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THE PROCESS OF PHOTOSYNTHESIS AND THE LIGHT REGIME OF SOME SWEET CHERRY CULTIVARS FROM THE NORTH-EAST OF ROMANIA

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Abstract
Sweet cherry has the highest intensity of photosynthesis among all stone fruit crops, thus, studies on the physiological processes of using lighting resources and improving photosynthetic activity provide a theoretical basis for increasing and stabilizing fruit production. The purpose of the present study was to evaluate the photosynthesis process by determining the amount of active photosynthetic pigments and the stomatal conductance at the leaf level, under the aspect of the light intensity at the tree canopy level. The research was carried out over two years (2022-2023) on three sweet cherry cultivars ('Van', 'Andreiaș' and 'Margonia'), in three distinct phenological stages, according to the BBCH scale (65-full flowering, 75-fruit growth and 89-fruit ripening), with samples taken from two areas of the tree canopy: the external, peripheral part and the internal part, close to the trunk. The obtained results revealed a higher content of chlorophyll a in the internal part of the canopy and during the observed phenophases, the total content of chlorophyll pigments (chlorophyll a and b) increased from 11.06 mg/100g F.W in the flowering stage and 12.96 mg/100g F.W in the fruit growth stage, up to maximum values of 25.84 mg/100g F.W at fruit ripening. The stomatal conductance had average values between 6.70 and 12.52 mmol/m²/s and in correlation with the light intensity, significant correlation coefficients (R^2) were obtained: 0.984 at 'Van', 0.978 at 'Andreiaș' and 0.928 at 'Margonia' cultivar. The intensity of the physiological processes varied depending on the light intensity, climatic conditions, cultivar and canopy area.

Keywords: canopy, chlorophyll, leaves, pigments, stomatal conductance.

1. INTRODUCTION

Sweet cherry (*Prunus avium* L.) is a valued and economically important fruit tree species that can be grown in a worldwide range of climatic conditions (Wenden et al., 2017). The sweet cherry is also unique among fruit trees because of its short flowering-to-harvest time, with the time between flowering and harvest being only 65 to 75 days. Thus, a substantial part of the carbohydrates required for the early growth of the crop comes from stored reserves but also from the ability of cherry leaves to photosynthesize in the marginal environmental conditions of early spring commonly found in temperate continental climates (Roper and Kennedy, 1986; Asanica et al., 2014). The physiological metabolism of fruit trees also depends on how the rootstock adapts to the grafted cultivars (Asanica et al., 2015), the genotype of the rootstock influencing the leaf and the content in chlorophyll pigments and the subsequent photosynthetic activity while the concentrations

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of metabolites and the physicochemical characteristics of the fruits are higher depending on the genotype (Perez et al., 1997; Goncavales et al., 2006).
Under natural conditions, leaf functional characteristics show high spatial variability depending on canopy shape (Frak et al., 2002). Microclimatic parameters such as stomatal conductance and light quality and physiological ones change simultaneously with light intensity at the canopy level. The different light environments cause seasonal changes both at the level of the leaves, associated with morphological, anatomical, chemical and physiological characteristics, and at the level of the whole plant mainly related to shoot architecture and biomass allocation patterns (Gonçalves et al., 2008).
According to studies of other stone fruit species, a defining factor that directly affects photosynthesis is the penetration of sunlight into the tree, thus the net photosynthetic capacity of leaves under insufficient light levels is 40 to 80% lower than that of leaves on the periphery or exterior of the canopy (Bondarenko, 2019). Photosynthesis is also influenced by the pigment content of the leaf mesophyll. Chlorophyll *a*, chlorophyll *b* and carotenoids are the main pigments that absorb photosynthetically active radiation, thus, photosynthetic productivity is minimized with the decrease in the concentration of these pigments in the leaves (Viljevak et al., 2013).
The aim of the present study was to evaluate the physiological parameters such as: pigment content and stomatal conductance at leaf level of sweet cherry trees in a classic orchard in correlation with the light intensity both from the periphery of the canopy and from the central part during the whole season of vegetation.

