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**HYBRIDIZATION OF CERTAIN FORMS OF SUBSPECIES BELONGING TO  
*G.HIRSUTUM* L. SPECIES, FORMATION OF BOLLS AND SEEDS IN THE BOLLS**

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**Abstract.** The analysis of hybridization results showed that the forms of *G.hirsutum* L. subspecies can be hybridized well with each other. In the studied hybrid combinations, the rates lower than 50,0% on the formation of hybrid bolls and full seeds in hybrid bolls were not observed. Currently, scientific researchers are facing urgent problems in creating varieties resistant to stress factors. Solution for these actual problems requires the use of wild, semi-wild, cultivated tropical subspecies and forms in the process of practical selection. In view of this, it is necessary to continue this research work consistently, to obtain rare recombinant forms and involve them in selection and breeding studies.

**Key words:** cotton, *Gossypium* L., *G.hirsutum* L., hybrid, wild, subspecies, intraspecific, recombinant.

**ГИБРИДИЗАЦИЯ ОТДЕЛЬНЫХ ФОРМ ПОДВИДОВ, ПРИНАДЛЕЖАЩИХ  
ВИДА *G.HIRSUTUM* L., ФОРМИРОВАНИЕ КОРОБОЧЕК И СЕМЯН В КОРОБОЧКАХ**

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**Аннотация.** Анализ результатов показал, что формы подвида *G.hirsutum* L. хорошо гибридизуются между собой. В изученных гибридных комбинациях показателей ниже 50,0% по образованию гибридных коробочек и полноценных семян в гибридных коробочках не наблюдалось. В настоящее время перед научными исследователями стоят актуальные проблемы создания сортов, устойчивых к стрессовым факторам. Решение этих актуальных проблем требует использования в процессе практической селекции диких, полудиких, культурных тропических подвидов и форм. В связи с этим необходимо последовательно продолжать данную исследовательскую работу, получать редкие рекомбинантные формы и привлекать их к селекционным исследованиям.

**Ключевые слова:** хлопчатник, *Gossypium* L., *G.hirsutum* L., гибрид, дикий, подвид, внутривидовой, рекомбинантный.

**INTRODUCTION**

It is known that the intra- and interspecific hybridization for the species

belonging to the *Gossypium* L. genus is one of the main factors that determine the



possibility of using them in practical selection and breeding as donors of beneficial traits in the of creation promising lines and varieties of cotton. Acceleration of selection-breeding processes is to solve the main problems that determine the perspective of the process of introgression of useful traits of wild species diversity into the genome of cultivated species.

In the process of hybridization, the high and low level of the formation of bolls and the seeds in the bolls depend on the phylogenetic closeness of the species and forms used in the hybridization. At the same time, hybridization results are closely related to flowering biology, one of the biological traits of cotton species.

It is known that the productivity of hybrid bolls formation and the seeds in hybrid bolls in intraspecific and interspecies hybridization of cotton species, whether the results are positive or negative, depends on the phylogenetic distance or closeness of the species involved in the hybridization and the physiological characteristics of the initial sources [4, 5, 10, 11, 12].

Transgressive forms were obtained as a result of hybridization of wild, ruderal, tropical, subtropical varieties and forms of the polymorphic *G. heraceum* L. and *G. arboreum* L. species belonging to the *Gossypium* L. genus. Valuable information was gathered as a result of conducting research on the possibility of using these forms as a source with unique traits in practical selection and breeding [6].

B.Kh. Amanov [1, 2, 3] carried out intraspecific hybridization of *G. barbadense* L. species of Peruvian cotton varieties and hybridization with *G. darwinii* Watt species. When *G. darwinii* Watt species was used as a mother form,

it was found that the rate of boll formation was low (33,3-40,0 %), and the percentage of seed formation in the boll was high (65,5-94,0 %).

According to the results obtained by crossbreeding intra-specific varieties of *G. hirsutum* L. [13], subs.*punctatum* var.*hopi*, subs.*mexicanum* var.*nervosum* (Jucatan), subs.*paniculatum* and subs.*mexicanum* var.*microcarpum palmerii* forms among wild and semi-wild subspecies and hybrids were hybridized and recombinants were received.

N.N. Nabieva [9] analyzed the results of formation of hybrid bolls and the seeds in the bolls on the basis of interspecific hybridization of Intraspecific cultivars belonging to the species *G. hirsutum* L. and *G. barbadense* L. species. As a result of research, unique recombinant forms were isolated and recommended for practical selection-breeding work.

Kh.A. Muminov [7, 8] studied the results of the formation of hybrid bolls and the seeds in the bolls on the basis of intra-and interspecific hybridization of *G. herbaceum* L. and *G. arboreum* L. intraspecific varieties. During the study transgressive forms with valuable economic traits were isolated and recommended as an initial source for practical selection-breeding studies.

**Methodology.** The experiments were carried out at the experimental field of the extension center and at the department of "Selection and Seed Production of Agricultural Crops and Cultivation of Medicinal Plants" of the Faculty of Agrobiology of the Andijan Institute of Agriculture and Agro-Technology.

**The object of the research** is *G. hirsutum* L. subs.*mexicanum* var.*microcarpum* f.*palmeri*, *G. hirsutum* L.



Table-1

The initial forms and the number of bolls and the percentage of the formation of full seeds in the bolls of F<sub>1</sub>-plants (2023)

№	Initial forms and F <sub>1</sub> -plants	Number of hybridization, piece	Number of formed bolls, piece	Boll formation, %	Number of seeds, piece		Formation of full seeds, %			
					full	full	$\bar{x} \pm S\bar{x}$	limit	S	V %
1	<i>G.hirsutum</i> L. subs. <i>mexicanum</i> var. <i>microcarpum</i> f. <i>palmeri</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Kelajak" cultivar	10	7	70,0	88	12	87,5 ± 0,48	88-91	1,6	1,9
2	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Kelajak" cultivar x <i>G.hirsutum</i> L. subs. <i>mexicanum</i> var. <i>microcarpum</i> f. <i>palmeri</i>	15	9	60,0	112	19	84,9 ± 0,29	84-86	1,1	1,4
3	<i>G.hirsutum</i> L. subs. <i>mexicanum</i> var. <i>microcarpum</i> f. <i>palmeri</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Genofond-2" cultivar	11	8	72,7	83	28	74,3 ± 0,36	71-76	1,4	1,7
4	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Genofond-2" cultivar x <i>G.hirsutum</i> L. subs. <i>mexicanum</i> var. <i>microcarpum</i> f. <i>palmeri</i>	10	9	90,0	124	28	83,4 ± 0,59	76-86	2,1	2,4
5	<i>G.hirsutum</i> L. subs. <i>mexicanum</i> var. <i>microcarpum</i> f. <i>palmeri</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Omad" cultivar	14	9	64,2	112	11	88,0 ± 0,53	85-94	2,0	2,3
6	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Omad" cultivar x <i>G.hirsutum</i> L. subs. <i>mexicanum</i> var. <i>microcarpum</i> f. <i>palmeri</i>	13	10	76,9	135	41	75,3 ± 0,49	73-85	1,8	2,2
7	<i>G.hirsutum</i> L. subs. <i>mexicanum</i> var. <i>microcarpum</i> f. <i>palmeri</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Sultan" cultivar	20	15	75,0	61	12	83,8 ± 0,54	81-85	1,6	1,8



8	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Sultan" cultivar x <i>G.hirsutum</i> L. subs. <i>mexicanum</i> var. <i>microcarpum</i> f. <i>palmeri</i>	11	8	72,7	113	21	82,2 ± 0,57	79-85	1,2	2,1
9	<i>G.hirsutum</i> L. subs. <i>punctatum</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Kelajak" cultivar	10	8	80,0	110	27	76,0 ± 0,67	73-80	2,4	2,7
10	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Kelajak" cultivar x <i>G.hirsutum</i> L. subs. <i>punctatum</i>	15	12	80,0	190	29	84,3 ± 0,61	83-91	2,1	2,5
11	<i>G.hirsutum</i> L. subs. <i>punctatum</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Genofond-2" cultivar	25	15	60,0	265	41	83,8 ± 0,49	81-88	1,4	1,8
12	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Genofond-2" cultivar x <i>G.hirsutum</i> L. subs. <i>punctatum</i>	12	12	100,0	102	15	87,7 ± 0,49	85-90	1,3	1,7
13	<i>G.hirsutum</i> L. subs. <i>punctatum</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Omad" cultivar	10	8	80,0	112	8	92,1 ± 0,53	90-95	1,6	1,9
14	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Omad" cultivar x <i>G.hirsutum</i> L. subs. <i>punctatum</i>	12	11	91,6	115	38	72,6 ± 0,37	71-75	1,3	1,8
15	<i>G.hirsutum</i> L. subs. <i>punctatum</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Sultan" cultivar	12	8	66,6	119	21	84,5 ± 0,28	83-86	1,0	1,2
16	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Sultan" cultivar x <i>G.hirsutum</i> L. subs. <i>punctatum</i>	15	9	60,0	132	9	93,5 ± 0,48	91-97	1,5	1,8
17	<i>G.hirsutum</i> L. subs. <i>punctatum</i> var. <i>gambia</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Kelajak" cultivar	11	8	72,7	81	28	74,1 ± 0,38	71-76	1,3	1,7
18	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Kelajak" cultivar x <i>G.hirsutum</i> L. subs. <i>punctatum</i> var. <i>gambia</i>	10	7	70,0	113	15	87,5 ± 0,47	83-89	1,6	1,9
19	<i>G.hirsutum</i> L. subs. <i>punctatum</i> var. <i>gambia</i> x <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Genofond-2" cultivar	15	12	80,0	189	29	86,3 ± 0,62	84-91	2,1	2,5
20	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Genofond-2" cultivar x <i>G.hirsutum</i> L. subs. <i>punctatum</i> var. <i>gambia</i>	13	10	76,9	129	38	76,3 ± 0,49	75-84	1,8	2,3



21	<i>G.hirsutum</i> L. subs. <i>punctatum</i> var. <i>gambia</i> <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Omad" cultivar	14	9	64,2	111	13	88,0 ± 0,52	87-91	1,6	1,9
22	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Omad" cultivar x <i>G.hirsutum</i> L. subs. <i>punctatum</i> var. <i>gambia</i>	20	14	70,0	185	38	82,7 ± 0,51	79-84	1,7	2,1
23	<i>G.hirsutum</i> L. subs. <i>punctatum</i> var. <i>gambia</i> <i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Sultan" cultivar	10	9	90,0	126	31	81,5 ± 0,66	77-84	2,1	2,3
24	<i>G.hirsutum</i> L. subs. <i>euirsutum</i> "Sultan" cultivar x <i>G.hirsutum</i> L. subs. <i>punctatum</i> var. <i>gambia</i>	10	9	90,0	125	27	81,4 ± 0,58	79-84	1,7	2,0



subs.*punctatum*, *G.hirsutum* L. subs.*punctatum* var.*gambia* forms belonging to *G.hirsutum* L. subspecies and also Kelajak, Genofond-2, Omad, Sul-ton varieties.

The method of the study is hybridization of subspecies of *G.hirsutum* L. in the experiment, and statistical analyses.

### Research results

It was found that *G.hirsutum* L. subspecies hybridized well, the formation of hybrid bolls was 60.0-100.0%, and the formation of full seeds in hybrid bolls was 72,6-93.5% (Table 1).

The higher rate of the formation of hybrid bolls in the studied subspecies was observed in the combination *G.hirsutum* L. subs. *eu-hirsutum* "Genofond-2" cultivar x *G.hirsutum* L. subs. *punctatum* (100,0%), while a low rate for this trait was noted in the combinations *G.hirsutum* L. subs. *eu-hirsutum* "Kelajak" cultivar x *G.hirsutum* L. subs. *mexicanum* var. *microcarpum* f. *palmeri*, *G.hirsutum* L. subs. *punctatum* x *G.hirsutum* L. subs. *eu-hirsutum* "Genofond-2" cultivar, *G.hirsutum* L. subs. *eu-hirsutum* "Sulton" cultivar x *G.hirsutum* L. subs. *punctatum* (60,0%).

The higher rate on the formation of full seeds in hybrid bolls obtained as a result of hybridization of *G.hirsutum* L. subspecies was observed in the combination *G.hirsutum* L. subs. *eu-hirsutum* "Sulton" cultivar x *G.hirsutum* L. subs. *punctatum* (93,5%); while a low rate was found in the combinations *G.hirsutum* L. subs. *eu-hirsutum* "Omad" cultivar x *G.hirsutum* L. subs. *punctatum* (72,5 %).

The analysis of hybridization results showed that the forms of *G.hirsutum* L. subspecies can be hybridized well with each other. In the studied hybrid combinations, the rates

lower than 50,0% on the formation of hybrid bolls and full seeds in hybrid bolls were not observed. Currently, scientific researchers are facing urgent problems in creating varieties resistant to stress factors.

Solution for these actual problems requires the use of wild, semi-wild, cultivated tropical subspecies and forms in the process of practical selection. In view of this, it is necessary to continue this research work consistently, to obtain rare recombinant forms and involve them in selection and breeding studies.

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## FORMATION OF YIELD PERFORMANCES IN THE SAMPLES OF FOREIGN COLLECTION BELONGING TO *CICER ARIETINUM* L. SPECIES

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**Abstract.** When the 36 samples of chickpea from the world collection were planted in the fall on 3 m<sup>2</sup> plots in 3 replications, and elements of productivity were analyzed, the weight of seedpods in one plant was 27.4-54.9 grams, the number of seedpods in one plant was 17.0-36.2 pieces, the weight of 1000 grains was 223.1-388.6 grams and average total yield was 340-789.0 g/m<sup>2</sup>. Of the 36 studied samples, 16 samples had a higher average total productivity, and it was found that they can yield more than 20 centners per hectare.

**Keywords:** chickpea, collection, budding, flowering, seedpod, productivity, trait, replication, genotype, weight of 1000 grains.

## ФОРМИРОВАНИЯ ЭЛЕМЕНТОВ УРОЖАЙНОСТИ ОБРАЗЦОВ НУТА ИЗ МИРАВОЙ КОЛЛЕКЦИИ ВИДА *CICER ARIETINUM* L.

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## Introduction

The chickpea plant is one of the important crops and is grown in more than 12 million hectares and in more than 100 countries worldwide [16]. It is known that chickpea grain is used as an important food for human consumption due to its rich content of complex carbohydrates, vitamins, minerals and phytochemicals [3]. The chickpeas are rich in crude protein and high in tryptophan amino acids. In the breeding of chickpea for yield and to meet the global demand for pea consumption, chickpea cultivation has significantly improved productivity, disease resistance, plant morphological parameters and adaptability traits in pea cultivation worldwide in the last three decades [4].

Correct phenotyping in chickpea breeding and its use in combination with molecular information in the genetic process will increase the selection efficiency [15]. In this case, the direct breeding process focused on physiological traits is considered more effective than the breeding focused on productivity traits. This term is used to describe the productivity and morphological variability of the genotype during the development periods under the influence of various external environmental factors [2]. Breeding based on conventional yield trait control is also based on yield analysis of tens of thousands of plants at the end of each cycle, which hides the effect of the trait of interest on chickpea yield. In contrast, breeding based on

yield-morphological traits has the advantage that enables to identify the traits substituting the traits of early season or of normal yield for the yield or yield-specific traits.

Chickpea plant, like all leguminous plants, activates the microbial functions of the soil, causing an increase in the amount of organic carbon in the soil [5]. The above-ground and below-ground biomass of the chickpea plant contributes directly to the increase of organic matter in the soil [14]. Above-ground biomass is especially important in increasing soil fertility and productivity. The leaf structure of the chickpea plant directly affects productivity by increasing the efficiency of photosynthesis. The effect of the increase in the efficiency of photosynthesis in the leaf on productivity is mentioned in the scientific literature [9].

P. Maharjan [10] believed that in the development of the selection process the new varieties and the genotype resistant to environmental factors are one of the most important aspects of environmental interaction. This is the difference between high and average yields for the most important agricultural crops (1:4 typical ratio) due to the tendency to increase the quantity and quality of yields depending on weather conditions.

Increasing the yield and adaptability of chickpea using different genetic resources is important for the breeding of new cultivars [1]. Chickpea



plant species with narrow genetic diversity are susceptible to pathogens or environmental stressors, which can lead to yield losses and significant reductions in adaptation characteristics [10]. The genetic diversity of the chickpea plant also varies significantly in terms of yield potential, short stem length, branch length, complex leaf structure, grain shape, seed size, and disease resistance [7]. Thus, the use of different genetic resources in increasing the yield of chickpea is important to select genetically diverse parents and to expand the genetic base of cultivated chickpea [11].

#### **The research objects and methods.**

Samples of the world collection of chickpeas of the international organization ICARDA were used as the research object. During the scientific research, the seeds of chickpea samples of the foreign collection were sown by 20 grams in 3m<sup>2</sup> area in the "Durmon" experimental field of the Institute of Genetics and Plant Experimental Biology, in 3 replications.

#### **The research results**

The productivity of the chickpea plant is one of the main important directions of the selection-breeding process. In the creation of the varieties with high genetic potential, it is expedient to combine genotypes that are resistant to negative factors of the external environment, have stable yield and high productivity.

In recent years, along with the increase in the genetic diversity of the chickpea plant, the problems in pea selection and breeding are being solved by analyzing the indicators of productivity, resistance to diseases and pests, as well as the physiological characteristics of the pea under the

influence of external environmental factors. Our research aimed to select genotypes with high yield using samples of foreign collections of peas and use them in selection-breeding processes.

The productivity of a chickpea plant depends on the number of seedpods produced. Our experiments show that the number of pods in one plant is one of the variable traits. The possibility of budding, flowering, pod formation in the chickpea plant is very high, but its preservation depends on the characteristics of the variety and agrotechnical practices and environmental conditions. Although the number of grains per plant is inextricably linked with the number of seedpods, variable productivity traits such as the number of grains per pod depend on the genotype of the collection sample being studied. The trait for number of seedpods does not significantly affect the yield rate. Laboratory studies have shown that most chickpea varieties and samples contain 2 grains per pod.

