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Changes in water content and carbohydrate metabolism in the annual shoots of contrasting plum varieties in the autumn period

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Abstract. A study of the water content in autumn revealed a significant 1.7-2.9-fold increase in the ratio of bound and free water in plum genotypes in November compared with September. At the same time, a significant increase in the content of sugars amount (by 9.9 % and 10.8 %) was noted in the bark of annual shoots n the plants of *Prunus salicina* and *Prunus* × *rossica* Erem. The outcome obtained suggest that the varieties of *Prunus* × *rossica* Erem. – Vetraz, Gek, Zlato Skifov, Kubanskaya Kometa and *Prunus salicina* – Nezhenka, Orlovskaya Mechta, Skoroplodnaya, Suvenir Vostoka and ELS 18476 have greater frost resistance in winter, because their adaptive processes were more intense in autumn.

1 Introduction

Central Russia is a zone of risky horticulture in terms of growing stone crops, in particular plums. This is due to the influence of unfavourable abiotic factors of winter, which reduce the productivity of cultivated varieties of this crop [1-5]. In the autumn period, with a decrease in metabolic activity, the protective mechanisms of plants are activated, as mentioned above, there is a decrease in tissue hydration and starch accumulation, followed by its hydrolysis [6], while the amount of sugars in plant tissues increases. Sugars increase the water-retaining capacity of protoplasmic colloids protecting against the formation of intracellular ice and excessive dehydration [7-9]. They increase as a result of starch hydrolysis. It is known that sugars, along with other osmolytics, in particular proline, increase the concentration of cell juice by increasing the level of bound water, which has a protective effect under conditions of negative temperatures [10]. In this regard, it remains relevant to study the in water content and carbohydrate metabolism of plum varieties in autumn due to their winter hardiness.

The aim of the research is to study the features of the water and carbohydrate metabolism of plums in the autumn.

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2 Material and methods

The research was conducted in 2021-2023 on the basis of the laboratory of physiology of fruit plants stability and experimental sites of Institute. 14 plum varieties of various genetic origin from the VNIISPK bioresource collection were used as research objects (Table 1).

The fractional composition of water noted in the bark of annual shoots of plum was determined by the Okuntsov-Marinchik method. The concentration of sucrose solution was determined refractometrically [7, 8].

The amount of starch noted in the bark of annual shoots was determined by the method [11]. The optical density of the solution is measured at 590 nm.

Table 1. Research objects and their origin

Variety	Origin
Prúnus doméstica	
Vengerka Belorusskaya	Delikatnaya × Stanley
Vengerka Zarechnaya	Michurinskaya × Krasnaya Desertnaya
Evrazia 21	From spontaneous hybridization of diploid Lacrescent
Zolotoye Runo	Skorospelka Krasnaya ×Viktoria
Stanley	D'Agen × Grand Duke
Prunus × *rossica* Erem.	
Vetraz	Elite 18/1 × Skoroplodnaya
Gek	Skoroplodnaya × Otlichnitsa
Zlato Skifov	From open pollination of Kubanskaya Kometa using mutagens
Kubanskaya Kometa	Skoroplodnaya × Pionerka
Prunus salicina	
ELS 18473	Skoroplodnaya from free pollination
Nezhenka	Skoroplodnaya × Kitayanka
Orlovskaya Mechta	Alyonushka from free pollination
Skoroplodnaya	Ussuriyskaya Krasnaya × Clajmax
Suvenir Vostoka	Zarya × Gigant (re-hybridization of first-generation varieties)

The amount of sugars amount noted in the bark of annual shoots was determined in three replicates based on a resorcinol reagent reaction [7, 8]. After cooling, the optical density was determined on a Bio-Rad SmartSpec Plus spectrophotometer at a wavelength of 520 nm [7, 12].

The activity of amylase will be determined by the colorimetric method of accounting for the amount of starch not hydrolyzed by amylase [13]. The method is based on the determination of the amount of starch cleaved by amylase at λ – 670 nm after treatment with 0.3 % iodine solution.

The obtained data were evaluated using statistics using single-factor analysis of variance ANOVA. The critical significance level was as-sumed to be 5 %.

