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EVALUATION OF MALE INDUSTRIAL HYBRIDS OF SILKWORM

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Abstract

This article reveals the results of the analysis conducted in 2018-2020 on the economical traits of F1 hybrid combinations obtained with the participation of C8 ngl and common lines and breeds balanced on l1 and l2 lethal genes which are located on the Z chromosome of mulberry silkworm. The male hybrids have a number of advantages over bisexual hybrids in terms of viability and silk fiber quality. In particular, in the combinations $L-66xC-8$ ngl.

Keywords: *mulberry silkworm, male hybrid, cocoon, embryonic viability, breed, post-embryonic viability, lethal gene.*

Introduction

In the production and processing of cocoons, the Republic of Uzbekistan ranks third in the world, producing more than 21000 tons of cocoons a year. In the last 3-4 years in our country, there has been a rapid development of the silk industry. But along with the achievements, there are certain shortcomings and problems in terms of the yield and quality of cocoon of each box of mulberry silkworm.

At the same time, it is required to intensify efforts to ensure the efficient use of infrastructure resources in these sector, improve breeding indicators, strengthen the feed base, as well as to establish systematical work intended to improve the quality and competitiveness of products [1].

Although more than a century has passed since only F_1 hybrids of silkworm were reared and its cocoon cultivation was introduced in the production of mulberry silkworms, the laws of the hybridization process and their combination ability in pure inter-bred and inter-system hybrids have not been fully determined yet. In particular, studies on obtaining male hybrids of silkworm and sexual dimorphism have not yet lost their relevance. Below we focus on some investigations on the hybridization of silkworm and the production of simple and complex industrial hybrids.

With the increasing demand for F_1 hybrids, two complex (involving 4 breeds) hybrids are now gaining popularity due to their undisputed superiority. Also, the polygenic effect in complex hybrids obtained as a result of double crossings is due to their “adaptability” to adverse conditions [2]. It means that the adaptation of the hybrid generation obtained from double crossbreeding is always higher.

However, in some experiments, as a result of selection of the genetic systems of mulberry silkworms, a decrease in egg productivity of lines with long silk fibers was observed [3].

C. Harada [4] stated that the manifestation of heterosis was 27,8% in cocoon shell weight; 25,9% in cocoon weight; 23% in hatching and 19% in fiber length.

India accounts for 90% of world silk production through hybrids obtained by crossing particularly the breeds polyvoltine and bivoltine, which have been used successfully for commercial purposes. [5]. The main purpose of the hybrids obtained by double crossbreeding is to achieve both qualitative and quantitative characteristics at the same time [6].

Indeed, in the preparation of a stock of parent breeds for the preparation of a hybrid for single crossing, the cost of maintaining a separate line, especially a pure breed with low fertility and low cocooning can be high. This requires an improvement in the cultivation of double-crossed hybrids, which combine the traits of 4 genetic breeds (systems) capable of producing a stable cocoon crop required for industry [7].

Above was some information on the differences between common and complex hybrids and their importance in production. However, it is possible to solve this problem by obtaining 100% male hybrids and using them in production. In this regard a number of studies have been conducted in China and the CIS countries. At present, the breeds with sex-lethals is partially used in practice. The problem of their low performance in egg production is characterized by the high cost in breeding these breeds [8].

In 1996, after negotiations with activists from the Zhejiang Agricultural Academy, valuable silkworm lines with balanced lethal genes created on the basis of complex genetic schemes were brought from the Russian SA. The main problem was then solved using two independent, patented technologies that were developed by incorporating the combined lethal gene into strains with high economic and technological traits [9,10].

Due to the specification in the preparation of male hybrid eggs, the value of eggs in this direction of the mulberry silkworm is theoretically 4 times higher than that of common breeds [11].

