The possibility and prospect of breeding wild silkworm of the Giant Peacock moth (*Saturnia pyri, Denis & Schiffermüller*, 1775), as a new branch of sericulture in Azerbaijan

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It is known that in modern times there is a great demand for wild silkworm silk and its products, especially in the field of biomedicine and nanotechnology, among domestic and foreign consumers. The presence of large areas of various forests and favorable conditions in Azerbaijan provides great opportunities for the cultivation of wild silkworms of the *Saturniidae* family for industrial purposes. In this regard, there is a need for a detailed study of the productivity of the breed, which actually lives in the green spaces of our country, i.e., the study of its biological and technological parameters in laboratory conditions. The article describes breeding experience of the Giant Peacock moth (lat. *Saturnia pyri*) in laboratory conditions at constant temperature (20-25°C) and humidity (80-85%) on the leaves of cherry plant (*Prunus avium* L., 1755). The prospects and possibilities for the future expanded cultivation and production of specific silk, which will be the first in Azerbaijan and the entire Transcaucasia, have also been assessed.

Keywords: Wild silkworm, Saturnia pyri eggs, silkworm hatching, silkworm molting, cocoon, sericulture

INTRODUCTION

Within the framework of the state program for the development of silk industry, specialists from our Sheki Regional Scientific Center of the National Academy of Sciences of Azerbaijan participated in various scientific events, analyzed the latest research articles to study methods for the improvement of sericulture and searched for methods to restore the former glory of Azerbaijani silk. In the articles of famous foreign scientists as well as young researchers, works devoted to non-traditional wild species of silkworms, about the aspects of using their silk in biomedicine, in smart and nano technologies were most frequently mentioned (Volova and et al., 2009), (Vepari and Kaplan, 2007), (Kasoju and et al., 2009).

Based on this, we began a detailed study of the possibility of breeding and obtaining silk of a local

species of wild moth in laboratory conditions. Giant peacock moth, Bombyx pyri or Saturnia pyri insects of the order Lepidoptera, family Saturniidae, genus Saturnia as a wild species that actually lives on the territory of our country (Efendi, 1971), became the object of our research. Although quite common in Azerbaijan, this species is very rare in Europe, for example, included in the Red Book of Ukraine and a number of southern regions of Russia (Shapoval, 2010) This moth has got its name due to the peculiarities of its colors: on each wing of the moth there is a disc-shaped ocular spot, similar to the spots on the tail of a peacock. S. pyri belongs to one of the largest species among all lepidopteran insects in Azerbaijan, and in the common folk this moth is called Butterfly-Soul (azerb. Ruh kəpənəyi), probably due to its large size and due to the fact that it mainly flies at night.

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LABORATORY RESEARCH

We were able to find a pair of mated Giant Peacock moths at night from May 28 to May 29, 2019 in the mountain village of Bash Shabalyd (azerb. Baş Şabalıd) in the Sheki district. These two moths were captured with a light trap and were carefully transferred into a cardboard box covered with gauze (Kumar and Shukurova, 2018). After fertilization, the male was taken away for further research, and the female was inactive. On May 30, 2019, the fertilized female started to lay the first group of eggs which consisted of 6 eggs, this process lasted until June 3, 2019 when the total number of eggs was 63. On June 4, the female moth died and was withdrawn for a detailed examination.

RESULTS OF VISUAL EXAMINATION OF MOTHS

Female Saturnia pyri: the length of body was 40 mm, the length of forewing – 85 mm and the wingspan – 170 mm. The body of the male was a bit larger – 45 mm, the length of forewing – 70 mm and the wingspan – 150 mm (Fig. 1). The inner (basal) part of the forewings of both moths is dark brown but the outer part is light grey, and both parts of the hindwings are light grey. There are dark straight inner lanes and light brown unevenly serrated outer lanes which divide all the wings into unequal transverse parts.