2. MATERIALS AND METHODS

Growing conditions

The study was carried out in the period 2022-2023 in a sweet cherry crop of the Research Station for Fruit Growing (RSFG) Iasi, located geographically in the North-East of Romania (47°20´N and 27°60´E). Weather conditions during the experiment were monitored by an *Agroexpert* system and the data obtained were compared with a database stored for 40 years (1969-2009), used as a multi-year reference value. Thus, during the studied period, the average annual temperature recorded was 12.0°C, +2.4°C higher than the multiannual average (1969-2009), and the total precipitation had values of 363.6 mm with a deviation of – 154.2 mm from multi-year values. The climatic conditions presented highlight the current global aspect of climate change.

Plant material

The biological material used comprises three cultivars of sweet cherry (‘Van’, ‘Andreiaș’ and ‘Margonia’), grafted on rootstocks of *Prunus mahaleb* L.. The trees of the mentioned cultivars were planted at a distance of 5 × 4 m without support or irrigation system and trained as a free palmette. The management of the culture was carried out in accordance with agronomic norms with the specificity of the sweet cherry fertilization technique and phytosanitary treatment (Quero-García et al., 2017).

Experimental protocol

Physiological determinations were performed at three different phenological stages, according to the scale BBCH (Biologische Bundesanstalt, Bundessortenamt and Chemical) of Meier, (2001) at 65 (full bloom); 75 (when fruits are approximately 50% of final size) and 89 (fruit consumption maturity) and in different areas of the canopy: internal (central) and external (peripheral). Leaf samples were collected from randomly selected branches from the full height of the trees.

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The rate of gas exchange through the leaf stomata, an indicator of the transpiration process, was determined by determining the stomatal conductance (gs) with a portable leaf porometer (Model SC-1, Decagon Devices Inc., Pullman, WA) using the method of determination of water vapor flux from the leaf surface to the atmosphere under local conditions presented by Pask and Pietragalla (2012). To obtain an average value, ten readings were performed on leaves from separate shoots at hourly intervals between 09:00 a.m and 07:00 p.m., in the three mentioned phenological stages.
The content of photosynthetic pigments of the type Chlorophyll *a* and Chlorophyll *b* in the leaves was determined by the spectrophotometric method described by Jităreanu (2007). The method consists in testing the light absorption capacity at wavelengths 647 and 663 nm absorbance analyzed by computer-aided spectrophotometer (Specord 210 PLUS UV-VIS Analytik Jena) of the acetone extract (1%) of pigments based on Lichtenthaler's equations (Lichtenthaler H.K., 1987; Welburn A.R., 1994):

$$Ca=12.25\times A663 - 2.79\times A647$$
$$Cb=21.50\times A647 - 5.10\times A663$$
$$Ca+b=7.15\times A663 - 18.71\times A647$$

Light intensity at tree canopy level was determined directly in the experimental field using AccuPAR LP-80 device in all tree height zones, in several time intervals of a day.
The data were statistically interpreted by the Duncan test with multiple intervals at $p \leq 0.05$, using SPSS software version 28 and by the standard deviation (STDEV) and the Pearson coefficient of variation (COVAR) (Botu and Botu, 2010)
To estimate the relationship between physiological determinations and light intensity, the correlation coefficient (R^2) was also calculated and interpreted (Taylor, 1990).

3. RESULTS AND DISCUSSIONS

The physiological response of sweet cherry leaves under natural conditions during the entire growing season revealed in all cultivars a total content of chlorophyll pigments that increased progressively starting from the 65 BBCH stage, where the leaves were at the beginning of their development (11.06 mg/100g), to maximum values at the stage of fruit ripening (25.84 mg/100g) (Table 1). Analyzing the two canopy areas, it is highlighted that the content of chlorophyll *a* has higher values in the internal area, compared to the peripheral part in all three evaluated phenophases, while the content of chlorophyll *b* shows higher values in the eternal part.
Chlorophyll pigments (chlorophyll *a* and chlorophyll *b*) are chromo-lipoprotein complexes involved in the physiological process of photosynthesis. Their presence in higher concentration in leaves helps photosynthetic efficiency (Asanica et al., 2015). Thus, among the studied cultivars, during the advancement in the vegetation stages, ‘Van’ recorded the maximum values during the 65 BBCH (11.77 mg/100g) and 75 BBCH (15.95 mg/100g) stages, while during the fruit ripening (89 BBCH), the maximum values were found at ‘Margonia’ (31.37 mg/100g).
The development of photosynthetic capacity in the sweet cherry was also consistent with its harvesting habits, depending on the time of fruit ripening.
In early spring, although the leaves are young and have limited leaf area and temperatures are lower, trees were still able to produce photoassimilates efficiently to meet the simultaneous demands of fruit and leaf growth. The results obtained were also consistent with other studies (Roper and Kennedy, 1986; Quentin et al., 2013), which indicated that sweet cherry leaves become

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photosynthetically competent early in their development and can thus supplement reserve carbohydrates for growth, early in the season.

