TRANSMISSION OF FOWL-POX BY MOSQUITOES.

By I. J. KLIGLER, PH.D., R. S. MUCKENFUSS, M.D., AND T. M. RIVERS, M.D.

(From the Hospital of The Rockefeller Institute for Medical Research.)

Plates 38 to 41.

(Received for publication, December 7, 1928.)

Fowl-pox, a common disease of the barnyard, annually recurs in epidemic form throughout numerous countries. The virus that produces the disease is an infectious agent (1) capable of passing through bacteria-tight filters (2), and, though not identical with other pox-producing viruses, probably is closely related to some of them. As a rule fowl-pox is not extremely fatal, yet the depression in the egg-laying activity of infected fowls leads to a great economic loss.

From the records of the epidemics of fowl-pox studied by Bollinger (1), Sanfelice (3), and others, it appears that the disease occurs for the most part during the spring and fall months. Summer epidemics, however, have been observed. In America, according to Beaudette (4), the disease is most prevalent during April and October.

Although the fact that fowl-pox is transmissible by direct inoculation (1) has long been recognized, the mode of its rapid spread under natural conditions has remained somewhat of a mystery. Contact infection undoubtedly can occur at times. Bollinger (1) reported that he obtained an infection in a healthy chicken by placing it in a yard with a sick fowl. Burnet (5), however, was able to obtain results similar to Bollinger's only when the majority of the feathers had been plucked from the healthy fowl before it was placed in contact with the diseased one. He was also able to infect a pigeon by feeding it virus, but, in order to assure a positive result, he mixed ground glass with the meal of wheat and virus. In this instance the lesions occurred in the mouth and esophagus and not on the comb, the usual place for the disease to manifest itself. Notwithstanding the results above described, the experience of most workers (Burnet (5)) has been that under laboratory conditions healthy fowls housed in the same cage with diseased birds do not contract fowl-pox. Indeed, Goodpasture (6) states that no method of artificial infection seems adequate to explain the rapid and thorough infestations which occur under natural conditions.

Some workers have attempted to incriminate (7) various insects as vectors of fowl-pox. Méggin (8) suggests that flies and other insects may play a rôle in the dissemination of the disease, but offers no convincing experimental evidence.
TRANSMISSION OF FOWL-POX BY MOSQUITOES

in support of his hypothesis. Farmers (4, 9) in various localities are of the opinion that mosquitoes are instrumental in the spread of the malady, but no records of experiments substantiating the idea have been found. Schuberg and Kuhn (10), however, report that in each of 3 tests they succeeded in transferring fowl-pox from diseased to normal chickens by means of interrupted feedings of Stomoxys calcitrans,—the flies starting their meal on an infected bird and completing it on a healthy one. These experiments were not extended, and it appears that they have aroused little, if any, interest.

The records of failures to transmit fowl-pox under the conditions indicated above, the occurrence of epidemics in the spring, summer, and fall, the fact that the lesions appear on exposed parts of the body (comb and wattles), and the experiments reported by Schuberg and Kuhn (10) indicated to us that biting insects may play an important rôle in the transmission of the disease. The experiments described in the present paper were undertaken to ascertain whether mosquitoes are of any significance in the spread of fowl-pox.

EXPERIMENTAL.

Methods and Materials.

Virus.—The strain of fowl-pox virus employed was originally obtained from Dr. Andervont and has been in this laboratory for approximately 2 years. Before starting our experiments we tested the activity of the virus by means of 2 serial passages in chickens, and also proved its purity by suitable tests in rabbits.

Chickens.—With a few exceptions the chickens employed in the experiments were White Leghorns. In working with fowl-pox one must remember that it is a natural disease of chickens and that immune or partially immune fowls are likely to be encountered. Consequently experiments should be repeated frequently or should be run in duplicate.

Mosquitoes.—Culex pipiens and Aedes aegypti were used. The former were either caught in houses or bred in the laboratory from larvae; the latter were hatched in the laboratory from dried eggs supplied by Dr. Boyd of the Rockefeller Foundation.

Cages.—The mosquito cages consisted of stiff wire frames made in the form of a cube and covered with ordinary cheese cloth. From one side of the cube the cloth extended in the form of a sleeve which served as the only opening to the cage and through which the mosquitoes or larvae were manipulated.