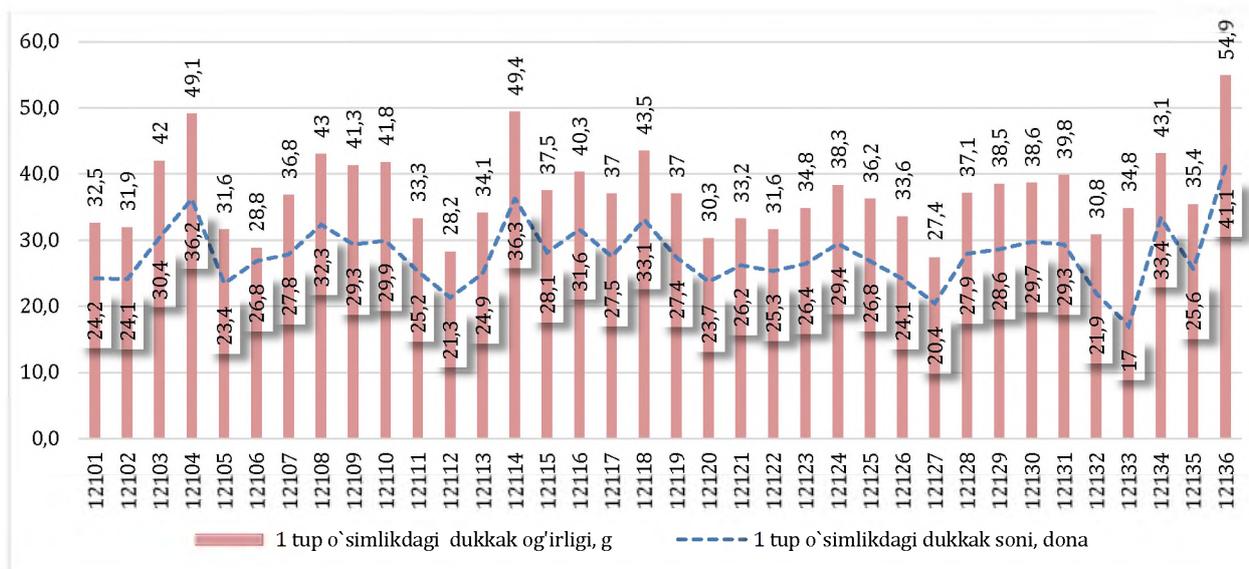
In our experiments, the weight of the seedpod per plant in the collection samples was from 27.4 grams to 54.9 grams. Among world samples of the chickpea collection, the highest index has been found in the samples 12136 (54.9 g), 12114 (49.4 g), 12104 (49.1 g), 12118 (43.5 g), 12134 (43.1 g), 12108 (43 g), 12110 (41.8 g), 12103 (42 g) and 12109 (41.3 g). In the rest of the samples of the collection, the weight of the seedpod per plant was less than 40. The lowest indicators was recorded in samples 12112 (28.2 g), 12106 (28.8 g) and 12127 (27.4 g) (Figure 1).

Among the productivity components, the highest indicator for the number of seedpods per plant was observed in sample 12104 (36.2 pieces),



while the lowest indicator was recorded in sample 12133 (17 pieces). It was found that the correlation of these two components according to the number and weight of seedpods per plant showed the highest results in the samples 12104 (number of pods 49.1; weight of pods 36.2 g), 12114 (number of pods 49.4 pieces; weight of pods 36.3 g), 12118 (number of pods 43.5 pieces; pod weight

33.1 g) and 12136 (number of pods 54.9 pieces; pod weight 41.1 g) compared to the rest of the samples. It was also observed that the adaptability of these samples to the soil climatic conditions was genetically resistant to external environmental factors and manifested the characteristics of resistance in the phenotype.



**Figure-1. The number and the weight of seedpods in the samples.**

The weight of 1000 grains is one of the important indicators of productivity enhancing components. The trait of the weight of 1000 grains in the chickpea plant is a quantitative trait that determines the yield and seed value of the samples based on their genetic characteristics. Controlling the genetic character of the sample according to this trait is a very difficult task and requires many years of research. The weight of 1000 grains in a chickpea plant varies depending on the favorable and unfavorable conditions of the external environment. In our experiments, when the weight of 1000 grains was determined, it showed 223.1-388.6 grams. In terms of the trait for the weight of 1000 grains, a relatively high value was

recorded in collection samples 12106 (283.7 g), 12118 (388.6 g) and 12131 (353.4 g), while the low value for this trait was 223.1 grams in sample 12116.

In the sample 12104 studied in our experiments, a high result was recorded for the number and weight of seedpods, but a low result was recorded for the weight of 1000 grains. It was observed that the reason for this was the number of seedpods per plant and the high number of grains in the seedpods. Similar results were observed in samples 12118 and 12136, the higher number of grains in the seedpod led to a decrease in the weight of 1000 grains. On the contrary, despite the low weight of 1000 grains, the productivity indicators were noted to be high in some samples. Among such

samples, the highest result of the yield was achieved in collection samples 12108, 12117, 12134, it was 702, 760, 742 g, respectively. Despite the relatively

low weight of 1000 grains of these samples, the higher number of seedpods and higher number of grains in seedpods resulted in higher yields.

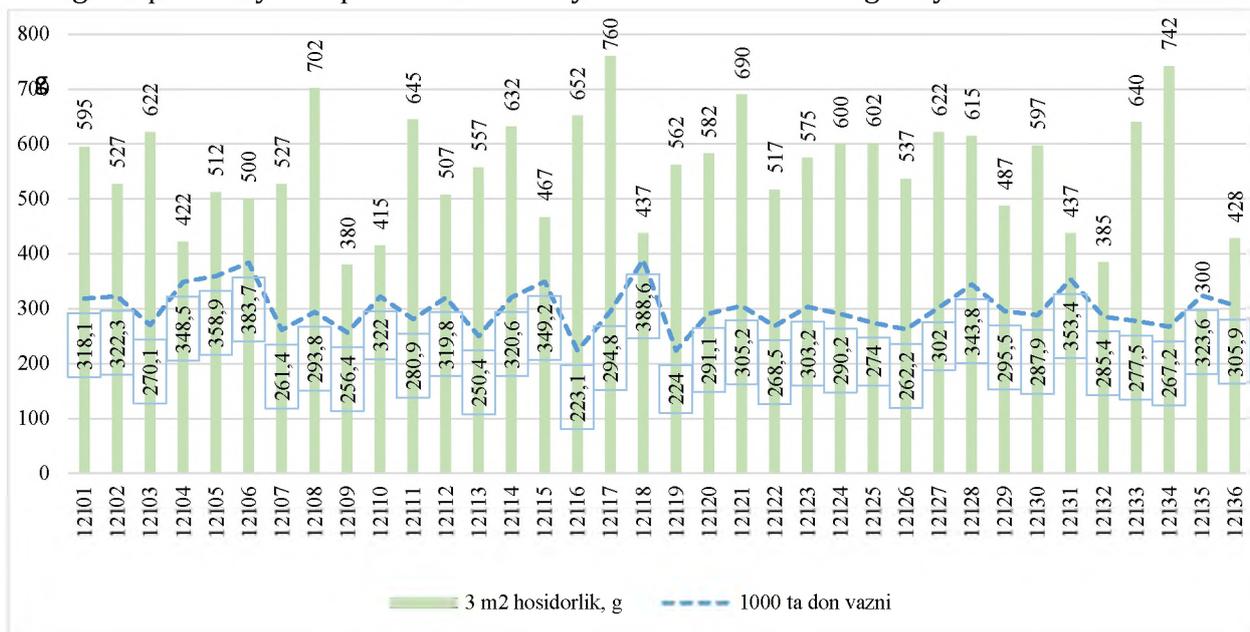


Figure 2. The weight of 1000 grains and yield performances of the samples

The relationship between the four traits studied during the research was determined in the samples 12108 (number of pods 32.3 pieces, pod weight 43 g, weight of 1000 grains 293.8 g, yield 702 g) and 12134 (number of pods 33.4 pieces, pod weight 43.1 g, the weight of 1000 grains 267.2 g, yield 742 g). In the remaining samples, a disparity between traits was observed (Table 1). For

example, in samples 12118 and 12104, the number of seedpods, the weight of seedpods and the weight of 1000 grains were relatively high, while the yield was low, 437 and 422 g. The reason for the low yield was the low number of plants per 3 m<sup>2</sup>. Similar results revealed an imbalance between the traits in samples 12103, 12116 and 12114.

Table-1

Yield performances of the samples

№	Catalogue number	Average total yield, 3 g/m <sup>2</sup>			
		$\bar{x} \pm S\bar{x}$	Limit	S	V %
1	12101	605,0 ± 5,6	590 - 630	21,7	3,6
2	12102	532,3 ± 6,5	510 - 560	25,4	4,7
3	12103	622,3 ± 3,2	610 - 635	12,5	2,0
4	12104	522,3 ± 5,8	500 - 545	22,5	4,3
5	12105	545,0 ± 8,7	512 - 580	34,0	6,2
6	12106	533,3 ± 9,07	500 - 570	35,1	6,5
7	12107	678,3 ± 4,5	660 - 695	17,5	2,5
8	12108	789,0 ± 4,2	772 - 805	16,5	2,0
9	12109	456,7 ± 5,3	440 - 480	20,8	4,5
10	12110	415,0 ± 1,2	410 - 420	5,0	1,2
11	12111	651,7 ± 1,97	645 - 660	7,63	1,1



12	12112	537,3 ± 7,0	507 - 560	27,31	5,0
13	12113	594,0 ± 4,9	577 - 615	19,31	3,2
14	12114	632,3 ± 1,9	625 - 640	7,5	1,1
15	12115	484,0 ± 4,27	467 - 500	16,5	3,4
16	12116	746,7 ± 1,5	740 - 752	6,11	1,1
17	12117	713,3 ± 2,6	705 - 725	10,4	1,4
18	12118	487,3 ± 4,5	470 - 505	17,5	3,5
19	12119	736,7 ± 3,2	725 - 750	12,5	1,7
20	12120	670,7 ± 5,6	647 - 690	21,8	3,2
21	12121	690,0 ± 1,2	685 - 695	5,0	0,7
22	12122	702,3 ± 1,1	697 - 705	4,61	0,6
23	12123	591,7 ± 3,9	575 - 605	15,2	2,5
24	12124	616,7 ± 4,5	600 - 635	17,5	2,8
25	12125	602,3 ± 1,9	595 - 610	7,5	1,2
26	12126	537,3 ± 8,3	505 - 570	32,5	6,0
27	12127	622,3 ± 5,8	600 - 645	22,5	3,6
28	12128	626,7 ± 6,3	610 - 655	24,6	3,9
29	12129	487,3 ± 1,9	480 - 495	7,5	1,5
30	12130	597,3 ± 1,9	590 - 605	7,5	1,2
31	12131	535,7 ± 6,4	510 - 560	25,0	4,6
32	12132	510 ± 3,87	495 - 525	15	2,9
33	12133	636,7 ± 5,2	615 - 655	20,	3,1
34	12134	743,3 ± 6,7	718 - 770	26,0	3,5
35	12135	340,0 ± 5,1	320 - 360	20,0	5,8
36	12136	359,7 ± 4,4	340 - 373	17,3	4,8

### Conclusion

The analysis of the obtained results of the studies showed that when 36 samples of chickpea from the world collection were planted on 3 m<sup>2</sup> plots in 3 replications in the autumn season, and analyzed the elements of the productivity of the chickpea plant, the weight of the seedpod in one plant was 27.4-54.9 grams, the number of seedpods in one plant was 17, 0-36.2 pieces, the weight of 1000 grains was 223.1-388.6 grams and average total yield was 340-789.0 g/m<sup>2</sup>. Out of 36 studied samples, 16 samples had higher total yield and it was determined that they could yield more than 20 centners per hectare. Samples of the collection with a complex combination of productivity traits were selected and recommended for selection-breeding work in practice.

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**MORPHOLOGICAL PROPERTIES OF COTTON PHYB RNAI LINES OBTAINED  
THROUGH SOMATIC EMBRYOGENESIS**

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**Annotation.** The goal of the study is to control the flowering process by inhibiting the functions of the PHYB genes that are responsible for cotton flowering using synthetic RNAi duplexes.

**Key world:** RNAi, Phytochrome B, Coker-312, cotton.

**МОРФОЛОГИЧЕСКИЕ СВОЙСТВА ЛИНИЙ РНК-ИНТЕРФЕРЕНЦИИ РНУВ ХЛОПКА,  
ПОЛУЧЕННЫХ В РЕЗУЛЬТАТЕ СОМАТИЧЕСКОГО ЭМБРИОГЕНЕЗА**

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**Аннотация.** Целью исследований является управление процессом цветения подавляя функции РНУВ генов ответственных за цветение хлопчатника с помощью синтетических RNAi дуплексов.

**Ключевые слова:** РНКи, фитохром В, Coker-312, хлопчатник.

**Введение**

Семейство фоторецепторов регулирует молекулярные процессы в растении, а также способствует экспрессии генов в ответ на световые сигналы окружающей среды. Предыдущие исследования показали, что в геноме тетраплоидного хлопчатника	генов фитохрома	идентифицированы четыре гена РНУА, два РНУВ и два РНУЕ (Abdurakhmonov et al., 2010). РНК-интерференция (РНКи) генов РНУА1 хлопчатника показала, что генотипы РНКи хлопчатника демонстрируют развитую корневую систему и вегетативный рост, раннее цветение, значительно увеличенную длину волокна и
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улучшение других основных характеристик волокна (Abdurakhmonov et al., 2014). Одним из представителей фитохромного гена фоторецептора красного света является ген RHYB, который ингибирует цветение растений. Например, световая сигнализация окружающей среды и ее трансдукция фитохромом В в длинные дни ингибируют цветение сорго, увеличивая экспрессию цветочных репрессоров длинного дня (Yang et al., 2014).

РНК-интерференция (РНК-и) - это процесс подавления активности определенных генов, и это происходит в клетках естественным образом. Впервые он был обнаружен в 1990-х годах у петунии (Krol et al., 1990), и до сих пор РНК-интерференция широко используется для регуляции экспрессии генов у растений, животных и микроорганизмов в качестве метода определения функции охарактеризованных *de novo* последовательностей (функциональная геномика). Для того чтобы индуцировать РНК-интерференцию, клетки должны быть трансформированы с использованием векторной конструкции, кодирующей малые интерферирующие РНК, специфичные для целевых генов-мишеней. Синтетические дуплексы олигонуклеотидов РНК-интерференции представляют собой один из эффективных методов индукции генно-специфических функциональных нокаутов, раскрывающих биологические функции генов-мишеней в живых организмах, включая растения. Активатор комплекса,

способствующего анафазе (APC), специфичный для CDH1, синтетический РНКи-дуплекс ингибирует функции гена CDH1 в клетке млекопитающих (Hideaki Naoe et al., 2010). Халконсинтаза (CHS), сигнальный этиленовый ген (EIN2), локус цветения С (FLC1) и дуплексы генов полифенолоксидазы (PPO) эффективно ингибировали функцию этих генов в модельном растении *Arabidopsis thaliana*. Однако эти дуплексы имеют длинные последовательности (от 120 до 741 п.н.), и их вставки в геном могут приводить к неспецифическим нокаутам генов и побочным эффектам.

Возможность получения неспецифических нокаутов генов в сложном аллотетраплоидном (AADD) геноме огромна, поскольку каждый ген имеет как минимум две ортологичные копии, полученные от предковых геномов А и D (Abdurakhmanov et al., 2010). Например, мы обнаружили по крайней мере четыре разных гена RHYA, два разных гена RHYB, RHYE, RHYC у тетраплоидного хлопчатника (Abdurakhmanov et al., 2010). Это затрудняет изучение точной биологической функции каждого гена фитохрома в подсемействе, как отмечалось и наблюдалось у Abdurakhmonov et al. (2014). Следовательно, здесь мы разработали короткие синтетические бинарные конструкции дуплекса РНКи, которые высокоспецифичны для генов фитохрома В хлопчатника, и соматически трансформировали RHYB-специфические короткие синтетические дуплексные РНКи кассеты в Coker-312, чтобы выявить функцию гена(ов) RHYB хлопчатника.



Конструкции векторов и методология ранее были описаны и запатентованы в Узбекистане (Abdukarimov et al., 2011). Здесь мы сообщаем о предварительных морфологических характеристиках новых линий RNAi Coker-312 («SynB»), трансформированных короткими синтетическими RNAi дуплексами, специфичными для гена(ов) РНУВ хлопчатника.

