their average monthly rates were registered by the automatic weather station "iMetos", located in the Experimental Base of IVE – Pleven.

Monitoring the grape ripening process from the studied varieties

During the grape ripening period (August – September), the dynamics of sugar accumulation (using a field refractometer) and the change in total acids, g/dm^3 (by titration with NaOH) in the grape juice were monitored in order to determine the reaching of technological maturity for each variety and the time of harvest (Ivanov et. al., 1979).

Phenol content determination of grapes from the studied varieties

The technological content of the phenolic compounds in grapes per varieties and their rate in the structural elements of the cluster (rachis, seeds, skins, berries) was determined at the onset of technological maturity and after harvesting. An average sample of 2 kg of grapes was taken from each variety, out of which, after removing the berries, 5 g of rachis (dried and cut into pieces), 5 g of seeds (grinded in a mortar), 5 g of skins (dried) and 10 g of berries (torn in a mortar and with crushed seeds) were weighed on an electronic scale. The prepared and weighed amounts were transferred to Erlenmeyer flasks. For the extraction of weighed quantities, 150 ml of extractant CH_3OH/HCl (1% v/v) was used (Stoyanov et. al., 2004; Stoyanov, 2007).

After the extraction in the liquid phase, the content was determined as follows:

- total phenolic compounds (TPC), g/dm^3 gallic acid – Singleton et Rossi method with Folin – Chiocalteu reagent (Ivanov et. al., 1979)
- flavonoid phenolic compounds (FPC), mg/dm^3 catechin equivalent – Sommers method (Chobanova, 2007)
- non-flavonoid phenolic compounds (NPC), mg/dm^3 coffee equivalent – Sommers method (Chobanova, 2007)
- monomeric anthocyanins, mg/dm^3 – spectrophotometrically by Ribereau-Gayon et Stonestreet method via pH changing (Ivanov et. al., 1979).

Statistical processing of the results

The data obtained from the analyses performed for the study period were subjected to statistical processing, represented by a mean value and a standard deviation (±SD). The program Excel 2007 (Microsoft Office) was used for determination.

RESULTS AND DISCUSSION

Meteorological indicators

The most important terroir factors that influence vine growth and development and quantity and quality of the grape harvest have been the climate and soil. The weather conditions during the growing season (temperature, precipitation rate, relative humidity of the air and solar radiation) are crucial. The frequency of adverse impacts is also essential – low winter temperatures, hail, prolonged droughts, etc. The climatic features of the region depend directly on its geographical location and the peculiarities of the relief. The meteorological conditions of the year have a direct influence on the content of all components of the chemical composition of grapes, including the phenolic substances (Abrasheva et. al., 2008; Jones, 2013; Leeuwen et. al., 2020).

In the first quarters of 2021 and 2022, in the area of the Experimental Base of IVE - Pleven, no critically low temperatures were recorded that would have negatively affected the physiology and development of the vine. Positive average monthly air temperatures were reported (Table 1). In 2021, the lowest average minimum temperatures were recorded in January (-12.1ºC) and February (-11.2ºC), and in 2022 in the months of January (-11.7ºC) and March (-11.3ºC). For the period January – March, the average maximum temperature in 2021 was 17.2ºC, and in 2022 it was 18.4ºC.

Table 1. Meteorological indicators for the period 01.01. – 31.03.2021 – 2022.

Year	Month	Air temperature °C			Σ av. Air humidity [%]	Σ av. Precipitation [mm/m^2]
		Σ av. min	Σ av. max	Σ average		
2021	January	-12.1	14.7	1.3	84	42.4
	February	-11.2	18.9	2.3	80	3.6
	March	-7.8	17.9	3.5	75	34.8
2022	January	-11.7	17.7	1.0	76	6.8
	February	-6.0	16.9	3.5	71	14.8
	March	-11.3	20.6	2.9	65	11.2

In 2021, the growing season of vine development began in the second decade of April, and in 2022, at the beginning of April, when the minimum air temperatures permanently remained positive. During the months of May and June, a gradual increase in the minimum, maximum and average temperatures was observed. Then the highest precipitation rate for the vine growing season in the area of cultivation was registered. The months of July and August traditionally have the highest air temperatures, with almost no precipitation. The reported temperatures in September had an increasing trend. The high temperatures and the lack of precipitation during the grape ripening period were the cause of atmospheric and soil drying. That was related to the acceleration of the grape ripening process, the earlier onset of technological maturity in some varieties and the grape harvest. In 2021, the recorded lower values of the minimum, maximum and average air temperatures in the months of May and June slowed down the vegetative development of the vines and led to a later harvest of the studied varieties (Figure 1).