Table 1. The content of chlorophyll pigments at the foliar level (RSFG Iasi, 2022-2023, n=3)

Cultivar	Canopy area	Chlorophyll *a*[2]			Chlorophyll *b*[2] (mg/100g)			Total chlorophyll		
		[1]65BBCH	79BBCH	89BBCH	65BBCH	79BBCH	89BBCH	65BBCH	79BBCH	89BBCH
Van	**Intern**	8.66 a	11.56 a	18.04 bc	3.10 ab	4.39 a	6.62 b	11.77	15.95	24.66
	Extern	7.88 b	9.35 bc	16.99 c	3.56 a	3.35 b	5.68 b	11.44	12.69	22.67
Andreiaș	**Intern**	7.67 b	10.16 ab	21.94 a	2.71 b	3.30 b	8.39 a	10.38	13.47	30.32
	Extern	7.45 b	9.50 bc	15.27 d	3.20 ab	2.88 b	5.67 b	10.65	12.39	20.94
Margonia	**Intern**	7.98 b	9.27 bc	22.29 a	3.19 ab	3.10 b	9.07 a	11.17	12.37	31.37
	Extern	7.62 b	8.20 c	18.60 b	3.34 ab	2.73 b	6.51 b	10.96	10.92	25.11
Average		**7.88**	**9.67**	**18.85**	**3.18**	**3.29**	**6.99**	**11.06**	**12.96**	**25.84**
STDEV		**0.43**	**1.12**	**2.77**	**0.28**	**0.59**	**1.42**	**0.51**	**1.68**	**4.16**
COVAR		**5.45**	**11.57**	**14.69**	**8.86**	**17.92**	**20.37**	**4.60**	**12.94**	**16.11**
Min.		**7.45**	**8.20**	**15.27**	**2.71**	**2.73**	**5.67**	**10.38**	**10.92**	**20.94**
Max		**8.66**	**11.56**	**22.29**	**3.56**	**4.39**	**9.07**	**11.77**	**15.95**	**31.37**

1-BBCH-Phenological growth stages (Meier, 2001): 65 (full flowering); 75 (fruit growth); 89 (fruit ripening);
2-Different letters after the number correspond with statistically significant differences for p 5% - Duncan test.

Stomatal conductance, an indicator of gas exchange during the physiological process of transpiration, oscillated in the environmental conditions of the years 2022-2023 from minimum values of 6.7 mmol/m²/s at the exterior canopy area of 'Margonia' in the 65 BBCH stage to maximum values the 89 BBCH stage at intern canopy area also of 'Margonia' cultivar (12.52 mmol/m²/s) (Table 2).

Table 2. The stomatal conductance at the foliar level (RSFG Iasi, 2022-2023, n=3)

Cultivar	Canopy area	Stomatal conductance[2] (mmol/m²/s)		
		[1]65BBCH	79BBCH	89BBCH
Van	**Intern**	8.65 a	7.27 a	10.88 bc
	Extern	7.81 ab	6.93 a	8.20 d
Andreiaș	**Intern**	8.35 a	7.50 a	11.43 b
	Extern	7.28 bc	7.24 a	9.14 d
Margonia	**Intern**	7.88 ab	7.00 a	12.52 a
	Extern	6.70 c	6.83 a	10.25 c
Average		**7.78**	**7.13**	**10.40**
STDEV		**0.71**	**0.25**	**1.56**
COVAR		**9.10**	**3.50**	**15.03**
Min.		**6.70**	**6.83**	**8.20**
Max		**8.65**	**7.50**	**12.52**

1-BBCH-Phenological growth stages (Meier, 2001): 65 (full flowering); 75 (fruit growth); 89 (fruit ripening);
2-Different letters after the number correspond with statistically significant differences for p 5% - Duncan test.