### Материалы и методы исследования

В данном исследовании в качестве растительного материала использовали соматически регенерируемый хлопок Coker-312 и его трансгенные линии. РНК-интерферированные растения получали с использованием оптимизированной в нашей лаборатории методики соматического эмбриогенеза, описанной Abdurakhmonov et al. (2014) в деталях. Для морфологических наблюдений мы получили несколько поколений (от T1 до T5) линий хлопка с РНК-интерференцией SynB и оценили их по сравнению с контрольными растениями дикого типа и с нулевой сегрегацией. Все эксперименты в полевых условиях и в теплицах проводились в случайном порядке, где мы собирали морфологические данные полевых наблюдений, а также образцы волокон. Образцы волокна были отправлены в центр тестирования качества волокна «SIFAT» для оценки качества волокна. РНКи растения-кандидаты, несущие синтетический дуплекс РНКи РНУВ, были проверены с использованием реакции ПЦР, которая специфически амплифицирует вставки дуплекса РНУВ из векторной конструкции и/или

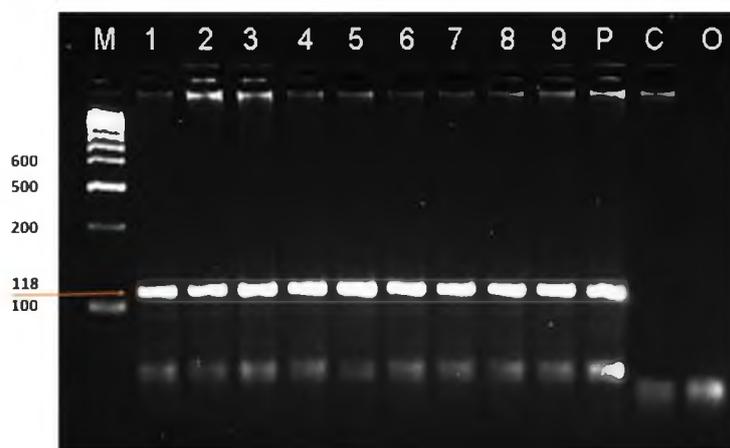
трансформированного растения. Для этого из тканей молодых листьев методом СТАВ выделяли геномные ДНК. Реакции амплификации проводили в объемах по 25 мкл, содержащих 2,5 мкл буфера 10xPCR с 1,5 мМ MgCl<sub>2</sub>, 1 мкл BSA, 0,5 мкл 25 мМ смеси dATP, dGTP, dTTP и dCTP, 2,5 мкл/50 нг/л каждого вектора RNAi, специфичного для вектора 35S-F /OST-R пары праймеров, 1 мкл 50 нг/мкл 1 ДНК-матрицы и 0,5 ЕД Taq - полимеразы (Sigma, США). Амплификацию проводили с начальной денатурацией при 94°C в течение 3 мин с последующими 45 циклами 94°C в течение 1 мин, 55°C в течение 1 мин и 72°C в течение 2 мин. Затем выполняли заключительное 5-минутное удлинение при 72°C. Для определения размеров продуктов ПЦР проводили электрофорез в 1,5% агарозном (Sigma) геле в 0,5x буфере TBE. Гели окрашивали бромистым этидием. Было взято около 110 п.н. продуктов ПЦР (рис. 1).

### Результаты исследования и обсуждение

Раннее цветение является очень важным признаком для всех сельскохозяйственных культур, включая хлопчатник. В этом исследовании РНК-интерференция была использована для подавления экспрессии гена РНУВ хлопчатника с целью изучения его биологических функций. Для этой цели мы выбрали очень четкие и специфические области размером 19–21 п.н. из фитохрома В хлопчатника и синтезировали кассеты РНКи, поместив интронную последовательность длиной 7–9 п.н. между смысловой и антисмысловой цепями (Abdukarimov et al., 2011). Эту

синтетическую кассету РНКи для фитохрома В встраивали в бинарный вектор рART27. Наша конструкция, которая была соматически трансформирована в Coker 312, несет синтетическую кассету РНУВ («SynB»), управляемую промотором 35S, и содержит маркер гена устойчивости к канамицину. Несколько RNAi растений «SynB» поколения T0 были получены с использованием метода соматического эмбриогенеза (Abdurakhmonov et al., 2014). Эти растения выращивали в тепличных горшках и собирали семена первого поколения (T1). RNAi растения SynB T1 высаживали случайным образом в тепличных условиях для проверки их предварительных морфологических характеристик. В качестве первого положительного результата мы заметили, что все RNAi растения «SynB» зацвели раньше по сравнению с контрольными растениями в теплице. Кроме того,

условиях продемонстрировали раннее цветение и набор много коробочек (рис. 2) и процентное содержание волокна линий SynB RNAi, Cocker-312 (контроль) и их гибридов с местными сортами (таб.1). В настоящее время проводятся исследования для проведения подробной молекулярной характеристики линий «SynB», включая 1) идентификацию числа копий геномных вставок кассет коротких дуплексов РНКи в трансгенных растениях, 2) количественную оценку уровня экспрессии генов РНУВ после трансформации по сравнению с нулевыми сегрегантами и генотипами дикого типа, а также мобилизацию эффектов РНУВ RNAi на фоне местного сорта(ов) посредством полового скрещивания, последующего обратного скрещивания и селекции. Наши предварительные результаты продемонстрировали возможную



после выращивания семян в тепличном эксперименте следующие поколения RNAi растений «SynB» выращивали в полевых условиях при обычном солнечном свете для проверки их агрономических и морфологических характеристик. Морфологическое наблюдение за RNAi растениями «SynB» в тепличных

биологическую функцию генов РНУВ хлопка и их агрономическое значение с использованием РНКи.

### Выводы

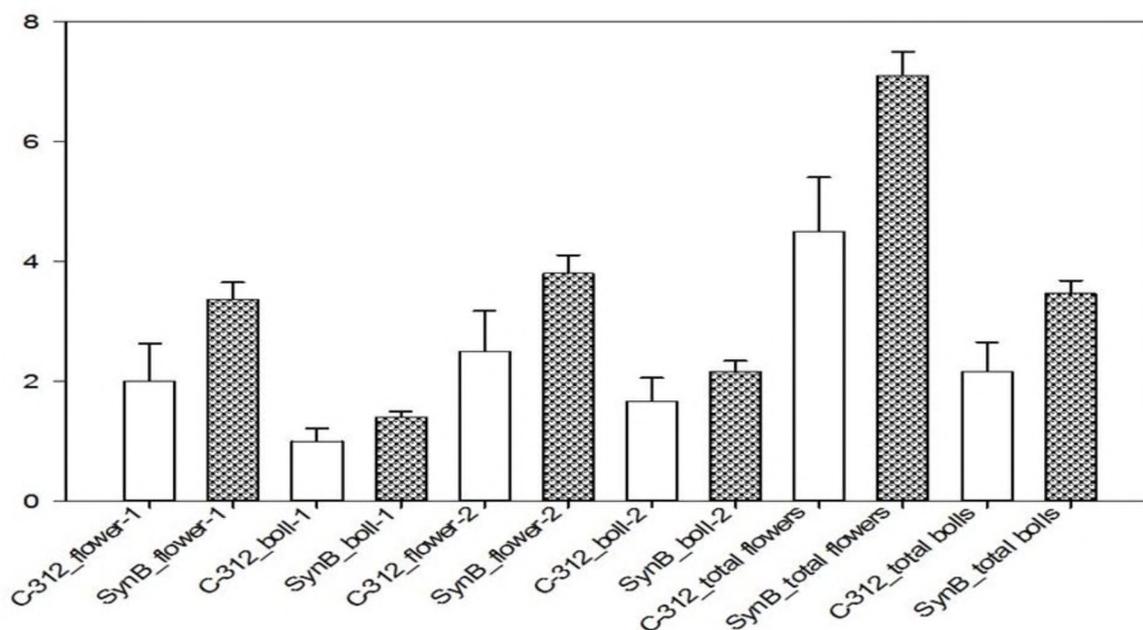
Наши усилия помогли точно отрегулировать функцию генов и связанные с ними агрономический важные признаки в сложных геномах растений, таких как



аллополиплоидный хлопчатник. Такая эффективная технология нокаута генов, легко применимая к любым цветковым растениям и сельскохозяйственным культурам, открывает новые возможности для манипулирования любым интересующим геном с высокой специфичностью и создания новых биотехнологических культур с «цисгенной» (на основе собственных генов растений, которые отличаются

от доступных «трансгенных» технологий с использованием чужеродных генов) природой, обеспечивающие ее экологическую безопасность.

**Рис. 1.** ПЦР-амплификация с парами 35S-прямой и OST-обратный праймеров для RNAi растений «SynB» T0. 1-9 «SynB» RNAi T0, С-контроль, Р-плазмида, О-нет смеси ДНК.



**Рис. 2.** Морфологическое наблюдение за растениями SynB RNAi T1 в условиях фитотрона. Цветки и коробочки хлопчатника подсчитывали дважды через две недели в период цветения.

**Таблица 1.**

**Процентное содержание волокна линий SynB RNAi, Cocker-312 (контроль) и их гибридов с местными сортами**

Level	No. of plants	Mean	STD	SEM	LSD	p-Value
ANB-2 <sup>e</sup>	13/3	33.35	2.05	0.49142	-0.49	0.1938
BC3F1(47xSynB) <sup>d,e</sup>	12/3	34.39	1.23	0.51149	-1.57	0.9629
Andijon-35 <sup>c,d</sup>	11/3	35.52	1.57	0.53423	-1.57	0.9541
BC3F1(52xSynB) <sup>b,c</sup>	12/3	36.09	2.67	0.51149	-0.96	0.5011
Shodlik-9 <sup>a</sup>	14/3	37.79	0.69	0.47355	0.802	0.0027 *
BC3F1(55xSynB) <sup>a,b</sup>	13/3	37.33	1.11	0.49142	0.312	0.0179 *
Cocker-312 <sup>c,d</sup>	8/3	34.94	3.2	0.62645	-2.32	1



SynB-T6 <sup>c,d</sup>	16/3	35.51	1.24	0.44296	-1.44	0.942
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УДК: : 57.9.632./36.37.4.01.08

## МОЛЕКУЛЯРНО-ГЕНЕТИЧЕСКАЯ ИДЕНТИФИКАЦИЯ ГРИБОВ РОДА *FUSARIUM* LINK., ПОРАЖАЮЩИХ МЯГКУЮ ПШЕНИЦУ (*TRITICUM AESTIVUM* L.)

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**Аннотация.** В комплексе мероприятий по борьбе с фузариозом важнейшую роль играет определение видового состава грибов, поражающих пшеницу. Для надежной видовой идентификации широкое распространение получил комплексный подход, включающий, помимо традиционных микробиологических методов, анализ нуклеотидных последовательностей маркерных генов и их сравнение со стандартными последовательностями, представленными в базе данных GenBank NCBI.

В течении 2012-2022 годов были проведены исследования в целях изучения грибов возбудителей болезней фузариозной корневой гнили и фузариозной гнили корневой шейки на посевах пшеницы в различных регионах Узбекистана. Было выделено 82 изолятов грибов рода *Fusarium*, которые были идентифицированы по морфологическим и микроскопическим признакам, а также был проведён молекулярно-генетический анализ выделенных изолятов грибов.



**Ключевые слова:** мягкая пшеница, фузариоз, грибы, корневая гниль, гниль корневой шейки, идентификация, молекулярная генетика, *tef-1α*, BLAST, NCBI.

## MOLECULAR GENETIC IDENTIFICATION OF FUNGI OF THE GENUS *FUSARIUM* LINK., AFFECTING BREAD WHEAT (*TRITICUM AESTIVUM* L.)

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Determining the species composition of fungi affecting wheat plays the most important role in the complex of measures to combat and control Fusarium diseases. For reliable species identification, an integrated approach, including, in addition to traditional microbiological methods, analysis of nucleotide sequences of marker genes and their comparison with reference sequences presented in the GenBank NCBI database is widely used.

During 2012-2022, research was carried out to study the fungi that cause Fusarium root rot and Fusarium root collar rot on wheat crops in various regions of Uzbekistan. 82 fungal isolates of the *Fusarium* genus were isolated, which were identified by morphological and microscopic traits, and in addition a molecular genetic analysis of these fungal isolates was carried out.

**Key words:** bread wheat, *fusarium*, fungi, root rot, root collar rot, identification, molecular genetics, *tef-1α*, BLAST, NCBI.

### Введение

В связи с тем, что население мира увеличивается с каждым днём, растёт и спрос на продукты получаемые из зерновых культур. Известно, что особое внимание уделяется созданию новых сортов мягкой пшеницы, адаптированных для различных почвенно-климатических условий, устойчивых к болезням и вредителям, обладающих высокой урожайностью и имеющих высокое качество зерна. Следует отметить, что *F. pseudograminearum*, *F. culmorum*, *F. graminearum* и *F. asiaticum* занимают лидирующие позиции среди 10 видов грибов, наносящих наибольший экономический ущерб сельскохозяйственным культурам. Поэтому актуально исследование основ молекулярно-биологических методов надёжной защиты растений

от фузариозных болезней, вызываемых микроскопическими грибами семейства *Fusarium*, в том числе выявление в них видов-возбудителей болезней. В связи с этим, надёжная защита растений от фузариозных болезней, вызываемых микроскопическими грибами рода *Fusarium*, в том числе использование современных молекулярно-биологических методов для определения видовой принадлежности возбудителей заболеваний относящихся к данному роду приобретает очень важное практическое значение.

Фузариозы пшеницы встречаются в мире везде, где возделывается эта культура. Растения могут поражаться грибами р. *Fusarium* во всех стадиях развития – от фазы



всходов до созревания и во время хранения зерна. Многие возбудители фузариозов пшеницы могут также сильно поражать ячмень, некоторые – кукурузу, овёс, рожь, рис, просо и злаковые травы.

Основными двумя формами болезни на пшенице являются 1) фузариозная гниль корней, корневой шейки и нижней части стебля (сокращённо – фузариозная корневая гниль пшеницы, ФКГ, ФКГП) и 2) фузариозная парша колосьев. В данной статье мы проанализируем ФКГП. Фузариозами поражаются также всходы пшеницы, что приводит к их загниванию, гибели и резкому снижению густоты стояния растений.

Возбудители фузариозов являются почвенными грибами и некоторые из них (напр., *F. culmorum*, *F. pseudograminearum*, *F. crookwellense*) могут сохраняться в почве в течение ряда лет в жизнеспособном состоянии с помощью хламидоспор. Источниками инфекции фузариозов для растений пшеницы являются растительные остатки; для некоторых видов (напр., для *F. graminearum*) основным источником болезни служат заражённые семена. При использовании инфицированных семян из них вырастают больные проростки.

Симптомы, круг поражаемых патогенами видов растений, цикл развития патогенов, источники инфекции, вредоносность фузариозов пшеницы и меры борьбы с ними подробно описаны в литературе [2, 3, 10-12].

В связи с тем, что пшеница является стратегической культурой и имеет огромное значение в

обеспечении пищевой безопасности всех стран мира, изучению её болезней, в том числе фузариозов, всегда уделялось и сейчас уделяется большое внимание. Хотя признанными возбудителями ФКГП являются считанные виды, в литературе приводится более 40 видов р. *Fusarium* в качестве возбудителей этих болезней и количество их постоянно увеличивается за счёт открытия новых видов или филогенетических линий внутри комплексов видов.

Ниже приводим более подробные сведения и критический анализ информации по этим видам. Виды р. *Fusarium*, о которых сообщали как возбудителях ФКГП, входят в состав 15 комплексов видов (КВ) р. *Fusarium*, а именно, КВ *F. buharicum* (FBSC), КВ *F. burgessii* (FBUSC), КВ *F. chlamydosporum* (FCSC), КВ *F. dimerum* (FDSC), КВ *F. fujikuroi* (FFSC), КВ *F. graminearum* (FGSC), КВ *F. heterosporum* (FHSC), КВ *F. incarnatum-equiseti* (FIESC), КВ *F. lateritium* (FLSC), КВ *F. nisikadoi* (FNSC), КВ *F. oxysporum* (FOSC), КВ *F. redolens* (FRSC), КВ *F. sambucinum* (FSAMSC), КВ *F. solani* (FSSC) и КВ *F. tricinctum* (FTSC) (табл. 1).

Наиболее распространёнными, сильными и агрессивными возбудителями гнили всходов и ФКГП являются *F. pseudograminearum*, *F. culmorum*, *F. graminearum* s. str. и некоторые изученные представители КВ FGSC. Пшеницу могут сильно поражать также *F. crookwellense*, *F. poae*, *F. acuminatum* и *F. avenaceum*, однако в сравнении с указанными выше тремя видами они считаются менее вирулентными, более зависимыми от погоды, имеющими более



ограниченные ареалы распространения [1, 4, 7, 8, 9].

### Объект и методы исследования

Образцы для микологической экспертизы были собраны во время экспедиции 2018-2022 г. в фермерские хозяйства культивирующие пшеницу различных регионов Узбекистана, с целью их дальнейшего исследования.

Микологическую экспертизу и изоляцию культур грибов из образцов больных растений проводили с использованием методик Хасанова и Глухой [5]. Идентификацию культур проводили по определителям, Booth C. [6] Leslie J.F., Summerell B.A. [10].

Выделение ДНК, ПЦР-амплификация и секвенирование. Выделение ДНК осуществляли с использованием набора для очистки геномной ДНК GeneJET (Thermo Fisher Scientific). Измерение количества и качества геномной ДНК проводили с помощью спектрофотометра NanoDrop Eight (Thermo Fisher Scientific, США), затем образцы ДНК хранили при -20°C до проведения ПЦР. Фрагмент гена *tef-1α* исследованных изолятов грибов рода *Fusarium* амплифицировали с использованием праймеров EF1 и EF2 с последующим секвенированием полученных ПЦР продуктов.

Для ПЦР использовали смесь реагентов для амплификации «Platinum™ Taq DNA Polymerase» (Thermo Fisher Scientific, США). ПЦР-смесь (25 мкл) содержала ДНК исследуемого штамма (4 мкл), 14,9 мкл dd H<sub>2</sub>O, 2,5 мкл 10xPCR-буфера, 0,75 мкл 50 мМ MgCl<sub>2</sub>, 0,5 мкл 10 мМ смеси dNTP, 0,5 мкл 10 мМ прямого праймера, 0,5 мкл мкл 10 мМ обратного праймера, 1,25 мкл удличителя KB, 0,1 мкл ДНК-полимеразы Platinum Taq.

Для ПЦР использовали следующую программу термоциклирования: начальная денатурация (95°C, 10 мин), денатурация (94°C, 10 с), отжиг (55°C, 30 с), элонгация (72°C, 1 мин). 40 циклов, финальная элонгация (72°C, 5 мин)

Полученные продукты ПЦР исследовали методом гель-электрофореза с буфером 1xTBE (pH 8,3) в 1,5% агарозном геле с доведением в гель раствора бромистого этидия (EtBr) 0,5 мкг/мл.