The dynamics of ripening of the studied Cabernet Sauvignon, Gamza and Rubin varieties.

The process of sugar accumulation and the reduction of organic acids in the berries have been the most intense at the onset of the ripening phase of the grapes, as thereafter it continues relatively evenly. The main components of the sugars accumulated during this period are glucose and fructose, as initially the glucose is dominant, but at the end the fructose predominates. The accumulation of phenolic substances in the berry also begin giving it the characteristic red color of varying intensity for the different varieties (Hellman, 2004; Shahood et. al., 2020). Upon reaching physiological maturity, the sugars and acids content in the berries and their ratio remains relatively constant, then, as a result of transpiration, the sugars increase and the acids decrease. In some varieties, the physiological maturity of the grapes coincides with the technological maturity. For most, however, the technological maturity occurs after the physiological (Abrasheva et. al., 2008).

It is important to take properly an average sample in order to monitor accurately and correctly the ripening process of the studied varieties and to determine the date for harvesting. Therefore, the average sample should include clusters from the different vines or parts of the vine or berries from the different clusters or parts of a cluster (Hellman, 2004; Mansfield, 2006).

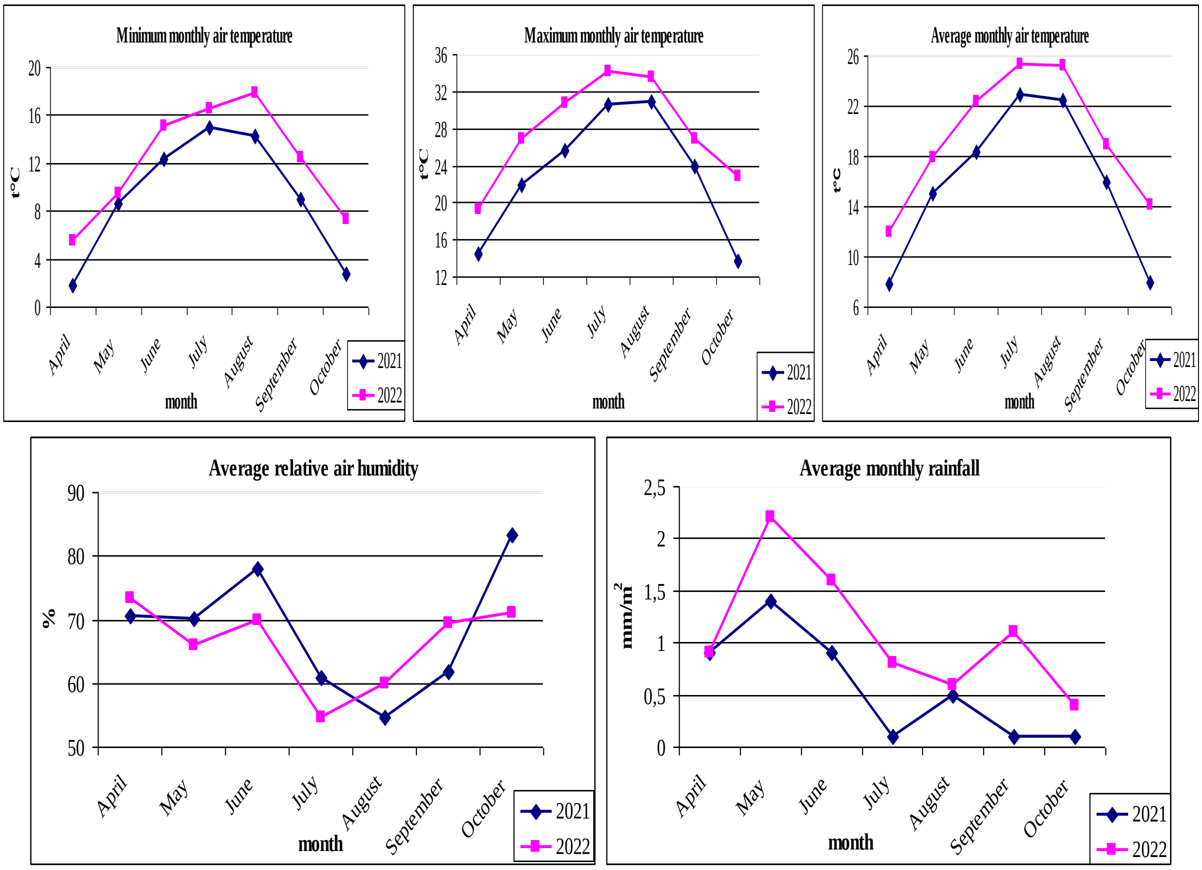


Figure 1. Meteorological indicators for the vegetation period 01.04. – 31.10.2021-2022.

