

A NOTE ON THE ACTION OF COMMON SALT ON THE LARVAE OF *STEGOMYIA FASCIATA*.

BY J. W. SCOTT MACFIE, M.A., M.B., CH.B.,

West African Medical Staff.

(PLATE XXXII.)

It has been pointed out by Graham* that measures designed to bring about the destruction of mosquito larvae may be divided into (a) measures intended to destroy the larvae, and (b) measures intended to destroy their food supply. He points out that very little attention has been paid to the second method, which entails an exact knowledge of the freshwater algae. Graham found that the suspended matter in water in which the larvae of *Pyretophorus costalis* were breeding could be precipitated by the addition of 3 per cent. of common salt, and that then the larvae became cannibalistic and destroyed one another. He considered that this phenomenon was due to the larvae being deprived of their natural food, the algae, which had been destroyed by the addition of salt. "In lesser concentration," he concluded, "salt appears to inhibit the growth of very young larvae, probably by diminishing the supply of food, but the development of fully-grown larvae appears to be hastened in a hypertonic medium, and they pass into and through the pupal stage with unusual rapidity."

The following experiments were undertaken with a view to determining to what extent the action of salt on mosquito larvae was due to the destruction of the natural food supply, and to what extent to the hypertonic nature of the solution. The larvae employed were those of *Stegomyia fasciata*. This species was selected both because its breeding places would be most easily treated with salt, should this substance prove of value as a larvicide, and on account of its importance in a country in which yellow fever is endemic.

Salinity of the Medium in which the Larvae were found.

At Lagos the larvae of *Stegomyia fasciata* are commonly found in water-pots and domestic utensils in the compounds of the native quarters of the town. A number of samples of water were obtained from this source, and an analysis was made of the amount of salt present in each. The percentage was found to vary considerably, ranging from 0.005% to 0.019% NaCl, and the average of six determinations was 0.012% NaCl.

This figure was found to be considerably below that for the water of the Lagos lagoon at the same season (September). The salinity of the water flowing beneath the Iddo bridge, to the North of Lagos, was found to be 0.026% NaCl: whilst that at the Magazine bridge, considerably nearer the sea, was 0.112% NaCl.

* Bull. Ent. Res. Vol. i, p. 51-52, 1910.

It may therefore be assumed as probable that the larvae of *S. fasciata* thrive best in a medium whose salinity is about 0.012 per cent. NaCl. In order to determine whether the higher percentage of salt present in the lagoon would prove injurious to them, three samples of water containing larvae from domestic utensils was selected, and salt was added to them until the salinity of each was 0.026, the same percentage as was found to be present in the lagoon water at the Iddo bridge. No perceptible action was observed on the larvae, which remained healthy and active during the succeeding four days. The percentage of salt in two of the samples was then raised to 0.112, as in the lagoon water under Magazine bridge. The larvae seemed to be affected somewhat by this degree of salinity, and tended to remain for long periods at the bottom of the jars. By the seventh day all those in the one sample were dead, and only one remained alive in the other. The last larva died on the eighth day.

Experiments with Solutions of 0.5 per cent. NaCl and upwards.

The foregoing preliminary experiments suggested that it might be of some interest to determine the effects of various strengths of salt solutions on the larvae of *S. fasciata*. In order to exclude the factor of precipitation, the experiments were in the first instance carried out in clean water.

For this purpose ten mature larvae were placed in each of six jars containing respectively 0.5 %, 1 %, 2 %, 3 %, 4 %, and 5 % NaCl solutions. The results of the experiments are shown in detail below :—

TABLE I.