Электрофорез проводили на системе горизонтального электрофореза СЭ-1 («Хеликон», Россия) при напряжении 110 В сила тока 80mA в течение 1 ч 20 минут. Продукты ПЦР визуализировали в УФ-свете и фотодокументировали с помощью гель-документирующей системы ВК-AG100 (Biobase Kings Co., Ltd, Китай).

Секвенирование ПЦР продуктов. Для определения нуклеотидных последовательностей гена *tef-1α* проводили секвенирование амплифицированных продуктов. Для этого фрагменты амплификации вырезали из агарозного геля и очищали с использованием набора PureLink™ Quick Gel Extraction Kit (Invitrogen, США). согласно методике производителя. Секвенирующие реакции выполняли с применением набора Big Dye® Terminator v 3.1 Cycle Sequencing Kit («Applied Biosystems», США) согласно методике производителя.

Реакция циклического секвенирования состояла из ddH<sub>2</sub>O - 3,5 мкл, BigDye - 1 мкл, 5x буфера для секвенирования - 2 мкл, секвенирующего праймера - 0,5 мкл,

очищенного ПЦР-продукта - 2 мкл. Для секвенирования использовали праймеры EF1 и EF2.

Для реакции циклического секвенирования использовали следующую программу термоциклирования: начальная стадия денатурации 96°C 10 минут; затем денатурацию при 96°C в течение 10 с, отжиг при 55°C в течение 10 с и элонгацию при 60°C в течение 3 мин повторяли в течение 40 последовательных циклов.

Продукты реакции секвенирования очищали от флуоресцентно меченных терминаторных нуклеотидов с использованием набора Dynabeads Sequencing Clean-Up Kit (Applied Biosystems, США). Капиллярный электрофорез продуктов реакции секвенирования ДНК проводили на генетическом анализаторе Applied Biosystems 3500 (Thermo Fisher Scientific).

### Результаты исследования

В течение 2012-2022 годов были проведены исследования в целях изучения грибов *F. graminearum* и *F. pseudograminearum* - возбудителей болезней фузариозной корневой гнили и фузариозной гнили корневой шейки на образцах пшеницы, зараженных фузариозной корневой гнилью и фузариозной гнилью корневой шейки. Выделенные культуры были идентифицированы по морфологическим и микроскопическим признакам. С целью выделения ДНК из их культур, они были приведены в состояние МСИ.

Регион гена IGS, перенесенный специфическими праймерами *Fusarium graminearum* Fgr-F, Fgr-R и *Fusarium pseudograminearum* FP1-1, FP1-2 был амплифицирован.

В процессе культурально-морфологической и молекулярно-биологической идентификации гриба *Fusarium graminearum* морфологически на 60% был правильно идентифицирован (рис.1).

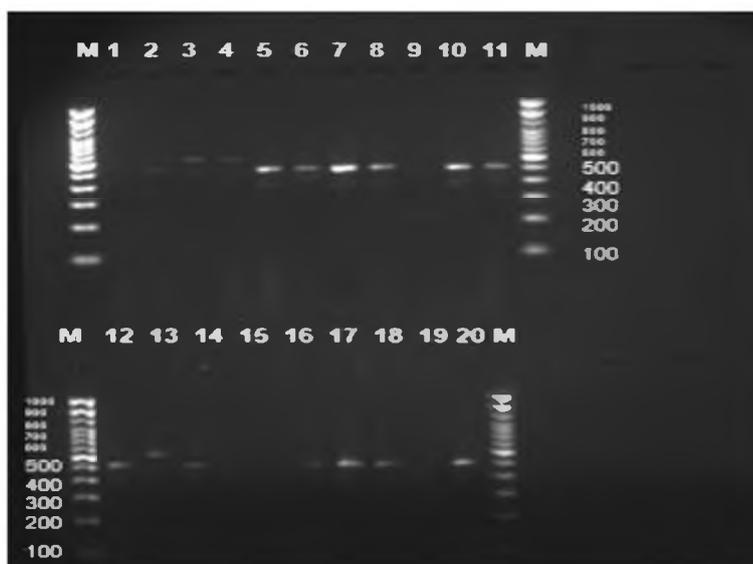


Рис.1. Электрофореграмма продуктов амплификации IGS ген региона, перенесенного на гриб *F. graminearum* специфическими Fgr-F и Fgr-R праймерами.



В целях изучения грибов *F. graminearum* и *F. pseudograminearum* - возбудителей болезней фузариозной корневой гнили и фузариозной гнили корневой шейки пшеницы в результате исследований, проведенных с помощью морфологических и молекулярно-генетических методов, не был выявлен гриб *F. pseudograminearum*. Молекулярно-генетическими анализами было подтверждено, что в Узбекистане корневую и фузариозную гнили корневой шейки пшеницы вызывает гриб *F. graminearum*.

С учетом сходства и изменчивости (полиморфности) по морфологическим признакам грибов рода *Fusarium*, выделенных из больных растений пшеницы, фрагмент их гена *tef-1α* был определен с помощью молекулярно-генетических методов.

Была осуществлена морфологическая и молекулярно-

генетическая идентификация моноспорных штаммов грибов рода *Fusarium*. Молекулярно-генетическая идентификация моноспорных штаммов грибов рода *Fusarium* имеет большое значение, поскольку макро- и микро морфологическая схожесть представителей этого рода и образование разных пигментов на различных питательных средах приводит к возникновению ряда проблем при их морфологической идентификации. Кроме этого, внутривидовые таксоны представителей грибов рода *Fusarium* многочисленны. Например, существует более 80 специализированных форм вида *Fusarium oxysporum*. Сравнение определения видовой принадлежности грибов рода *Fusarium* морфологическим и молекулярно-генетическим методом показало совпадение результатов этих методов на 56% (табл. 1).

**Таблица 1**  
**Морфологические и молекулярно-генетические анализы грибов рода *Fusarium*, выделенных из больных растений пшеницы**

№	Область, откуда выделены грибы	Виды грибов, определенные морфологическим методом	Виды грибов, определенные молекулярно-генетическим методом
1	Бухарская	<i>F. poae</i>	<i>F. poae</i>
2	Джизакская	<i>F. oxysporum</i>	<i>F. equiseti</i>
3	Ташкентская	<i>F. graminearum</i>	<i>F. graminearum</i>
4	Ташкентская	<i>F. oxysporum</i>	<i>F. oxysporum</i>
5	Сурхандарьинская	<i>F. proliferatum</i>	<i>F. proliferatum</i>
6	Ташкентская	<i>F. graminearum</i>	<i>F. culmorum</i>
7	Наманганская	<i>F. solani</i>	<i>F. solani</i>
8	Бухарская	<i>F. graminearum</i>	<i>F. graminearum</i>
9	Сырдарьинская	<i>F. oxysporum</i>	<i>F. incarnatum</i>
10	Ташкентская	<i>F. oxysporum</i>	<i>F. equiseti</i>
11	Бухарская	<i>F. poae</i>	<i>F. poae</i>
12	Сырдарьинская	<i>F. poae</i>	<i>F. poae</i>
13	Кашкадарьинская	<i>F. graminearum</i>	<i>F. culmorum</i>
14	Андижанская	<i>F. sporotrichioides</i>	<i>F. tricinctum</i>
15	Ташкентская	<i>F. sporotrichioides</i>	<i>F. tricinctum</i>
16	Бухарская	<i>F. poae</i>	<i>F. poae</i>



## Заклучение

Таким образом, неправильная систематическая и таксономическая идентификация видов может привести к неправильной диагностике болезней, встречаемых у сельскохозяйственных растений и, в результате - к неверному и неэффективному выбору мер борьбы, что может привести к потерям урожая.

Фрагмент гена *tef-1α* исследованных изолятов грибов рода *Fusarium* амплифицировали с использованием праймеров EF1 и EF2 с последующим секвенированием полученных ПЦР продуктов. Полученные последовательности фрагмента гена *tef-1α* были проанализированы в биоинформатической программе BLAST NCBI для определения видовой принадлежности. Сравнение определения видовой принадлежности грибов рода *Fusarium* морфологическим и молекулярно-генетическим методом показало совпадение результатов этих методов на 56%.

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THE EFFECT OF DROUGHT STRESS ON LEAF CHLOROPHYLL CONTENT IN  
COTTON CULTIVARS

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**Abstract.** The article presents the results of the analysis of chlorophyll and its components in the leaves of medium fiber cotton (*G. hirsutum* L.) genotypes grown under optimal water supply (control) and water deficit (experimental) conditions. Based on the analysis of chlorophyll a, chlorophyll b, total chlorophyll and carotenoid in leaf tissues of cotton genotypes, KK-1086, L-N1 and L-141 cotton variety samples are unstable in water deficit compared to optimal water supply conditions, Catamarca 811, L-45, Zangi ota, C-417, Mehnat and Namangan-77 cotton samples were found to be stable.

**Key words:** *G. hirsutum* L., cotton, water regime, water deficit, chlorophyll a, chlorophyll b, total chlorophyll, carotenoid.

ВЛИЯНИЕ СТРЕССА ЗАСУХИ НА СОДЕРЖАНИЕ ХЛОРОФИЛЛА В ЛИСТЬЯХ СОРТОВ  
ХЛОПЧАТНИКА

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**Аннотация.** В статье представлены результаты анализа хлорофилла и его компонентов в листьях генотипов хлопчатника средневолокнистого (*G. hirsutum* L.), выращенных в условиях оптимального водообеспечения (контроль) и водного дефицита (эксперимент). На основании анализа содержания хлорофилла а, хлорофилла b, общего хлорофилла и каротиноидов в тканях листьев генотипов хлопчатника образцы сортов хлопчатника КК-1086, Л-Н1 и Л-141 нестабильны при дефиците воды по сравнению с оптимальными условиями водообеспечения, образцы хлопка Catamarca 811, Л-45, Занги ота, С-417, Мехнат и Наманган-77 оказались стабильными.

**Ключевые слова:** *G. hirsutum* L., хлопок, водный режим, дефицит воды, хлорофилл а, хлорофилл b, общий хлорофилл, каротиноид.

INTRODUCTION

The global climate changes that observed in the world cause an increase in air temperature in the biosphere, and

hot winds caused by a sharp decrease in relative humidity in the summer months cause atmospheric and soil dryness. In



the modern era, when the water problem is serious, the creation and implementation of water-saving agricultural technologies, including the creation of cotton varieties that are resistant to soil and atmospheric drought and with a high coefficient of efficient water use, are considered to be the most relevant tasks of the world cotton industry.

The scientific research is being carried out to combine traditional genetic breeding methods with physiological research to create modern varieties of cotton, which is one of the main agricultural crops in the world. In this regard, in addition to medium fiber varieties of cotton, which occupy the main area of cotton cultivation, it is necessary to use sources of the cotton gene pool with high technological indicators of fiber and resistance to environmental stress factors, determine the response of varieties, lines and hybrids of cultivated cotton species to water deficiency based on morphological characteristics, isolation of resistant genotypes and involvement in breeding work, paid to the creation of drought-resistant varieties of this valuable industrial crop.

As in many regions of the world, water scarcity remains one of the pressing problems in Central Asia, especially in Uzbekistan [5]. Considering that the agriculture of our republic is based on irrigated agriculture and the main crop is cotton, the created cotton varieties are valuable – because they must have high economic characteristics, as well as be resistant to adverse environmental conditions.

One of the important tasks is to study the resistance of cotton varieties to drought and to determine the dependence of the physiological and biochemical

parameters of the genotype on water deficiency. For the development of cotton production in our republic, it is necessary that the created cotton varieties be productive and have high fiber quality, as well as be resistant to abiotic stresses, including the drought.

Cotton is a valuable industrial crop cultivated in many regions of the world. This plant is also an industrial crop grown in developed and developing countries of the world [1].

Cotton varieties belonging to the medium fiber type *G. hirsutum* L. are cultivated as the main field crop in 77 countries of the world, covering an area of about 32.0 million hectares and grown in various soil and climatic conditions. The global cotton trade is about US\$20.0 billion per year [2].

The cotton ginning and processing industries, the textile industry, etc. are the main source of employment for millions of people and make up a significant share of the GDP of many countries such as Uzbekistan, Australia, Greece, India, China and Pakistan [3, 4]. Uzbekistan ranks the 5th in the world in the production of cotton and 4th in the export of raw cotton, therefore it is one of the largest cotton-producing countries in the world. About 93 percent of the country's cotton fields are sown with medium fiber varieties of cotton [4, 5].

Like other countries, Uzbekistan faces severe drought problems due to lack of irrigation water.

The main problem of declining cotton yields is the lack of irrigation water. Therefore, the creation of new varieties that can withstand the conditions of water scarcity is one of the most urgent tasks facing cotton growing. The current global climate change will exacerbate water scarcity in the future.



Given that these changes will continue in the near future; water scarcity is becoming a serious obstacle to crop production worldwide. In this situation, the need to create varieties that are resistant to water shortages increases [6].

According to the data, the global average temperature is expected to increase by 2–4 °C, the amount of precipitation will decrease by 30%, which will have a strong negative impact on productivity and water resources in 2050 [7]. Thus, drought is a major food security issue, pointing to the need to develop crop varieties that thrive in water scarcity [8, 9].

The lack of water is one of the factors that adversely affect the growth and development of plants and, in turn, productivity. The depletion of water resources absorbed by the roots of plants, as well as the large amount of energy spent on the absorption of water, directly affect the yield of cotton in irrigated agriculture. Therefore, the study of plant drought resistance is one of the main issues of concern to farmers in the field of cotton cultivation, as well as in many other areas of agricultural crops [10, 11].

Some scientists claim that cotton is a drought-resistant crop. However, as a result of the drought, the yield of cotton, like other crops, is significantly reduced. The lack of water has a significant negative impact on the morphological and physiological characteristics and productivity of cotton [12, 13, 14].

Some researchers point out that under conditions of water scarcity and high temperatures, high genetic variability occurs in cotton, and that this variability is maintained by genetic factors [15, 16].

Currently, studies on the development of varieties resistant to water deficiency have been studying on the characteristics of cotton resistance to drought, high temperatures, insects, pests and diseases in connection with morphoeconomic and physiological traits (17). For the effective selection of drought-resistant varieties, the management of genetic differentiation according to various morphoeconomic traits was implemented [18, 19].

The lack of water is a factor that has a strong negative impact on the physiological mechanism of cotton. Long periods of drought can be detrimental to cotton. The highest level of water demand is observed during the flowering period, while in the early and late stages of flowering this requirement is relatively low. The loss of crop elements under the influence of constant dehydration also leads to a significant loss in yield [20]. The evolution of structures tolerant to water deficiency is important for understanding the diverse set of phenotypic traits studied under drought conditions. Molecular biologists have developed transgenic approaches to identify drought-tolerant genes [21].

As a reaction to the conditions of water deficiency, initially, at a very early date, the plant shows a sharp change towards slowing down the process of increasing the leaf plate, however, it is noted that the process of photosynthesis does not change significantly.

By slowing down the process of increasing the surface of the leaf plate, the level of consumption of carbohydrates and energy in the process of metabolism in the tissues of the plant organism is reduced, and it is assumed that the saved energy and nutrients can be directed to



the root system. Thus, it was noted that the root system of the plant has a low sensitivity to the effects of drought compared to the growth and development of the aerial part [22].

It has been studied that the morphophysiological characteristics of a plant with a lack of water can be assessed by the water-holding capacity of the leaf, leaf surface, permeability of the leaf mouth, the size of the leaf mouth and other indicators [23].

The relative water content is reported to be the most important indicator of water status in plants [24]. A unique relationship has been identified between relative water content and seed cotton yield under drought conditions [15].

Water deficiency tolerant genotypes reduce water loss by reducing leaf area and stomatal opening. The morphophysiological traits that are considered the most effective criteria for identifying high yielding genotypes under drought conditions include cell membrane stability index, chlorophyll A content, and relative water content [16, 17].

Under the influence of abiotic stress factors, such as salinity, high temperature and water deficiency, the level of production of reactive oxygen species (free radicals, such as peroxide, superoxide) increases in plant tissue cells, which, in turn, leads to large-scale functioning, disturbances in cells and disruption of photosynthesis. This situation is called oxidative stress and is one of the main causes of disorders that occur in the plant organism as a result of the deterioration of external environmental conditions [15].

The decrease in photosynthesis is associated with the main components of

chloroplasts, which directly limit the photosynthetic potential of the plant [5]. The chlorophyll is one of the main components of chloroplasts. Chlorophyll pigments "a" and "b" in the composition of chlorophyll are considered important in the process of photosynthesis, the result of which depends on the growth and development of the plant [16].

The changes in the amount of total chlorophyll, chlorophyll "a" and "b" in plants have been studied [8, 12, 14, 9, 11]. It was found that the amount of chlorophyll "a", chlorophyll "b" and total chlorophyll decreased to a greater extent in sunflower varieties under drought conditions compared to the optimal water regime [17, 18].

It has been found that the total chlorophyll content decreases from 29% to 42% in both varieties of olives when grown under drought conditions compared to optimal water supply conditions [18, 19]. Cotton is characterized by a decrease in chlorophyll in drought conditions [19].

It is reported that the decrease in the content of chlorophyll under drought conditions is associated with the degradation of chlorophyll in the process of photooxidation [4, 9, 11].

As a result of their experiments, a number of scientists noted that the lack of water negatively affects the development of plants, and this, in turn, leads to a decrease in plant productivity by up to 50% [5, 6, 22, 23].

The problem of creating cotton varieties with a high level of plant productivity stability requires a comprehensive study of the relationship between growth, resistance to adverse environmental factors and cotton productivity. Because the physiological and biochemical processes in a plant



organism depend on the biological characteristics of the plant and environmental conditions. In other words,

### MATERIAL AND METHODS

In our study, KK-1795 (D-2), KK-1796 (D-3), L-1000 (D-4), S-9006 (D-5), KK-1086 (D-7) belong to *G. hirsutum* L. type), Catamarca 811 (D-12), C-9008 (D-14), Hapicala 19 (D-20), C-2025 (D-31), SAD-35-11 (D-34), L-45 (D-26), Father Zangi (D-27), KK-602 (D-32), C-4769 (D-25), O-30 (D-21), L-N1 (D-18), L-141 (D-19), Saenr Pena 85 (D-28), C-417 (D-39), Mekhnat, Andijan-35, Navbakhor-2, Ishonch, Tashkent-6, C-6524 varieties and the samples Namangan-77 were taken as objects, and under the conditions of the field experiment - control, that is, the optimal water supply (irrigation scheme 1-2-1, total volume of water used for irrigation, 4800-5000 m<sup>3</sup>/ha with seed water) and experiment, i.e. water deficit (irrigation scheme 0-1-0, the total amount of water used for irrigation was sown in four replications with seed water 1200-1500 m<sup>3</sup>/ha).