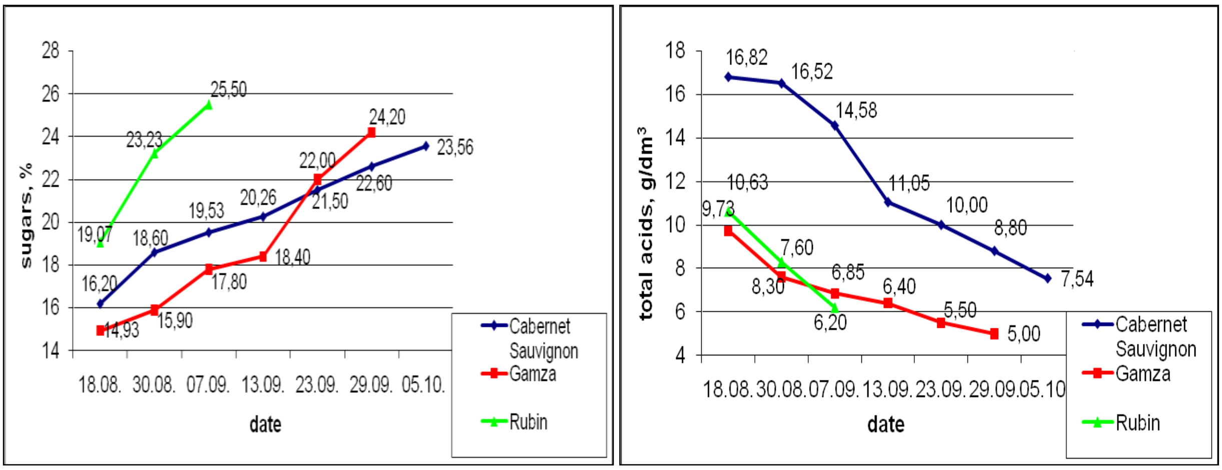
During the ripening period of the studied varieties, the dynamics of sugar accumulation and the change in total acids were monitored. The observations began in the second ten days of August (18.08.2021 and 19.08.2022) and ended with the harvesting of the grapes from the respective varieties.

Despite the weather specifics of 2021 and the delay in the vegetative development of the vines during the months of May and June, the ripening process proceeded normally, as a result of the atmospheric and soil drying that occurred during the following period July – September. The change in sugars and acids was going on without a deviation from the normal course. The grapes had the typical composition of the respective variety, despite the later harvest (Figure 2).

