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Studying the Content of Starch Correlated With Resistance to Low Winter Temperatures in Some Grapevine Varieties

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Abstract

Cryoprotectants are known as substances that are used for protection of biological tissue from freezing damage. Among the most important cryoprotectants are carbohydrates such as glucose, fructose, sucrose raffinose, starch. Starch is a carbohydrate commonly found in the plant tissue, and its concentration is in close relationship with other carbohydrates. This study is about determination of the starch concentration in four *Vitis vinifera* varieties: Vranec and Smederevka (subconvarietas balcanica Negr.) and Cabernet sauvignon and Chardonnay (subconvarietas gallica Negr.). The concentration of starch in vines is related to their point of cold hardiness. The study shows that the concentration of starch in balcanica varieties is lower than in gallica varieties. Therefore, the Vranec and Smederevka grapevine varieties are not resistant to low temperatures against varieties Cabernet sauvignon and Chardonnay which are tolerant.

Key words: Vitis vinifera, cryoprotectants, starch, low winter temperatures.

Introduction

Starch and soluble sugars (sucrose, glucose, fructose and myo-inositol) are the two main forms in which grapevine stores carbohydrate reserves (Mc Artney 1998). It is shown that the concentrations of starch and soluble sugar in all parts of the grapevine vary measurably over the growing season depending on vine phenology. According to Winkler and Williams, starch reserves are at their highest point in all aerial parts of the vine (canes, cordons, trunk and roots) at the end of the growing season. In the dormant season, starch concentrations start to decrease while the concentrations of soluble sugars start to increase (Winkler and Williams 1945, Williams 1996). These changes are related to the development of winter hardiness, where the increase of the

concentrations of soluble sugar from starch conversion acts as cryoprotectant against cold hardiness injury (Hamman et al. 1996). The conversion of carbohydrate reserves (starch) in the spring period is caused by the enzymatic transformation of starch into soluble sugars which can be observed by xylem fluxes of sucrose, glucose and fructose, with glucose as dominant (McArtney 1998).

An important factor in the increasing hardiness may be the sugar accumulation. Jennings and Carmichael's study from 1975 showed us that the concentration of sugar is not related to the changes in dormancy status but to the weather changes.

Sugar accumulation in acclimating plants can have many functions. Carbohydrates are known to lower the freezing point and to increase the osmotic potential. This kind of changes could reduce the amount of dehydration during extra-cellular freezing and save the cell from damage (Levit, 1980).

The agro-technical procedure is another important factor that impacts the concentration of carbohydrates in the dormant period. From the Winkler and Williams' study we can see that the defoliation and irrigation are of great importance for the cold hardiness of the vines.

Materials and methods

*Plant material***.** Four types of vine were analysed, two low resistant (Vranec and Smederevka) and two high resistant (Cabernet sauvignon and Chardonnay) grape varieties. Grapevines canes for the analysis were taken from the Macedonian national collection of grapevine trees. The experiment was made in 3 months period, from December 2011 to February 2012. The samples were taken 2 to 3 days after the lowest temperature has been broadcast, following the weather forecast of National Hydrometeorological service (location Zajcev rid).

The agro-technical procedures in the vineyard were standard. There was no irrigation during the vegetation period and there was no fertilisation in the vineyard.

The samples (vine canes) taken from the vineyard were cut in smaller pieces 5cm long, placed in vegeglas containers and dried in an oven for 24 hours at 105 ºC ±2 ºC. The dried canes were ground into fine powder with an electric mill.

For the starch analysis, 0.1g of ground tissue was extracted with 10ml of 80% ethanol at 80 ºC for 10 min. The samples were centrifuged at 2500rpm x 5min and the supernatant was thrown out. The plant material was reflux to evaporate the residual ethanol. 10 ml 0.1N NaOH was added to the material and extracted in water bath for 45min at 90 ºC. The samples were cooled down to 20ºC and neutralised with 0.1N HCl. The neutralised sample was transferred into 50ml volumetric flask. 1 ml 0.1 M iodine solution and 1ml of H_2O_2 were added and filled up to the mark with d. H_2O. After shaking them thoroughly the solution in the test tubes was allowed to stand for an hour for the starch iodide complex to be developed. The blank solution contains everything except the analysed sample. The developed starch iodide complex was spectrophotometrically read at 580nm. Starch purchased from Merch was used as the

standard for the construction of a calibration curve (R=0.9992) and the concentrations are expressed as starch equivalents (mg/g).

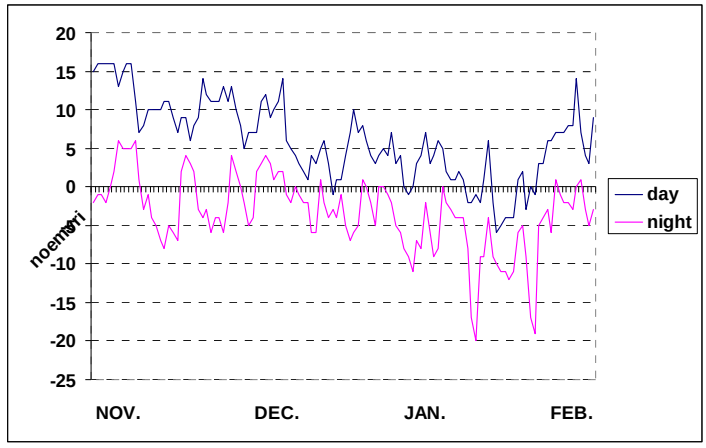


Fig.1. Day and night temperatures in the period of 5 months during winter 2011-2012.
Dnevne i noćne temperature u petomjesečnom periodu tokom zime 2011-2012. godine

Results and discussion

The resistance of the vine is closely related to the quantity of starch in the plant. During the dormant season, the concentration of starch varied among the different types of vine. After analysing the 4 varieties of Vitis vinifera (2 gallica and 2 balcanica varieties), the results that have been obtained in the period of 3 months are presented in Table 1. All the samples were analysed in triplicates.