At the same time, plants under simulated drought conditions were grafted with only one watering at the beginning of flowering. On both backgrounds, the same agrotechnical work was carried out. When studying the drought resistance of cotton varieties, the amount of total chlorophyll, chlorophyll "a", chlorophyll "b", carotenoid pigments was determined by physiological parameters under conditions of optimal water supply (soil moisture 68% during flowering) and water deficit. (soil moisture 50% during flowering) was determined [25].

### RESULTS AND DISCUSSION

Determining the amount of chlorophyll is of great importance in studying the resistance of plants to adverse environmental factors. Because ensuring the overall productivity of plants

the realization of the genetic potential is determined by the level and limitations of the main environmental factors.

The experiment determined the amounts of chlorophyll 'a,' 'b,' and carotenoids in plant leaves. In this case, samples came from three to four leaves of the cotton plant, starting the count from the point of growth in field conditions. Placing each leaf of 50 mg in a test tube followed. Each leaf sample underwent homogenization in 5 ml of 95% ethyl alcohol solution (Sumanta et al., 2014).

The homogenate centrifugation took a speed of 5000 for 12 min. An Agilent Cary 60 UV-Vis spectrophotometer at 664, 649, and 470 nm determined the amounts of chlorophyll 'a,' 'b,' and carotenoids in the resulting extract. Based on this indicator, the amount of chlorophyll 'a,' 'b,' and carotenoids in cotton plant leaves was calculated using the following equation [25]:

$$\text{Chlorophyll 'a' (mg/g)} = 13.36 \times A_{664} - 5.19 \times A_{649}$$

$$\text{Chlorophyll 'b' (mg/g)} = 27.43 \times A_{649} - 8.12 \times A_{664}$$

$$\text{Carotenoid (mg/g)} = 1000 \times A_{470} - 2.13 \times \text{Chlo 'a'} - 97.63 \times \text{Chlo 'b'}/209$$

$$\text{Total Chlorophyll} = \text{Chlo 'a'} + \text{Chlo 'b'}$$

The determined levels of adaptation of cotton cultivars to water deficit conditions employed the method according to Eberhart and Russell [26].

The statistical analysis of the parameters of the studied traits under conditions of water deficit and optimal water supply, obtained on the basis of experiments, was carried out in EXSEL 2010 using ANOVA in the Stat View 5.0 program. is mainly related to the amount of chlorophyll and its components.

The decrease in photosynthesis is associated with the main components of chloroplasts, which directly limit the photosynthetic potential of the plant.



Chlorophyll is one of the main components of chloroplasts. Chlorophyll pigments "a" and "b" in the composition of chlorophyll are considered important in the process of photosynthesis, the result of which depends on the growth and development of the plant.

The main reason for the decrease in the amount of chlorophyll with a lack of water is the slowdown in photosynthetic activity. Low concentrations of photosynthetic pigments and reduced photosynthetic potential limit plant productivity.

The amount of chlorophyll in a leaf is one of the most important indicators from a physiological point of view. It is believed that the loss of chlorophyll in conditions of water deficiency is caused by the death of primary plant cells [27,

28, 29, 30, 31, 32, 33, 34, 35, 36]. In our experiments, when studying chlorophyll "a" from chloroplast pigments during the flowering-harvesting period of genotypes and samples of Upland cotton under conditions of optimal water supply, Upland cotton had the highest indicators at the level of the 3rd leaf in varietal samples Andijan-35, Zangi Ota and C-417 (13.84 mg/g, 13.03 mg/g and 14.75 mg/g, respectively), and the lowest values were found in L-N1 and L-141 varieties (8.72 mg/g and 8.95 mg/g).

Under conditions of water deficit, it was found in the highest values in Mehnat, Tashkent-6, C-6524 and Namangan-77 varieties, and in the lowest values were (5.73 and 6.90 mg/g, respectively) in KK-1795 and C-9006 cotton samples.

**Table 1**

**Indicators of chlorophyll a content in plants of Upland cotton genotypes under various water conditions**

Varieties and samples of Upland cotton	Optimal			Drought			Levels of Adaptation (%)
	Mean	SD	SE	Mean	SD	SE	
KK-1795	11,29	2,07	1,19	5,73	0,94	0,54	-49,27
KK-1796	11,75	1,63	0,94	7,05	1,11	0,64	-39,99
L-1000	11,66	1,13	0,65	8,73	2,79	1,61	-25,15
C-9006	11,98	1,45	0,83	6,90	1,15	0,66	-42,45
KK-1086	10,00	1,79	1,03	7,80	1,10	0,64	-22,01
Catamarca 811	11,93	0,95	0,55	10,32	2,14	1,24	-13,49
C-9008	10,59	1,03	0,59	8,07	1,25	0,72	-23,80
Hapicala 19	12,01	1,41	0,81	7,53	1,11	0,64	-37,29
C-2025	11,79	1,87	1,08	9,30	1,56	0,90	-21,15
SAD-35-11	11,02	3,74	2,16	8,83	2,47	1,43	-19,91
L-45	12,16	0,50	0,29	10,71	2,38	1,38	-11,89
Zangi ota	11,10	0,87	0,51	9,81	1,02	0,59	-11,63
KK-602	12,22	2,38	1,38	9,89	0,92	0,53	-19,12
C-4769	12,31	1,28	0,74	8,26	1,30	0,75	-32,89
O-30	10,15	2,16	1,25	8,95	0,50	0,29	-11,83
L-N1	8,72	3,24	1,87	10,04	1,56	0,90	15,09
L-141	8,95	3,20	1,85	10,87	0,96	0,56	21,46
Saenr Pena 85	11,87	1,03	0,60	10,29	0,19	0,11	-13,29
C-417	13,84	0,92	0,53	10,75	2,63	1,52	-22,30
Mekhnat	11,94	2,23	1,29	11,06	1,21	0,70	-7,36
Andijan- 35	13,03	0,11	0,06	9,80	1,53	0,88	-24,83
Navbahor-2	10,30	2,57	1,49	8,41	2,21	1,28	-18,38
Ishonch	12,63	0,97	0,56	9,36	1,24	0,71	-25,90



Tashkent- 6	10,48	3,06	1,76	11,97	1,36	0,78	14,27
C-6524	10,38	4,35	2,51	11,97	2,36	1,37	15,27
Namangan-77	12,56	3,77	2,17	11,29	0,56	0,32	-10,07

According to the analysis of the coefficient of adaptability, cotton samples KK-1795, C-9006, Hapicala 19 and C-4769 showed a strong sensitivity to water deficiency by the chlorophyll a marker and a weak sensitivity to Mekhnat variety. According to the results of the experiment, it can be said that the varieties Catamarka a 811, L-45, Zangi ota, O-30, Mekhnat and Namangan-77 were more adapted to the conditions of water deficit than other varieties in terms of chlorophyll a.

An analysis of our results on the determination of chlorophyll b showed that under conditions of optimal water

supply, that is, in the control variant, the amount of chlorophyll b in the leaves of the experimental group of the cotton variety was the highest in the samples of varieties C-9008, Navbahor-2 and Namangan-77 (respectively 5, 57 mg/g, 5,04 mg/g and 7,30 mg/g), and the lowest indicator of the trait was noted in KK-1796 sample and amounted to 2.83 mg/g. (table 2)

Against the background of water stress, the highest indicators in the group of ridges were noted in the Andijan-35 variety, the average indicator was 6.30 mg/g, and the lowest indicators in samples KK-1795 and KK-1796, and it formed 2.03 and 2.25. mg/g.

**Table 2**

**Indicators of chlorophyll b content in plants of Upland cotton genotypes under various water conditions**

Varieties and samples of Upland cotton	Optimal			Drought			Levels of Adaptation (%)
	Mean	SD	SE	Mean	SD	SE	
KK-1795	4,02	1,06	0,61	2,03	0,42	0,24	-49,55
KK-1796	2,83	0,61	0,35	2,25	1,02	0,59	-20,74
L-1000	3,58	1,69	0,98	2,52	0,88	0,51	-29,60
C-9006	3,59	0,35	0,20	3,15	0,08	0,05	-12,29
KK-1086	3,98	0,56	0,33	2,71	0,53	0,31	-31,95
Catamarka 811	3,03	0,42	0,24	2,39	0,65	0,38	-21,04
C-9008	5,57	0,80	0,46	2,96	0,47	0,27	-46,79
Hapicala 19	4,56	0,70	0,40	3,14	0,61	0,36	-31,00
C-2025	4,41	0,09	0,05	2,87	0,47	0,27	-34,90
SAD-35-11	3,38	0,90	0,52	3,75	0,92	0,53	11,16
L-45	3,08	0,29	0,16	3,14	0,83	0,48	1,94
Zangi ota	3,76	0,45	0,26	3,51	0,50	0,29	-6,56
KK-602	3,97	1,11	0,64	3,48	0,46	0,26	-12,40
C-4769	4,12	0,46	0,27	3,39	0,51	0,30	-17,63
O-30	4,00	0,69	0,40	2,81	0,16	0,09	-29,85
L-N1	4,28	0,79	0,45	2,56	0,56	0,32	-40,32
L-141	3,67	0,82	0,47	3,88	0,84	0,49	5,69
Saenr Pena 85	4,46	0,68	0,39	3,80	0,20	0,11	-14,95
C-417	3,98	0,32	0,18	3,44	0,83	0,48	-13,66
Mekhnat	3,95	0,58	0,34	4,05	0,65	0,37	2,42
Andijan- 35	4,75	0,51	0,30	6,30	1,58	0,91	32,64
Navbahor-2	5,04	0,97	0,56	4,14	0,61	0,35	-18,00
Ishonch	4,27	0,50	0,29	3,89	0,44	0,26	-8,99
Tashkent- 6	4,11	0,92	0,53	3,86	0,06	0,04	-5,88



C-6524	4,93	1,37	0,79	3,79	1,07	0,62	-23,28
Namangan-77	7,30	2,15	1,24	3,55	0,31	0,18	-51,40

According to the analysis of the coefficient of adaptability, cotton varieties KK-1795, C-9006, L-N1 and Namangan-77 showed a strong sensitivity to water deficiency in chlorophyll b and a weak sensitivity to L. -45 sample. It was noted that varieties of varieties SAD-35-11, L-45, Zangi ota, L-141, Mekhnat and Andijan-35 in traits of chlorophyll b are more adapted to water deficit conditions than other varieties.

The lowest values of total chlorophyll were found in samples L-N1

and L-141 (12.13 mg/g and 11.77 mg/g, respectively), and the highest values were found in varieties C-417 and Namangan-77 medium staple cotton under optimal water supply conditions (18.11 mg/g and 17.89 mg/g, respectively).

In plants of med Upland lines of cotton under conditions of water deficiency, the highest values were 16.2 mg/g for C-6524, and the lowest values were for accessions KK-1795 and KK-1796 (8.21 mg/g and 9.11 mg/g, respectively) (table 3).

**Table 3**

**Indicators of total chlorophyll content in plants of Upland cotton genotypes under various water conditions**

Varieties and samples of Upland cotton	Optimal			Drought			Levels of Adaptation (%)
	Mean	SD	SE	Mean	SD	SE	
KK-1795	14,97	3,10	1,79	8,21	1,36	0,78	-45,13
KK-1796	15,06	2,17	1,25	9,11	1,41	0,81	-39,49
L-1000	14,78	2,76	1,59	12,02	3,66	2,11	-18,72
C-9006	15,66	1,80	1,04	10,03	1,20	0,69	-35,98
KK-1086	13,81	2,26	1,30	11,10	1,57	0,90	-19,61
Catamarca 811	15,28	1,32	0,76	13,47	2,80	1,61	-11,86
C-9008	15,38	1,81	1,05	10,87	1,70	0,98	-29,32
Hapicala 19	16,34	2,06	1,19	10,67	1,68	0,97	-34,72
C-2025	16,25	1,79	1,03	12,27	1,99	1,15	-24,49
SAD-35-11	14,50	4,62	2,67	12,30	3,33	1,92	-15,20
L-45	15,56	0,72	0,42	14,72	3,19	1,84	-5,41
Zangi ota	14,38	1,31	0,76	13,78	1,51	0,87	-4,14
KK-602	16,74	3,41	1,97	13,88	1,30	0,75	-17,08
C-4769	16,25	1,73	1,00	11,29	1,81	1,04	-30,53
O-30	13,47	2,61	1,51	11,62	0,62	0,36	-13,77
L-N1	12,13	4,01	2,32	13,01	2,07	1,20	7,26
L-141	11,77	4,00	2,31	15,48	1,78	1,03	31,49
Saenr Pena 85	15,71	1,70	0,98	13,88	0,33	0,19	-11,61
C-417	18,11	1,24	0,72	14,08	3,43	1,98	-22,23
Mekhnat	15,41	2,79	1,61	14,97	1,76	1,02	-2,87
Andijan- 35	17,18	0,63	0,36	14,28	2,80	1,61	-16,90
Navbahor-2	14,23	3,54	2,04	11,84	2,81	1,62	-16,76
Ishonch	16,61	1,40	0,81	12,79	1,65	0,95	-22,98
Tashkent- 6	13,64	3,97	2,29	15,90	1,42	0,82	16,58
C-6524	13,73	5,73	3,31	16,02	3,42	1,98	16,67
Namangan-77	17,89	5,90	3,41	14,60	0,77	0,45	-18,38

According to the analysis of the coefficient of adaptability, samples of cotton varieties KK-1795, KK-1796, S-9006 and Hapicala a showed a strong sensitivity to water deficiency in traits of total chlorophyll, while variety L-N1 and varieties Menkhnat showed low sensitivity. The varietal samples L-45, Zangi ota, L-N1, L-141 and Menkhnat proved to be resistant to water deficit conditions in comparison with other varieties in terms of total chlorophyll.

When studying the content of carotenoids in plants of Upland cotton genotypes under various conditions of the water regime, the lowest values were 3 mg/g in samples KK-1795, KK-1086, C-9008, O-30, L-H1. and L-141 under conditions of optimal water supply in small quantities, and the highest value was found in samples C-417 at 3.72 mg/g (table 4).

**Table 4**  
**Indicators of carotenoid content in plants of Upland cotton genotypes under various water conditions**

Varieties and samples of Upland cotton	Optimal			Drought			Levels of adaptation (%)
	Mean	SD	SE	Mean	SD	SE	
KK-1795	2,98	0,54	0,31	1,71	0,33	0,19	-42,59
KK-1796	3,48	0,44	0,25	2,11	0,63	0,36	-39,45
L-1000	3,48	0,27	0,16	2,50	0,95	0,55	-28,38
C-9006	3,29	0,37	0,21	1,80	0,55	0,32	-45,25
KK-1086	2,62	0,40	0,23	2,37	0,32	0,19	-9,69
Catamarca 811	3,59	0,31	0,18	3,23	0,66	0,38	-10,16
C-9008	2,88	0,33	0,19	2,47	0,14	0,08	-14,22
Hapicala 19	3,38	0,44	0,25	2,27	0,23	0,13	-32,88
C-2025	3,47	0,01	0,01	2,87	0,31	0,18	-17,32
SAD-35-11	3,08	1,07	0,62	2,79	0,68	0,39	-9,39
L-45	3,47	0,24	0,14	3,34	0,81	0,47	-3,57
Zangi ota	3,24	0,04	0,02	2,83	0,31	0,18	-12,76
KK-602	3,43	0,64	0,37	2,78	0,13	0,08	-19,12
C-4769	3,31	0,09	0,05	2,57	0,42	0,24	-22,45
O-30	2,90	0,74	0,43	2,57	0,23	0,13	-11,36
L-N1	2,34	0,78	0,45	2,63	0,48	0,28	12,19
L-141	2,81	0,80	0,46	2,94	0,19	0,11	4,51
Saenr Pena 85	3,22	0,28	0,16	3,15	0,11	0,06	-2,20
C-417	3,72	0,33	0,19	3,22	0,66	0,38	-13,52
Mekhnat	3,52	0,45	0,26	2,94	0,45	0,26	-16,52
Andijan- 35	3,51	0,20	0,11	2,59	0,19	0,11	-26,13
Navbahor-2	3,15	0,67	0,39	2,33	0,45	0,26	-26,09
Ishonch	3,39	0,33	0,19	2,60	0,36	0,21	-23,16
Tashkent- 6	3,06	0,92	0,53	3,44	0,29	0,17	12,35
C-6524	3,03	1,03	0,60	3,60	0,64	0,37	18,67
Namangan-77	3,46	1,04	0,60	3,24	0,35	0,20	-6,24

Under conditions of water deficit, the highest value was 3.60 mg/g for the cotton variety C-6524, and the lowest value was found for cotton varieties KK-

1795 and C-9006 (1.71 mg/g and 1.80 mg/g).

According to the analysis of indicators of the coefficient of



adaptability, the samples of cotton varieties KK-1795, KK-1796, C-9006 and Hapicala have a strong sensitivity to water deficiency as a sign of carotenoid content, while L-N1, L-141 and Saenr Pena 85 have low sensitivity. The resistance of cultivars SAD-35-11, L-N1, L-141, Namangan-77 and Saenr Pena 85 to water deficit conditions was noted to be stable in comparison with other cultivars in terms of the carotenoid index.