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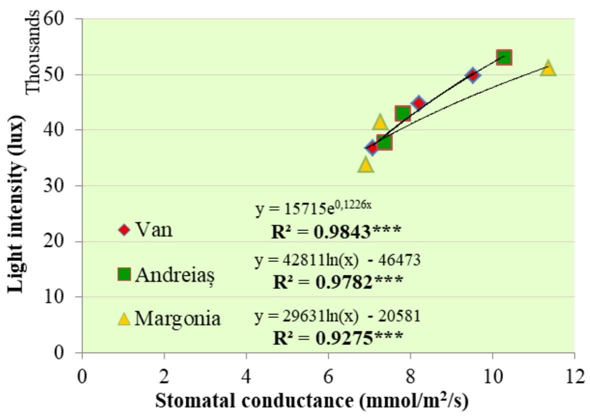
Seasonal changes in stomatal conductance in fruit trees and stomatal responses to dehydration in sweet cherry cultivars thus seem to be influenced by fruit development and ripening, with the highest values of this indicator being in this phenophase. Lower stomatal conductance values may reveal a protective mechanism against water and thermal stress, thus allowing plant water conservation and improving plant water use efficiency (Yoon and Richter, 1990). Variation between cultivars shows statistically significant differences only in stage 89 BBCH, oscillating according to the study of Blanco et al. (2019) depending on environmental conditions, tree architecture and cultivar variability.
The light intensity evaluated during the vegetation period at the level of the canopy did not show major differences, the type of canopy ensuring a uniform distribution of the light of the entire trees. Light intensity during the evaluated stages varied depending on the temperature recorded at the time of their phenological development. The average light intensity values were 43,515.8 lux.
In addition to temperature, light intensity is one of the primary environmental factors affecting plant growth and development, predominantly influencing the photosynthetic chain and the photorespiratory system (Pallardy, 2010). Thus, the correlation between light intensity and stomatal conductance, graphically represented in Figure 1 revealed a positive and a high correlation coefficients at all three cultivars. The relationship of light dependence with the content of chlorophyll pigments (Figure 2) followed the same trend, photosynthesis intensifying with the increase of light intensity, at ‘Van’, a distinctly significant coefficient R^2=0.6412 was obtained, while at ‘Andreiaș’, R^2=0.8778 and at ‘Margonia’, R^2=0.8831, highly significant coefficients were obtained.



**-positive correlation - distinctly significant
***-positive correlation- high significant

Figure 1. Dependence of light intensity to the stomatal conductance

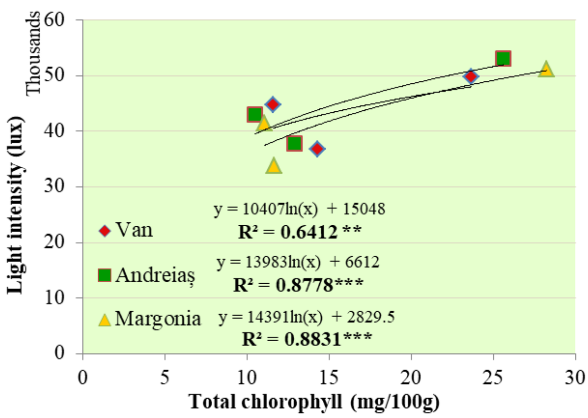


Figure 2. Dependence of light intensity to the total chlorophyll content

The dependence of light variability on stomatal conductance and pigment content increased proportionally with the seasonal change of phenological stages, from flowering to fruit ripening. These results amplify the influence of seasonal climate changes in plant physiological properties that is important in assessing the fluxes of water, energy and CO_2 between fruit trees and the atmosphere. The significant correlations between physiological indicators with light intensity showed that the variables increase simultaneously through an exact linear rule (Ratner, 2009).

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4. CONCLUSIONS

The studied sweet cherry cultivars are useful resources for further research on the physiological processes taking place at the level of the leaf and the influence of light intensity on them.

The most intense photosynthetic activity at the foliar level occurred in the vegetative stage of fruit ripening, which in sweet cherry also overlaps with the beginning of the accumulation of reserve substances and the differentiation of fruit buds for the following year. The ratio of chlorophyll *a* to chlorophyll *b* was 3/1 over the entire growing season.

The samples taken from the intern area of the tree canopy recorded an intensification of both photosynthetic activity and stomatal conductance, compensating for possible light deprivation.

5. ACKNOWLEDGEMENTS

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