Between the lanes on each wing there is a large eyespot of a dark blue color. On the upper part of the eyespot the hair scales sharply decrease – this small part is transparent, above that there is a slightly wide rufous (reddish brown) strip in the shape of a crescent, then there is a white crescent above it and another rufous thin one on the top of that. On the bottom of the eyespot there is a light brown crescent. Finally, the outermost border of the whole oval is a black line (Zolotarev and et al., 1940).

A light stripe runs along the edge of the wings, which gradually becomes slightly darker (cream color) towards the border of the wings. At the base of the wing there is a wide black strip that stretches to the apex, then breaks off. The body is massive, covered in thick hairs, divided into three parts: head, thorax and abdomen.

There are eyes, antennae and mouth on the head (Fig. 2 and 3). The antennae are bipectinate and have a feather-like shape, they are composed of segments with long slender lateral processes on both sides. The processes on the antennae of female moth are thinner and have denticles, on the contrary, the processes of male are quite conspicuous, and they resemble a feather. Therefore, the female is easily distinguished from the male due to the structure of the antennae. However, the length of the antennae of both moths was almost the same. (Symonds et al., 2012).





Fig. 1. Female *Saturnia pyri*; wings in a wide (1) and seated position (2).

The organs of vision are located on the both sides of the head – two large dark compound eyes, each of which consists of many separate simple eyes – ommatidia. The mouthparts of the moths are underdeveloped (Mikhailov and Gershenson, 1958).

The thorax of *Saturnia pyri* consists of three segments, fused together: prothorax, mesothorax,

and metathorax. Each of these segments has a pair of jointed legs that are covered in scales; the legs end up with small paws. There are claws on the paws which enable the moth to cling to the surfaces (Fig. 4).



Fig. 2. A specific eyespot on the wings.



Fig. 3. Saturnia pyri antennas: male (1) female (2).

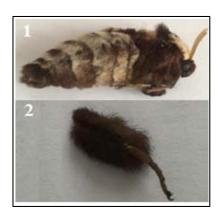


Fig. 4. Corpus (1) and leg (2).

The mesothorax and metathorax carry a pair of well-developed wings which are made up of two chitinous layers, the veins that pass through the wings support them by providing mechanical strength.

The abdomen is multi-segmented, it consists of nine segments linked together by soft flexible tissues. It should be mentioned that the abdomen of male looked thicker because the segments were more closely linked and the scales were longer which gave an additional volume, also the striped color of abdomen was more distinguished. On the contrary, the abdomen of female was more elongated. At the end of the abdomen are the anus and external genitalia. The end of the abdomen of female is blunt and is covered in hairs. The end of the male abdomen is conical and has genital appendages with setae. So, this is their distinguishing feature.

Moreover, several moth eggs of different days from different fragments were also taken away for the examination. The eggs were examined for 10 days, right up to their hatching.

RESULTS OF EGG EXAMINATION

The eggs of *S. pyri* are oval, slightly flattened, they have an average length of 2.5 mm and an average width of 1.5 mm (Fig. 5). Due to the small amount of eggs, they were weighed in groups and it was possible to approximately calculate the mass of one egg (6 mg) that appeared in the first egglaying, the egg weight in all subsequent egg-laying was approximately 4 mg.



Fig. 5. Eggs after laying (1) and hatching (2).

Upon a detailed examination, the eggs have a greenish-white chitinous shell but the coating substance which glues the eggs to each other is greyish-brown, so that is why the eggs are greyish-brown in appearance. It was seen under the microscope that

the eggshells were penetrated by many filamentous thin tubules where the gas exchange was likely to occur, also there was a point hole (micropyle) through which the sperm penetrated, and the egg was fertilized. The egg was filled with greenish semi-liquid contents – egg white. The color of the egg changes and becomes lighter with the development of the embryo because a transparent yolk membrane emerges under the shell, and eventually a third serous membrane appears under it. After hatching, the eggs are empty, dry and have a corroded pole of the shell.

