Influence of the chemical composition of mulberry on the growth dynamics of silkworm larvae

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Abstract. In countries where sericulture is well-developed, there is a pressing need to create new systems, varieties, and hybrids of the Morus alba L. species, which is the only food source for the mulberry silkworm Bombyx mori L. These new systems should be suitable for the care of silkworms, and their nutritional and chemical composition should be studied. The introduction of these new systems into production is considered a crucial problem that needs to be addressed. Research experiments were carried out in 2013-2017 at the Tashkent State Agrarian University and the Silk Research Institute. The results showed that in 2017, the new varieties contained 75.1-74.3% water, while in 2018, the water content was 76.0-77.3%. Meanwhile, the Tajikskaya bezsemyannaya variety showed an increase in this indicator, reaching 75.2-76.2%. However, these results were still lower than the benchmark of 98.8-101.4%. The amount of protein is one of the most important indicators of mulberry leaf composition.

1 Introduction

In countries with developed sericulture network, creation of new systems, varieties and hybrids of Morus alba L. species, which is the sole food of mulberry silkworm Bombyx mori L., and their nutritional and chemical composition are suitable for care of silkworms, and their widespread introduction into production is an urgent problem of today [1-3]. In this direction, in countries such as Japan, the People's Republic of China, South Korea and Vietnam, where the science of sericulture has developed, interesting scientific research is being conducted on the selection of new varieties of mulberry with high nutritional value and nutritional value, suitable for natural climatic conditions of different seasons, using the resources of the existing mulberry gene pool wisely [4-6].

In Uzbekistan today, extensive measures are being taken to develop cocooning, especially to create new mulberry varieties suitable for silkworm hybrids, and to establish mulberry groves suitable for different seasons based on them. It is worth noting that it is necessary to improve the productivity of mulberry varieties, research the mechanisms of the influence of new mulberry varieties on the cocoon productivity and post-embryonic viability of mulberry silkworm industrial hybrids, and further develop scientific research

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and innovation to increase their efficiency. Research scientists [1, 2, 7] emphasized that paternal and maternal forms are of great importance in the creation of new hybrid combinations of mulberry and substantiated that the productivity of new generations with whole leaves has its effect on the next generations. Mulberry breeder scientists [8, 9] emphasized the permissibility of paying special attention to their morphobiological, economically valuable characters in their research, and paid special attention to these characters when creating new mulberry varieties. It is possible to create mulberry varieties with high nutritional value suitable for different seasons based on the gene pool of more than 240 mulberry trees from different regions of the Institute of Sericulture Research of Uzbekistan. Several scientists [6-10] analyzed the chemical composition of the new selection systems created at the Scientific Research Institute of Sericulture and assessed the nutritional value of mulberry varieties. Leading scientists [7-10] studied the effect of new breeding systems of mulberry on economic valuable traits of silkworm and noted their importance in industrial cocoon production.

2 Materials and methods

Research experiments were carried out in 2013-2017 at the Tashkent State Agrarian University and the Silk Research Institute. New selection numbers of mulberry were grown on the plots of "Mulberry experimental unitary enterprise" under the institute, and every year (2013, 2014, 2015) the growth and development of selection numbers and morphophysiological signs were monitored [1-7].

Selection numbers #2-02, #3-02, #4-02, #5-02, #7-02 were selected for experiments. "Tadjikskaya bez semyannaya" variety was selected as a comparative variety (Fig. 1). The research work consisted of the following experiments [5-8]:

- determination of morphological characteristics of new selection numbers of mulberry:
- determination of characteristics and productivity of leaves of new selection numbers of mulberry;
- determination of the chemical composition of leaves of new selection numbers of mulberry;
- study of the characteristics of the assimilation of the leaves of selective numbers by the silkworm:
- to determine the effect of selection numbers on the biological indicators of hybrid

Industrial hybrids of mulberry silkworm "Ipakchi 1 x Ipakchi 2", "Ipakchi 2 x Ipakchi 1", which are widely distributed in Uzbekistan, were reared with the leaves of 5 selection numbers of mulberry selected for experiments. Worm care was carried out in the special ecological worm houses of the Silk Research Institute under the same hygrothermal conditions [8-10].