Tab. 1. Starch dynamic in the canes of different grapevines (measured in %/DW)
Dinamika skroba u lastaru različitih sorti vinove loze (mjereno u %/DW)

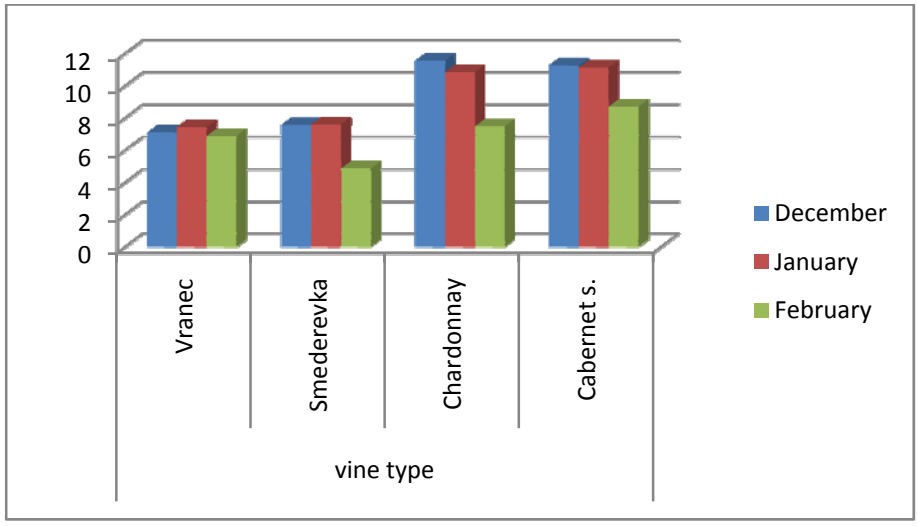
Month	Varieties			
	Vranec	Smederevka	Chardonnay	Cabernet sauvignon
December	7,09	7,56	11,55	11,27
January	7,45	7,60	10,84	11,13
February	6,87	4,88	7,48	8,71

The results shown in Table 1. are measured in percent for dry weight of the vine cane material.

Data from the analysis showed that the highest concentration of starch during the winter period is in December at the beginning of the dormant season. In this period, the concentration of starch in the high resistant types was between 11.27 – 11.55 % DW. On the other hand, the concentration of starch in the low resistant types was between 7.09-7.56 % DW. After the winter period, along with the temperature fall, the

concentration of starch decreased. In the period from January to February, they reached their minimum point. The concentration of starch in January was from 7.4 - 7.6% DW for the low resistant types and for the high resistant types from 10.8 - 11.2% DW. The lowest results were in February where the concentration of starch was from 4.8 - 6.8% DW for Vranec and Smedervka and for Chardonnay and Cabernet s. from 7.4 - 8.7% DW.

All these results about the starch dynamic are shown in Graph 1.



Graph. 1. Starch dynamic between different vine canes during the dormant season
Dinamika skroba u lastaru različitih sorti vinove loze tokom sezone mirovanja

Conclusion

We can conclude from this study that the concentration of starch is associated with the weather changes. The correlation between the temperature and starch value from December to February showed that the highest contents of starch were obtained during the highest temperatures and the lowest when the temperatures were at their minimum. The grapevine varieties resistant to low winter temperatures (Cabernet sauvignon and Chardonnay) accumulated more starch than the susceptible grapevine varieties (Vranec and Smederevka).

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Ispitivanje sadržaja skroba u vezi sa otpornošću na niske zimske temperature kod nekih sorti vinove loze

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Sažetak

Krioprotektanti su poznati kao tvari koje štite biološko tkivo od oštećenja nastalog smrzavanjem. U najvažnije krioprotektante ubrajaju se ugljeni hidrati kao što su glukoza, fruktoza, sukroza, rafinoza, skrob, itd. Skrob je ugljeni hidrat koji je tipičan za biljno tkivo, a njegova koncentracija je usko vezana za druge ugljene hidrate. Ovo istraživanje se bavi određivanjem koncentracije skroba u četiri sorte *Vitis vinifera*: Vranec i Smederevka (*subconvarietas balcanica* Negr.) kao i Cabernet sauvignon i Chardonnay (*subconvarietas gallica* Negr.). Koncentracija skroba u vinovoj lozi je vezana za njenu tačku otpornosti na niske temperature. Istraživanje pokazuje da je koncentracija skroba u sortama *balcanica* niža od *gallica* sorti. Stoga sorte Vranec i Smederevka nisu otporne na niske temperature nasuprot sortama Cabernet sauvignon i Chardonnay koje su otporne.

Ključne riječi: Vitis vinifera, krioprotektanti, skrob, niske zimske temperature.

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