When analyzing the general indicators of the physiological

characteristics of plants of medium fiber cotton genotypes under various conditions of the water regime, it was found that the amount of chlorophyll a, the amount of chlorophyll b, the total amount of chlorophyll, the amount of carotenoids, and the amount of chlorophyll content in plants decreases under conditions of water deficiency compared to conditions of optimal water mode.

**Table 5**

**General indicators of physiological characteristics of Upland cotton genotypes in plants under different water regimes**

№	Physiological traits	Optimal			Drought		
		Mean	SD	SE	Mean	SD	SE
1	Amount of chlorophyll a in plants	11,41	1,20	0,24	9,37	1,60	0,31
2	Amount of chlorophyll b in plants	4,18	0,89	0,18	3,36	0,84	0,16
3	Total chlorophyll content in plants	15,19	1,56	0,31	12,78	2,06	0,40
4	Carotenoid content in plants	3,22	0,33	0,06	2,73	0,48	0,09
5	Chlorophyll granules in plants	47,45	3,24	0,64	44,33	4,26	0,84

The main reason for the decrease in the amount of chlorophyll under drought stress is the slowdown in photosynthetic activity. At the same time, swelling of chloroplast membranes, impaired vesiculation of lamellae, and accumulation of lipid droplets were noted [18, 24, 28, 29, 37].

Low concentrations of photosynthetic pigments and reduced photosynthetic potential limit plant productivity. The amount of chlorophyll in a leaf is one of the important parameters from a physiological point of view. It is believed that the loss of chlorophyll under conditions of water deficiency is caused by the destruction of plant mantle cells [23, 27, 29, 33]. Carotenoids, which are the main components of plant chloroplasts, protect plants from photooxidation under stress [5, 17, 19, 26, 31, 34].

**Conclusion**

Based on the analysis of the indicators of chlorophyll and its components in various conditions of the water regime of cotton compared to the conditions of optimal water supply, the cotton samples of the varieties KK-1086, L-N1 and L-141 are unstable, Catamarka 811, L-45, Zangi ota, C-417, the samples of cotton varieties Mekhnat and Namangan-77 were stable.

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## SOME BIOCHEMICAL PERFORMANCE OF COLLECTION SAMPLES BELONGING TO

### *HELIANTHUS ANNUUS* L.

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**Annotation.** This article reveals the results of the analysis on seed oil content, total nitrogen and protein content of the seeds of sunflower samples of the *Helianthus annuus* L. species in the period of the years 2021-2023. The amount of oil in the seeds of the local Jakhongir variety was lower (33,4-33,8%) compared to the rest of the samples, and according to this indicator, sample 9853 (Russia) had the highest result (53,4-54,4%) among other sunflower varietal samples. In addition, according to the three-year analysis results of the total nitrogen and protein content of seeds, sample 9843 (Turkey) showed a



low indicator for total nitrogen content (2,6-2,9-3,2%) and the highest average indicator for protein content was recorded in 30837 (Australia; 21,6%), 33673 (France; 21,03%), 9848 (Russia; 21,13%) samples and recommended as a donor for breeding research.

**Key world:** Sunflower, collection, sample, analog, hydrolysis, total nitrogen, total protein, oil acid, coefficient of variation.

## НЕКОТОРЫЕ БИОХИМИЧЕСКИЕ ПОКАЗАТЕЛИ КОЛЛЕКЦИОННЫХ ОБРАЗЦОВ, ПРИНАДЛЕЖАЩИХ *HELIANTHUS ANNUUS L.*

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**Аннотация.** В статье представлены результаты анализа на содержание масла в семенах, содержание общего азота и белка в семенах образцов подсолнечника вида *Helianthus annuus L.* в период 2021-2023 гг. Количество масла в семенах местного сорта Джухонгир было ниже (33,4-33,8%) по сравнению с остальными образцами, причем по этому показателю наиболее высокий результат имел образец 9853 (Россия) (53,4-54,4%) среди других сортов образцов подсолнечника. Кроме того, по результатам трехлетнего анализа содержания общего азота и белка в семенах образец 9843 (Турция) показал низкий показатель по содержанию общего азота (2,6-2,9-3,2%) и самый высокий средний показатель по содержанию белка отмечен у 30837 (Австралия; 21,6%), 33673 (Франция; 21,03%), 9848 (Россия; 21,13%) образцов поэтому рекомендован в качестве донора для селекционных исследований.

**Ключевые слова:** Подсолнечник, коллекция, образец, аналог, гидролиз, общий азот, общий белок, масляная кислота, коэффициент вариации.

### Introduction

Sunflower (*Helianthus annuus L.*), belonging to the Compositae family, is an important oilseed crop worldwide. Currently, the demand for animal and vegetable proteins is increasing in the world, but animal proteins are expensive in terms of market price and environmental impact. Compared to other sources of plant protein, sunflower seeds are low in anti-nutritional factors.

Sunflower seeds are mainly used for their oil, but like other oilseeds, the residue after oil extraction is a valuable product because of its high protein

content. Sunflower protein mainly consists of albumins and globulins and therefore has high solubility [11].

The oil content of *Helianthus annuus L.* sprouts was reported to be positively correlated with the linoleic acid content of the oil. Based on the comparison of the data on the minimum and maximum temperatures of sunflower germination, it was concluded that there is an advantage of growing varieties with a low initial temperature for good seed germination when planting in early spring [1].

Proteins are the main and most important biological tool that cannot be replaced. Lack of protein weakens the body, the metabolism becomes more difficult, immune system weakens, the growth process stops, the work of internal secretion is disturbed, and other negative conditions are observed. While an excess of protein causes a change in sensitivity of the nervous system, malfunction of liver, kidney and other internal organs [3; 11].

Fats participate in metabolic processes of all substances in the body and have the effect of accelerating the metabolism. Most of the fat in the human body is used as energy material. Some fats enter the cell membrane and take part in its construction. In addition, lipids affect the use of proteins, mineral salts and vitamins in the body. With an increase in the amount of fat in the diet, it increases the release of calcium and magnesium salts and fatty acids from the body and reduces their absorption into the body, causing a decrease in the accumulation of calcium and phosphorus in bones [2, 9, 16, 17].

According to the information on vegetable oils, vegetable oils contain saturated and unsaturated fatty acids, and these fatty acids include oleic, lipoic, and linolenic fatty acids. The chemical composition of vegetable oils mainly consists of glycerides – 95,0-98,0%, free fatty acids – 1,0-2,0%, phosphatides - 1-2,0%, sterols – 0,3-0,5%, vitamins and carotenoids [8].

Taoufik Hosni et al. [15] analyzed the oil content and fatty acid content of 22 local sunflower samples in his research. The average oil content of sunflower plant samples was from 35,33% to 59,67%.

The sunflower plant has an oil content of 38,0% to 50,0% depending on the variety [10]. Sunflower oil contains a large amount of unsaturated fatty acids. About 70,0% of sunflower oil is linoleic acid. Other fatty acids in sunflower oil: myristic acid (m. 1 %), palmitic acid (3-6 %), stearic acid (1-3 m.), arachidic acid (0.5-4 %), oleic acid (14 g.). -35 wt%) and linolenic acid (< 1.5%) [13].

A study was conducted to investigate the effects of conventional magnesium and nano-magnesium fertilizers to improve yield characteristics in *Helianthus annuus* L. plants. The results of the experiment showed a significant improvement in parameters such as the number of days required for 50% flowering, sunflower head diameter, number of seeds per head, pollination rate, seed weight and oil content of these plants [14].

#### **The object and the methods of research**

The research was carried out in the experimental area of the "Biology" and "Genetics and Evolutionary Biology" departments of the Faculty of Natural Sciences of the Chirchik State Pedagogical University and in the "Molecular Biology and Bioinformatics" scientific laboratory.

**The object of the research** is foreign varietal collection samples: 9859 (USA), 30837 (Australia), 33673 (France), 9843 (Turkey), 30835 (Turkey), 9853 (Russia), 9848 (Russia) samples belonging to *Helianthus annuus* L. species and local Jakhongir (Uzbekistan) variety.

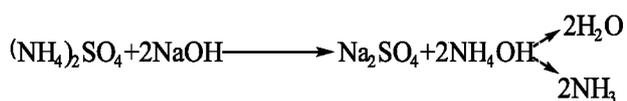
#### **Determination of total protein.**

One of the methods for determining the amount of total proteins is the Keldal method. According to this, the amount of total protein is calculated by determining the amount of nitrogen. The essence of the method is to hydrolyze the organic



substances in the sample with the help of concentrated sulfuric acid (amine groups in the protein) to form ammonium sulfate salts.

After the hydrolysis is complete, the ammonium sulfate formed is treated with sodium hydroxide to convert it to ammonia.



Ammonia or ammonium hydroxide formed as a result of neutralization is absorbed into sulfuric acid solution.

The remaining acid is titrated with an alkaline solution. The amount of nitrogen is calculated from the amount of ammonia calculated. An accurate sample for analysis is weighed from the average ground homogeneous sample of the studied sample into a test tube, the error rate should not exceed 0,1%. The sample is quantitated in a Keldal flask. Then the continuation of the experiment is carried out according to the instructions [5].

Processing of the obtained results: the mass fraction of nitrogen (X) in the analyzed sample is calculated by the formula as a percentage of the mass of the sample by the volume after the titration of the amount of ammonia passed through dilute sulfuric acid.

$$X = \frac{(V_1 - V_0) * K * 0.0014}{m} * 100\%$$

$V_0$ - volume of 0.1 mol/L sodium hydroxide solution used to titrate the excess 0.1 mol/L sulfuric acid solution in the sample experiment, ml.

Determination of the amount of oil in the seed. Seed degreasing was carried out in Sok-Slet first in acetone and then in ethyl ether. After removing the degreased samples from the apparatus, they were dried and the masses of flour and filter paper were measured and the difference

between the mass before and after degreasing was found. Based on this difference in mass, the amount of oil in the seed was determined as a percentage [6].

### Results of research

Biochemical performance of the sunflower samples selected for the study were analyzed, that is, when the indicator of oil content in seeds was compared according to the results of 2021-2023, it was observed that the oil content of seeds in the studied samples differed sharply from each other. According to the results of the biochemical analysis, the oil content of the seeds was 33,4-34,1% on average for the years in the standard local Jakhongir variety. The average indicator for other samples for this trait was 31,9-54,4% (Table 1).

The oil content of sunflower seeds planted in 2021 ranged from 31,9±0,51 to 54,4±0,87%. For example, in the sample 9853 (Russia), the highest index of oil content in seeds was 54,4±0,87%, and the coefficient of variation was 3.55%, while the lowest index was 31,9±0,51 % in the foreign sample 9859 (USA) (Table 1).

The oil content of sunflower seeds planted in 2022 ranged from 33.4±0.48 to 53.99±0.14%. In sunflower samples, the highest indicator of oil content in seeds was 53.99±0.14% in sample 9848 (Russia), and the coefficient of variation was 1.87%, and the lowest indicator for this trait was noted in the local Jakhongir (Uzbekistan) variety. This indicator ranged from 33.4±0.48% to 33.8±0.66% in 9859 (USA) samples (Table 1).

According to the results of the third year, a similar situation was observed in terms of the oil content of the seed. As for the results of the analysis of 2023, sample 9853 (Russia) had the highest index of



53,4±0,48%, coefficient of variation of 2,84% compared to other samples, and the lowest index of sample 9859 (USA) was 32,8±0,49%, coefficient of variation

was 2,96%. Similar values were also observed in the remaining sunflower samples (Table 1).

**Table-1**

**Oil content of seeds of sunflower plant samples (in 2021-2023)**

№	Samples	Oil content, % in 2021			Oil content, % in 2022			Oil content, % in 2023		
		$\bar{x} \pm S\bar{x}$	S	V %	$\bar{x} \pm S\bar{x}$	S	V%	$\bar{x} \pm S\bar{x}$	S	V %
1	Jakhongir (Uzbekistan)	33,8±0,56	1,24	3,67	33,4±0,48	1,07	3,20	34,1±0,62	1,31	3,23
2	9843 (Turkey)	40,8±0,73	1,64	4,01	39,7±0,82	1,83	4,60	40,3±0,64	1,53	3,48
3	9859 (USA)	31,9±0,51	1,13	3,54	33,8±0,66	1,48	4,37	32,8±0,49	1,14	2,96
4	30835 (Turkey)	41,3±0,71	1,58	3,82	39,5±0,28	0,61	1,56	40,9±0,67	1,44	3,57
5	30837 (Australia)	44,4±0,76	1,69	3,80	40,0±0,43	0,96	2,39	43,6±0,71	1,57	3,24
6	33673 (France)	44,9±0,20	0,46	1,01	41,7±0,73	1,61	3,85	42,3±0,52	1,24	3,08
7	9853 (Russia)	54,4±0,87	1,93	3,55	51,3±0,69	1,54	3,01	53,4±0,48	1,02	2,84
8	9848 (Russia)	51,9±0,91	2,03	3,91	53,9±0,14	0,97	1,87	52,6±0,39	1,08	1,92

The results of the total nitrogen and protein content of the sunflower samples selected for the study (2021-2023) were analyzed. It was observed that the total amount of nitrogen and protein in the seed content of the studied sunflower samples was not significantly different from each other. According to the results of the research analysis in 2021, the indicator for total nitrogen content of seeds was high (3,5%) in the samples 30837 (Australia), 33673

(France), 9848 (Russia), and the low indicator (2,6-2,9-3,2%) for this trait in the period of 2021-2023 was noted in 9843 (Turkey) sample. In addition, if we pay attention to the biochemical analyzes of 2021 on the indicator of protein content, a high rate (21,8%) was found in samples 30837 (Australia), 33673 (France), 9848 (Russia) for this trait. A low indicator (18,1%) for the trait on protein content was observed in sample 9843 (Turkey) (Table 2).

**Table-2**

**Total nitrogen and protein content in sunflower plant samples (in 2021-2023)**

Total nitrogen content, % (in 2021)		Protein content, % (in 2021)		Total nitrogen content, % (in 2022)		Protein content, % (in 2022)		Total nitrogen content, % (in 2023)		Protein content, % (in 2023)	
$\bar{x} \pm S\bar{x}$	V%	$\bar{x} \pm S\bar{x}$	V %	$\bar{x} \pm S\bar{x}$	V%	$\bar{x} \pm S\bar{x}$	V %	$\bar{x} \pm S\bar{x}$	V%	$\bar{x} \pm S\bar{x}$	V %
<b>Jakhongir (Uzbekistan)</b>											
3,08±0,10	5,8	19,2±0,31	2,7	4,1±0,20	8,3	20,2±0,15	1,7	3,4±0,15	7,6	18,9±0,26	2,4



9843 (Turkey)											
2,9±0,12	6,9	18,1±0,17	1,6	3,2±0,19	10,3	19,2±0,23	2,1	2,6±0,12	8,1	19,8±0,59	5,1
9859 (USA)											
3,15±0,20	11,1	19,7±0,44	3,8	4,0±0,21	9,1	18,8±0,33	3,0	2,7±0,27	17,3	20,2±0,38	3,2
30835 (Turkey)											
3,3±0,21	10,9	20,8±0,35	2,8	4,1±0,23	9,7	21,6±0,29	2,7	3,1±0,23	12,9	19,6±0,56	4,9
30837 (Australia)											
3,5±0,36	17,8	21,8±0,35	2,6	4,2±0,21	8,5	22,6±0,57	4,3	3,0±0,26	14,8	20,6±0,46	3,8
33673 (France)											
3,5±0,12	5,7	21,8±0,45	3,5	4,0±0,13	5,1	20,5±0,50	3,9	3,1±0,21	11,6	20,8±0,62	5,1
9853 (Russia)											
3,3±0,25	12,2	20,6±0,51	4,3	3,9±0,12	5,0	21,1±0,55	4,5	2,8±0,17	10,7	19,9±0,21	1,8
9848 (Russia)											
3,5±0,15	7,2	21,8±0,35	2,7	3,9±0,16	6,7	20,4±0,50	4,2	4,1±0,32	13,5	21,2±0,66	5,3

A similar situation was observed in terms of total nitrogen and protein content of sunflower seeds planted in 2022. The average indicator for total nitrogen content was 3,2-4,2%, the coefficient of variation was 8,5-10,3%. In sunflower sample 30837 (Australia), the highest indicator for total nitrogen content was 4,2%, the coefficient of variation was 8,5%, while the relatively low indicator for this trait was 3,2% on average in sample 9843 (Turkey), the coefficient of variation was 10,3%. At the same time, the highest rate for protein content was noted in sample 30835 (Turkey) 21,6%, and the lowest rate for this trait was 18,83% in sample 9859 (USA) (Table 2).

In sunflower samples planted in the third (2023) year, there was a decrease in total nitrogen and protein content of seeds. For example, when the trait of total nitrogen content was analyzed, there was a decrease noted in the samples 9843 (Turkey) (2,6%), 9859 (USA) (2,72%), 30837 (Australia) (3,03%), 9853 (Russia) (2,8 %) compared to the results of 2021-2022. The total nitrogen content of the local Jakhongir variety was 3,4%. In addition, a slight

decrease in protein content was observed in the local Jakhongir variety (18,9%), while the protein content index was 21,2% in sample 9848 (Russia), and the coefficient of variation was 5,32%.

### Conclusion

The analysis of the obtained biochemical studies showed that the oil in the seed content, total nitrogen and protein content of the sunflower samples of the *Helianthus annuus* L. species were analyzed in the period 2021-2023. The amount of oil in the seeds of the local Jakhongir variety was lower (33,4-33,8%) compared to the rest of the samples, and the sample 9853 (Russia) had the highest result (53,4-54,4%) according to this trait. In addition, according to the three-year results of the total nitrogen and protein content of seeds, sample 9843 (Turkey) showed a low total nitrogen content (2,6-2,9-3,2%) and the highest average protein content was recorded in 30837 (Australia; 21,6%), 33673 (France; 21,03%), 9848 (Russia; 21,13%) samples and recommended as a donor for selective research.