Fig. 1. Selection numbers #2-02, #3-02, #4-02, #5-02, #7-02 for the experiment.

Preliminary research begins with determining the chemical composition of the leaves of new selection numbers of mulberry. The composition of chemical substances in the leaves of the new selection numbers and the comparative variety and their amount per 100 g of dry matter was determined in the spring season of 2017-2018 [4-7, 10].

For this purpose, enough leaf samples were taken from the breeding numbers in the spring period in the middle of the 5th age of worms to obtain 100 g of dry leaves from each number. The total moisture content was determined based on the difference between the initial weight of the leaf and the weight in the fully dried state [4-6]. Total nitrogen, protein, crude ash, \(\beta\)-sitosterol, polyprenols and vitamin A content of the main nutrients were determined in the certification laboratory of the Institute of Plant Substances Chemistry of the UzFA.

In our further studies, we devoted ourselves to determining the influence of selection numbers on the growth dynamics of silkworm larvae and the length of the larval period. In 2015-2017, the influence of the leaves of the new selection numbers of the Ipakchi 1 x Ipakchi 2 hybrid on the growth dynamics of worms and the duration of the larval period was determined based on the spring test worm feeding [3-5]. Each year, the worms of the

Ipakchi 1 x Ipakchi 2 industrial hybrid were fed separately from the first day of the 1st year to the last day of the 5th year with leaves of different selection numbers according to variants (Fig. 2).



Fig. 2. Maintenance of Sipakchi 1 x Sipakchi 2 hybrid with leaves of different selection numbers.

Growth dynamics was calculated based on the difference between the weight of worms on the first day of the IV age and the first day of the V age. For this, at each age, 40 worms were weighed on a scale and the average value was determined (Fig. 3).



The first day of age 4



First day of age 5

Fig. 3. Weighing worms.

The duration of the worm period was determined by calculating the number of days from the first year to the fifth year of the Ipakchi 1 x Ipakchi 2 hybrid, which was being cared for according to variants [1-5]. On the basis of the obtained results, it was concluded that which selection number has a positive effect on the growth dynamics of worms and the length of their young, as well as its advantage over the comparative mulberry variety [1-7, 9].

3 Results and discussion

In our country, cocooning has been practiced for many years, silkworm feeding is mainly carried out in the spring season. But the natural climatic conditions of Uzbekistan allow feeding silkworms in late summer and early autumn. As a result of the conducted research, the scientists of the Silk Industry Institute proved that it is possible to breed silkworms repeatedly in the mountain regions of our republic. In the rest of the regions, due to extremely high air temperature and low humidity, feeding of mulberry silkworm is somewhat complicated. Based on this, there was a demand for varieties and hybrids of mulberry suitable for feeding silkworms in different seasons.

The newly released varieties of mulberry are suitable for the harsh continental climate of different regions of our republic, are resistant to severe frosts and are characterized by high leaf yield. Food for mulberry silkworm is an external factor, and a mulberry leaf that enters the worm's body undergoes a complete transformation as a result of being absorbed into the worm's body and decomposed. At the same time, feed has a direct effect on the growth and development of worms, and the adaptability of the silkworm organism to the content of the feed is observed. Due to the different chemical composition of mulberry leaves, the nutritional level is naturally not the same. It certainly depends on the mulberry variety, the origin of the variety, the ancestors that were originally selected as breeding material, and the intensity of photosynthesis occurring in the mulberry leaves. The dynamics of silkworm growth and development is influenced by the quantity and quality of this or that substance in different mulberry varieties. As a result, worms in some cases grow slowly, the duration of youth is prolonged and they do not gain enough weight for their age.

Mulberry varieties should differ and be superior to other previously created varieties in terms of their leaf yield and other morpho-physiological characteristics, as well as the chemical composition of the leaf. Determination of these indicators is carried out by chemical analysis of the leaf. Here, the amount of nitrogen in the leaf plays a big role. Because this important component is involved in protein biosynthesis in the silk gland of the worm. In addition, the amount of carbohydrates in the leaf and their quality are factors that determine the viability of the silkworm and the productivity of the cocoon. We tried to determine the chemical composition of the new varieties No. 3-02 (Jararik 9) and No. 7-02 (Jararik 10) created in our research.