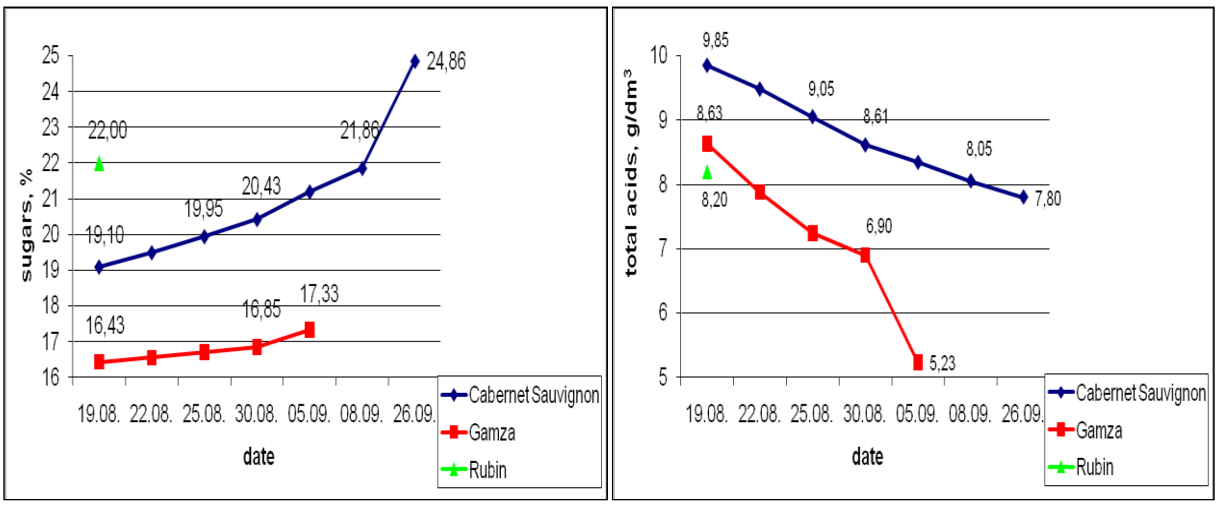
During the study period, the fastest sugar accumulation and the earliest technological maturity were recorded in the Rubin variety. The harvest was carried out the earliest – at the end of August (2022) or at the beginning of September (2021). The high sugar accumulation has been a specific feature of the variety. The Gamza and Cabernet Sauvignon varieties were ripening later and they were harvested at the end of September or at the beginning of October. With Cabernet Sauvignon, a smooth increase in sugars was observed every year. In the Rubin and Cabernet Sauvignon varieties, a high sugar content with retention of higher total acids was found that, was influenced by the weather conditions – the drying during the ripening period. The highest sugar accumulation in the Rubin and Gamza varieties was found in 2021, while for Cabernet Sauvignon in 2022.

In 2021, the grapes of the Rubin variety were picked on September 7^{th}, with a sugar content of 26.50±0.00% and total acids 6.20±0.11 g/dm^3. In the other two varieties, the ripening process occurred normally, but with a delay. The grapes of the Gamza variety were picked on September 29th, with indicators of 24.20±0.17% sugars and 4.90±0.10 g/dm^3 of total acids. With it, there was a sharp increase in the sugar accumulation and a smooth change in acidity. In the Cabernet Sauvignon variety, the reverse trend was observed – a smooth increase of sugars and a sharp decrease of total acids at the beginning of monitoring the ripening process, as thereafter their change was weaker. During the grape harvest (October 5th), the monitored indicators had the rates of 23.56±0.25% and 9.54±0.19 g/dm^3, respectively (Figure 2a).

In 2022, the Rubin variety was harvested the earliest, on August 19th, with a sugar content of 22.00±0.17% and total acids of 8.20±0.04 g/dm^3. Due to the rainfall at the beginning of September and the risk of the grapes rotting, the Gamza grape harvest was held earlier than the usual period for the variety – on September 5th. The grapes were picked at low sugars 17.33±0.11% and total acids 5.23±0.04 g/dm^3. The Cabernet Sauvignon variety was harvested the latest – on September 26th. The dynamics of grape ripening showed a gradual increase of sugars and a decrease of the acid ratio. During the vintage, the monitored indicators had values of 24.86±0.11% and 7.80±0.13 g/dm^3, respectively (Figure 2b).



a) 2021



b) 2022

Figure 2. Dynamics of sugars and total acids in the grapes from the studied red varieties during the period 2021 – 2022.

Phenol content determination of grapes from the studied varieties Cabernet Sauvignon, Gamza and Rubin

The phenolic components are unevenly distributed in the various cluster parts and their concentration strongly depends on the variety. Their content is the highest in the rachis and seeds, followed by the skins and the fleshy part. The predominant representatives of NPC are the phenolic acids, and of FPC – catechin, epicatechin and gallocatechin. The phenolic acids are mainly localized in the skins and in the fleshy part. In the red varieties, flavan-3-ols and proanthocyanidins are the main phenols in the grape seeds, and anthocyanins in the skins (Godevac et. al., 2010; Xia et. al., 2010; Gomez Gallego et. al., 2012; Shi et. al., 2016; Hornedo-Ortega et. al., 2020). Generally, the flavonols are present in the skins, at an amount dependent on the variety, and in 3-O-glycoside forms. The main flavonol in the red varieties is quercetin-3-O-glycoside (Hornedo-Ortega et. al., 2020).

The phenolic components contained in the grapes of the red varieties pass into the wine during vinification. The anthocyanins reserve in the skins is particularly important for determining the color of the future wine. Their content depends on a number of factors such as varietal specificity and the climatic conditions of growing (Shi et. al., 2016; Gombau et. al., 2020).