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## ANALYSIS OF PHOTOSYNTHETIC PIGMENTS IN FOREIGN SAMPLES BELONGING TO *LATHYRUS SATIVUS* L. SPECIES

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**Abstract.** This article presents the results of the analysis of the amount of chlorophyll "α", chlorophyll -"β", total chlorophyll, carotenoid pigments in the leaves of foreign collection samples of *Lathyrus sativus* L. species and in the leaves of common chickpea varieties taken for control. In the analysis, the highest indicator of the amount of chlorophyll-"α" pigment in the bud formation phase was  $21.47 \pm 0.71$  mg/g in the sample Bio 520x1330, and the highest indicators in the amount of chlorophyll "α" pigment in the flowering phase was  $21.37 \pm 0.11$  mg /g in the sample Ratan x IG 135481, the highest result for the amount of chlorophyll "α" pigment in the ripening phase was observed in the Bio 520x1330 sample,  $11.92 \pm 0.05$  mg/g. In the analyzed grass chickpea samples, the highest indicator of chlorophyll-"β" content in the bud formation phase was  $12.79 \pm 0.70$  mg/g in the Bio (520 x Bio)x273 sample, and the highest index of the chlorophyll-"β" pigment content in the flowering phase was in the Ratan x2125 sample,  $16.51 \pm 0.31$  mg/g, the highest index for this trait in the ripening phase was  $17.21 \pm 0.17$  mg/g in the sample Bio (520 x Bio)x 274. In the analyzed samples, the highest index of total chlorophyll in the bud formation phase was  $31.56 \pm 0.56$  mg/g in the Bio 520 x Bio??) x 274 sample, and the highest index of the total chlorophyll pigment in the flowering phase in the Bio (520xBio)x 274 sample,  $34.47 \pm 0.16$  mg/g, while in the ripening phase, the highest value for this trait was noted in the Ratan x 2125 sample,  $25.62 \pm 0.09$  mg/g, the coefficient of variation was 0.59%. In the foreign samples of *Lathyrus sativus* L. species, the higher value for carotenoid content was noted in the budding phase,  $5.57 \pm 0.13$  mg/g in the Ratan x IG 135481 sample, while in the flowering phase, the carotenoid content was higher than the others in the Prateek x IG 140034 sample, 6.02 mg/g, in the ripening phase the highest index was noted in the Prateek x IG 140034 sample a (5.33 mg/g) compared to the rest.

The results of the conducted research show that the concentration of chlorophyll--"α", chlorophyll-"β", total chlorophyll and carotenoid pigments in the analyzed samples and varieties during the growing season was slightly low, while it was maximally high during the flowering phase and a partial decrease was observed in indicators during the ripening phase of the plant.

**Key words.** Chickpea, phase, anthocyanin, photosynthetic pigment, spectrophotometric analysis, light absorption, chlorophyll-a, chlorophyll-b, total chlorophyll, carotenoids.

## АНАЛИЗ ФОТОСИНТЕТИЧЕСКИХ ПИГМЕНТОВ В ЗАРУБЕЖНЫХ ОБРАЗЦАХ, ПРИНАДЛЕЖАЩИХ К ВИДУ *LATHYRUS SATIVUS* L.

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**Аннотация.** В статье представлены результаты анализа количества хлорофилла «α», хлорофилла-«β», общего хлорофилла, каротиноидных пигментов в листьях зарубежных коллекционных образцов вида *Lathyrus sativus* L. и в листьях сортов нута обыкновенного, взятых для контроля. При анализе наибольший показатель количества пигмента хлорофилла «α» в фазе бутонобразования составил  $21,47 \pm 0,71$  мг/г у образца Bio 520x1330, а самые высокие показатели количества пигмента хлорофилла «α» в фазе цветения составила  $21,37 \pm 0,11$  мг/г у образца Ratan x IG 135481, наибольший результат по количеству пигмента хлорофилла «α» в фазе созревания отмечен у образца Bio 520x1330 -  $11,92 \pm 0,05$  мг/г. В проанализированных образцах травяного нута наибольший показатель содержания хлорофилла-«β» в фазе бутонобразования составил  $12,79 \pm 0,70$  мг/г в образце Bio (520 x Bio)x273, а самый высокий показатель содержания хлорофилла-«β» Содержание пигмента в фазе цветения составило в образце Ratan x 2125  $16,51 \pm 0,31$  мг/г, наибольший показатель по этому признаку в фазе созревания составил  $17,21 \pm 0,17$  мг/г в образце Bio (520 x Bio)x 274. В анализируемых образцах наибольший показатель общего хлорофилла в фазе бутонобразования составил  $31,56 \pm 0,56$  мг/г у образца Bio 520 x Bio??) x 274, а самый высокий показатель общего пигмента хлорофилла в фазе цветения у образец Bio (520 x Bio) x 274 -  $34,47 \pm 0,16$  мг/г, тогда как в фазе созревания наибольшее значение по данному признаку отмечено у образца Ratan x 2125 -  $25,62 \pm 0,09$  мг/г, коэффициент вариации составил 0,59%. У зарубежных образцов вида *Lathyrus sativus* L. более высокое значение содержания каротиноидов отмечено в фазе бутонизации -  $5,57 \pm 0,13$  мг/г у образца Ratan x IG 135481, тогда как в фазу цветения содержание каротиноидов было выше, чем у образца Ratan x IG 135481. Остальные в образце Prateek x IG 140034 -  $6,02$  мг/г, в фазе созревания наибольший показатель отмечен у образца Prateek x IG 140034 а ( $5,33$  мг/г) по сравнению с остальными.

Результаты проведенных исследований показывают, что концентрация хлорофилла-«α», хлорофилла-«β», суммы хлорофиллов и каротиноидных пигментов в анализируемых образцах и сортах в период вегетации была незначительно низкой, а в период вегетации была максимально высокой. В фазу цветения и частичное снижение показателей наблюдалось в фазу созревания растения.

**Ключевые слова.** Нут, фаза, антоциан, фотосинтетический пигмент, спектрофотометрический анализ, светопоглощение, хлорофилл-«α», хлорофилл-«β», общий хлорофилл, каротиноиды.

## Introduction

Due to the growing population of the world, the demand for nutritional protein is increasing, which requires the selection and breeding of samples with a high yield level, resistant to various diseases by studying the physiological characteristics of the reproduction of leguminous and cereal plants by planting samples from the world collection.

Grass chickpea (*Lathyrus sativus* L.) is a cool-season legume grown mainly for food in the Indian subcontinent and Ethiopia, and as a food and fodder crop in other parts of the world. *Lathyrus sativus*

L. has various beneficial agronomic properties such as tolerance to salinity, drought, insect and biotic stresses, and good growth in swampy, semi-arid and problematic soils. At the same time, as a leguminous crop, it enriches the soil with nitrogen. These properties of the plant show it as a useful crop for maintaining agricultural productivity and popularizing it in changing climates. In terms of nutritional value, these crops are very rich in protein, ranks second only to soybeans, and provide balanced amino acids to poor people in the countries where they are consumed, together with



cereals. In addition, it increases the supply of L-homoarginine nitric oxide (NO), which has heart-healthy potential, and improves endothelial functions in the body by activating cardiovascular activity [1, 6, 13].

It is known that green plants have the ability to accumulate organic matter and release molecular oxygen into nature. This process is closely related to the photosynthetic activity of plants. Photosynthesis is an important physiological process that determines plant growth, development, productivity, and crop quality [2, 5, 8, 14].

One of the most important physiological processes observed in plants is the synthesis of organic substances directly related to the process of photosynthesis. Changes in the rate of photosynthesis are associated with the main components of chloroplasts, which determine plant photosynthetic productivity [7].

Photosynthesis plays an important role in increasing productivity along with oxygen production. Therefore, man provides himself with nutrients by creating an artificial biocenosis in the fields with the help of solar energy and agricultural practices, thereby increasing productivity [3].

Along with chlorophyll pigments found in plants, there are red, yellow, and orange pigments, i.e., carotenoids, and these pigments participate in absorbing the light rays necessary for photosynthesis, releasing molecular oxygen, and protecting the chlorophyll molecule from strong light effects [4].

Photosynthetic pigments are substances with very diverse chemical structures; they exist in the form of porphyrin pigments (chlorophyll-“α”, chlorophyll-“β”), carotenoids, anthocyanins [10].

The ratio of chlorophyll-“α” and chlorophyll-“β” in land plants was used as an indicator of their response to shaded conditions.

Ethyl is a safer solvent than acetone or methanol, but is not often used for chlorophyll analysis, although it is flammable when it contains chlorophyll-“α”, chlorophyll-“β”, and is not very toxic and suitable for laboratory use [12].

**Research object** is (Bio 520 x Bio) x273a, Pratek x IG 140034a, Ratan x 2125, Jabbouleh, 1330 x 2125, Ratanx 1307, Ratan x IG 135481, (Bio 520 x Bio) x 273b, Prateek x IG 140034b, Bio 520x1330 foreign collection samples belonging to *Lathyrus sativus* L. species and common chickpea varieties Lalmikor and Polvon.

**Research methods** are the analyses of the amounts of chlorophyll-“α”, chlorophyll-“β” and carotenoids in the leaves of the plant under the experiment. For this, samples were taken from the 3-4 leaves of the plant, counting from the growing point of the plant under field conditions. 50 milligrams of each leaf was placed in a test tube. Each leaf sample was homogenized in 5 ml of 95% ethyl alcohol solution. The homogenate was centrifuged at a speed of 5000 for 12 minutes. The amounts of chlorophyll “α”, chlorophyll “β” and carotenoids in the resulting extract were determined by an Agilent Cary 60 UV-Vis spectrophotometer at 664, 649 and 470 nm.. Based on this indicator, the amounts of chlorophyll “α”, chlorophyll “β” and carotenoids in plant leaves were calculated using the following equation [9].

$$\text{chlorophyll -“ } \alpha \text{” [mg/g]} = 13.36A_{664} - 5.19 \cdot A_{649},$$

$$\text{chlorophyll -“ } \beta \text{” [mg/g]} = 27.43A_{649} - 8.12 \cdot A_{664}$$

$$\text{carotenoid [mg/g]} = (1000A_{470} - 2.13 \cdot \text{chlo } \alpha - 97.63 \cdot \text{chlo } \beta) / 209$$

$$F (\text{Mg/g}) = (V \cdot S) / P$$

The statistical analysis of the amounts of total chlorophyll, chlorophyll "α", chlorophyll "β" and carotenoid content of foreign collection samples belonging to *Lathyrus sativus* L. species obtained on the basis of research was carried out based on the ANOVA program.

### Research results

During the study, the results of the obtained amounts of chlorophyll "α", chlorophyll "β", total chlorophyll and carotenoid content in the leaves of the plant in the bud formation, flowering and ripening phases of the foreign collection samples belonging to *Lathyrus sativus* L. species were analyzed (Fig. 1). The amount of chlorophyll "α", chlorophyll "β", total chlorophyll and carotenoid content produced in the photosynthesis in the plant leaves of foreign collection samples belonging to grass chickpea (*Lathyrus sativus* L.) species was analyzed spectrophotometrically during the bud formation phase (diagram 1).

The amount of chlorophyll "α" in *Lathyrus sativus* L. samples was noted to be different. In particular, the amount of chlorophyll "α" was  $17.49 \pm 0.21$  mg/g in the Lalmikor variety, and the coefficient of variation was 2.04%; in the Polvon variety, it was  $17.43 \pm 0.38$  mg/g, and the coefficient of variation was 3.76%. The highest indicator for this trait in foreign samples was  $21.47 \pm 0.71$  mg/g in the Bio 520x1330 sample, the coefficient of variation was 5.73%, while the lowest indicator for this trait was  $13.09 \pm 0.44$  mg/g in the Ratan x 2125 sample, and the coefficient of variation was found to be 5.77% respectively (diagram 1).

In the studied grass chickpea samples, the highest indicator of

chlorophyll "β" content was found to be  $12.79 \pm 0.70$  mg/g in the sample Bio (520 x Bio)x273, the coefficient of variation was 9.49%, while the lowest indicator was  $5.28 \pm 0.22$  mg/g in the sample Ratan x IG 135481, and the coefficient of variation was 7.22% respectively. In Lalmikor and Polvon varieties taken as a control option, this indicator was  $4.08 \pm 0.22$  mg/g to  $9.89 \pm 0.50$  mg/g, and the coefficient of variation was 8.79-9.33% (diagram 1). Total chlorophyll content was analyzed in foreign samples of *Lathyrus sativus* L. and in domestic common pea cultivars. In particular, the lowest indicator of total chlorophyll content in foreign samples was found to be  $21.16 \pm 0.29$  mg/g in the Ratan x 2125 sample, the coefficient of variation was 2.4%, while the highest indicator was  $31.56 \pm 31.56$  in the Bio 520 x Bio??)x274 sample, 0.56 mg/g, the coefficient of variation was 3.05%. In the Lalmikor variety of common chickpea taken as a control, this indicator was found to be  $27.38 \pm 0.59$  mg/g, correspondingly, the coefficient of variation was 3.71%, in the Polvon variety it was  $21.51 \pm 0.17$  mg/g, the coefficient of variation was up to 1.35%.

As a result of the analysis of carotenoid content in grass chickpea (*Lathyrus sativus* L.) variety samples, the highest indicators were recorded in the Ratan x IG 135481 sample,  $5.57 \pm 0.13$  mg/g, the coefficient of variation was 4.04%, and the lowest indicators were determined in the sample Bio 520 x Bio??)x273,  $2.47 \pm 0.12$  mg/g, the coefficient of variation was observed to be 8.5%.

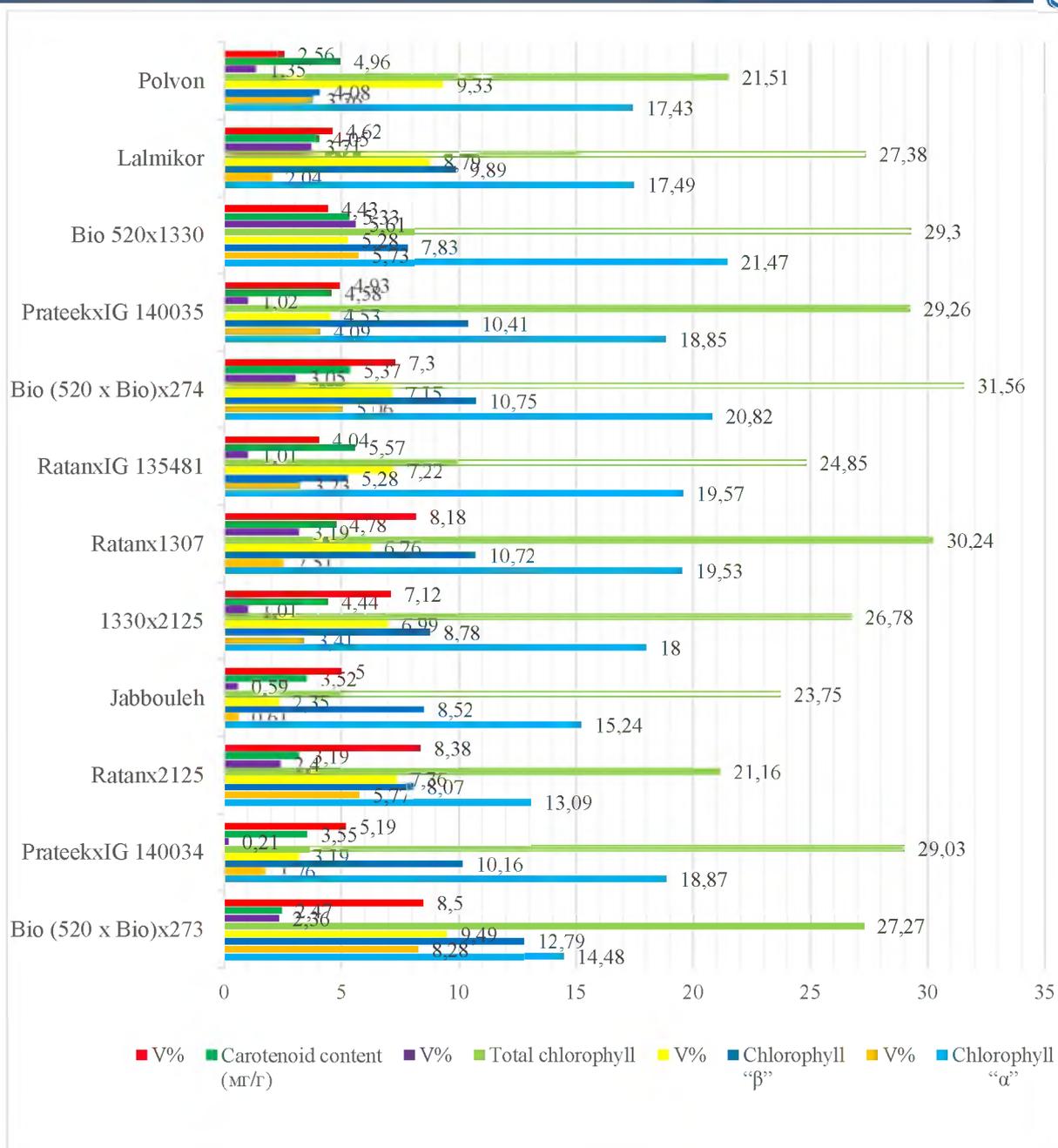


Diagram 1. Analysis of photosynthetic pigments in the budding phase of foreign samples belonging to *Lathyrus sativus* L. species.