In the course of research, mulberry varieties were maintained on the basis of the same agrotechnical rules. Changes in the amount of water and chemical substances in the leaves of the cultivars, sensitivity of the cultivars to various factors of the external environment in the spring period, and a number of morpho-physiological signs were studied. In particular, we will provide information on the chemical composition of new varieties in this direction. For chemical analysis, leaf samples were taken and analyzed in the spring season in the middle of the fifth instar of the caterpillars (Fig. 4). During 2017 and 2018, a leaf sample of 100 g was taken from each variety at 8 o'clock in the morning during spring worm feeding, i.e., in the middle of the fifth instar of worm development. The leaves were dried and chemical analysis was carried out. In the chemical analysis, moisture content, crude ash content, total nitrogen, crude protein (proteins) and, for the first time, secondary polyprenol, β-sitosterol and vitamin A indicators were determined. Table 1 shows the amount of

chemicals in the leaves of mulberry cultivars during the spring 2017 and spring 2018 seasons.



Fig. 4. Leaf samples of new mulberry cultivars.

Table 1. Mulberry leaf #3-02 (Jararik 9) and #7-02 (Jararik 10) varieties during 2017.

Mulberry varieties and sample number	ıre,		Based on absolute dry matter, %					
	Worm feeding season	Total moisture,	proteins	nitrogen content	application	poly-prenols	ß- cytosterol	vitamin A ME
Example 5 No. 3-02 (Jararik 9)	spring	75.1	22.5	4.32	12.2	0.03	0.023	35859.6
	compa rison relativ e, %	99.9	98.2	102.9	88.4	300.0	121.1	206.7
Example 6 No. 7-02 (Jararik 10)	spring	74.3	22.3	4.02	11.1	0.03	0.027	37005.0
	compa rison relativ e, %	98.8	97.4	95.7	80.4	300.0	142.1	213.3
Example 8 Tajikskaya bez semyannaya (comp.)	spring	75.2	22.9	4.20	13.8	0.01	0.019	17345.25
	%	100.0	100.0	100.0	100.0	100.0	100.0	100.0

From the data in Tables 1 and 2, it can be seen that the chemical composition of the leaves of mulberry varieties Jararik 9 (sample 5) and Jararik 10 (sample 6), which participated in the analysis in 2017-2018, is significantly different from the comparative Tajikskaya bezsemyannaya variety. The amount of water in the new varieties was 75.1-74.3% (2017) and 76.0-77.3% (2018), while in the Tajikskaya bezsemyannaya variety, this indicator increased to 75.2-76.2% was equal. This is a higher result than the benchmark of 98.8 percent to 101.4 percent. One of the most important indicators of the composition of mulberry leaves is the amount of protein. The more protein there is in the leaf, the more biosynthesis of sericin and fibroin proteins takes place in the silk gland of silkworms fed on such leaves. This important indicator is equal to 22.3-22.5% (2017) and 22.0-22.5% (2018) in our new varieties, which is 108.4-110.8% higher than the comparative variety in 2018 level is the results of selection work.

Table 2. Mulberry leaves No. 3-02 (Jararik 9) and No. 7-02 (Jararik 10) varieties during 2018.

			Based on absolute dry matter, %					
Mulberry varieties and sample number	Worm feeding season	Total moisture, %	proteins	nitrogen content	application	poly-prenols	ß-cytosterol	vitamin A ME
Example 5 No. 3-02 (Jararik 9)	spring	76.0	22.0	3.53	11.34	0.03	0.026	35849.4 1
	ison relative	99.7	108.4	130.7	84.6	100.0	173.3	239.4
Example 6 No. 7-02 (Jararik 10)	spring	77.3	22.5	3.60	11.05	0.03	0.025	36966.0
	ison relative	101.4	110.8	133.3	82.4	100.0	166.7	246.8
Example 8 Tajikskaya	spring	76.2	20.3	2.70	13.41	traces	0.015	14977.3 6
bez semyannay a (comp.)	%	100.0	100.0	100.0	100.0	-	100.0	100.0

