

**CHARACTERISTICS OF THE REFLECTION SPECTRA OF GREEN PLANTS IN THE PRESENCE OF ENVIRONMENTAL STRESSES**

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**Abstract:** Reflection spectra demonstrating the optical properties of leaves provide information on reflectance of some specific wavelength. These spectra allow to define the color of the leaves by chromatic coordinates and in addition also the wavelength of the "dominant" color of the leaves. The shape of the reflection spectra of leaves as well as the values of certain ratios exhibit typical changes between analyses leaves demonstrated structural and functional modifications as a result of adaptation to different light environment. The measurements were carried out with three types of leaves (sun, half-shade and shade) of two different pear varieties: Santa Maria and Abbas.

**Keywords:** *emission spectra, fruit trees, plants, reflection spectra; wavelength of the "dominant" color*

### Introduction

Chlorophylls are green photosynthetic pigments which allow plants to get energy from light. In the process of growth plants are exposed to a variety of stressors that directly or indirectly affect the function of photosynthetic apparatus in leaves [1], [6], [7]. High temperature and high light combined with other stresses and pollution reduce the activity of photosynthetic apparatus. Spectral characteristics, fluorescence and reflectance, describing the possible changes and damages to the structure and function of photosynthetic apparatus in the presence of environmental stress, provide data on the quantum conversion of solar and photosynthetic activity in leaves. Plant adaptations to different exposed light environment during their growth affect development of the entire plant and particularly to chloroplasts and their structure, thylakoid arrangement as well as the relative amounts of the photosynthetic pigments, the chlorophylls and carotenoids. Thus these adaptations implicate both structural and functional differences [7], [8]. Sun leaves with their sun chloroplasts (low and narrow grana stacks) possess higher values for the ratio Chl a/b and lower values for the weight ratio total chlorophylls to total carotenoids, ratio  $(a+b)/(x+c)$ , as compared to shade leaves with their shade chloroplasts (broad and high grana stacks) [6]. Within a tree crown there also exist the leaves of the north crown of trees termed blue-shade leaves which differ in their reception of light quality and quantity from sun and shade leaves. These leaves are receiving only blue sky light but never full sun light [8]. In addition, trees possess half-shade leaves which are in the shade during the major part of the day, but that also receive full sunshine for a short period during the course of the day. The purpose of this study is to evaluate the activity of photosynthetic apparatus of some fruit trees in Tirana area in the presence of environmental stresses to that are exposed.

### Material and Methods

**Plants.** Measurements were carried out with leaves selected on three kinds of positions (sun - south part of crown, blue shade - north part and half-shade/shade - inside a tree crown) of two different varieties of pear Santa Maria and Abbas that belong to the group of species

*Pyrus Communis* L pears and family of a rose. Both varieties are characterized by a very good harvest without serious problems of diseases and pests, fit well in dry land without water, field and hill. The Santa Maria pear is a summer variety resistant under suitable conditions for at least three months; ripening period is on June, are characterized by an average grain size and yellow color. Abbas variety is resistant to two months; the grain size is larger than the first variety with color yellow to red, ripening period is late August early September. In contrast to the first variety Santa Maria, variety Abbas has a greater productivity on hill than on field.

**Pigment determination.** Leaf pigments were extracted with 100% acetone in the one circular piece of 9mm in diameter cut from the leaves using a mortar. The pigment extracts were centrifuged for 5 min at 500 X g in glass tubes to obtain the fully transparent extract. The pigment contents, Chl a, Chl b and total carotenoids, were determined spectrophotometrically from acetone extract using the extinction coefficients and equations re-determined by Lichtenthaler [4], [5]. The represented values are the mean of six determinations from six leaves.

**Reflectance spectra.** Leaf reflectance (R) was recorded from upper side of the leaf in a spectral range from 400nm to 800nm with a spectral resolution of 2nm with a spectrophotometer equipped with an integrating sphere attachment [2], [7]. Leaf reflectance spectra were recorded against barium sulphate as a white reference standard. Leaves were placed on black velvet used as a background which has a reflectance less than 0.5% over the spectral range of measurements. Reflectance (R) was represented as the ratio of the radiation intensities reflected by the leaf sample and the white standard respectively. The leaf spectra were taken in the intercostal fields between the larger leaf veins. These spectra represent an integrated signal over several square centimeters.

