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INFLUENCE OF LARVAL NUTRITION ON PHYSIOLOGY OF PROPAGATION OF VARIOUS MOSQUITOES

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In the foreground of investigations on mosquitoes important from the medical viewpoint have been for sometime already questions of the formation and differentiation of strains. Of special interest in this are physiological peculiarities and characteristics whose genotypic character is still in dispute. For the purpose of analyzing these characteristics, there have been studied in detail primarily two environmental factors, the climatic conditions whose importance has been again stressed recently by Martini (1), and the influence of nutrition. Climatic and microclimatic factors may develop their effectiveness both in the larval and in the imago stages but the influence of nutrition makes itself felt primarily during the development of the larvae.

Weidling (2) has already investigated in our institute the relations between the amount of food and the imagoes (size, number, and development of the ovarioles) of various diptera and Hecht (3) later carried out experiments on physiology of nutrition in connection with the autogenous propagation of Culex pipiens. This is intended to be a brief report on our own recent experiments in the latter direction. A detailed review of this work will be made later.

In continuation of the experiments by Hecht, we utilized different foods, in addition to changing temperature, in raising larvae of C. pipiens. The larvae were usually fed with an infusion of hay and the mosquitoes thus fed also came to deposit eggs without having absorbed any blood. The amount of food has an influence on the number of eggs deposited but not on the autogenous production of eggs as such. Infusion of hay would appear to be a food relatively high in protein but it would be difficult to analyze the individual components as a function of the developing microfauna and microflora.

According to a recent communication by Boissezon (4), feeding with boiled lentils is alleged to be highly suitable for stimulating autogenous propagation of C. pipiens because of the high protein content. I did em-

ploy this type of feeding but observed no advantage as compared to the infusion of hay. Feeding with lentils has developed especially large animals with a voluminous fat body but without any perceptible influence on the egg development and yolk formation.

This more or less vegetarian nutrition was contrasted by a purely animal nutrition with dried blood and pulverized calf-liver. Both types of feeding help the larvae to thrive and produce strong imagoes. The quickest growth is produced by feeding with liver. The young mosquitoes are very lively and suck much easier than those raised on a vegetarian diet (the females in some batches declined any absorption of blood) and eggs matured quicker and more intensively in the autogenous batches.

If the descendance of the mosquitoes fed with liver was again fed with liver, manifestations were observable in the subsequent generations which I am inclined to interpret as degeneration. Mortality and sensitivity of the larvae increased; in some batches only a few larvae reached pupation and subsequently developed into imagoes. Of these imagoes almost all showed egg development but the latter became arrested in various stages of yolk formation. Only a very small percentage of the mosquitoes was able to mature and deposit eggs. Raising with infusion of hay of the larvae obtained from these eggs immediately eliminated the manifestations of degeneration. The mortality of the larvae disappeared and ovarian development was uniform. Even in breeding with very small mosquitoes left hungry, a large part of the females not only reached autogenous egg development but also deposited them. The nutrition of the larvae with purely animal food and especially with such a particular and rich food as liver very probably does not correspond to natural conditions. We may therefore assume that such feeding may trigger a passing stimulation in some direction but will result in degenerative manifestations in the long run. So far we know very little on the nutrition of mosquito larvae under natural conditions and the assimilation of the individual nutritional components. However, we can say that the condition and behavior of mosquitoes may be regulated by environmental influences which are quite remote such as the nutrition of the parent mosquitoes during the larval stage.

It seemed evident to employ feeding with liver also for other mosquito larvae. Experiments with C. fatigans at first were unsuccessful. We did succeed in raising them without difficulty when using a combined feed of liver and blood. This resulted in normally copulative males and females but an autogenous egg development did not take place. However, those batches were fed with more blood than liver and even the former was given only in the last larval stage. From my experiences with C. pipiens, I can say that any feeding of this type which is to be successful, must begin earlier. The females of C. fatigans displayed a special hunger for blood. Subsequent to a blood meal, normal deposition of eggs took place. The first batch deposited (two weeks after sucking) was bred further. The larvae again received blood and liver. As of this moment (seven days after emergence), the females show a perceptible growth of the eggs and appreciable yolk deposition whereas no egg development could be determined in the

paternal generation. Whether the development of complete eggs will take place cannot yet be affirmed.

