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THE DYNAMICS OF FOLIAR ASSIMILATORY PIGMENT FRACTION OF *HIPPOPHAE RHAMNOIDES* L. VARIETIES GROWN UNDER CONTROLLED CONDITIONS

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INTRODUCTION

Foliar chlorophyll pigments, represented mainly by chlorophylls a and b, are metallo-porphyrins, which are complex combinations of magnesium and iron porphyrins. Generally, in the tissues of green plants chlorophyll a is found in higher concentrations compared with chlorophyll b. So it is considered that the ratio chlorophyll a/ chlorophyll b is about 3:1 or even 3 : 2 (Burne and others, 1977).

In the individual development of plant chlorophylls are being biosynthesized and are being accumulated in leaves in different ways. In the leaves of herbaceous plants, except grass plants, and in the leaves of the woody plants, chlorophylls are being biosynthesized and are being accumulated continuously, reaching a maximum value during flowering, after which decrease quantitatively, their formation and accumulation in leaves being directly dependent on the intensity of plant metabolism (Tarragon, 1992).

Sea buckthorn (*Hippophae rhamnoides* L.) is a highly branched and thorny shrub, spread in Romania from sand to mountain coasts, making sometimes groves and thick bushes and large bushes. The leaves are linear lanceolate, from green-gray on the top to silvery white on the bottom.

The copies of this shrub are less demanding in terms of soil and rainfall, loving high light, growing generally in places where other plants cannot vegetate (eroded hill sides, salty land, rocky areas and also plains of the rivers).

The objective of this paper is to determine the foliar pigments chlorophyll fractions with the specific components (chlorophyll a, chlorophyll b) in five varieties of sea buckthorn (Auras, Silvia, Serpent, Tiberius and Victoria), from May to September, 2009. Our choice of study is based on the fact that the chlorophyll pigments represent the main antenna of the superior plants to capture and transform the light energy into chemical energy incorporated into biosynthesized products during the process of photosynthesis.

MATERIAL AND METHODS

The analyzed plant material consists of leaves harvested monthly from late spring to early autumn (from May to September) in 2009, from five varieties of sea buckthorn cultivated in Romania: Auras, Silvia, Serpent, Tiberiu and Victoria. The leaves were taken from the shoots of one year from the bushes grown under controlled conditions in the greenhouse located at SC FRUCTEX S.A. Bacau (20-22°C temperature, drip irrigation every 30 minutes, warm sandy soil). The plant material randomized collected from shoots from May to September 2009, was analyzed through biochemical tests in order to determinate chlorophylls a and b.

The determination of the chlorophyll pigments is made by Mayer-Bertenrath spectrophotometric method, modified by Ştirban and Fărcuş (Hager and Meyer-Bertenrath, 1966). The method consists, in principle, in the extraction of the assimilating pigments in acetone in several stages, a process followed by their spectrophotometric determination, at wave lights proper to the maximum absorption for each pigment separately. The results represent the average of three determinations and are reported to the quantity of fresh plant material used to determine the biochemical indicator investigated.

RESULTS AND DISCUSSIONS

The environmental factors such as temperature, radiation, water availability strongly influence the bioactive compounds from plants (Weston and Barth, 1997).

The measurements about the content of assimilating pigments assessed within May to September 2009 at the variety Auraş, show an upward dynamic of the process of biosynthesis of chlorophylls a and b (fig. 1).

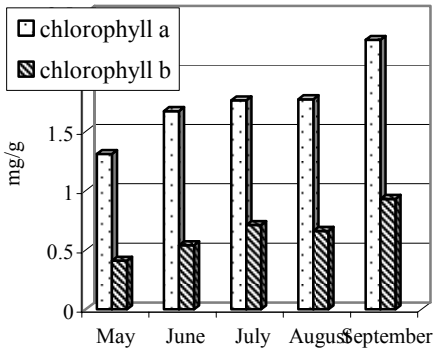

Fig. 1. The variation of the content in assimilating pigments in leaves collected from the variety **Auraş**

We note that during July and August there are not recorded significant differences, behaviour which could be explained by the fact that during this period of time the temperature, radiation and availability of water didn't change so to significantly influence the process of photosynthesis carried out by the unit leaf plants tested, expressed practically by biochemical the changes of the biochemical indicators studied.The amplitudes regarding chlorophyll a is between a minimum of 1.31 mg/g recorded in May and a maximum of 2.27 mg/g in September. On the biosynthesis of chlorophyll b, the minimum and maximum value are found in the same months as in the case of chlorophyll a, the absolute values being of 0.41 mg/g and 0.93 mg/g.

