

Original Research Article

<https://doi.org/10.20546/ijcmas.2023.1212.015>

## The Effect of Soil Salinization on Quantitative Indicators of Photosynthetic Pigments of Vicia

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### ABSTRACT

#### Keywords

Vicia, pigments, chlorophyll (Chl), carotenoid (Car), factor analysis, soil salinity level, salt resistance

#### Article Info

##### Received:

18 October 2023

##### Accepted:

22 November 2023

##### Available Online:

10 December 2023

This article contains information on the effect of soil salinization on quantitative indicators of photosynthetic pigments of plants. The hairy vicia plant (*Vicia villosa* Roth) was chosen as the object of research. This plant has been studied in conditions of weak, medium and highly saline soils. The results showed that an increase in soil salinity led to a decrease in the amount of chlorophyll (Chl) «a», an increase in the content of chlorophyll (Chl) «b» and carotenoids (Car). It was found that during the growing season, the leaves of plants in the blooming phase contain a large amount of pigments. In slightly saline soil, the content of chlorophyll «a» in plant leaves was 5.42 mg/g, and in highly saline soil -4.88 mg/g. The content of (Chl) «b» increased from 1.89 mg/g to 3.19 mg/g, respectively. This is exactly the result observed for carotenoids (Car), recording an increase of 1.69 mg/g from 1.45 mg/g. When determining the amount of photosynthetic pigments in plants in saline soil conditions, it was considered advisable to use leaves during the blooming period.

### Introduction

Photosynthetic plant pigments are involved in the formation, transmission and conversion of sunlight into internal energy. Increasing the function and intensity of photosynthetic pigments in plants, with the help of which it becomes possible to more effectively absorb moisture and mineral nutrients in the soil, which, in turn, helps to increase plant

productivity. Photosynthetic pigments play an important role in the study of photosynthesis at the molecular, ecosystem and biosphere levels.

According to Dymova and Golovko (2019), 300 million tons of chlorophyll are renewed three times a year and 100 million tons of carotenoids are synthesized. If the number of chlorophylls in one chloroplast of light-loving plants is 1-2 x 10<sup>9</sup>, then

it is noted that plants growing in the shade have five times less. R.E. Glick and A. Melis noted that the minimum amount of chlorophylls for the effective functioning of the photosynthesis apparatus is 132 molecules, and that their number is influenced by physiological and environmental conditions of the external environment (1).

The activity of photosynthetic pigments is directly related to sunlight. The results of studies to determine the effect of light on the amount of chlorophylls carried out on barley showed that the amount of chlorophyll «a» in the control was 0.590 mg/g, in a leaf aged in the dark. In 42 hours, its amount was -0.266, and in 72 hours - 0.181 mg/g and the chlorophyll index «b», respectively: 0.190; It turned out to be 0.146 and 0.102 mg/g (2). From these data, it can be seen that the lack of sunlight affected the plant as a stress factor.

The amount of photosynthetic pigments is influenced by many factors, including heavy metals. According to the definition given in Lobkova's research, the presence of  $\text{Ni}^{2+}$ ,  $\text{Co}^{2+}$ ,  $\text{Cu}^{2+}$ ,  $\text{Pb}^{2+}$  acetates in the cultivation medium in all concentrations affects the quantitative and qualitative characteristics of chlorophylls «a» and «b», which indicates a violation of the photosynthesis process. Data on the change in the ratio of chlorophylls «a/b» allow us to conclude that chlorophyll «a» degrades in relation to chlorophyll «b» when exposed to metals (3).

However, in vitro model experiments have shown that a number of carotenoids, for example, astaxanthin, are capable of forming complex compounds with metals such as  $\text{Ca}^{2+}$ ,  $\text{Cu}^{2+}$ ,  $\text{Pb}^{2+}$ ,  $\text{Zn}^{2+}$ ,  $\text{Cd}^{2+}$  и  $\text{Hg}^{2+}$  (4). The formation of a metal-chelate complex not only changes the absorption spectrum of the carotenoid, but also increases its ability of scavenging (intercept) free radicals (5).

The chlorophyll content in the dry mass of plants ranges from 1.0 to 17 mg/l and, as noted, is 1.5 times higher in herbaceous plants compared with trees. It was noted that the ratio of carotenoids to

chlorophylls increases as we move north (6). The chlorophyll content in plant leaves is twice as high as in their stalk (7).