The development of the eggs of Saturnia pyri began immediately after the egg laying and did not have a resting stage (diapause). We tried to work without tactile contact and sudden movements with the experimental subjects during our experiments on growing S. pyri. The first 4 days after egg laying - during the period called embryo overturning (blast kinesis), the eggs were kept at the room temperature (22-24°C). In the evening of June 3, the eggs in a plastic box with good ventilation and protection from other insects were placed in the garden, in a specially designated self-made structure – in a mini greenhouse, where the natural conditions for incubation were created in the open air. Our mini greenhouse was placed in the shade where the air temperature was 24-26°C, it could reach up to 30° C in the afternoon. In order to maintain the microclimate inside the greenhouse, we daily placed a fresh branch of cherries or pears and a 30 ml container with water. Air was well accessible to the eggs throughout the incubation period. All the time, the eggs remained perfectly still on that paper surface where they were laid by the moth (Sinitsky et al., 1952).

Age I. The first hatching took place on June 8, 2019 and this process lasted 3 days, so the caterpillars in the greenhouse were of different ages during the whole control time. The approximate number of hatched and viable ones: 50/63. The larvae, that hatched on the first two days (the majority) were about 6-7 mm long and weighed about 4 mg, while the ones hatched on the next day were 5-6 mm long (Fig. 6)

At the first age, the caterpillar was black and had four rows of warts where long light brown setae protrude. The branches of elm, oak, plum, cherry and pear were offered as food sources; the young shoots of cherry turned out to be optimal for them, but they also fed on elm, pear and plum branches (Klimiashvili- Nutsibidze, 1952).

From June 12 to June 15, the sleep of caterpillars was noted – this is a stationary state in which the front part of the caterpillar's body is raised, the old chitin capsule is sliding from the head and during this period which lasted 1-2 days, the feeding stops. It should be noted that further feeding was continued in the laboratory in a portable small greenhouse.



Fig. 6. *Saturnia pyri* at the first age (5-7 mm).

Age II. The first molting occurred from June 14 to June 16: the caterpillars achieved a gap on the dorsal side of the old skin by making wavy movements. Caterpillars of the second age had changed their body color to taupe, the warts throughout their body became orange and more conspicuous, and their head and anal segments acquired a brown color (Fig. 7).



Fig. 7. Saturnia pyri at the second age (26-30 mm).

About 1-2 hours after the molting, the caterpillars actively began to feed on young branches of

cherries and plums. They excreted a lot of granular feces. By the end of the second age, the length of caterpillars was $\sim 26\text{--}30$ mm. Since June 19, the activity of most of the caterpillars had stopped, they fell asleep.

Age III. The second molting took place on June 21 as a result of which the caterpillars partially shed the old skin and the old cranial capsule came off. Their new skin was light green with small black dots, yellow warts and quite long hairs. In addition, their head had solidified and hardened, so became a bit larger. In less than two and a half hours, the caterpillars started to eat again. The feeding continued with young branches of pears, cherries and plums. The length of the caterpillars by the end of this age reached ~ 35-40 mm (Fig. 8).



Fig. 8. Saturnia pyri at the third age (35-40 mm).

At that moment, there were caterpillars of two ages (II and III) in the experimental greenhouse. On June 23, 5 caterpillars of the third age were released on the plum tree for the control study. On June 26, the remaining caterpillars of the third age fell asleep and their sleep lasted two days.

Age IV. The third molting occurred from June 27 to June 28, 2019. During the fourth stage of development, the caterpillars acquired a green body color and sky blue warts with long hairs and spikes filled with liquid. Also, a long yellow stripe occurred on the side of the body. The length of the caterpillars was $\sim 45\text{-}55$ mm (Fig. 9). On July 3-5, the caterpillars of this stage stopped eating food, some of them strongly clung to the branches, others to the gauze that covered the box. They took a tense elongated shape; the color of their warts became slightly lilac.