The analyses performed for the leaves collected from the variety Silvia regarding the chlorophyll a content has a similar aspect of the variation curve with the variety Auraş, except that the amplitudes of values, particularly in May and June are reduced (fig. 2).

Minimum value recorded in May is 0.95 mg/g and a maximum of 2.12 mg /g is recorded in September.

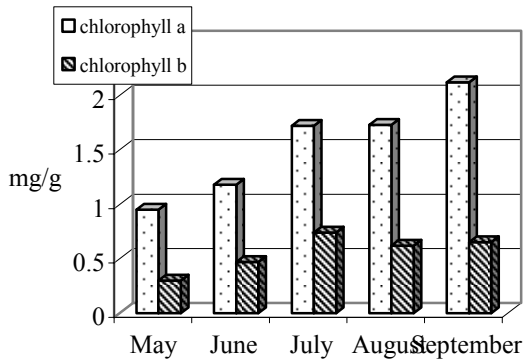

Fig. 2. The variation of the content of assimilating pigments in leaves collected from the variety **Silvia**

The amount of chlorophyll b increases up to July, when the biosynthetic capacity is maximum (0.74 mg/g), then decreasing slightly, it stabilizes at around 0.63 mg/g in August and September.

Evaluating the results of the dynamics of chlorophyll a biosynthesis at the variety Serpenta indicates a minimum value recorded in May (0.84 mg/g), and then in June the biosynthetic capacity increases by about 70%. In July and August the amount of chlorophyll a is constantly higher than in May with 90%.

The greatest amount of chlorophyll a is recorded in September when, compared with May, the amount of pigment increases by 109%. The curve resembles the varieties discussed above (fig. 3).

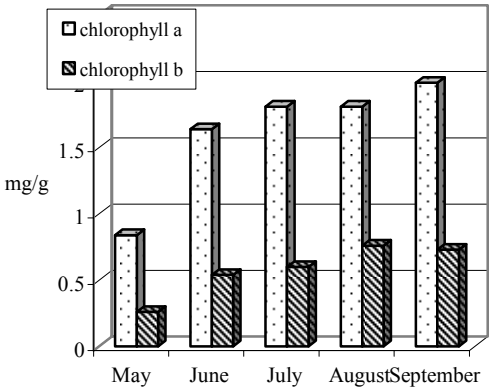

Fig. 3. The variation of the content in assimilating pigments in leaves collected from variety **Serpent**

Examined during the five months the amount of chlorophyll b increases, noticing the quantity of this fraction doubled in June (0.54 mg/g) compared to May (0.24 mg/g), and the biosynthetic capacity records unimportant variations in the continuation of the investigated time. The content of the chlorophyll a pigments of the variety Tiberius has a dynamic different from that of varieties Auras, Silvia or Serpent. Analyzing the graphical representation of the results achieved we notice the reduction of the biosynthetic capacity in the first investigated period of time, so that in June it is recorded the lowest value of the investigated concentration indicator (1.22 mg/g). The trend highlighted by our results for the coming months is generally to increase the capacity of biosynthesis of chlorophyll a.With no significant differences in amplitudes, the recorded chlorophyll b content falls in the values 0.40 mg/g - 0.59 mg/g (fig. 4).

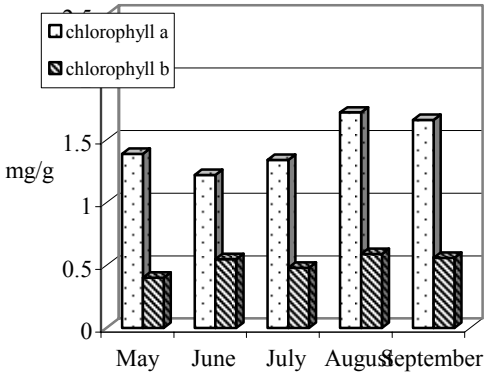

Fig. 4. The variation of the content in assimilating pigments in leaves collected from variety **Tiberius**

The graphical representation of the dynamics of the chlorophyll a content of the variety Victoria shows a general trend in increasing in the process of biosynthesis (fig. 5). It is noticeable the result of the assessment made in July, when the chlorophyll a content has maximum value (2.03 mg/g).

Regarding the content of chlorophyll b, up to July, the biosynthetic capacity increases with a constant rate constant and for the further investigated period the biosynthesis capacity of this pigment reduces.