3 Results and Discussion

In the autumn period, one of the essential signs of plant adaptability to seasonal changes is the dynamics of the ratio of bound water to free water [6-8, 14]. As a result of the conducted research, it was found that in annual plum shoots, by the beginning of winter, the amount of bound water increased by 1.2 times and the free water decreased by 1.8 times. At the same time, the total hydration decreases by 5.2 % in *Prunus* × *rossica* Erem. varieties, by 7.0 % in *Prúnus doméstica* varieties and by 18.7 % in *Prunus salicina*. In September, the lowest ratio of bound water to free water from 1.3 to 2.9 was noted while in November the maximum ratio from 2.3 to 7.7 was noted (Fig. 1). It is known that an increase in bound water in plant tissue by the beginning of winter is associated with a change in the conformation of hydrophilic substances. At the same time, there is a weakening of metabolic processes, a change in the composition of protoplasm and an accumulation of osmoprotective substances (sugars) that affect the state of the water regime and frost resistance of plants [6-8, 14]. So in November, the ratio of bound water to free water increased by 2.9 times in *Prunus × rossica* Erem., by 1.7 times in *Prunus salicina* and by 2.2 times in *Prúnus doméstica* compared to September (Fig. 1). Thus, as a result of the conducted research, a significant increase in bound water in annual shoots of plum varieties was found by the beginning of winter, which reduces the risk of ice formation inside plant cells under conditions of low-temperature stress in winter, because bound water has a reduced freezing point compared to free water.

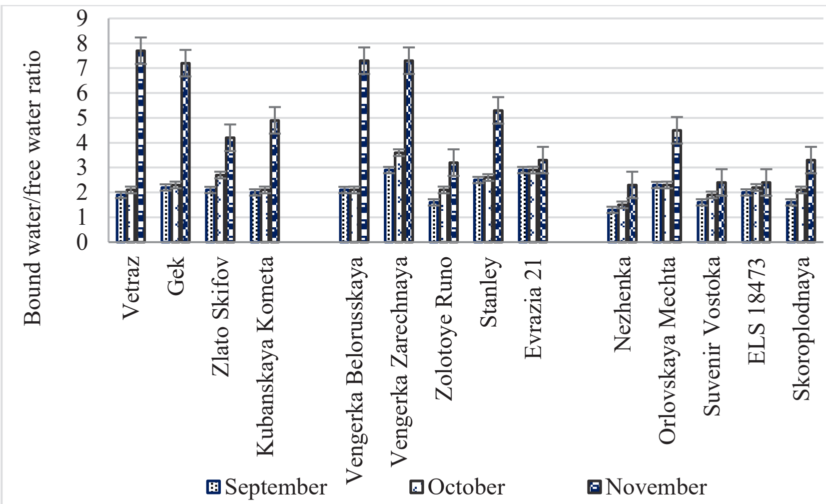

Fig. 1. The ratio of bound water to free water in annual plum shoots in the autumn, %

In the autumn period, with a decrease in metabolic activity, the protective mechanisms of plants are activated, as mentioned above, there is a decrease in tissue hydration and starch accumulation, followed by its hydrolysis, while the amount of sugars in plant tissues increases. So in September, the studied varieties of *Prunus × rossica* Erem. and *Prúnus doméstica* had a maximum amount of starch in the tissues of the bark of annual shoots of 0.968 and 0.972 mg/cm^3, respectively (Fig. 2). At the same time, there was observed higher amylase activity of 143.27 and 157.05 starch mg/hour per·g of the enzyme catalyzing starch hydrolysis (Fig. 3). *Prunus salicina* had an average starch content of 0.72 mg/cm^3 in the bark of annual shoots and the amylase activity was 141.76 starch mg/hour per·g. In October, during hydrolysis, a decrease by 3 times in the amount of starch in the studied plum varieties was noted compared with September against a background of a decrease by 6.6 times in the activity of the enzyme amylase in the bark of annual shoots. It is important to note that in October the starch hydrolysis was more intense in *Prunus salicina* varieties due to higher amylase activity (29.23 starch mg/hour per·g) compared with *Prunus doméstica* (18.12 starch mg/hour per·g) and *Prunus × rossica* Erem. (19.63 starch mg/hour per·g). The tendency of decreasing starch levels and amylase activity in annual plum shoots was also observed in November. This month, there was a 3.9-fold decrease in the amount of starch (Fig. 2) in the bark of annual shoots of plum varieties and the activity of the enzyme amylase by 22.8 times compared with September. At the same time, it should be said that by the end of autumn, in the varieties *Prunus salicina* and *Prunus × rossica* Erem. the intensity of starch hydrolysis is 2.2 and 1.9 times lower, respectively, compared with the varieties *Prúnus doméstica*. In the varieties of *Prúnus doméstica* Vengerka Belorusskaya, Zolotoye Runo and Stanley, a higher amylase activity of 9.85 starch mg/hour per·g in November (Fig. 3) indicates a longer exchange process during the hardening period.

