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Development Phases of the Larva of the Black Soldier Fly Hermetia Illucens on the Metabolic Byproducts of Monogastric Animals and Birds

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Abstract

Processing of organic waste by insect larvae is a relevant and promising direction in biotechnology. The phases of fly development were studied, a comparative assessment of development phases was carried out on chicken droppings and pig manure. The terms of the complete biological cycle of the fly on different substrates were compared. The studies were carried out on specially prepared substrates with the addition of a probiotic preparation culture, namely a strain of soil spore bacteria Bacillus subtilis, which were added to reduce the content of harmful gases, shorten the decomposition time, as well as for disinfection from pathogenic microorganisms. On chicken droppings, the biological process, including all development phases, proceeds faster than on pig manure.

Disciplinary: Agricultural Sciences (Entomology).

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1 Introduction

The processing of organic waste, obtained from farm animals and poultry, mainly pigs and chickens, is currently relevant. The most developed and promising biotechnology direction is the use of insect larvae, especially Diptera species.

The black soldier fly from the Stratiomyidae family belongs to the species of Diptera. Droppings and manure are the main food of the larvae. With their help, the natural disposal of organic waste is carried out. Large larvae that are at the pre-pupa stage can be used to feed animals and birds. Insects, when processing pig manure and chicken droppings, convert residual proteins

and other nutrients into their own biomass. It will be considered a high-quality protein feed. All this solves the problem of manure and droppings disposal [6].

Now, a promising direction in agriculture is the production of animal feed with the help of insects. As is known, the microflora of manure and droppings changes with the help of larvae, as well as a decrease in the number of harmful bacteria [10, 11].

The high nutritional value of fly larvae is in the content of a large amount of protein and calcium. Also, with a high growth rate of larvae, a significant amount of protein and fat accumulates [2, 3].

Larvae can be used for the processing of various organic waste: pig waste, bird litter, food waste, animal manure, bird droppings. After processing, the resulting substrate can be used for growing plants. For agro-industrial biotechnologies, this determines the prospects of this species of dipterous [9, 11].

The purpose of our research is to study the development phases of the larva of the Black soldier fly Hermetia illucens on the metabolic byproducts of monogastric animals and birds.

Research tasks is to study in detail the development phases of the Black soldier fly on pig manure and chicken droppings, and compare the terms of the complete biological cycle of the fly on different substrates.

2 Material and Methods

The research was funded by RFBR and Chelyabinsk Region, project number 20-416-740008\20 "Development of a method for increasing the degree of biological usefulness of the processing waste products of monogastric animals and birds using Hermetia illucens larvae" and conducted in the conditions of the Department of Animal Husbandry and Poultry Breeding of the Institute of Veterinary Medicine of the South Ural State Agrarian University.

Figure 1: Insectarium for keeping flies, mating and laying eggs

For this study, flies were taken from one insectarium (Figure 1) and the entire life cycle of a fly was studied from egg-laying to hatching of a new fly. The phases of fly development on different substrates were considered in great detail, there are five of them.

All the data were analyzed, and the whole biological process was accompanied by photographs.

The studies were carried out on specially prepared substrates with the addition of a probiotic preparation culture, namely a strain of soil spore bacteria Bacillus subtilis, which were added to reduce the content of harmful gases, shorten the decomposition time, as well as for disinfection from pathogenic microorganisms. Two containers were taken, one with pig manure (group 1), the second with chicken droppings (group 2), Bacillus subtilis hay bacterium was added to them.

The resulting substrate was placed in a dark place and stored for a day at a temperature of 22°C. After the expiration of time, the first experimental substrate (pig manure + Bacillus subtilis,) and the second experimental substrate (chicken droppings + Bacillus subtilis) were brought to the optimal humidity of 78% set in a series of experiments earlier. This humidity was constantly maintained to obtain a positive effect of growing larvae.

It should be noted that the optimal temperature for the development of larvae in the room is 27°C, which was monitored daily during the experiment. Natural lighting was mainly used with additional lighting during laboratory work.

The larvae were cultured in plastic containers on tables, 3-day-old grown larvae were introduced on the surface of the substrate, at the rate of 150 g of larvae per 5 kg of substrate [1].

3 Results and Discussion

Before proceeding to a more detailed study of the development phases of the Black soldier fly, we would like to note that its appearance and behavior are similar to a wasp, although it differs in one pair of wings, does not have a sting, but has a dark monochrome body color.

The adult Black soldier fly is a rather gentle creature, despite the larvae' ability to survive in harsh conditions, it flies very little inside the insectarium cell. It does not bite, does not sting, has a weakly pronounced and poorly developed jaw apparatus. After hatching from the pupa, the fly does not eat. It drinks only water, because of the licking mouthparts. Its food is protein reserves, which accumulate when it is in the larval stage. This serves as a source of energy for its life. Like other flies, it does not tolerate diseases. It mates only with males and lays eggs. It lives a little time, about five sometimes up to eight days.