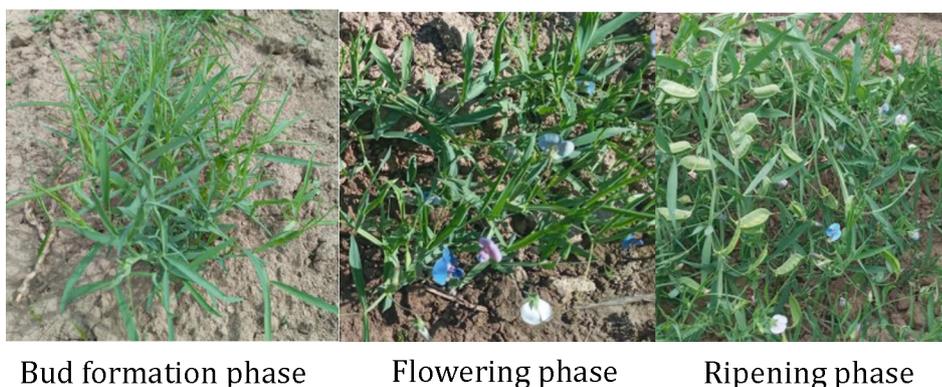


Figure-1. Vegetation period of foreign collection samples belonging to grass chickpea (*Lathyrus sativus* L) species.



The amount of chlorophyll-“ $\alpha$ ”, chlorophyll-“ $\beta$ ”, total chlorophyll and carotenoid content produced by photosynthesis in the foreign collection samples of grass chickpea (*Lathyrus sativus* L) species was analyzed spectrophotometrically in the flowering phase of plant leaves (diagram 2).

In the grass chickpea samples, the lowest indicators of chlorophyll-“ $\alpha$ ” was noted to be  $16.34 \pm 0.13$  mg/g in the sample Bio (520 x Bio)x273, the coefficient of variation was 1.33%, and the highest indicators for this trait in the sample Ratan x IG 135481 was  $21.37 \pm 0.11$  mg/g, and the coefficient of variation was 0.88% respectively. In terms of this trait, the indicators in the local Polvon variety were  $20.51 \pm 0.03$  mg/g, the coefficient of variation was 0.24%, while the slightly higher indicators were found in the Lalmikor variety,  $21.34 \pm 0.33$  mg/g, and the coefficient of variation was 2.65 % respectively.

The highest indicators of chlorophyll-“ $\beta$ ” content in foreign grass chickpea samples were  $13.55 \pm 0.23$  mg/g in the Bio (520 x Bio)x274 sample, the coefficient of variation was 2.99%, and the indicators in the Ratan x 2125 sample were  $16.51 \pm 0.31$  mg/g, the coefficient of variation was 3.21%, while the lowest indicator was  $7.48 \pm 0.39$  mg/g, the coefficient of variation was 9.09% in the Prateek x IG 140034 sample. In the Lalmikor variety taken as a control, the indicators were  $8.28 \pm 0.42$  mg/g, the coefficient of variation was 8.74%, and in the Polvon variety it was  $11.29 \pm 0.15$  mg/g, the coefficient of variation was 2.37% (2 -diagram).

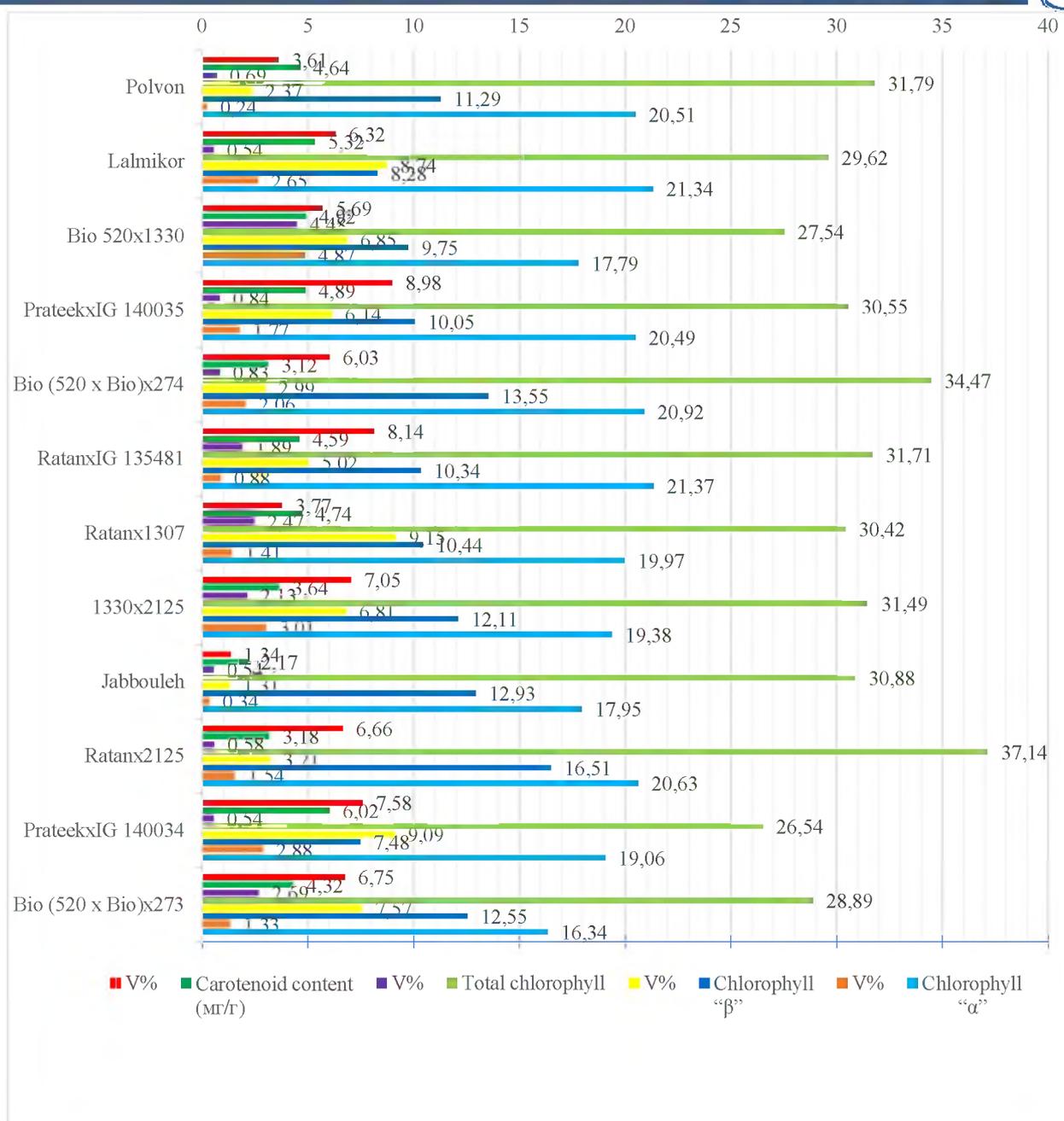
In the collection samples belonging to the *Lathyrus sativus* L. species, the

carotenoid content in the Jabbouleh sample was slightly lower than other samples (indicator  $2.17 \pm 0.02$ , coefficient of variation 1.34%), while in the Prateek x IG 140034 sample, it was higher than the rest (6.02 mg/g, respectively, the coefficient of variation 7.58%). The lowest indicator of carotenoid content was 4.64 mg/g in Polvon variety taken as a control, and the coefficient of variation was 3.61%.

Total chlorophyll content was analyzed in foreign samples belonging to *Lathyrus sativus* L. species and in domestic common chickpea cultivars. The total chlorophyll content in the studied samples and varieties ranged from  $26.54 \pm 0.08$  mg/g to  $34.47 \pm 0.16$  mg/g. The lowest indicator for this trait was  $26.54 \pm 0.08$  mg/g in the foreign Prateek x IG 140034 sample, the coefficient of variation was 0.54%, while the highest indicator in the Bio (520xBio)x274 sample was  $34.47 \pm 0.16$  mg/g, the coefficient of variation was 0.83%.

It was found that in common chickpea variety Lalmikor taken as a control, the indicators were  $29.62 \pm 0.16$  mg/g, coefficient of variation 0.54%, while in Polvon variety the indicator was  $31.79 \pm 0.13$  mg/g, coefficient of variation was 0.69%.

A spectrophotometric analysis was carried out on the amounts of chlorophyll-“ $\alpha$ ”, chlorophyll-“ $\beta$ ”, total chlorophyll and carotenoid content produced as a result of photosynthesis in the leaves of plants of the foreign collection samples of grass chickpea (*Lathyrus sativus* L.) species and common chickpea varieties during the pod formation phase (Table 3).



**Diagram 2. Analysis of photosynthetic pigments in foreign samples of *Lathyrus sativus* L. species during the flowering phase.**

The lowest indicators of chlorophyll-“α” among foreign samples were  $8.02 \pm 0.26$  mg/g in the Bio (520 x Bio)x273 sample, and the coefficient of variation was 5.62%, while in the Bio (520 x Bio)x274 sample the indicators were  $8.10 \pm 0.27$  mg/g, the coefficient of variation was up to 5.74%, the highest indicator for this trait was  $11.92 \pm 0.05$  mg/g, and the coefficient of variation was

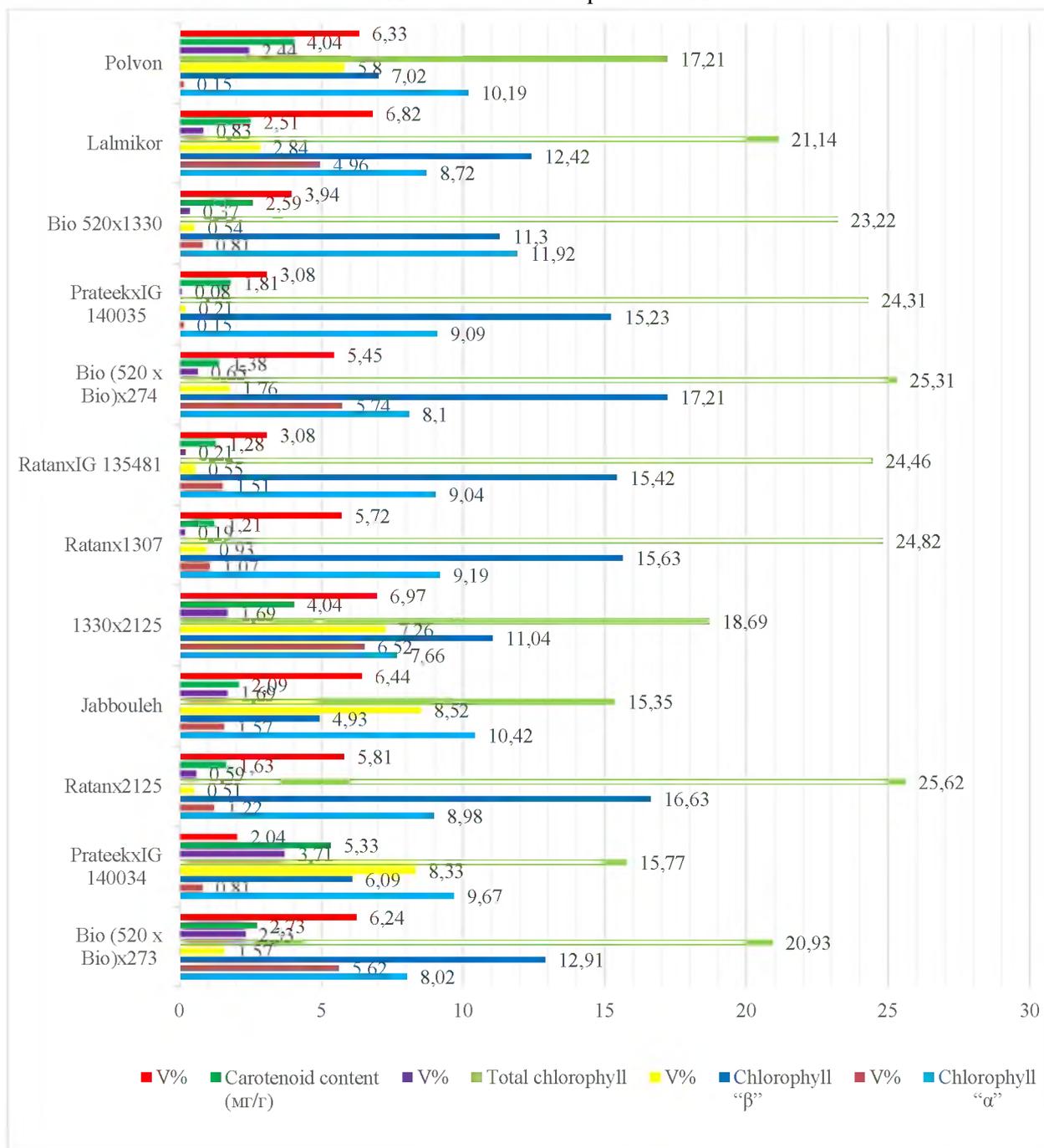
0.81% in the Bio 520x1330 sample. The amount of chlorophyll-“α” in Lalmikor and Polvon varieties taken as a control was found to be from  $8.72 \pm 0.25$  mg/g to  $10.19 \pm 0.01$  mg/g, and the coefficient of variation was 0.15- 4.96% respectively.

The highest indicator for the amount of chlorophyll-“β” among foreign chickpea samples was  $17.21 \pm 0.17$  mg/g in the Bio (520 x Bio)x274 sample, and



the coefficient of variation was 1.76%, and this indicator was  $15.63 \pm 0.08$  mg/g in the Ratan x 1307 sample, and the coefficient of variation was up to 0.93%, while the lowest value was  $4.93 \pm 0.24$  mg/g in the Jabbouleh sample, and the coefficient of variation was 8.52%. It was

observed that the parameters of the Polvon variety taken as a control were  $7.02 \pm 0.23$  mg/g for this trait, and the coefficient of variation was 5.8%, and in the Lalmikor variety it was  $12.42 \pm 0.20$  mg/g, and the coefficient of variation was up to 2.84%.



**Diagram 3. Spectrophotometric analysis of photosynthetic pigments of foreign samples belonging to *Lathyrus sativus* L. species in the ripening phase.**

In the collection samples belonging to *Lathyrus sativus* L. species, the average carotenoid content was  $2.59 \pm 0.06$  mg/g,

and the coefficient of variation was 3.94%. The Ratan x1307 sample selected for the study had a slightly lower value



(1.20 mg/g, coefficient of variation 5.72%) compared to other samples, while the Prateek x IG 140034 sample had a higher value (5.33 mg/g, coefficient of variation 2.04%) compared to the rest. Lalmikor variety, taken as a control, had the lowest carotenoid content of 2.51 mg/g, its coefficient of variation was 6.82%.

Total chlorophyll content was analyzed in foreign samples of grass chickpea and in domestic common chickpea varieties. The total chlorophyll content in the studied samples and varieties ranged from  $15.35 \pm 0.15$  mg/g, the coefficient of variation from 1.69% to  $25.62 \pm 0.09$  mg/g, and the coefficient of variation to 0.59%. The lowest indicator for this trait was found to be  $15.35 \pm 0.15$  mg/g, coefficient of variation 1.69% in the foreign Jabbouleh sample, while the highest indicator was  $25.62 \pm 0.09$  mg/g, coefficient of variation was 0.59 % in the Ratan x2125 sample. In Polvon and Lalmikor varieties taken as a control, these indicators were recorded from  $17.21 \pm 0.24$  mg/g to  $21.14 \pm 0.10$  mg/g, accordingly, the coefficient of variation was 0.83-2.44%.

### Conclusion

When the amount of chlorophyll "α", chlorophyll "β", total chlorophyll, carotenoid pigments was analyzed in the leaves of foreign collection samples belonging to *Lathyrus sativus* L. species and in common chickpea varieties taken for control, the highest indicator of the amount of chlorophyll "α" pigment in the budding phase was  $21.47 \pm 0.71$  mg/g in the sample Bio 520x1330, the highest result in the amount of the pigment chlorophyll "α" in the flowering phase was observed to be  $21.37 \pm 0.11$  mg/g in the sample Ratan x IG 135481, while in the ripening phase the highest result for

the amount of chlorophyll "α" pigment was observed in Bio 520x1330 sample  $11.92 \pm 0.05$  mg/g. In the studied grass chickpea samples, the highest indicator of chlorophyll "β" in the budding phase was  $12.79 \pm 0.70$  mg/g in the Bio (520 x Bio)x273 sample, and in the flowering phase the highest indicator of chlorophyll "β" pigment was  $16.51 \pm 0.31$  mg/g in the Ratan x2125 sample, in the ripening phase the highest indicator for this trait was  $17.21 \pm 0.17$  mg/g in the Bio (520 x Bio)x274 sample. In the analyzed samples, the highest indicator of the amount of total chlorophyll in the bud formation phase was  $31.56 \pm 0.56$  mg/g in the sample Bio 520 x Bio??)x274, in the flowering phase the highest indicator in the amount of total chlorophyll pigment was  $34.47 \pm 0.16$  mg/g in the sample Bio (520xBio)x274, while in the ripening phase the highest indicator for this trait was recorded in the Ratan x2125 sample,  $25.62 \pm 0.09$  mg/g, the coefficient of variation was 0.59%. In foreign samples belonging to *Lathyrus sativus* L. species, the highest indicator for carotenoid content was found to be  $5.57 \pm 0.13$  mg/g in Ratan x IG 135481 sample in the budding phase, in the flowering phase Prateek xIG 140034 sample showed 6.02 mg/g indicators which were higher than the others, also in the ripening phase the Prateek xIG 140034 sample had a higher value (5.33 mg/g) than the rest.

The results of the conducted research show that the indicators for the concentration of chlorophyll-"α", chlorophyll-"β", total chlorophyll and carotenoid pigments in the analyzed samples and varieties during the growing season were slightly low, while they were maximally high during the flowering phase and a partial decrease in the



indicators during the ripening phase was noted.

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**Согласно решению Высшей  
аттестационной комиссии  
Республики Узбекистан от 31 марта  
2023 года № 332/5/6 публикация  
основных научных результатов  
диссертаций по биологическим  
наукам включена в перечень  
рекомендуемых национальных  
научных изданий.**

**According to the decision of the Higher  
Attestation Commission of the  
Republic of Uzbekistan dated March  
31, 2023 No. 332/5/6, the publication  
of the main scientific results of  
dissertations in biological sciences is  
included in the list of recommended  
national scientific publications.**