It was found that the amount of pigments in plants depends on the type of plant, climatic conditions, temperature, environmental situation, and genetic characteristics. It was found that the amount of chlorophylls and carotenoids in desert vegetation is 1.5-2 times less than in the forest habitat.

According to the authors, it was noted that determining the level of chlorophyll is one of the important criteria determining whether plants are adapted to life in nature. In particular, it was noted that the determination of chlorophyll level plays an important role in the diagnosis of plant adaptability (8; 9; 10).

The amount of carotenoids is one of the important criteria determining the resistance of plants to stressful conditions (11). It was recognized that the selection of genotypes based on the number of photosynthetic pigments is an important factor in increasing plant productivity (12).

An increase in the amount of carotenoids leads to an increase in the plant's resistance to stress factors. Carotenoids are an essential component of the pigment systems of all photosynthetic organisms (13; 14). Carotenoids occur both in the free state and in the form of glycosides; they are able to bind non-covalently to proteins and membrane lipids (13; 15) It has been established that carotenoids are important antioxidants that limit the level of free radicals and reduce Lipid peroxidation processes, which inhibits aging and loss of seed viability (16; 17; 18).

The *antioxidant* function of carotenoids is that they are able to prevent damage caused by the formation of triplet chlorophyll and singlet oxygen (19; 20). Also, carotenoids serve as substrates for the synthesis of Abscisic acid (5). It is noted that quantitative and qualitative indicators of photosynthetic pigments are among the effective

indicators for assessing the physiological state of plants and the activity of the photosynthetic apparatus (21).

It can be seen from the above information that the amount of photosynthetic pigments depends on the biological characteristics of plants and external factors. The sensitivity of pigments to stressful conditions is especially important when choosing and evaluating plants. This is important when choosing salt-tolerant plants.

Since soil salinization acts on plants as a stress factor, reducing productivity by 10-20% in slightly saline soils; at average salinity - by 20-50%; and in severe salinization it decreases by 50-80% (22).

Based on this, it is of scientific and practical importance to determine the amount of photosynthetic pigments of plants in saline soil conditions. Leguminous crops occupy an important place in this place. One of the important biological properties of these crops is associated with increasing soil fertility and improving its physical condition.

In studies conducted by researchers, licorice (*Glycyrrhiza glabra* L.) is noted among legumes as a phytomeliorant, salt-resistant, soil fertility-enhancing crop (23; 24). Similar studies were conducted with hairy vicia (*Vicia villosa* Roth) and its tolerance to salinity was recognized (25). The Mirzachel-1 variety, created by individual selection in conditions of soil salinization from this species, is nutritious, it is recommended for planting in saline soil as a siderate (26; 27).

The main reason for choosing vicia was salt resistance and the amount of photosynthetic pigments in the ontogenesis of a saline environment was not studied. Based on this, this study was conducted, since the quantity and quality of photosynthetic pigments in plants under stress is an important criterion determining the adaptability of plants to the external environment, including the level of soil salinity.

## **Object and methods of the research**

The plant of the hairy vicia (*Vicia villosa* Roth) of the Mirzachel-1 variety was chosen as the object of the experiment. The experiment was conducted in an experimental field with weak, medium and highly saline soils located on the territory of Gulistan State University. Great importance was attached to the amount of dry salt residue to determine the degree of salinity of the soil of the experimental site. The amount of chlorophylls («a, b») in weakly, medium and highly saline soils of the vicia plant was determined during the phenological phases. To quantify chlorophyll, about 1.0 g (exact weight) of crushed raw materials were placed in a heat-resistant flask with a 100 ml reflux refrigerator and extracted with 96% ethyl alcohol (30 ml each) in a boiling water bath for 30 minutes. The extraction was repeated two more times. The resulting extracts were filtered into a 100 ml volumetric flask and brought to the mark with 96% ethyl alcohol after cooling. 5 ml of the resulting extract was transferred to a measuring flask with a capacity of 25 ml and brought to the mark with 96% ethyl alcohol. The optical density was measured using a SF-2000 spectrophotometer at 470, 649 and 664 nm. The comparison solution was 96% ethyl alcohol (28). The obtained primary data on the amount of chlorophyll were statistically analyzed using the SPSS-17 program. On the basis of this program, arithmetic averages of quantitative indicators and factor analysis were performed (29).