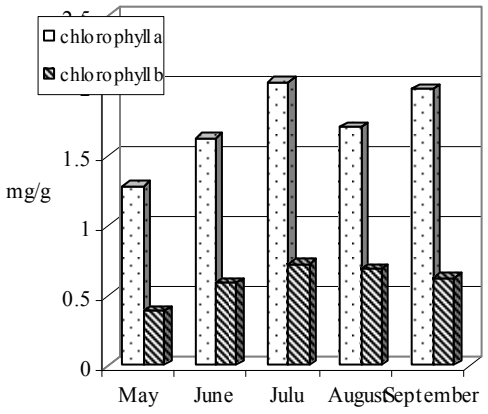

Fig. 5.The variation of the content in assimilating pigments in leaves collected from variety **Victoria**

Thus, the chlorophyll is continuously renewed in plant leaves, in the range of 2 - 3 days at a rate of 40% (Burzo and others, 2004).

The relationship between chlorophyll a and b is a parameter that provides information on the amount of light received by plants hence its growth and development.

At the superior heliofile plants, the chlorophyll a predominate quantitatively in leaves compared with chlorophyll b and when the ratio chlorophyll a/chlorophyll b is in favour of chlorophyll a, the plant is considered directly light-loving, presenting practically an exhibition of favourable light.

The evolution of the ratio chlorophyll a / chlorophyll b at the five varieties of sea buckthorn recorded values between 2.2 and 3.4, always in favour of chlorophyll a, thus showing that the varieties of Hippophae rhamnoides studied prefer sunny places (table 1).

The results we obtained are in agreement with the special literature, which states that at the superior plants the quantitative predominance of chlorophyll a, compared with chlorophyll b, shows gives to the ratio chlorophyll a /chlorophyll b values of approximately 3 : 1 or 3 : 2, giving indications on optimum lighting level for a good photosynthetic activity and hence their growth and development (Burne and others, 1977); it is specified in this direction the fact that the ratios chlorophyll a /chlorophyll b reach higher values in the leaves of heliofile plants compared with those of ombrofile plants (Murariu, 2002).

Table 1. The dynamic of the ratio chlorophyll a/chlorophyll b in leaves of seabuckthorn dynamically harvested, at different varieties

Harvest time/ varieties of seabuckthorn	May	June	July	August	September
Auraş	3,1	3,0	2,4	2.6	2.4
Silvia	3,1	2,5	2,3	2.7	3.2
Serpenta	3,2	3,0	3,0	2.3	2.7
Tiberiu	3,4	2,2	2,7	2.9	2.9
Victoria	3,3	2,7	2,7	2.4	3.1

The highest values of the ratio chlorophyll a/chlorophyll b of the examined specimens is found in May, at the beginning of the vegetation period of plants, a stage of the development process of growth of leaves and shoots (the range of biosynthesis and accumulation of chlorophylls in the chloroplast).

CONCLUSIONS

The average content of chlorophyll pigments of the studied varieties of Hippophae (leaves harvested from shoots of one year), as a biochemical parameter of interpretation of the intensity of the photosynthesis process, presents specific quantitative variations for each studied variety.

We find similar dynamics but different amplitudes of the content of chlorophyll pigments, which is why we believe that having similar behaviour in terms of the studied varieties: Auraş and Serpenta, on one side, and Silvia and Victoria, on the other side.

The variety Tiberius is individualized from the point of view of the ability of biosynthesis of the analyzed chlorophyll pigments. In general, it is remarked an intensification of the processes of biosynthesis of the content of chlorophyll pigments from May to September.

The ratio chlorophyll a/chlorophyll b exceeds the value 3 to all the varieties in the first month of our experimental model, moment that signifies the beginning of the period of vegetation of seabuckthorn, of leaves and shoots growth, namely the period of biosynthesis and accumulation of chlorophylls in the chloroplast. Furthermore we notice a reduction of the ratio values in all the studied varieties, with the tendency to return towards the end of time that ranges the experiment.

Through the values of the ratios chlorophyll a /chlorophyll b the studied sea buckthorn varieties prove to be loving of well - light places.

ABSTRACT

The content and the composition of the bioactive compounds in plants depend on family, genus, species, genotype, climatic conditions, soil quality, harvest time etc..

This paper presents the results of a study about biosynthesis and the accumulation of chlorophylls a and b in leaves collected from five varieties of sea buckthorn (*Hippophae rhamnoides* L.) in dynamic. Our results show an increase in the biosynthetic process of foliar chlorophyll pigments, and the values of the ratio chlorophyll a / chlorophyll b strengthen the description of varieties studied as heliofile varieties.

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