Especially noteworthy is the reaction of stegomyia fasciata to feeding with blood and liver during the larval stage. Development took place smoothly. The imagoes were quite obviously larger than those of our institute batches fed with infusion of hay. The first batch produced twenty-six males and eleven females. One of the females was examined very soon after emergence for demonstration of copulation. Three females were permitted to suck blood and normally deposited eggs after four days. Moreover, two females had almost matured eggs upon examination of their ovaries and five females showed the beginning of yolk deposition. It is almost certain that at least two of the prematurely examined females would have reached deposition. All females had copulated. The F_1-generation showed, four days after emergence, the beginning of egg development and minor yolk deposition. This proves at least a disposition for autogenous egg development also for S. fasciata.

Anopheles maculipennis has always been known as necessarily hematophage. Development of eggs without absorption of blood has not yet been observed. In a similar manner as for culex and stegomyia, I raised newly emerged larvae of the strain messeae by feeding some with blood and some with liver. At a mean temperature of 26°, pupation of the larvae fed with liver took place 10 days after emergence and one day later for those fed with blood. The size of a number of mosquitoes selected at random and measured was above the mean size determined from specimens caught in the open and far above that produced by our other feeding methods such as infusion of hay, or "piscidin." The strain atroparvus showed no difference in the rapidity of development as compared to messeae. Only the mortality of the larvae is greater for messeae than for atroparvus. The mosquitoes of the strain messeae raised with liver displayed greater activity than those fed with blood. Most of the females could be induced without difficulty to suck on rabbits and those with liver were more proficient in this than those fed with blood. These findings agree with those communicated recently by Roubaud (5). The latter obtained larger and more aggressive individuals through feeding with spleen than by feeding a vegetarian diet.

A number of females raised by me showed a rather considerable growth of the ovary without prior absorption of blood. Egg and nutritional cells separated very perceptibly and yolk formation began in the egg cells. The degree of egg development and of yolk deposition was irregular. It was more pronounced among the specimens fed with liver than among those fed with blood. The beginning of autogenous egg development on the basis of larval nutrition can therefore be observed also in A. maculipennis.

The feeding experiments of anopheles pursued a particular objective. We know that it has not been possible so far to induce the strain messeae to copulate in captivity. An attempt to induce the females to copulate by feeding the larvae with liver which doubtlessly stimulates the vitality of

the mosquitoes, has so far not been successful in my experience. Histological examination in vivo shows that the females do develop normally spermatozoae but that the latter frequently are completely immobile and very small in number whereas specimens caught in the open (at the moment I have available for comparison only females of atroparvus) always show very highly mobile seminal cells. The behavior of the spermatozoae was the same in the animals fed with blood and liver. There is no doubt that the absence of copulation in messeae raised in the laboratory must be ascribed to the females just as in the non-copulative batches of C. pipiens. It is therefore possible that the absence of a component in the larval nutrition causes the behavior of the spermatozoae (if the latter is truly abnormal) and consequently the lack of inclination to copulation.

Eggs matured normally in messeae females after a blood meal. A second blood meal was declined. Deposition of non-fertilized eggs has been observed so far once. The insects behaved otherwise like non-copulative Culex females. In the latter, the actual deposition is retained. Non-fertilized eggs are laid infrequently. In many cases the females die before they lay eggs.

Summary: Experiments with C. pipiens and fatigans, stegomyia fasciata and anopheles maculipennis messeae indicated that raising of the larvae with a purely animal feed (blood and calves liver) as compared to a primarily animal (sic) feed (infusion of hay) has a positive influence on the size of the imagoes and increases their vitality. Continuation of this larval nutrition produced manifestations of degeneration in subsequent generations of C. pipiens which disappeared again on feeding with infusion of hay. Autogenous propagation is favored in C. pipiens by feeding the larvae with liver. In the other mosquitoes examined, the influence of the larval nutrition produces the beginning of ovarian development with egg development and yolk deposition without prior blood meal.

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