## **Results of the research and their discussion**

Photosynthetic pigments are important for plant growth and development. Scientific sources note that the level of soil salinity caused a violation of the structure of chloroplasts. It has been found that chlorosis occurs in plant leaves due to soil salinization as a result of chlorophyll hydrolysis by chlorophyllase. Therefore, it is very important to preserve the supply of chlorophyll in plants in saline soil, since they are involved in the absorption of light energy and its conversion into chemical energy of organic matter. According to the literature, it is

known that 1 g of chlorophyll binds about 2 thousand kcal of solar energy per year, which is equivalent to about 0.5 kg of dry biomass. It has been recorded that under extreme conditions, the levels of chlorophyll «a» are more fragmented compared to chlorophyll «b» (30; 31).

Primary data on the effect of soil salinity on the amount of photosynthetic pigments in the plant are presented in the table (Table 1). Analyzing the results obtained, it can be seen that the amount of chlorophyll «a» in the leaves of the vicia plant decreased significantly with increasing soil salinity. This situation was observed at all phenological phases. The amount of chlorophyll «a» in slightly saline soil was 2.89 mg/g, in medium saline — 2.83 mg/g, in highly saline — 2.60 mg/g. The amount of chlorophyll «b» was 1.58 mg/g in slightly saline soil, 2.07 mg/g in medium saline soil and 1.71 mg/g in highly saline soil. The results obtained are consistent with the data of the authors (32), who found a decrease in the chlorophyll «a/b» ratio in lichens when exposed to heavy metals and suggested that the decrease in chlorophyll a content can be explained by the conversion of chlorophyll «a» to chlorophyll «b» as a result of oxidation of the methyl group of ring II to aldehyde when exposed to metals.

It is known that chlorophyll «a» partially and chlorophyll «b» are completely located in the antenna complex of the photosystem, and their share is recorded as 55-65% of the complex. It follows that the ratio of chlorophyll «a» to «b» is one of the important indicators. According to our data, in conditions of slightly saline soil, the ratio of chlorophyll «a/b» was equal to 1.83, 1.59 in medium saline soil and 1.50 in highly saline soil. From these data, it can be seen that an increase in soil salinity led to a decrease in the ratio of chlorophylls. Carotenoids are important components of photosynthetic pigments, are involved in the process of light absorption and perform an important function in protecting the photosynthetic apparatus. In recent years, studies related to photosynthetic pigment have interested

many researchers (33; 34). Carotenoids are one of the important criteria determining plant resistance to stress. It was noted that they can be used to determine the resistance of plants to stressful conditions (for example, drought) (35), especially during long-term storage of fruits, the amount of carotenoids contained in the leaves serves to assess their condition (36). In conditions of slightly saline soil, the amount of carotenoids was 0.71, in moderately saline soil -0.82, and in highly saline soil - 0.86 mg/g. From these data, it can be seen that an increase in soil salinity also led to an increase in the amount of carotenoids. This condition can be explained by an increase in the content of carotenoids under stressful conditions.

It was under stress that the increased level of carotenoids was also noted by other researchers (6; 11). Thus, an increase in the level of soil salinity affected plants under stressful conditions, as a result of which the number of carotenoids increased. The ratio of chlorophylls and carotenoids determines the carotenoid compound. From the data shown in the table, it can be seen that an increase in the level of soil salinity led to a decrease in the ratio of carotenoids and chlorophylls. The equivalent of 8.37 mg/g in slightly saline soil was 7.79 mg/g in medium saline soil and -6.36 mg/g in highly saline soil. The effect of the soil salinity level on the amount of pigments in % is shown in Figure 1.

The content of chlorophyll «a» in highly saline soil decreased by 10.29% compared with slightly saline soil, and the content of chlorophyll «b» in highly saline soil increased by 8.22%. This result was recorded with respect to carotenoids and it was noted that in highly saline soil, their content increased by 20.01% (Fig. 1).

The results of a factor analysis conducted to determine the effect of soil salinity on the amount of photosynthetic pigments showed that (Table 2) the factor load for chlorophyll «a» in slightly saline soil was 0.994, for chlorophyll «b» - 0.991, for the sum of chlorophylls - 0.994, and for the sum of carotenoids - 0.993.