The amount of total nitrogen in the leaves of Jararik 9 and Jararik 10 varieties was also significantly higher compared to the comparative variety - 95.7-102.9% (2017) and 130.7-133.7% (2018), respectively.Now, an important new information of our research work, that is, for the first time in our scientific work, the composition of secondary metabolites in mulberry leaves was studied. If we pay attention to Tables 1 and 2, we can see that the amount of polyprenol, β-sitosterol and vitamin A in Jararik 9 and Jararik 10 during the years 2017-2018 is significantly higher than the comparative Tajikskaya bezsemyannaya variety. If taken as a percentage of the comparator, we can see that the content of polyprenol is up to 300.0% and the content of β-sitosterol and vitamin A is up to 173.3% and 246.8%, respectively. Experiments carried out to study the chemical composition of new high-yielding varieties of mulberry showed differences in the amount of nutrients and secondary metabolites in the leaves of mulberry varieties. In the spring season, the leaves

are rich in the most necessary nutrients, and therefore, for the normal nutrition, growth and development of worms, mulberry leaves must first of all be rich in moisture and protein.

According to the results of experiments in this direction, the chemical composition of the leaves of new varieties directly depends on their genotype and origin. Because under the same conditions, the significant nutrient content of the leaves grown in the same field is significantly higher than the comparative standard Tajikskaya bezsemyannaya variety, indicating that it is possible to grow quality silkworm cocoons from these mulberry varieties.

Along with eating and digesting food, its effect on the growth and development of worms is of great physiological importance. Mulberry silkworm growth is inseparable from development. Because these two physiological processes are completely dependent on each other. The growth of the organism is realized mainly by the occurrence of three processes: 1. cell reproduction; 2. cell growth; 3. increase in the mass of intercellular derivatives. Silkworm growth can be determined in two ways: 1. measuring the length of silkworms by young; 2. Determining the weight of worms by young. In our experiments, we decided to determine the effect of the leaves of new mulberry varieties on the growth and development of silkworms by changing the weight of the worms by young.

Analyzing the dynamics of worm weight gain in Table 3, it can be noted that the leaves of the new selection numbers of mulberry have a significant positive effect on the growth dynamics of industrial hybrid worms Sipakchi 1 x Sipakchi 2 at 4-5 years of age. According to the average results of 2015-2017, it can be observed that the highest dynamics of growth of worms in #3-02 (0.614 g) and numbers #2-02 and #7-02 was the growth dynamics of worms (0.607 g). Also, on the first day of the 4th year of 2016 and 2017, according to the weight of worms #7-02 and #3-02 were 0.319-0.291 grams, and on the first day of the 5th year, the above selection numbers were 0.919 - 0.854 grams. The average three-year growth dynamics of silkworms maintained with the comparative Tajikskaya bezsemyannaya variety was equal to 0.546 g. The dynamics of growth of the weight of these worms certainly indicates the effectiveness of the candidate selection numbers for the new variety.

Table 3. Dynamics of increase in worm weight of Silky 1 x Silky 2 hybrid (2015-2017).
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Selection numbers of mulberry	IV - the weight of worms on the first day of life $\overline{X} \pm \overline{x}$, g	V - the weight of worms on the first day of life $\overline{X} \pm \overline{x}$, g	Growth dynamics of worms $\overline{X} \pm \mathbf{C} \ \overline{x}$, g
No. 2-02	$0.250 * \pm 0.014$	0.826 ± 0.059	$0.607 * \pm 0.074$
#3-02	$0.291 * \pm 0.021$	0.854 ± 0.098	0.614 * ±0.067
#4-02	0.252 ± 0.017	0.817 ± 0.080	0.565 ** ±0.063
No. 5-02	0.273 * ±0.021	0.851 *±0.070	0.578 * ±0.028
No. 7-02	$0.319*\pm0.006$	0.919 * ±0.040	0.607 * ±0.037
Comparative (Tajikskaya bezsemyannaya)	0.213 ± 0.017	0.756 ± 0.089	0.546 ± 0.078