NaCl.	0.5 %	1 %	2 %	3 %	4 %	5 %
Oct. 12, 11 a.m.	10 L.	10 L.	10 L.	10 L.	10 L.	10 L.
" 1 p.m.	9 L., 1 P.	10 L.	9 L., 1 P.	10 L.	1 L., 9 dead L.	All dead
" 3 p.m.	9 L., 1 P.	10 L.	9 L., 1 P.	7 L., 3 dead L.	All dead.	
" 5 p.m.	9 L., 1 P.	10 L.	9 L., 1 P.	6 L., 1 dead L.		
" 11 p.m.	9 L., 1 P.	10 L.	9 L., 1 P.	All dead.		
Oct. 13.	7 L., 3 P.	7 L., 3 P.	8 L., 1 P., 1 dead L. Three larvae almost inert.			
Oct. 14	6 L., 4 P.	7 L., 2 P., 1 M.	4 L., 1 P., 3 dead L.			
Oct. 15	5 L., 2 P., 3 M.	6 L., 3 M.	All dead.			
Oct. 16	5 L., 2 P.	6 L.				
Oct. 17	5 L., 2 M.	4 L., 2 dead L.				
Oct. 18	5 L.	4 L.				
Oct. 19	5 L.	4 L.				
Oct. 20	5 L.	3 L., 1 dead L.				
Oct. 22	5 L.	1 L., 2 dead L.				

L = Larva, P = pupa, M = adult mosquito.

The effects of the different solutions on the larvae varied directly with their strengths. In 5 % NaCl all the larvae had died within two hours, in 4 % within four hours, and in 3 % within twelve hours. In 2 % NaCl solution all had died within three days; in 1 % during the ten days for which the experiment was continued, five larvae died, four pupated and hatched, and one remained alive at

the conclusion of the observations. The lowest concentration, 0.5 % NaCl, seemed to have but little effect; five larvae pupated and hatched, and five remained alive at the end of the experiment; none died.

The rapidity with which the larvae were killed by the stronger solutions seems to prove that, at any rate as low as 2 or 3 % NaCl, the action on the mature larvae of *S. fasciata* is an osmotic one, and is not dependent on the destruction of algae, nor on the deprivation of the insects of their natural food supply.

The action of the salt solution is not, however, appreciably altered by the presence of debris, etc., as is proved by the following experiment. A large number of larvae and pupae in their natural medium were placed in a jar, and enough salt added to bring up the strength of the solution to 2%. Six hours later many of the larvae were dead, and twelve hours after this all had perished. A number of the pupae hatched during the first two days, but on the third day all those that remained were dead.

Experiments with more dilute solutions of salt.

Although solutions of salt of 2 % and upwards appeared to produce an inimical effect on the larvae of *S. fasciata* by direct osmotic action, more dilute solutions in clear water had a much less pronounced effect. It was considered possible, however, that the latter solutions might cause a precipitation of the organic constituents of the natural fluid in which the mosquito larvae were found, and might thus kill, or at any rate inhibit the growth of, the larvae by depriving them of food as was suggested by Graham.

Mature larvae were therefore distributed into glass jars, and the salinity of the natural medium, which contained 0.012 % NaCl, was increased by the addition of salt to 0.10 %, 0.15 %, 0.20 %, 0.25 %, and 0.30 % respectively. Larvae were also introduced into a solution of 0.32 % NaCl in pure water for comparison. The results of these experiments are shown below.

TABLE II.

NaCl.	0.10 %	0.15 %	0.20 %	0.25 %	0.30 %	0.32 % in pure water.
Oct. 11.	20 L.	20 L.	10 L.	20 L.	20 L.	10 L.
Oct. 12.	17 L., 3 P.	13 L., 2 P., 5 dead L.	5 L., 1 P., 4 dead L.	9 L., 4 P., 7 died L.	9 L., 1 P., 10 dead L.	7 L., 1 P., 2 dead L.
Oct. 13.	11 L., 9 P.	9 L., 4 P., 2 dead L.	2 L., 3 P., 1 dead L.	4 L., 6 P., 3 dead L.	8 L., 2 P.	5 L., 3 P.
Oct. 14.	8 L., 12 P.	3 L., 9 P., 1 dead P.	5 P.	1 L., 7 P., 2 M.	2 L., 7 P., 1 M.	5 L., 3 P.
Oct. 15.	2 L., 9 P., 8 M., 1 dead P.	1 L., 8 P., 3 M.	2 P., 3 M.	4 P., 3 M., 1 dead L.	7 P., 1 M., 1 dead L.	3 M., 5 dead L.
Oct. 16.	1 L., 8 P., 2 M.	3 P., 3 M., 3 dead P.	2 M.	2 P., 2 M.	2 P., 5 M.	
Oct. 17.	3 P., 4 M., 2 dead P.	3 M.		1 M., 1 dead P.	1 M., 1 dead P.	
Oct. 18.	1 P., 2 M.					
Oct. 19.	1 M.					