**Colorimetry.** Evaluation of the visual impression of a leaf sample was assessed by the chromaticity coordinates in the CIE 1931 color space which allow defining quantitative links among wavelengths in the electromagnetic visible spectrum and physiological perceived colors in human color vision [2]. The coordinates x and y, which define a visual color in the CIE 1931 color space chromaticity diagram, were determined using the reflectance data and the color matching functions for daylight illumination (D65). The brightness (values between 0 = dark and 100 = completely bright), the dominant wavelength (the wavelength characteristic for the color of the sample determined by the intersection point with a curved outer boundary line of the line connecting the achromatic point, i.e., "white" with  $x = y = 0.33$ , and the detected color point), and the color saturation (percentage of distance of the color point between the achromatic point and the boundary line: 100% at the spectrum locus, 0% at the achromatic point) were determined too.

## Results

**Photosynthetic pigments.** For leaves of Santa Maria and Abbas pear variety selected in the period from May to October of 2015, can be demonstrated that the contents of Chl a, Chl b, Chl (a+b) and carotenoids are higher on May, as the period with optimum conditions compared to June that can be considered a stress period due to the highest solar radiation and higher temperatures. Also the photosynthetic pigment contents of chlorophylls and carotenoids on the period May-June in both varieties, Santa Maria and Abbas, represented

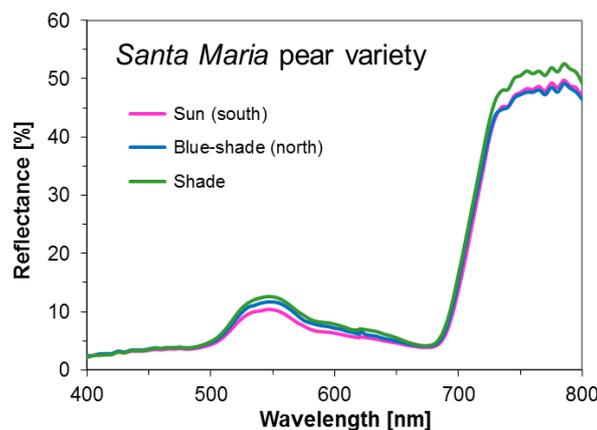
higher values in sun leaf (south part of crown tree) than other leaf types (Tab. 1).

**Table 1.** Levels of Chl a+b and total carotenoids (x+c) per leaf area unit as well as the pigment ratios Chl a/b and chlorophylls (a+b) to carotenoids (a+b)/(x+c) between sun, blue-shade, shade/half-shade leaves of *Santa Maria* and *Abbas* variety trees. Mean values of 6 determinations per leaf-type.

Leaf-type	Chl a+b (mg dm <sup>-2</sup> )	Carotenoids (mg dm <sup>-2</sup> )	Chl a/b	(a+b)/(x+c)
<i>Santa Maria</i> - May				
Sun	6.85 ± 0.20	1.32 ± 0.22	2.68 ± 0.09	5.17 ± 0.37
Blue-shade	5.91 ± 0.26	1.09 ± 0.05	2.43 ± 0.13	5.44 ± 0.36
Half-shade/shade	5.52 ± 0.35	1.03 ± 0.09	2.36 ± 0.11	5.53 ± 0.23
<i>Abbas</i> - June				
Sun	5.92 ± 0.50	1.04 ± 0.11	2.62 ± 0.26	5.25 ± 0.74
Blue-shade	4.93 ± 0.46	1.02 ± 0.15	2.38 ± 0.43	5.47 ± 0.31
Half-shade/shade	4.52 ± 0.35	0.73 ± 0.09	2.35 ± 0.21	5.97 ± 0.42

The ratios of the photosynthetic pigments, Chl a/b and (a+b)/(x+c), that reflect the light adaptation of the photosynthetic apparatus (Lichtenthaler 2013) shown different values in the three leaf types. In sun leaves the mean values of the ratio Chl a/b are higher as compared to blue-shade and shade leaves (Tab. 1). Sun leaves displayed lower values of the ratio (a+b)/(x+c) as compared to two other leaf types (Tab. 1). These differences can be related to the effect of stress in the period of June as well as to the specific characteristics of analysed varieties.