 $^{^*}$ - Pd = 0.999

In the above experiment, the weight of the worms was analyzed between 4-5 years of age, and in the next table 4, the weight of Ipakchi 1 x Ipakchi 2 and Ipakchi 2 x Ipakchi 1 hybrids on the fifth day of the fifth age was studied. It is not in vain to analyze the weight of worms on this day of the 5th year, because the maximum peak of mulberry silkworm

 $^{^{**}}$ - Pd = 0.889

nutrition corresponds to these days. If the mulberry leaves are nutritious and high-quality during this period, the ground is created for the production of abundant and high-quality cocoons. The numbers in Table 4 show that the weight of worms at the age of 5 of two hybrids is 3.68-3.86 g in selection number #2-02 based on the 3-year average; 4.41-4.45 g in selection number #3-02; 4.13-4.16 g in selection number #4-02; 4.25-4.32 g in selection number #5-02. However, it was 4.29-4.33 g in selective number #7-02 and 4.00-4.06 g in the comparative version. It was found that the indicators of new numbers were 3.3-10.3% higher than the comparative one. These numbers also indicate that the leaves of the new selection numbers of mulberry have a positive effect on the growth and development of the silkworm.

Table 4. Caterpillars fed on leaves of different mulberry varieties weight on the fifth day of the fifth year.

		Weight of worms on the fifth day of V-age, g						
Varieties of mulberry	Years	Silkworm 1 x Silkworm 2 hybrid	q relative to the ratio, %	Silkworm 2 x Silkworm 1 hybrid	q relative - compared the ratio,%			
	2015	3.99	100.8	3.91	100.3			
	2016	3.81	93.3	3.80	95.0			
No. 2-02	2017	3.26	93.8	3.87	93.9			
	$\overline{X} \pm C \overline{x}$,	3.68±0.21	96.6	3.86±0.03	96.5			
	2015	4.28	108.1	4.21	107.9			
#3-02	2016	4.39	104.0	4.32	108.0			
(Jararik 9)	2017	4.69	111.1	4, 7 0	114.1			
	$\overline{X} \pm C \overline{x}$,	4.45±0.12	109.6	4.41±0.14	110.3			
	2015	4.00	101.0	3.98	102.1			
	2016	4.18	99.1	4.11	99.8			
#4-02	2017	4.31	102.1	4.29	104.1			
	$\overline{X} \pm C \overline{x}$,	4.16±0.09	102.5	4.13±0.09	103.3			
No. 5-02	2015	4.12	104.0	4.08	104.6			
	2016	4.37	107.1	4.30	107.5			
	2017	4.48	106.2	4.37	106.1			
	$\overline{X} \pm C \overline{x}$,	4.32±0.10	106.4	4.25±0.08	106.3			
#7-02 (Jararik 10)	2015	4.11	103.8	4.00	102.6			
	2016	4.39	107.6	4.31	107.8			
	2017	4.48	106.2	4.57	110.9			
	$\overline{X} \pm C \overline{x}$,	4.33±0.11	106.7	4.29±0.16	107.3			

	g				
	2015	3.96	100.0	3.90	100.0
Comparative (Tadj.bez semyannaya variety)	2016	4.0 8	100.0	4.00	100.0
	2017	4.22	100.0	4.12	100.0
	$\overline{X} \pm C \overline{x}$,	4.08±0.07	100.0	4.00±0.06	100.0

4 Conclusions

The new selection numbers of mulberry leaves have a significant positive effect on the growth and development of industrial hybrid worms Sipakchi 1 x Sipakchi 2. The weight gain dynamics of the worms fed with these leaves were found to be higher than those fed with the comparative Tajikskaya bezsemyannaya variety. Specifically, the growth dynamics of worms were found to be the highest in selection numbers #3-02, #2-02, and #7-02.

Moreover, the weight of the worms on the fifth day of the fifth age was found to be higher in the new selection numbers, indicating the positive effect of these leaves on the growth and development of the silkworms. The weight gain in the new selection numbers was found to be 3.3-10.3% higher than the comparative variety. Therefore, the use of these new selection numbers of mulberry leaves can be considered an effective strategy for enhancing the growth and development of silkworms, leading to abundant and high-quality cocoon production.

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