At the Shandong Sericulture Research Institute, scientists conducted experiments on the breeds of silkworm whose sex was marked during the egg period. They statistically analyzed the ratio of light yellow and gray eggs in determining the genotype of the generation obtained with the participation of a common, sex-unmarked native Jinsong breed with Romanian breeds using cross-breeding in F_1-F_3 generation. In F_1 generation, all eggs were gray in color. In F_2 generation, there was ratio of light yellow eggs of 1/8 (**??**) with gray eggs 7/8 (**??**) [12].

In order to reduce the cost of hybrids developed by Yaotao and several scientists, they conducted research on obtaining unique hybrids by hybridization of female clones obtained by cloning the silkworm with the participation of lethally balanced breeds [13].

A group of Korean scientists have also conducted research on sex-marked breeds of silkworm in cocoon stage. These breeds include, in particular, the biparental Hanbyeolnue created based on Jam 319 breed of Japanese origin and Jam 320 breed of Chinese origin, and sexually marked in cocoon stage [14].

Moreover, in the years 2012 - 2014, M. Panayotov et al worked on sex-marked hybrid combinations. They created new hybrid combinations from 19, 1013, Magi 2, and Lea 2 breeds that were well-hatched, normal, and almost free of diseases [15].

Despite the fact that the first works in this area have been carried out for a long time, a number of scientists have been conducting research and studies at the Uzbek Sericulture Research Institute. In particular, their thorough scientific research eventually resulted in obtaining 100% male hybrids such as *Turon1, Turon 2.* In addition, U.N. Nasirillaev et al obtained a new translocation in the genotype of large-cocoon breeds "Guzal" and "Marvarid", which allowed to mark their sex in the egg stage, that is, light yellow eggs become male and gray eggs become female. [16].

According to a 3-year (2012-2014) study conducted by B.U. Nasirillaev et al., in sex-marked Line 1 mar. and Line 2 mar.lines egg production was high in the egg stage, and they were found to lay eggs in the range of 687–801 pieces [17].

As B.U.Nasirillaev et al stated, *Line 2 mar.* (sex-marked at egg stage), *Line 3 mar.* (sex-marked by cocoon color) and a sex-controlled F_1 industrial hybrid combination involving above lines were created, and in this hybrid combination, egg hatching rate was 97,0%, worm viability was 93,8%, and morbidity was 1,1%. It was concluded that translocation in the genotype of the Line 2 mar. system did not adversely affect the viability of worms [18].

The aim of the study was to make a comparative analysis of the viability and cocoon productivity of industrial hybrids involving C-8 ngl breed of silkworm balanced by double lethal genes and the common breed, and also the lines of silkworm.

1 Materials and methods

As an object of the study were selected C-8 ngl breed of silkworm with balanced double lethal genes and common “Marvarid” breed, and also Line 66 lines, and first-generation industrial hybrids were obtained with their participation. In hybrid combinations, the common breed and lines participated as the maternal parent component, while only the C-8 ngl breed participated as the male parental component. Hybrids Uzbekistan-5 regionalized in the Republic was taken as control variant against these hybrids and the following indicators were identified:

1. reproductive indicators;
2. hatching and viability traits;
3. cocoon productivity;
4. technological indicators.

Larva of breeds and lines selected for the experiment were reared at a temperature of 26–27° C and a relative humidity of 70–75% at their young instars, and at a temperature of 24–25 ° C and a relative humidity of 65–70% at adult instars. For 1 box of silkworm larva, a 1000 kg of mulberry leaves were used. The worms were fed on the leaves of mulberry varieties Jararik 4, Jararik 5 and Jararik 6. At the 2^{nd} instars, worms were counted by 250 pieces in 3 replications.

2 Results and discussion

The most important criterion in the evaluation of industrial hybrids is the viability of eggs and worms. This is because in the first generation, due to heterosis, silkworms are more resistant and more viable than the parental forms. Based on this, in our experiments we determined the viability of silkworms and their disease susceptibility in the fifth instars (Table 1).