In adults, sexual dimorphism is observed, females are larger than males in body size, but not significantly larger, although they may be the same since the length of the body varies depending on the food consumed by the larvae. As can be seen from Figure 2, females have a body length of 15-20 mm, black body color, with white tarsus and tibiotarsus. The wings are dark, the structure is dense, covered with membranes. Regardless of gender, they have a short and wide head, with wide-set eyes, with long antennae that are almost 2 times the length of the head.

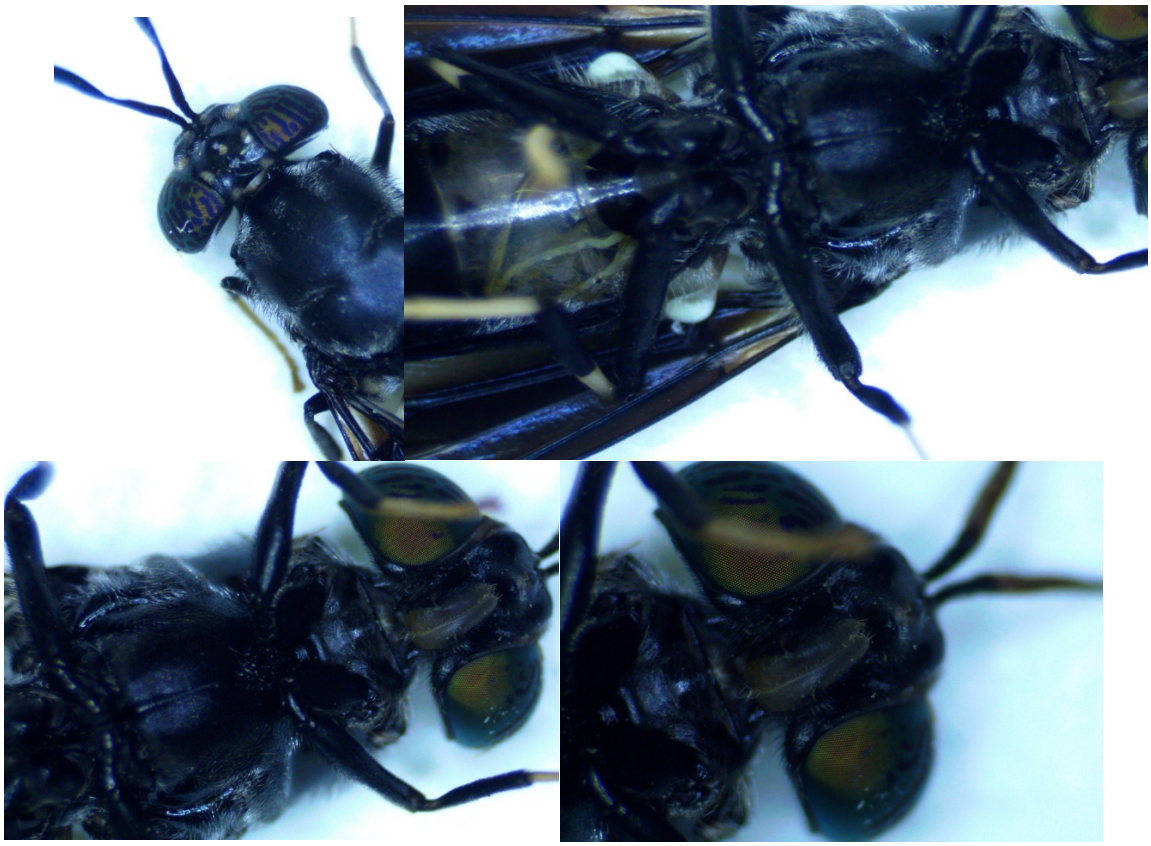

Figure 2: Black soldier fly (magnified under a microscope)

Mating takes place in the insectarium; we see this in Figure 3. Conditions close to nature are kept in it, which means like in the wild. In natural light and in sunny weather, mating occurs most intensively.

Figure 3: Mating flies in the insectarium

The metamorphosis of the development of most insects takes place in several stages, and the Black soldier fly is no exception. The entire life cycle includes all the traditional stages, mainly insects have four of them, and the Black soldier fly has five of them (Figure 4). This is the first distinguishing feature of flies from other insects. Then we studied each stage in detail, as well as the entire life cycle of the insect on different substrates and compared these phases with each other.

In the beginning, we would like to note that the life cycle from egg to egg is 42-45 days. Larvae need a moist and nutritious environment for development. To do this, the female after mating is looking for a more favorable environment for laying eggs. In this case, for laid eggs, this is the place of sponges or planks. After the female finds a suitable place for laying, she lays about 500-800 eggs at a time and if all the conditions are met their number increases.