The results for the phenolic content in the investigated red varieties Cabernet Sauvignon, Gamza and Rubin are presented in Table 2. Different reserve in the structural elements of the cluster of the varieties was observed in terms of TPC, FPC and NPC. The phenolic composition of the grapes of the studied varieties was influenced both by the differences in the weather conditions of the year, and by the varietal characteristics, specificities and potential. The content of the investigated phenolic components in all three varieties increased in the order of berries < skins < rachis < seeds. The varieties showed different levels of anthocyanins. Their content in the skins was higher compared to that in the berries. The berries and skins of all varieties from the 2021 vintage were richer in anthocyanins compared to the 2022 harvest. The Rubin variety had the highest content of phenolic compounds and anthocyanins, followed by Cabernet Sauvignon, and the lowest ratio was found in Gamza. The reason for this could be the genetic origin of the Rubin variety obtained by intraspecific hybridization. The differences in the phenol content of the studied varieties could also be due to both the stem training system used for their cultivation and the loading of the vines. The results showed that the Rubin and Cabernet Sauvignon varieties grown on a medium-stem training system and with a greater loading of the vines had a higher phenolic content than the Gamza variety, grown on a low-stem system and loaded with the least winter eyes.

The Cabernet Sauvignon grapes, vintage 2021, were distinguished by a higher phenolic ratio in relation to all studied phenolic components in all grape structural elements. Despite the later vegetative development of the vines that year, the berries, skins, rachis and seeds contained more TPC, FPC, NPC and anthocyanins compared to the 2022 harvest. An exception was observed only in the rachis, where the 2022 harvest was richer in TPC.

The berries of the Gamza variety contained more TPC and anthocyanins in 2021, and FPC and NPC in 2022. The skins from the 2021 harvest had a higher phenolic content in all investigated components compared to that from the 2022 harvest. In 2022, the rachis was richer in TPC and NPC, while FPC was more in 2021. The seeds of the variety had a similar reserve of TPC in both years of the study, but more FPC and NPC in 2021.

In 2022, the berries (with the exception of anthocyanins) and the rachis of the Rubin variety had a better ratio of TPC, FPC and NPC. The skins of the 2021 harvest contained more TPC, FPC and anthocyanins, and NPC in 2022. The seeds of the variety were richer in TPC and FPC in 2022 and in NPC in 2021.

Table 2. Phenolic content of grapes from the studied red varieties, for the period 2021 – 2022.

Structural elements of the cluster	Indicators	Cabernet Sauvignon		Gamza		Rubin	
		2021	2022	2021	2022	2021	2022
Berries	TPC, g/dm^3 gallic acid	0.64 ±0.00	0.52 ±0.00	0.40 ±0.00	0.37 ±0.00	0.75 ±0.00	1.10 ±0.03
	FPC, mg/dm^3 catechin equivalent	1120.73 ±5.39	917.28 ±5.48	642.15 ±6.81	2570.48 ±2.62	1435.98 ±2.88	1983.96 ±17.58
	FPC, mg/dm^3 coffee equivalent	88.65 ±0.81	58.69 ±1.12	68.48 ±0.88	187.67 ±0.62	109.48 ±0.23	136.38 ±5.52
	Anthocyanins, mg/dm^3	209.17 ±0.61	119.26 ±2.45	149.81 ±6.99	92.03 ±0.80	177.82 ±0.60	162.92 ±0.10
Skins	TPC, g/dm^3 gallic acid	0.93 ±0.00	0.66 ±0.20	0.80 ±0.00	1.32 ±0.00	1.58 ±0.00	1.42 ±0.00
	FPC, mg/dm^3 catechin equivalent	1654.65 ±2.32	863.49 ±0.96	1575.27 ±11.35	934.59 ±3.98	2809.34 ±7.25	1727.88 ±31.45
	FPC, mg/dm^3 coffee equivalent	130.57 ±0.26	80.75 ±6.92	168.20 ±0.93	143.50 ±0.92	206.53 ±0.42	112.33 ±4.54
	Anthocyanins, mg/dm^3	494.58 ±0.18	272.15 ±0.89	409.27 ±7.38	192.03 ±0.80	699.56 ±3.76	444.57 ±2.25
Rachis	TPC, g/dm^3 gallic acid	1.23 ±0.00	1.30 ±0.00	1.45 ±0.00	2.37 ±0.00	2.00 ±0.00	4.10 ±0.01
	FPC, mg/dm^3 catechin equivalent	1685.03 ±3.11	1358.30 ±32.96	2033.13 ±4.30	1543.93 ±8.08	3474.34 ±1.88	5391.33 ±10.70
	FPC, mg/dm^3 coffee equivalent	99.49 ±0.63	66.72 ±0.05	80.84 ±0.28	91.30 ±0.16	191.28 ±0.13	201.22 ±0.43
Seeds	TPC, g/dm^3 gallic acid	2.66 ±0.10	2.30 ±0.00	3.67 ±0.00	3.97 ±0.00	2.88 ±0.13	4.30 ±0.00
	FPC, mg/dm^3 catechin equivalent	4839.38 ±2.47	3508.57 ±2.93	7171.33 ±16.63	5565.48 ±7.26	6048.45 ±31.72	6557.48 ±3.30
	FPC, mg/dm^3 coffee equivalent	171.64 ±0.84	125.00 ±0.43	217.78 ±1.56	187.66 ±0.62	411.95 ±0.13	335.74 ±1.98