**Table.1** The effect of soil salinity on the amount of photosynthetic pigments in the vicia plant

Degree of soil salinity	Chl«a»	Chl«b »	Chl«a/b»	Chl«a»+ «b »	Carotenoids	Chl/ Carotenoids
<b>Weak</b>	2.89±0.56	1.58±0.31	1,83±0.05	4.48±0.88	0.71±0.15	8.37±1.38
<b>Minimum</b>	0,02	0,01	1,12	0,03	0,08	0,38
<b>Maximum</b>	8,26	4,98	2,33	13,24	2,18	28,56
<b>Medium</b>	2.83±0.55	2,07±0.52	1,59±0.09	4.90±0.95	0,82±0.17	7,79±1.30
<b>Minimum</b>	0,01	0,01	0,19	0,02	0,08	0,25
<b>Maximum</b>	8,12	11,00	2,25	13,23	2,75	25,60
<b>Strong</b>	2.60±0.50	1.71±0.34	1,50±0.06	4.31±0.85	0,86±0.18	6.36±1.04
<b>Minimum</b>	0,01	0,01	0,95	0,02	0,10	0,20
<b>Maximum</b>	7,59	5,39	2,00	12,98	2,86	20,75

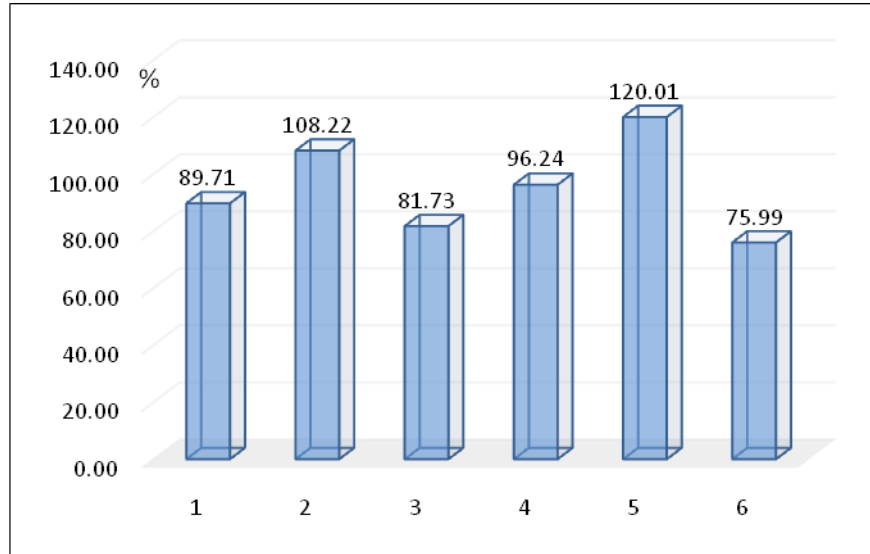
**Table.2** The results of factor analysis of photosynthetic pigments of the vicia plant

№	Photosynthetic plant pigments	Factor loads depending on the level of soil salinity		
		Weak	Medium	Strong
<b>1</b>	<b>Chl«a»</b>	0,994	0,939	0,995
<b>2</b>	<b>Chl«b»</b>	0,991	0,812	0,993
<b>3</b>	<b>The ratio of chl «a» and «b» («a/b»)</b>	0,041	-0,027	0,124
<b>4</b>	<b>Total Chl's («a/b»)</b>	0,994	0,988	0,996
<b>5</b>	<b>Amount of carotenoids</b>	0,993	0,955	0,992
<b>6</b>	<b>Ratio of total Chl to carotenoids (Chl/Car)</b>	-0,267	-0,138	-0,196

**Table.3** Factor load of morphological features of vicia in conditions of soil salinization

Morphological features of the plant	Phases	Factor loads depending on the level of soil salinity		
		Слабо	Среднее	Сильно
<b>In the leaves</b>	<b>Germination</b>	0,4826	0,4009	0,5354
	<b>Branching</b>	<b>1,2147</b>	<b>1,0621</b>	<b>1,0939</b>
	<b>Blossoming</b>	<b>2,0186</b>	<b>1,9594</b>	<b>2,0849</b>
	<b>Bean formation</b>	<b>1,2439</b>	<b>1,0779</b>	<b>1,1766</b>
	<b>Ripening</b>	-0,1620	0,4806	-0,1868
<b>In the stalks</b>	<b>Branching</b>	-0,7729	-0,7686	-0,1868
	<b>Blossoming</b>	0,6724	-0,7077	-0,7707
	<b>Bean formation</b>	-0,7465	-0,7959	-0,618
	<b>Ripening</b>	-0,9196	0,9380	-0,7287
<b>In the inflorescences</b>	<b>Blossoming</b>	-0,6319	-0,9280	-0,951
<b>In the beans</b>	<b>Ripening phase</b>	-0,8823	0,9596	-0,614