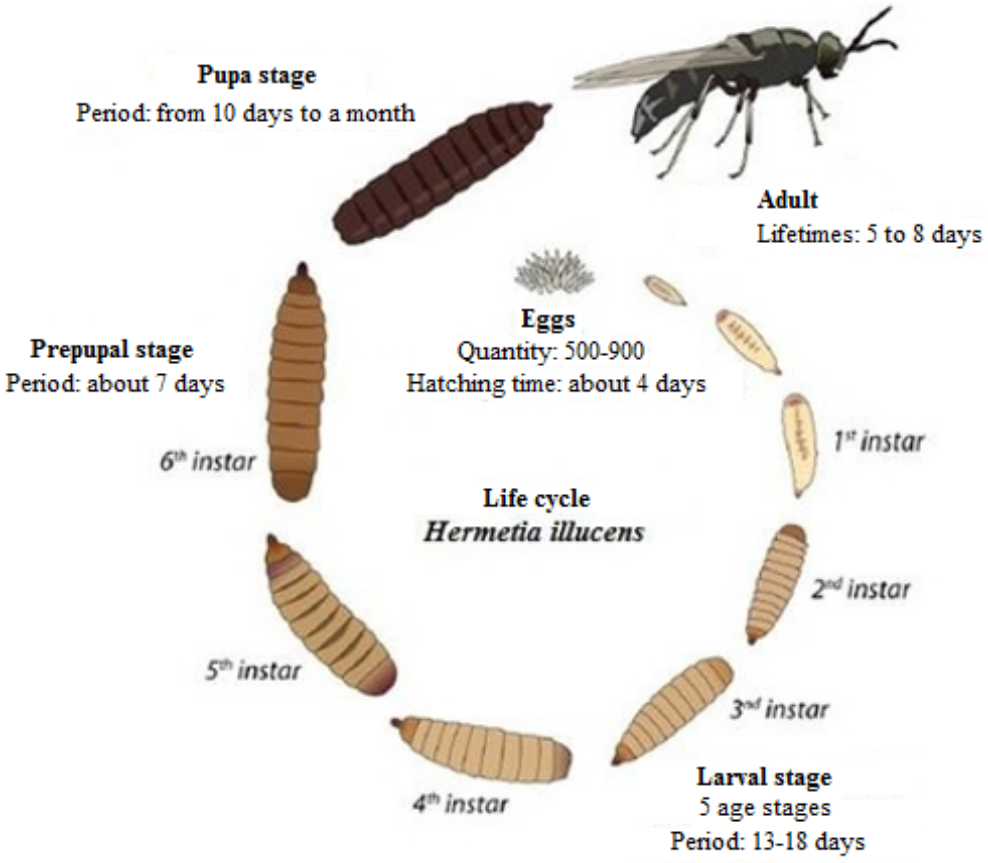

Figure 4: The life cycle of the Black soldier fly

For laying, it chooses a place closer to food for its offspring. In our research, we used meat broth and honey syrup. Laying lasts no more than 7 days. Egg laying is influenced by housing conditions, mainly temperature, humidity, lighting, feeding does not affect, as in other insects and animals.

Many factors, such as lighting, humidity, temperature and feeding, affect the growth, development, and feed biomass when breeding the Black soldier fly. If we consider the habitat of the fly, then we can say that the insect is unpretentious. And its larvae are omnivorous and develop at different temperatures in their environment. The temperature can range from 20 to 50°C, and the humidity can range from 40 to 90%. For an adult insect, the temperature should be 30°C, and the humidity 70%. We also consider both natural and artificial lighting and the availability of drinking water. If the optimal parameters of the microclimate are observed, the hatching ability of eggs reaches 80 percent or more. Antonov et al. [4] argue that the minimum lighting required for mating adult flies should be 70 mmol/m^2/s, the maximum – 100 mmol/m^2/s. The temperature for laying eggs is 27°C, and the relative humidity of the environment is 60%.

As we have already noted, the entire life cycle of the Black soldier fly should be about 45 days, but it may not correspond and will be delayed since ideal housing conditions will not be created, since breeding depends on external environmental conditions, and they greatly affect the development of the organism, mainly it is air temperature - 26-30°C, relative humidity up to 80%, natural lighting - 0.5-1.2%, availability of drinking water and abundant nutrition. The conditions should be close to the natural habitat to increase the percentage of hatching ability of eggs. In this case, the temperature in the insectarium was 27-29°C (at a norm of 26-30°C), the air humidity was 60-65% at a norm (75-77%), in this regard, a large waste of pupae was obtained, they simply dried out due to lack of humidity, and the laid eggs also dried quickly. Although, as we have already said, the insect is unpretentious, and the larvae are omnivorous and are able to develop at different temperature fluctuations from 18 to 41°C and humidity from 40 to 90 % [5, 7, 8].