L = Larva; P = pupa; M = mosquito.

It will be observed that during the first 24 hours the effect of the saline solutions in the natural medium, estimated by the death of the larvae, was proportional to the strengths of the solutions used. In the experiment with clear water the action was not marked until three days later. It is possible that the injurious effects in the former cases may have been due to the clogging of the tracheae of the larvae by precipitates. The deaths during the first 24 hours cannot at any rate be attributed to starvation.

All the larvae in these experiments had either died, or pupated and hatched, by the 9th day. The number that died was roughly in proportion to the strength of the saline solution. Thus in the 0·10 % solution none died as larvae, but in the 0·30 % eleven of the original twenty died in this condition.

NaCl.	0·10 %	0·15 %	0·25 %	0·30 %
Hatched	17	9	8	8
Died as pupae	3	4	1	1
Died as larvae	0	7	11	11

Very young larvae appeared to be more adaptable than mature larvae to a change in the concentration of the medium. Solutions containing 0·10 %, 0·20 %, and 0·30 % NaCl did not appear to have any appreciable effect on them.

Experiments with Alum Solutions.

If, as has been suggested, the inimical action of common salt on mosquito larvae is due to the precipitation of the minute algae on which they feed, it might be supposed that alum would have a more pronounced effect owing to its well-known clarifying action on impure waters. It was thought possible also that the gelatinous precipitate might interfere with the respiratory processes of the larvae, for it had been observed that in a pool containing innumerable larvae great numbers had died when some toad spawn was deposited in it. The spawn floated on the surface of the water and spread out like a veil, the eggs being connected by fine threads of mucus in which the mosquito larvae had become entangled and had died.

Three experiments were carried out in which potash alum was added to turbid water containing larvae so as to bring the strengths of the solutions up to 0·05 %, 0·10 %, and 0·20 % respectively. Ten mature larvae were used in each experiment.

The addition of the alum produced a copious precipitate which slowly settled to the bottom of the jars, but there was no immediate effect on the vitality or activity of the larvae. In the jar containing 0·05 % solution five larvae pupated and hatched, one larva died on the 7th day, and four that had pupated died on the third and fourth days. The last pupa hatched on the 12th day. Alum in this concentration appeared to have little or no direct effect. In the jar containing 0·10 % solution only one mosquito hatched, and the other nine larvae died between the third and the seventh days. In the third jar containing 0·20 % solution seven larvae died between the fourth and the seventh days, and three

that had pupated died on the third, fourth, and sixth days respectively. It was evident therefore that alum in these concentrations had no peculiar action on the larvae.

In more dilute solutions in clean water the results were inconclusive. Of ten mature larvae in a 0.01 % solution three pupated and hatched, and seven died as larvae; in a 0.02 % solution exactly the same results occurred; and in a 0.03 % solution two pupated and hatched and eight died as larvae. All had either hatched or died by the eighth, ninth, and tenth day respectively.

The action of a medium containing salt and alum in equal parts was tried in another series of experiments. The results, however, were similar to those obtained with a corresponding concentration of common salt alone.

Summary of Results.

The foregoing experiments would seem to prove that, in solutions of 2 % and upwards, the action of common salt on the larvae of *Stegomyia fasciata* is due to the hypertonicity of the solution. In more dilute solutions the destruction of the natural food supply of the larvae may have some influence. Alum, however, which clarifies water more efficiently than common salt, has no peculiar action on the larvae.

In Lagos the larvae of *Stegomyia fasciata* are found most abundantly in water contained in domestic utensils, and in the large pots in which the natives store up water for drinking and cooking. It would be of great advantage if common salt could be used as a larvicide in these cases; for not only would the water not be rendered unfit for use in cooking, as it is by the application of kerosene, but also evaporation would tend to increase the strength of the larvicide, and repeated applications would be unnecessary. It would also be possible to keep stores of water in the compounds without danger. From the experiments described, it would appear that sufficient salt would have to be added to each vessel to bring the concentration of the solution up to 2 % NaCl in order to ensure the destruction of the larvae.