In 2021, the Rubin variety, with the exception of the seeds, had the best content of FPC in the rest of the structural elements of the grape. In that order, it was followed by Cabernet Sauvignon, in terms of berries and skins, and Gamza for rachis. The seeds of the Gamza variety contained the most TPC (3.67±0.00 g/dm^3), but the berries (0.40±0.00) and the skins (0.80±0.00) contained almost twice less than Rubin.

More significant differences were observed in the content of FPC. The same trend was noticed as with TPC. The reserve of the Rubin variety with FPC was the greatest in the berries (1435.98±2.88 mg/dm^3), the skins (2809.34±7.25 mg/dm^3) and the rachis (3474.34±1.88 mg/dm^3). In the seeds of the Gamza variety FPC were the most (7171.33±16.63 mg/dm^3), which was almost twice compared to Cabernet Sauvignon. The berries and the skins of the Rubin variety contained twice as much FPC as Gamza, and the rachis – from Cabernet Sauvignon.

Again Rubin had the highest content of NPC, in all structural elements of the cluster, as in the rachis (191.28±0.13 mg/dm^3) and the seeds (411.95±0.13 mg/dm^3) their amount was twice as high compared to the other varieties. Cabernet Sauvignon had the lowest NPC content in the skins (130.57±0.26 mg/dm^3) and the seeds (171.64±0.84 mg/dm^3), and Gamza - in the berries (68.48±0 .88 mg/dm^3) and the rachis (80.84±0.28 mg/dm^3).

In the three varieties, the anthocyanin content in the skins was several times higher than in the berries. The berries and the skins of the Rubin variety had the best anthocyanin reserve (177.82±0.60 and 699.56±3.76 mg/dm^3, respectively), followed by Cabernet Sauvignon (209.17±0.61 and 494.58±0.18 mg/dm^3, respectively), and it was the lowest for Gamza (149.81±6.99 and 409.27±7.38 mg/dm^3, respectively).

In 2022, the Rubin variety again had the best content of TPC in the structural elements of the cluster. The Gamza variety contained the least TPC in the berries. The rest of the cluster elements showed an increasing trend in the order Cabernet Sauvignon < Gamza < Rubin. The higher content of TPC in the skins, clusters and seeds of Gamza compared to Cabernet Sauvignon was due to the earlier harvest, where the grapes were picked before reaching phenolic maturity. In the berries of the Rubin variety (1.10±0.03 g/dm^3), TPC was almost twice as much as in Cabernet Sauvignon (0.52±0.00 g/dm^3) and three times as much as in Gamza (0.37± 0.00 g/dm^3). The Rubin and Gamza varieties had a similar content in the skins and the seeds (respectively 1.42±0.00 g/dm^3, 1.32±0.00 g/dm^3 and 4.30±0.00 g/dm^3, 3.97±0.00 g/dm^3), which was almost twice that of Cabernet Sauvignon (0.66±0.20 g/dm^3 and 2.30 ±0.00 g/dm^3). The content of TPC in the rachis of Cabernet Sauvignon (1.30±0.00 g/dm^3) was twice less than that of Gamza (2.37±0.00 g/dm^3) and four times that of Rubin (4.30± .00 g/dm^3).