Age V. From July 6-10, the old cranial capsule came off and the old skin was torn - so, the fourth

molt had occurred. During this molt, three caterpillars could not completely get rid of the old skin since it did not fully tear on the back and caused the constriction of their body leading to their death.



Fig. 9. Saturnia pyri at the fourth age (45-55 mm).

The color of the caterpillars that passed to the fifth age gradually turned into yellowish-green, starting in July 12. Their massive legs and prolegs were clearly seen, the head width was about 9 mm and the labrum (upper lip) was quite large. Caterpillars grew rapidly, as they consumed a large amount of foliage: we provided them with 6-7 ripe branches of cherry (~30-35 cm in size) per day. It should be noted that the average length of their body at the fifth age was 90-100 mm, weight – 10.0-10.5 g (Fig. 10).

Another color change took place by July 16, right before pupation. The color of the caterpillars changed, starting from the head, and became yellowish orange, this time there was no molting. The caterpillars gradually stopped eating, their body became dull and sluggish, it felt like they shortened in length. They were moving anxiously and jerking the leaves, it was clear that they were looking for a convenient place to spin themselves a cocoon. As soon as they found such a place, they calmed down and began to excrete excrement and mucus, and then they started to spin their cocoons. The caterpillars began the process by pulling together several leaves around their body with the help of silk threads, thus forming a comfortable bed for themselves. Then, they weaved the inner surface of the pulled leaves and in 2 hours, the caterpillars literally were not visible.

Cocoon. There were several freshly spun cocoons in our laboratory on July 18 (Fig. 11). They were of an elongated oval shape (pear-shaped),

dark brown in color, with a diameter of ~ 22.7 mm and a length of ~ 54 mm. Their walls consisted of a loose outer shell and a dense inner shell which is formed by strongly glued loops of silk thread. In the future, we will continue experiments to determine the structure and shape of the threads, to find out the causes of some subjective facts.



Fig. 10. Saturnia pyri at the fifth age (90-100 mm).



Fig. 11. Cocoon (90-100 mm).

Controlled Silkworms: Out of 3 silkworms of the third age that were transplanted to the plum tree on June 23, just one specimen was found on July 6 (Fig. 12). That caterpillar was reddish green in color, quite large in size, its length was ~ 10.5 cm. On that day, it was going down the trunk of the tree, probably in search of a place to pupate. To protect it, the caterpillar was moved to a well-ventilated container with a dry plum branch.

2 hours later that evening, the body of the caterpillar contracted as it neither was eating nor excreting feces. It was clearly seen how a silk thread was secreted from a silk-secreting papilla, due to which the caterpillar was able to connect several leaves and began to weave the outer (main) part of the cocoon. In almost 4 hours, the caterpillar was no longer visible, only a dark brown, shaggy, pear-shaped cocoon was seen between the leaves.



Fig. 12. The control one (105 mm).

Examination of excrements: Excrements (feces) were in the form of prismatic hexagons with a star-shaped cross section. The size of the granule increased with the development of the caterpillar. The color of the excrements was dark blue green, they unfolded in warm water secreting a greenish tint and a specific smell.

CONCLUSIONS

In the article, we described the biological characteristics of *Saturnia pyri*, the period of growth and development of the caterpillar from hatching to pupation *in vitro*. Our experiment allows us to make the following conclusions:

- 1. It is possible to breed *Saturnia pyri* in laboratory conditions.
- 2. To provide food to the caterpillars, the leaves of cherry which grows well in our forests and gardens can be used.
- 3. The following feeding regimen must be observed, depending on the age of the caterpillars: at I II ages, feeding must be 1-2 times a day, III age 3 times a day, IV age 4 times a day and V age 5 times a day.
- 4. The optimal feeding rate by age per 100 caterpillars: I age 30-35 g, II age 180-200 g, III age 1000-1500 g, IV age 3500-4000 g, V age 14000 g.
- 5. To fully satisfy the stocks of fodder leaves for further mass breeding of this species, it is necessary to plant cherry plantations, which is very low-budget in the natural conditions of our country.
- 6. Throughout the entire feeding period, it is necessary to maintain the temperature within 20°-25° C, with a relative humidity of 80-85%.
- 7. The control caterpillar, which fed and grew in