The Survival of Mosquito Larvae in Temporary Pools.

It often happens that small pools of water, such as those illustrated by the accompanying photographs (Pl. XXXII), collect beneath the taps of tanks and at the open ends of drains, and into these mosquito larvae are apt to be swept. Such pools soon dry up, the water seldom lying in them for more than an hour or two, and they may thus escape the attention of the Sanitary Inspectors. The soil round them is, however, permanently wet: and if, as is usually the case, the pools are renewed once a day or more frequently by the tanks or drains being used, it is quite possible that mosquito larvae might develop in them. In some cases the finer soil washes away leaving a mass of irregularly-shaped stones in the interstices between which water may lie concealed all day long. Such pools may occasionally account for the presence of mosquitos in a house near which no breeding place can be located.

With a view to determining to what extent mosquito larvae were capable of surviving intermittent desiccation of this kind, the conditions obtaining under a

tank like that shown in Pl. XXXII. fig. 2 were reproduced in the laboratory by means of tins in the bottom of which small holes were pierced. The tins were half filled with soil and small stones, and an inch of water was added on the top. The drainage was arranged so that the surface of the soil became exposed after about one hour. The artificial pools thus formed were filled up every morning, and allowed to drain away as described. Thus a free surface of water was present for about one hour each day only. The soil, however, remained moist throughout the day.

Into such artificial pools pupae, mature larvae, and young larvae of *Stegomyia fasciata* were introduced. In the case of pupae the majority always hatched in the course of three or four days. In one experiment started with ten pupae, four had hatched within the first 24 hours, three more during the next 24 hours, and two more during the third day; only one pupa perished. Mature larvae were found to be much less resistant to desiccation. They not infrequently pupated, but in our experiments none of the pupae hatched. Young larvae also survived only a short time, and did not undergo their normal development. Some of them however, remained alive until the sixth day; and it is possible that had the pools been renewed more often, or allowed to persist a little longer, as no doubt they often do under natural conditions, the larvae might have matured, pupated and even hatched out into mosquitos.

The chief danger of such occasional pools therefore is that pupae, washed into them from other pools or from the gutters of bungalows, may complete their development in them. But as even immature larvae may also live for a number of days in such situations, it would perhaps be a wise precaution to treat these places with larvicides.



Fig. 1. Temporary pool at the open ends of a drain.

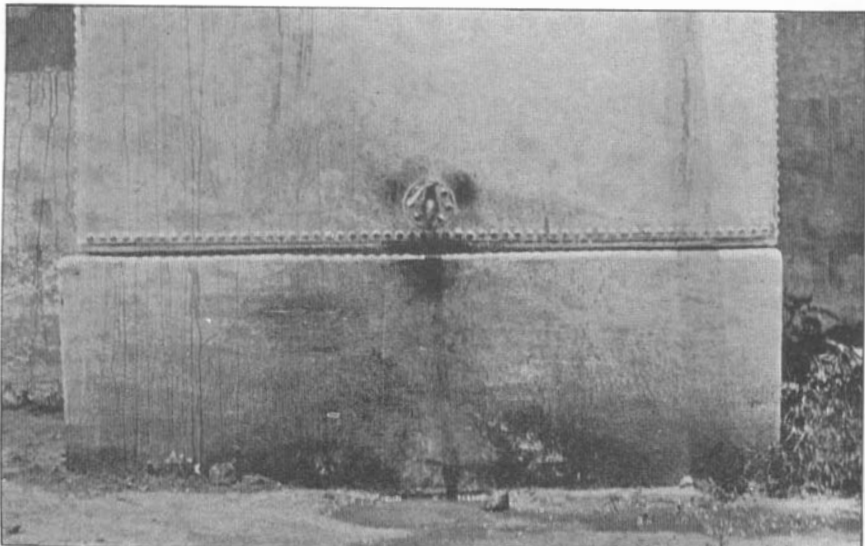


Fig. 2. Water-tank, showing the pools that collect under the tap.

BREEDING PLACES OF *STEGOMYIA FASCIATA*, LAGOS, S. NIGERIA.