Figure 5: Planks and sponges for laying eggs

Egg laying is not a long period, the fly lays eggs for 4-5 days, the life span of an adult is only 5-8 days, then the flies die. Although we have observed that this process can be delayed for a longer time up to 20 days or more, this happens at low temperatures and insufficient weak lighting, flies become less active, mate poorly, are sedentary, sit, drink water, and if the fly is fed with honey syrup, then their life span increases and, consequently, the process. We also noticed that flies live much longer, but do not mate. From the moment the flies hatched from the pupa until all the flies died, the time could be up to a month. As we have noticed, the life expectancy of the adults is small, and the main function is reproduction, this is the meaning of the life of this fly. Flies lay eggs in the most suitable places for the survival of their offspring, in this case, we see in Figure 5 - these are sponges moistened with water or honey syrup, as well as planks that were placed over meat broth or honey syrup, and best of all these are honeycombs (Figure 6). Eggs do not need to be collected from them, they can be immediately laid in a prepared substrate, without growing in a nutrient medium, so there is less loss of eggs, and the larvae feed at first at the expense of honeycombs made of wax.

Figure 6: Honeycomb for laying eggs

Staying in the egg state is the shortest development cycle. The egg collection is shown in Figure 7. Eggs were collected and transferred to a nutrient medium for growing larvae, it consisted of steamed compound feed (oat, barley). It takes only three days for the egg to metamorphose into a tiny larva. In our experiment, the exit of the larva from the egg was from three to five days. Only the larvae that have hatched from the eggs are very small white, in the form of thin threads that are barely visible to the naked eye. After a couple of days, the larvae that appeared, which had a white or light yellow color and a yellow-brown head, had a body length of about 3 mm.

Figure 7: Egg collection

At the first stage of the research, we studied the biological development cycle, according to which, the first stage of development, a slight growth of larvae, lasted about 14 days, more often not even more than 5 days, and the initial size was 5 mm. The larva is very difficult to detect, since it hides from direct sunlight, rushing into the depths of the nutrient medium from which it was born. The second stage is 10 days, an active increase in body size up to 10-12 mm. In Figures 8 and 9 we see the third stage (pre-pupa) - 8-12 days, larvae eat a lot, grow quickly, size up to 20 mm. From Figure 9 it can be seen that the larva body is light yellow, you can even say golden in color,

consists of segments, the head is clearly visible. In Figure 10, we see the first molt of the larvae, in which the initial color changes and becomes dark brown. The shell becomes rigid and dense. For its development in the thickness of the nutrient substrate, the larva gains the calories it needs. It becomes fat, large, and the color changes from a pale shade to brown. Pupation takes place – this is a new stage – the pupal stage (Figures 11 and 12). This is the most passive stage of the insect's life, it lasts 10-11 days, during which the skin of the larva hardens (pupal cover, puparium) and a pupa is formed. Recent changes are taking place inside the puparium. The larva's tissues are disintegrating, and all its systems are being restructured. After that, an adult individual (imago) is formed. An adult fly comes out of the pupal chamber. The breeding cycle repeats again [6, 7].

Figure 8: Larvae in the pre-pupal phase

Figure 9: Larva (magnified under a microscope)

Figure 10: Molting of the pre-pupa

Figure 11: Larva in the pupal stage

Figure 12: Larva in the pupal stage (magnified under a microscope)

The length of adult larvae reaches an average of 25-27 mm, it depends on the substrate, in this case, the larvae develop faster and better on chicken droppings, they are more active compared to pig manure (Figure 13). For better processing of organic waste, we added Bacillus subtilis hay bacterium to chicken droppings and pig manure.

Figure 13: Larvae on pig manure and chicken droppings

The life cycle of the Black soldier fly averages 6-7 weeks. Our studies have shown that the total life expectancy of the Black soldier fly was 42 days on chicken droppings and 45 days on pig manure. We put together each separate phase of development from egg-laying to fly hatching and got this period.

4 Conclusion

Having studied in detail all the phases of the Black soldier fly development on pig manure and chicken droppings from egg to adult, we added up the duration of all phases and concluded that the duration of the complete biological cycle of the fly on different substrates corresponds to this period of 6-7 weeks. Our research has shown that the life cycle on chicken droppings is less by 3 days than on pig manure. It was 42 days on chicken droppings, and 45 days on pig manure. Also, based on the conducted research, we can shorten or increase the biological cycle of the fly, since it depends on several factors, mainly temperature, humidity, lighting, feeding and watering.

The addition of Bacillus subtilis bacteria, in turn, contributes to favorable conditions for keeping larvae and shortens the decomposition time of droppings and manure, as well as disinfects pathogenic microorganisms.

5 Availability of Data and Material

Data can be made available by contacting the corresponding author.

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