Technique of Feeding.—Individual mosquitoes were caught in wide test-tubes, 8” x 1”. The mouth of each tube was then covered with cheese cloth, which was held in place by a rubber band. Since mosquitoes do not feed readily under these conditions, they were starved for 24 hours prior to the experimental meal. Feeding
was further facilitated by first holding the gauze-covered end of the tube containing
the mosquito towards the source of light. The mosquito was attracted by the
light and came to rest on the gauze. The tube was then manipulated without
disturbing the mosquito so that the gauze-covered mouth of the tube was placed
over the area of the comb or wattle to be fed upon. If the mosquito was hungry
and its proboscis gently came in contact with the skin of the chicken, feeding
promptly ensued. Sometimes a great deal of patience was required. As a rule,
however, no great difficulty was encountered in getting the mosquitoes to feed on
or near lesions.

The feeding once begun could be interrupted at any time by gently lifting the
tube. To prevent infection arising from the apparatus, immediately after the
feeding had been interrupted the gauze was removed, the lip of the tube was
washed with alcohol and dried, and a clean piece of gauze was placed over the
mouth of the tube. At different intervals of time following the infectious meal
the mosquito was made to refeed on normal chickens. During the process of
refeeding the insect often interrupted its meal either because of an unsatisfactory
probe or because the operator moved the tube. If the interruption was brought
about so gently that the mosquito remained on the gauze the meal was imme-
diately resumed at a new place. The natural or imposed interruptions proved to
be important because in this manner a single mosquito has been shown to be able
either to infect a fowl in several places or to infect more than one fowl.

Each tube and each chicken were numbered and records were kept of the time
and place of the different feedings. After the completion of each meal the tubes
containing the mosquitoes were placed upright in a wire basket and covered with
a moist towel. In order to determine the survival time of the virus, the infected
mosquitoes were fed on normal chickens at intervals of 3 or 4 days. At various
intervals of time following the infecting meal, mosquitoes were also killed, mac-
erated, and injected into the comb or wattles of a fowl.

Eighteen experiments were conducted, 10 with Culex pipiens and 8
with Aedes aegypti. The results are shown in the following protocols.

Culex pipiens.

Experiment 1.—Sept. 26, Culex 2 fed on the infected comb of Chicken 46,
drawing whitish fluid. 1 hour later the insect made several insertions of its
proboscis, without feeding, on the left side of the comb of Chicken 47. Oct. 5, 4
small vesicles were observed at the points where the mosquito had attempted to
refeed. Oct. 8, lesions larger (Fig. 1). Fluid from two of the vesicles was removed
by means of a capillary pipette and inoculated on the scarified comb of Chicken 56.
4 days later the comb of Chicken 56 showed definite lesions which progressed and
endured for many days; Figs. 4 and 5 are photographs of Chicken 56 taken a week
and a month, respectively, after inoculation. Oct. 10, lesions still increasing in
size (Fig. 2). Oct. 22, lesions still present (Fig. 3). Oct 25, lesions regressing.
Experiment 2.—Sept. 26, Culex 3 fed on the infected comb of Chicken 46, drawing blood. 45 minutes later the insect refed on the right side of the comb of Chicken 47. No lesions developed at the points of refeding.

Experiment 3.—Sept. 26, Culex 4 fed on the infected wattle of Chicken 45, drawing blood. 45 minutes later the mosquito refed on the right wattle of Chicken 47. No lesions developed at the points of refeding.

Experiment 4.—Sept. 28, Culex 5 fed near lesions on the infected comb of Chicken 46, drawing blood, and 15 minutes later refed, 1 bite, on the left wattle of Chicken 51. Oct. 5, a vesicle appeared on the wattle of Chicken 51 at the point where the mosquito had refed 7 days previously. Oct. 11, lesion larger (Fig. 6). Oct. 22, small secondary nodules were observed on the wattle (Fig. 7). The chicken was tested, Nov. 10, for immunity to fowl-pox virus.

Experiment 5.—Sept. 28, Culex 6 fed near lesions on the infected comb of Chicken 46, drawing blood. 15 minutes later the insect refed, 3 bites, on the left comb of Chicken 51. Oct. 5, 2 vesicles were noticed on the comb of Chicken 51, where the mosquito had refed. Oct. 11, lesions increasing in size (Fig. 6). Oct. 22, lesions still increasing in size (Fig. 7). Oct. 26, lesion regressing. Nov. 10, Chicken 51 was reinoculated with fowl-pox virus. Abortive lesions appeared rapidly and were gone within 9 days after the reinoculation.

Experiment 6.—Oct. 15, Culex 7 fed on the infected comb of Chicken 57, and 1 hour and 30 minutes later refed, 2 bites, on the left wattle of Chicken 59. Oct. 25, on the left wattle, at the points where the mosquito had refed, 2 vesicles were observed. The lesions progressed and were active for many days (Fig. 8). Nov. 17, definite evidence of healing.