Table 1. Embryonic and post-embryonic viability of breeds and hybrids (2018-2020)

Breeds and hybrids	**Years**	**n**	**Egg hatching, %**	**Larva viability, %**	**Morbi-dity percen-tage, %**
Line 66 (++)	2018	4	95,5±1,09	89,9±1,39	4,1±0,17
	2019	4	97,7±0,88	87,1±1.18	6,6 ±0,71
	2020	4	96,6±0,80	88,4±0,40	4,53±0,70
	Average		**96,6±0,83**	**88,5±0.80**	**5,07±0,77**
Marvarid (++)	2018	4	97,5±1,27	91,9±1,77	3,2±0,60
	2019	4	98,0±0,58	90,8±0,78	5,6±0,53
	2020	4	96,0±0,90	95,2±0,46	2,7±1,16
	Average		**97,2±0,60**	**92,6±3,86**	**3,83±0,89**
	2019	4	(46,0) 92,0±1,03	89,2±1,24	3.3±0,81
	2020	4	(44,6) 89,3±1.45	83,7±0,70	2,4±0,63
	Average		**92,5±2,04**	**87,6±2,0**	**3,3±0.54**
	2019	4	(45,0) 90,0±2,0	77,7±4,54	8,1±0,31
	2020	4	(40,0) 80,0±2,0	98,0±0,81	2,0±0,53
	Average		**88,8±4,77**	**89,7±6,15**	**4,3±1,9**
Istikbol (**F_1 100% »**) ($+l_1$ $+l_2$)	2018	4	(48,9) 97,8±0,31	93,8±0,74	2,5±0,64
	2019	4	(47,65) 95,3±0,09	91,3±0,97	1,9±1,02
	2020	4	(46,3) 92,6±0,80	94,9±0,51	2,3±0,122
	Average		**95,2±1,5**	**93,3±1,06**	**2,2±0,17**

Uzbekistan 5 (F$_1$ +,> control) (++)	2018	4	96,0±0,98	91,0±0,89	3,2±0,38
	2019	4	90,3±3,52	88,0±0,89	4,2±0,15
	2020	4	87,6±2,80	93,3±2,94	2,17±0,26
	Average		**91,5±2,47**	**90,7±1,55**	**3,19±0,58**

If we look at the results of "Istikbol" hybrid among the tested industrial hybrids, we can see that its hatching rate, worm viability, and viability indicators in the V-instars and pupa period are significantly higher than in Line-66xC8 ngl hybrid combination. In particular, the highest indication on egg hatching of this hybrid was observed to be 95,2 %, that is, 6,4 % higher than in Line-66xC8 ngl hybrid combination. This indicates that there was almost no difference from pure maternal parent breed "Marvarid". In terms of the trait of worm vaibility, the highest limit was 93,3 % (Istikbol), the lowest limit was 87,6 % (C-8 ngl).

Close dependence of the percentage of disease incidence in silkworms along with the agronomic techniques of silkworm rearing, and also with the parental breeds and their genotype was reflected in the figures. In other words, the disease incidence made 2.2% due to the heterosis factor observed in "Istikbol" hybrid that was obtained from crossbreeding of the large-cocoon "Marvarid" breed with $C-8$ ngl breed.

Several sex-marked breeding lines of mulberry silkworm have been developed, which, as a result of crossbreeding with breeds and lines with distinctive high farm traits, have incorporated the traits maternal breed in F$_1$ generation male hybrids. Therefore, new hybrids have high indicators of silkiness.

Prior to the creation of new hybrid combinations, the 2018-2020 experiments identified cocoon productivity traits of $C-8$ ngl, L-66 lines and "Marvarid" breed and the hybrid combinations obtained with their participation (Table 2).