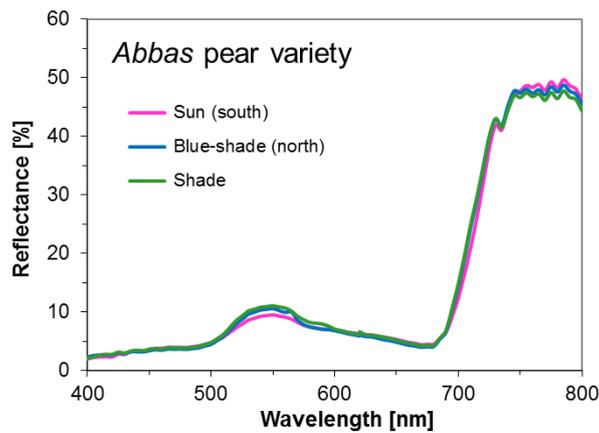
**Reflectance spectra.** Reflectance spectra of the three types of leaves of both pear varieties exhibited a higher reflectance between 500nm and 650nm, in the green-to-orange range of the spectrum, and mainly at wavelengths between 680nm and 740nm in the near infra-red. In addition reflectance spectra exhibited a low reflectance between 400nm and 500nm in blue part of visible spectra and also near 680nm in red part of visible spectra (Fig. 1, Fig. 2). The observed variations correspond to the absorption region of the in-vivo chlorophyll bands.



**Fig. 1.** Reflectance spectra of the sun (south part), blue-shade (north part) and shade/half

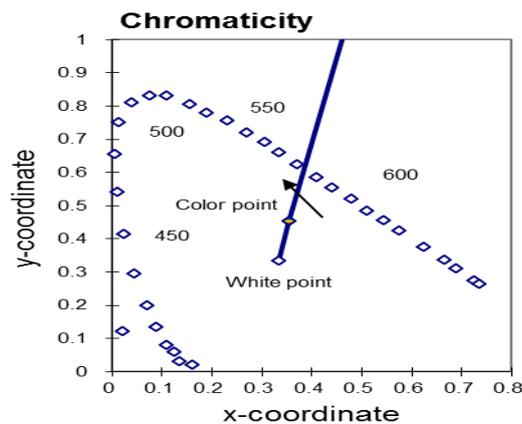
shade leaves of *Santa Maria* pear variety on May (upper side). Mean of 6 reflectance spectra per leaf-type.

The reflection spectra of two varieties exhibit the highest value in the green-orange range of the spectrum of shade leaves compare to two other leaf types. Also, could be observed a blue shift of the “red edge” (inflection point of the rise of signal at wavelengths between 680nm and 740nm) towards shorter wavelengths to the shade leaves. These variations among three types of analysed leaves are related to the chlorophyll content being lower in shade leaves and higher in sun leaves (Tab. 1). The higher values of reflectance in the green-orange range of the spectrum detected in the leaves of *Santa Maria* variety compared to the *Abbas* variety could be explained by the differences on chlorophyll content too. Higher signals of reflection spectra of shade leaves of *Santa Maria* pear variety on May (Fig. 1), a period with optimal growth conditions, could be related to the higher leaf water content as comparing to the sun and blue-shade leaves.



**Fig. 2.** Reflectance spectra of the sun (south part), blue-shade (north part) and shade/half shade leaves of *Abbas* pear variety on June (upper side). Mean of 6 reflectance spectra per leaf-type.

**Colorimetry.** The reflectance spectra of the leaf samples of three leaf types on both pear varieties were used to define the color as x and y chromaticity coordinates in the CIE 1931 color space (Fig. 3).