Experiment 7.—Oct. 15, Culex 8 fed on the infected comb of Chicken 57, and 1 hour and 30 minutes later refed, 1 bite, on right side of the comb of Chicken 59. Oct. 22, a vesicle was noticed on right side of comb at the point where the insect had refed. The lesion progressed and was active for many days. Nov. 17, definite evidence of healing.

Experiment 8.—Oct. 18, Culex 9 fed on the infected comb of Chicken 57. 30 minutes later the insect finished its meal on the right wattle of Chicken 59. Oct. 25, at the points where the mosquito had bitten Chicken 59, 2 lesions, which progressed and evidenced activity for many days, were seen. Nov. 17, definite evidence of healing.

Culex 9 was kept alive for further refeedings.

Oct. 22, Culex 9 refed on the right wattle of Chicken 63. No lesions developed. This chicken was tested, Nov. 10, and found to be immune to fowl-pox.

Oct. 25, Culex 9 refed on the right side of comb of Chicken 66. A questionable lesion appeared, Oct. 29, and regressed in a few days. This chicken was tested, Nov. 10, and found to be immune to fowl-pox.

Oct. 30, Culex 9 was macerated and injected into the comb of Chicken 69. No lesion developed. This chicken was tested, Nov. 10, and found to be immune to fowl-pox.
It appears that the refeedings of *Culex* 9 on Oct. 22 and 25 took place on immune fowls. The chicken into which the macerated insect was injected also seems to have been immune.

**Experiment 9.**—Oct. 18, *Culex* 10 fed on the infected comb of Chicken 57, and 30 minutes later finished its meal on the left side of the comb of Chicken 59. Oct. 25, 2 small lesions were observed on the left side of the comb of Chicken 59. These lesions progressed and remained active for many days (Fig. 8). Nov. 17, definite evidence of healing.

*Culex* 10 remained alive and was refed on 3 occasions.

Oct. 25, *Culex* 10 refed on the right side of the comb of Chicken 63. No lesions developed. This chicken was tested, Nov. 10, and found to be immune to fowl-pox.

Oct. 31, *Culex* 10 refed on the right side of the comb of Chicken 69. No lesions developed. This chicken was tested, Nov. 10, and found to be immune to fowl-pox.

Nov. 1, *Culex* 10 refed on the anterior part of the right wattle of Chicken 70. After being refed the mosquito was macerated and injected into the right side of the comb of the same fowl. No lesion developed at the point of injection, but on Nov. 5, 2 definite lesions were observed at the points where the insect had refed. These lesions progressed and were photographed, Nov. 7 (Fig. 10). On Nov. 9, 1 of the lesions was excised and fixed in Zenker's fluid. Stained sections showed a typical fowl-pox lesion with Bollinger bodies in the epithelial cells (Fig. 14). Material from the other lesion was inoculated on the scarified comb of Chicken 73. Typical fowl-pox lesions developed.

The lesion on Chicken 70 that was not excised and from which material was removed for transfer began to regress on Nov. 12 and by Nov. 19 was completely healed. The lesion appeared quickly and healed rapidly. It is possible that Chicken 70 was partially immune.

From the experiments recorded above it is evident that *Culex* mosquitoes are able to transmit fowl-pox from infected to susceptible normal chickens. In 7 of 9 tests, infection occurred in the susceptible fowls which had been bitten by mosquitoes that had fed on infected birds 15 minutes to 2 hours previously. In 1 of 2 experiments, *Culex* 10, in which the mosquitoes remained alive for a number of days, a typical infection was produced in a fowl 14 days after the insect had taken its infective meal. The negative results obtained with the other mosquito, *Culex* 9, were probably due to the fact that the chickens on which it refed were immune to fowl-pox.

*Aëdes aegypti.*

Experiments similar to those with *Culex* mosquitoes were conducted with *Aëdes*. The results are recorded below.
Experiment 10.—Oct. 22, Aedes 1 fed on the infected comb of Chicken 57. Two hours later the insect completed its meal on the left wattle of Chicken 63. No lesions developed. This chicken was tested, Nov. 10, and found to be immune to fowl-pox.

Oct. 24, Aedes 1 refed on the left wattle of Chicken 66. No lesion developed. This chicken was tested, Nov. 10, and found to be partially immune to fowl-pox.

Oct. 30, Aedes 1 refed on the left wattle of Chicken 68. On Nov. 5, a lesion appeared where the insect had fed, and subsequently developed into a typical fowl-pox or contagious epithelioma wart.