Table 2. Cocoon productivity of breeds and hybrids (2018-2020)

Breeds and hybrids	Years	n	Cocoon weight, g	Cocoon shell weight, mg	Silkiness, %
Line 66 (++)	2018	90	1,75±0,002	416±1,2	23,8±0,17
	2019	90	1,95±0,022	459±0,9	23,5±0,62
	2020	90	1,69±0,023	407±6,6	24,0±0,25
	Average		**1,79±0,07**	**427±1,6**	**23,7±0,14**

Marvarid (++)	2018	90	2,23±0,026	513±4,2	23,0±0,32
	2019	90	2,57±0,017	589±2,9	22,9±0,09
	2020	90	2,27±0,015	514±3,8	21,7±0,21
	Average		**2,35±0,09**	**538±2,52**	**22,5±0,51**
	2019	90	2,07±0,050	498±14,2	24,1±0,12
	2020	90	1,94±0,120	445±2,3	22,9±0,29
	Average		**1,96±0,13**	**456±21,77**	**23,2±0,48**
	2019	90	1,78±0,005	460±6,3	25,8±0,4
	2020	90	2,16±0,010	569±9,9	26,3±0,29
	Average		**2,08±0,15**	**510±13,7**	**25,2±0,86**
Istikbol (F_1 100% ») ($+l_1$ $+l_2$)	2018	90	2,13±0,036	502±9,3	23,6±0,52
	2019	90	2,2±0,06	514±7,6	24,2±0,97
	2020	90	1,93±0,050	496±8,1	25,6±0,26
	Average		**2,08±0,08**	**504±5,35**	**24,5±0,59**
Uzbekistan 5 (F_1 +,>control) (++)	2018	90	2,15±0,014	526±4,6	24,5±0,47
	2019	90	2,15±0,013	529±4,2	22,0±0.24
	2020	90	2,09±0,031	430±9,5	21,5±0,13
	Average		**2,13±0,02**	**495±14,21**	**22,6±0,92**

Table 3. Cocoon productivity of parental components and F_1 hybrids

Breeds and hybrids	**Years**	**n**	**Productivity of single box**	
			Cocoon yield, kg	**Cocoon shell yield, kg**
Line 66 (++)	2018	90	69,0	16,2
	2019	90	67,6	16,1
	2020	90	73,5	15,7
	Average		70,0	16,0
Marvarid(++)	2018	90	88,5	20,4
	2019	90	102,1	23,5
	2020	90	93,4	21,1
	Average		94,6	21,7

	2019	90	83,9	20,2
	2020	90	65,3	14,9
	Average		74,5	17,2
	2019	90	56,0	14,5
	2020	90	69,7	18,4
	Average		69,6	17,7
Istikbol **(F_1 100% »)** ($+l_1$ $+l_2$)	2018	90	86,3	20,3
	2019	90	90,2	21,2
	2020	90	76,3	19,6
	Average		84,3	20,4
Uzbekistan 5 **(F_1 +,>control)** (++)	2018	90	84,5	20,7
	2019	90	76,9	18,9
	2020	90	76,9	15,8
	Average		79,1	18,5

3 Conclusion

Based on the results of experiments, it can be concluded as follows:

1. There was no preference observed for parental traits in terms of the percentage of silkiness of hybrid generation. In hybrid combinations, the predominance over parental traits was mainly reflected in viability, growth, and developmental characteristics of silkworms.

2. The cocoon productivity in the new male hybrids is at the level of the bisexual Uzbekistan-5 hybrid (2,08-2,13 g) and proved to be able to compete with the most productive, comparative bisexual, large-cocoon hybrid in terms of cocoon shell yield.

3. F_1 male hybrid combinations presented higher cocoon productivity indicators, even at the level of tetra-hybrid despite being a common breed, i.e a hybrid from crossing of just two breeds. Of course, these figures reveal that lethal genes in the hybrid genotype do not have a negative effect on productivity. In addition, the introduction of industrial hybrids in the practice of silk production, which give 100% male generation, will allow to growing additional raw silk without increasing production costs.

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