Thus, starch, being a reserve energy substance, accumulates in the organs of plum plants when preparing plants for winter. So the maximum amount of starch in the bark of annual plum shoots was noted in early autumn against the background of higher amylase activity. With a decrease in the average daily air temperature, the content of polysaccharide in the vegetative organs of plum varieties decreases as a result of its hydrolysis with the participation of the enzyme amylase, followed by an increase in sugars. It was noted that starch hydrolysis in the autumn period was more intense in the varieties of *Prunus* × *rossica* Erem. and *Prunus salicina*.

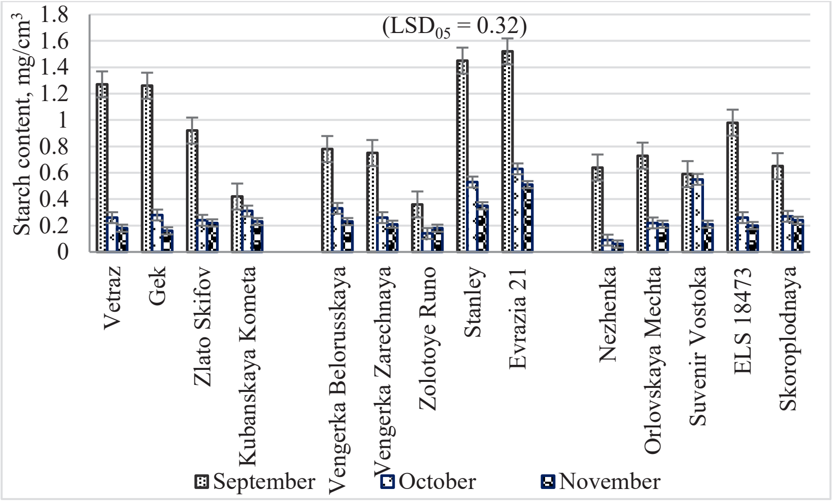

Fig. 2. Starch content in the bark of annual plum shoots in autumn, mg/cm^3

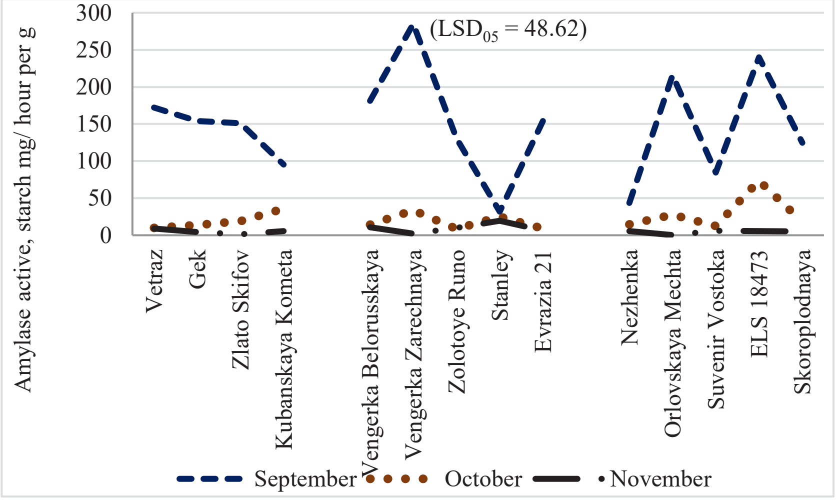

Fig. 3. Amylase activity in the bark of annual plum shoots in autumn

In the autumn period, significant differences between plum varieties were noted in the accumulation of sugars in the bark of annual shoots. Thus, the sugar content of *Prúnus doméstica* in the bark of annual shoots in September was 16.3 % and 22.2 % less than that of *Prunus salicina* and *Prunus* × *rossica* Erem. varieties, respectively. In the autumn period, significant differences between plum varieties were noted in the accumulation of sugars in the bark of annual shoots. Thus, the sugar content of domestic plums in the bark of annual shoots in September was 16.3 % and 22.2 % less than that of *Prunus salicina* and *Prunus* × *rossica* Erem. varieties, respectively (Fig. 3). In October, the varieties of domestic plum also contained low-molecular carbohydrates in the bark of annual shoots by 12.2 % and 14.0 % less than the varieties of *Prunus salicina* and *Prunus* × *rossica* Erem, respectively. At the same time, sugars accumulated more intensively in the bark of *Prunus salicina* varieties Orlovskaya Mechta, Suvenir Vostoka and ELS 18476, *Prúnus doméstica* varieties Evrazia 21 and *Prunus* × *rossica* Erem. Kubanskaya Kometa. The tendency to increase the amount of sugars in annual plum shoots was also observed in November. At the same time, *Prunus* × *rossica* Erem varieties accumulated low-molecular carbohydrates in the bark of annual shoots by 10.8 % and *Prunus salicina* varieties by 9.9 % more than *Prúnus doméstica*. To a greater extent, sugars accumulated in the bark of *Prunus salicina* varieties: Orlovskaya Mechta, Suvenir Vostok and ELS 18476; *Prúnus doméstica*: Vengerka Zarechnaya, Evrazia 21; and *Prunus* × *rossica* Erem.: Vetraz, Zlato Skifov and Kubanskaya Kometa (Fig. 4). By November, in general, the amount of sugars in the bark of annual shoots of the studied plum varieties increased 1.5 times compared to September.