Cabernet Sauvignon had the lowest content of FPC, in all structural elements of the cluster. A significantly higher amount of FPC was recorded in the skins, rachis and seeds of the Rubin variety compared to the other two. In these structural elements of the cluster, the change per varieties increased in the order Cabernet Sauvignon < Gamza < Rubin. The Gamza berries contained more FPC (2570.48±2.62 mg/dm^3) than Rubin (1983.96±17.58 mg/dm^3) and Cabernet Sauvignon (917.28±5.48 mg/dm^3). The content of FPC in the skins of Cabernet Sauvignon (863.49±0.96) and Gamza (934.59±3.98 mg/dm^3) was similar and twice less than Rubin (1727.88±31.45 mg /dm^3). The same was observed in the rachis, where in the Rubin variety (5391.33±10.70 mg/dm^3) the content was almost 4 times higher than in the other two varieties. The seeds of Rubin (6557.48±3.30 mg/dm^3) and Gamza (5565.48±7.26 mg/dm^3) had twice as much FPC compared to Cabernet Sauvignon (3508.57±2.93 mg /dm^3).

That year (2022), Cabernet Sauvignon had the lowest content of NPC in all structural elements of the cluster. For the berries and the skins, the highest values were for the Gamza variety, respectively 187.67±0.62 mg/dm^3 and 143.50±0.92 mg/dm^3, and for the rachis and seeds it was Rubin, respectively 201.22±0.43 mg/dm^3 and 335.74±1.98 mg/dm^3. NPC in the Cabernet Sauvignon berries (58.69±1.12 mg/dm^3) were almost twice less than Rubin (136.38±5.52 mg/dm^3) and three times less than in Gamza (187.67±0.62 mg/dm^3). The skins of Cabernet Sauvignon (80.75±6.92 mg/dm^3) had almost twice less reserve than Gamza (143.50±0.92 mg/dm^3). The rachis and seeds of Cabernet Sauvignon (66.72±0.05 mg/dm^3 and 125.00±0.43 mg/dm^3, respectively) had almost three times less NPC content, compared to Rubin (201.22±0.43 mg/dm^3 and 335.74±1.98 mg/dm^3, respectively).

The tendency was maintained for the berries and skins of the Rubin variety to be characterized by the best anthocyanin content (respectively 162.92±0.10 mg/dm^3 and 444.57±2.25 mg/dm^3), followed by Cabernet Sauvignon (respectively 119.26±2.45 mg/dm^3 and 272.15±0.89 mg/dm^3), and the lowest values were in Gamza (respectively 92.03±0.80 mg/dm^3 and 192.03±0.80 mg/dm^3).

CONCLUSION

From the research results it could be summarized:

- During the winter months, no critically low temperatures were recorded in the cultivation area, which would have negatively affected the physiology and development of the vine. The vegetation period of vine development in 2021 began in the second decade of April, and in 2022 at the beginning of April, when the minimum air temperatures permanently remained positive. The high temperatures during the ripening period and the lack of precipitation were the cause of atmospheric and soil drying that, accelerated the grapes ripening process.
- Despite the delayed vegetative development of the vines during the months of May and June 2021, the ripening of the grapes proceeded normally. The change of sugars and acids went on without a deviation from the normal course and the grapes had the typical composition of the respective variety, despite the later harvest.
- The fastest sugar accumulation with the preservation of relatively high total acids was recorded in the Rubin variety. The high sugar content was a characteristic feature of the variety, while the high acidity was affected by the drying during the ripening period. The Gamza and Cabernet Sauvignon varieties were later ripening and the grape harvest was carried out at the end of September and at the beginning of October, with a gradual increase in sugars and a decrease in total acids.
- The highest sugar accumulation in the Rubin and Gamza varieties was found in 2021 while for Cabernet Sauvignon, it was in the grapes from the 2022 harvest.
- The phenol content in the structural elements of the variety was influenced by both the weather conditions of the year and the varietal features, specificities and potential.
- The varieties had different reserves of TPC, FPC and NPC in the cluster, growing in the order of berries < skins < rachis < seeds. The Rubin variety had the highest phenol content due to its intraspecific hybrid origin, followed by Cabernet Sauvignon, and the lowest was in Gamza.
- In 2022, the higher ratio of TPC in the skins, rachis and seeds of the Gamza variety compared to Cabernet Sauvignon was due to the earlier harvest, where the grapes were picked before reaching phenolic maturity.
- In all three varieties, the anthocyanins content in the skins was significantly higher compared to that in the berries. The berries and skins of the Rubin variety had the best anthocyanins content, followed by Cabernet Sauvignon and Gamza.

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