**Fig.1** The effect of the soil salinity level on the amount of photosynthetic pigments of the vicia plant, %.



**Note:** The first column is Chl «a»; the second column is Chl «b»; the third is the ratio of Chl «a/b»; the fourth is the total Chl s; carotenoids of the fifth column; the sixth column is the ratio of Chl to carotenoids (Chl /car).

These indicators determined the amount of photosynthetic pigments. For this reason, this factor can be called the “pigment” factor. The same result was recorded in conditions of medium and highly saline soils. The factor load of chlorophyll «a» was 0.939 in medium salinity soil and 0.995 in heavy salinity soil. These data indicate a strong correlation between chlorophylls and carotenoids.

Since factor analysis is based on correlation analysis, the results obtained are consistent with the work of the researcher (37) on the correlation of morphological features in plants.

It can be seen from the table that the amount of pigments (chlorophylls «a», «b» and carotenoids) had a high load at different levels of soil salinity. Now let's determine the proportion of morphological features of the vicia plant based on these factors. These data are presented in the table below (Table 3). From the data in the table it can be seen that the leaf of the plant has the greatest load of morphological features of plants. It can be seen that the factor load in the germination phase was 0.4826 (in slightly saline soil), 1.2147 in the branching phase, 2.0186 in the blossoming phase, 1.2439 in the bean formation phase and -0.1620 in

the maturation phase. After germination, the number of pigments in the leaf increased. The highest rate was recorded in the blossoming phase. This result was recorded in soils of medium weeds (1.9594) and strong salinity (2.0849). Thus, the amount of pigments in the vicia c plant was highest in the blossoming phase and decreased in the maturation phase (-0.1620). This situation was observed at all levels of soil salinity.

In general, according to the data provided, it can be noted that the level of soil salinity affected the amount of photosynthetic pigments of plants. In highly saline soil, the amount of chlorophyll «a» decreased by 10%, and the amount of chlorophyll «b» increased. The same situation was observed for the carotenoids.

This means that the level of soil salinity acts as a stress factor. It was found that the amount of photosynthetic pigments in the vicia plant has a maximum value mainly in the leaf during the blossoming phase. In slightly saline soil, the amount of chlorophyll «a» in the leaf was 5.42 mg/g, the amount of chlorophyll «b» was 1.89 mg/g, and the amount of carotenoids was 1.45 mg/g. The ratio of carotenoids to chlorophyll was 5.53 mg/g.

In medium saline soil, the amount of chlorophyll «a» leaf pigments was 5.32 mg/g, chlorophyll «b» — 4.05 mg/g, carotenoids — 1.64 mg/g, and in highly saline soil these indicators were — 4.88;3.19; 1.69 mg/g. These data show that an increase in soil salinity caused an increase in chlorophyll «b» and carotenoids while reducing chlorophyll «a».

The level of soil salinity affected the amount of photosynthetic pigments. The amount of chlorophyll «a» decreased in highly saline soil compared to slightly saline soil, the amount of chlorophyll «b» and carotenoids increased.

Most photosynthetic pigments are located in the stalk and on the leaf. In slightly saline soil, the amount of chlorophyll «a» in leaves is 5.42 mg/g, while in highly saline soil -4.88, chlorophyll «b» was 1.89-3.19 mg/g, respectively. The amount of carotenoids changed from 1.45 in slightly saline soil to 1.69 mg/l in highly saline soil.

In saline soil conditions, it was considered advisable to determine the amount of photosynthetic pigments in the leaves of plants in the blossoming phase.

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**How to cite this article:**

Ismoilova Karomatkhon Makhmudjonovna, Kuliyeu Tojiddin Hamdamovich, Sultonova Nigorakhon Muxammadkodiurovna and Karimova Shoiru Bakhodirkizi. 2023. The Effect of Soil Salinization on Quantitative Indicators of Photosynthetic Pigments of Vicia. *Int.J.Curr.Microbiol.App.Sci*. 12(12): 118-126. doi: <https://doi.org/10.20546/ijcmas.2023.1212.015>