Thus, in the autumn period, during the hardening of plum varieties due to starch hydrolysis, there is an intensive accumulation of sugars, which play a protective role in conditions of low-temperature stress in winter.

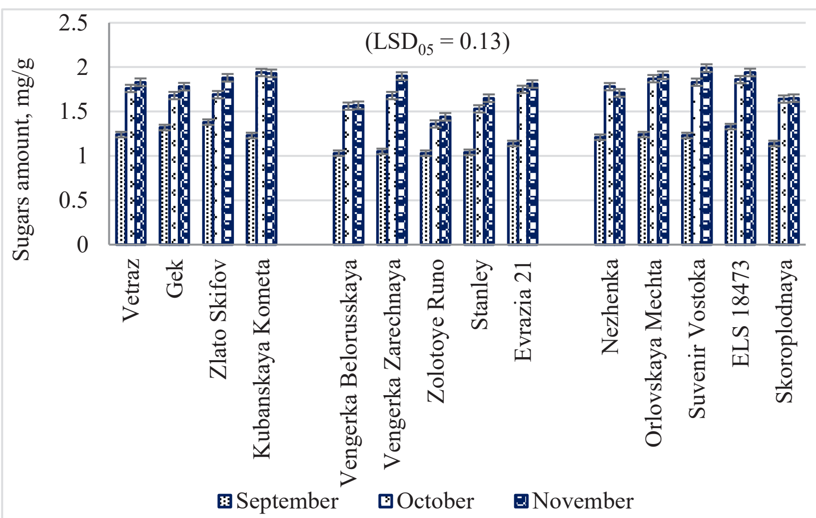

Fig. 4. Sugars content in the bark of annual plum shoots in autumn, %

We have analysed the correlation between physiological and biochemical parameters and meteorological conditions of the autumn period. According to Table 2, a significant negative correlation was found between the minimum and average daily temperature with the content of bound water and the amount of sugars, as well as a significant positive correlation was observed with starch content. A positive correlation was noted between the amount of precipitation and sugar content, as well as a significant negative correlation with starch content was noted. A strong correlation was revealed between the determined physiological and biochemical parameters of plum adaptability during the hardening period in autumn. Shows a strong positive correlation between the content of bound water and the sum of sugars ($r = 0.82$). A high negative correlation was found between the content of bound water and starch content ($r = -0.83$).

Table 2. Correlation coefficient (r) between the content of bound water, starch, sugars in annual plum shoots and meteorological conditions in autumn

Physiological and biochemical indicators of adaptability	Minimal air temperature, °C	Average daily air temperature, °C	Precipitation amount, mm
Bound water content, %	-0,99	-0,93	0,83
Starch content, mg/cm^3	0,97	0,82	-0,99
Sugars content, mg/g	-0,98	-0,83	0,99

4 Conclusion

As a result of the studies it has been found that plum varieties are characterized by an increase in bound water against the background of a decrease in the overall hydration of annual shoots. The bound water/free water ratio increased significantly by the beginning of winter in all varieties. At the same time, a significant increase in the content of low molecular weight osmoprotectors in the bark of annual shoots by the beginning of winter was noted for the varieties of *Prunus salicina* and *Prunus × rossica* Erem. In addition, the varieties of these two species showed greater plasticity of changes in the amount of starch in the tissues of the bark of annual shoots against the background of increased activity of the enzyme amylase. These results suggest that the varieties of *Prunus × rossica* Erem. – Vetraz, Gek, Zlato Skifov, Kubanskaya Kometa and *Prunus salicina* – Nezhenka, Orlovskaya Mechta, Skoroplodnaya, Suvenir Vostoka and ELS 18476 have greater frost resistance in winter, because their adaptive processes were more intense in autumn. Among the genotypes of the *Prúnus doméstica*, Vengerka Belorusskaya and Evrazia 21 should be noted, which showed a higher intensity of metabolic processes during the hardening in autumn.

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