- the natural environment, had a shorter development cycle. The pupation process of this caterpillar was 10 days earlier than the experimental group.
- 8. To obtain the cocoons of good quality, the caterpillars should defecate for the last time before pupation. After that, they can be placed in separate paper bags with adequate ventilation, where they can weave their cocoons in a clean environment.
- 9. To maintain the best quality of the cocoons, they must be collected in 15 days after the beginning of the pupation.

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Vəhşi ipək qurdu olan armud saturniyasının (*Saturnia pyri, Denis & Schiffermüller*, 1775), Azərbaycanda ipəkçiliyin yeni bir qolu kimi yetişdirilməsi ehtimalı və perspektivləri

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Məlumdur ki, müasir dövürdə yerli və xarici istehlakçılar arasında, xüsusilə biotibb və nano texnologiyalar sahəsində, vəhşi (yabanı) ipəkqurdu ipəyinə və ondan hazırlanan məhsullara böyük tələbat var. Azərbaycanda geniş meşə ərazilərinin və əlverişli şəraitin olması *Saturnidae* ailəsinə aid vəhşi ipəkqurdularının sənaye məqsədləri üçün yetişdirilməsinə böyük imkanlar yaradır. Bununla əlaqədar olaraq, ölkəmizin yaşıl ərazilərində mövcud olan vəhşi ipəkqurdu növünün bioloji və texnoloji göstəricilərinin laboratoriya şəraitində araşdırılmasına, istehsal gücünün ətraflı öyrənilməsinə ehtiyac var. Məqalədə müəyyən temperatur (20-25°C) və nəmlik (80-85%) şəraitində, armud saturniyası (lat. *Saturnia pyri*) ipəkqurdunun gilas ağacı

(*Prunus avium* L., 1755) yarpaqları ilə laboratoriyada yetişdirilməsi təcrübəsi təsvir edilmişdir. Həmçinin, məqalədə ilk dəfə olaraq *Saturnia pyri* ipəkqurdunun və ondan alınan xüsusi ipəyin gələcəkdə Azərbaycanda və bütün Zaqafqaziya bölgəsində geniş istehsal mümkünlüyü və səmərəliliyi qiymətləndirilmişdir.

Açar sözlər: Vəhşi (yabanı) ipəkqurdu, Saturnia pyri yumurtaları, ipəkqurdunun yumurtadan çıxması, ipəkqurdunun qabıq dəyişməsi, barama, ipəkçilik

Возможности и перспективы разведения дикого шелкопряда грушевой сатурнии (Saturnia pyri, Denis & Schiffermüller, 1775), как нового направления шелководства в Азербайджане

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Известно, что в настоящее время среди отечественных и зарубежных потребителей существует большой спрос на шёлк диких шелкопрядов, особенно в области биомедицины и нанотехнологий. Наличие больших лесных массивов и благоприятные условия Азербайджана создают большие возможности для выращивания в промышленных целях диких шелкопрядов, принадлежащих семейству Saturnidae. В связи с этим возникает необходимость изучения в лабораторных условиях биологических и технологических характеристик дикого шелкопряда, живущего в зеленых районах нашей страны, а также детального изучения его производственной мощности. В статье описывается опыт разведения грушевой сатурнии (лат. Saturnia pyri) в лабораторных условиях при постоянной температуре (20-25°С) и влажности (80-85%) на листьях растения черешни (Prunus avium L., 1755). Также оценены возможности и перспективы расширенного выращивания грушевой сатурнии и производства в будущем «дикого» шелка, который будет первым в Азербайджане и во всем Закавказском регионе.

Ключевые слова: Дикий шелкопряд, яйца Saturnia pyri, вылупление шелкопряда, линька шелкопряда, кокон, шелководство