**Fig 3.** Calculation of chromaticity parameters by the leaf reflection spectra and CIE

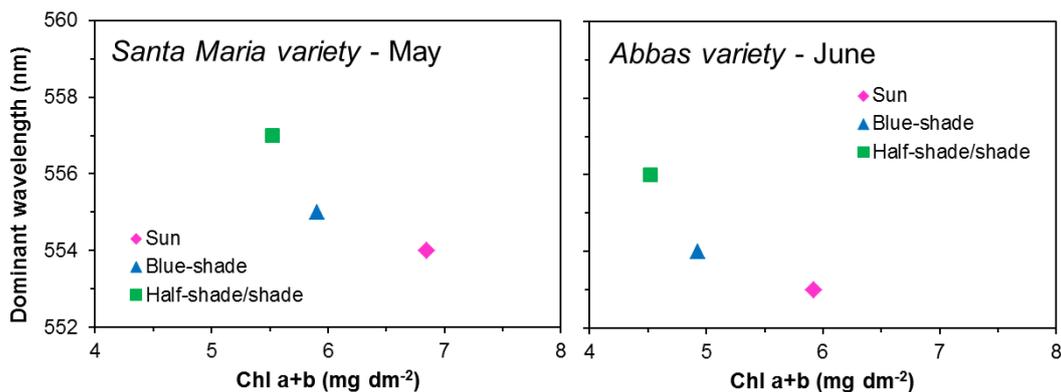
chromaticity color diagram using line centered from white point (0.33, 0.33): chromaticity coordinates  $x = 0.35$ ;  $y = 0.45$ , brightness  $Y = 10.84$ , dominant wavelength 561 nm and color saturation 42.7%.

Colorimetric data of two varieties displayed differences showing grater values for Abbas variety than Santa Maria variety (Tab. 2) following the variations on chlorophyll content in these two varieties actually being higher on leaves of Santa Maria variety and lower on leaves of Abbas variety (Tab. 1). Also, differences can be detected on CIE parameters like the brightness, the dominant wavelength and the color saturation of each variety depending by leaf type (Tab. 2). The values of these three parameters increased while the pigment content decreased from sun leaf to shade leaf.

**Table 2.** Colorimetric determination according CIE 1931 for the leaf samples: sun, blue-shade, shade/half-shade leaves of *Santa Maria* and *Abbas* variety trees. Mean from 6 reflectance spectra per leaf-type.

Leaf-type	x-coordinate	y-coordinate	Brightness Y	Dominant wavelength (nm)	Color saturation (%)
<i>Santa Maria</i> – May					
Sun	0.33	0.44	7.65	554	31.5
Blue-shade	0.34	0.44	8.62	555	34.2
Half-shade/shade	0.34	0.45	9.30	557	38.1
<i>Abbas</i> – June					
Sun	0.33	0.44	7.83	553	32.1
Blue-shade	0.33	0.44	8.66	554	33.7
Half-shade/shade	0.34	0.45	9.43	556	34.5

In the cases of Santa Maria variety on May and Abbas variety on June, where content of Chl a+b represented higher values in sun leaves (south part of crown tree) and lower values on shade leaves (inside a tree crown), can be noticed a decrease of the dominant wavelength with increasing of chlorophyll content of different type of leaves (Fig. 4).



**Fig 4.** Decrease of the dominant wavelength with increasing of chlorophyll content of different type of leaves, from shade leaf to sun leaf, of the Santa Maria variety on May and Abbas variety on June.

### Conclusions

Photosynthetic pigments content of leaves selected from two pear varieties, at Tirana area (Zhurje) represented higher value on optimal growth conditions on May, adequate moisture and suitable irradiance compare to June characterize by stress conditions such as higher light, higher temperature and dry. Pigment contents on the period May-June in both analysed varieties, Santa Maria and Abbas, displayed higher values in sun leaf (south part of crown tree) than other leaf types. The values of the pigment ratio Chl a/b were higher while of the ratio  $(a+b)/(x+c)$  are lower in sun leaves as compared to blue-shade and shade leaves in both varieties.

The reflection spectra of two varieties exhibited the lowest value in the green-orange range of the spectrum of sun leaves compare to blue-shade and shade leaves and a blue shift of the “red edge” towards shorter wavelengths to the shade leaves demonstrating that the reflectance signals of leaves are determined by leaf pigment content and pigment absorption properties. The observed variations between analysed pear varieties, Santa Maria and Abbas, could be explained by the differences on chlorophyll content too.

Colorimetric parameters of two varieties exhibited grater values on Abbas variety than Santa Maria variety following existed variations on chlorophyll content being higher on leaves of Santa Maria variety and lower on leaves of Abbas variety. The values of CIE parameters like the brightness, the dominant wavelength and the color saturation of each variety increased while the pigment content decreased from sun leaf to shade leaf displaying dependency by leaf type.

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