After Aedes 1 had fed, Oct. 30, on Chicken 68, it was macerated and injected into the left side of the comb of Chicken 68. On Nov. 16, a typical fowl-pox lesion was observed at the point where the injection was made.

Experiment 11.—Oct. 22, Aedes 2 fed on the infected comb of Chicken 57, and 30 minutes later completed its meal on the left side of the comb of Chicken 63. No lesions developed. This chicken was tested, Nov. 10, and found to be immune to fowl-pox.

Experiment 12.—Oct. 24, Aedes 3 fed on the infected comb of Chicken 57. The insect finished its meal 1 hour later on the left side of the comb of Chicken 66. No lesions developed. This chicken was tested, Nov. 10, and found to be partially immune to fowl-pox.

Oct. 26, Aedes 3 refed on the left side of comb of Chicken 67. No lesions developed. This chicken was tested, Nov. 10, and found to be immune to fowl-pox.

Oct. 29, Aedes 3 refed on the posterior portion of the right side of the comb of Chicken 68. On Nov. 7, 3 vesicles appeared where the mosquito had fed and later developed in typical fowl-pox warts. Nov. 12, photographed (Fig. 9).

Oct. 31, Aedes 3 was macerated and injected into the right wattle of Chicken 72. No virus of fowl-pox could be detected in the wattle excised 9 days later.

Experiment 13.—Oct. 24, Aedes 4 fed on the infected comb of Chicken 57, and 1 hour later completed its meal on the left side of the comb of Chicken 66. No lesions developed. This chicken was tested, Nov. 10, and found to be partially immune to fowl-pox.

Oct. 26, Aedes 4 refed on the right side of the comb of Chicken 67. No lesions developed and the chicken was later found to be immune.

Experiment 14.—Oct. 25, Aedes 5 fed on the infected wattle of Chicken 51, and a few minutes later finished its meal on the right side of the comb of Chicken 66. No lesions developed. This chicken subsequently was found to be immune to fowl-pox.

Experiment 15.—Oct. 26, Aedes 6 began its meal on the infected wattle of Chicken 61 and finished it, 2 bites, on the right wattle of Chicken 67. A doubtful lesion appeared, Nov. 3, and disappeared within a few days. Chicken 67 was tested, Nov. 10, and found to be immune to fowl-pox.

Oct. 28, Aedes 6 refed in the center of the right side of the comb of Chicken 68.
No lesions appeared. Chicken 68 was definitely susceptible (see Experiments 10 and 17).

Oct. 31, Aedes 6 refed on the left wattle of Chicken 69. No lesions developed and the fowl was later found to be immune.

Nov. 3, Aedes 6, with Aedes 8, was macerated and injected into the right side of the comb of Chicken 72. No lesions developed.

Experiment 16.—Oct. 26, Aedes 7 fed on the infected wattle of Chicken 61, and a few minutes later finished its meal on the left wattle of Chicken 67. On Nov. 1, a small lesion appeared which persisted for only 3 days. Chicken 67 was tested, Nov. 10, and found to be immune to fowl-pox.

Oct. 29, Aedes 7 refed on upper part of the right wattle of Chicken 68. No lesion developed. The fowl was definitely susceptible (see Experiments 10 and 17).

Oct. 31, Aedes 7 refed on the central portion of the right side of the comb of Chicken 69. No lesion developed. The fowl was subsequently shown to be immune to fowl-pox.

Nov. 3, Aedes 7 was macerated and injected into the left wattle of Chicken 72. No fowl-pox virus was demonstrated in the wattle excised 9 days later.

Experiment 17.—Oct. 26, Aedes 8 fed on the infected wattle of Chicken 61. A few minutes after the meal was interrupted it was completed on the upper part of the left wattle of Chicken 67. No lesion appeared. Chicken 67 was shown subsequently to be immune.

Oct. 29, Aedes 8 refed on the lower part of the right wattle of Chicken 68. On Nov. 3, 2 small nodules were observed at the points where the insect had refed. Nov. 12, lesions photographed (Fig. 9). These nodules developed into typical fowl-pox lesions.

Nov. 1, Aedes 8 refed on the posterior part of the right wattle of Chicken 70. No lesion developed. This chicken was certainly partially susceptible at the time it was bitten (see Experiment 9).

Nov. 3, Aedes 8, with Aedes 6, was macerated and injected into the right side of the comb of Chicken 72. No lesion developed.

The results of the experiments with Aedes mosquitoes were not as uniformly positive as were those with the Culex. Eight Aedes mosquitoes were fed on infected chickens and later, at different intervals of time following the infective meal, were refed on healthy fowls. Of the 8 refeedings that were made within 2 hours of the infective meal only 2, Aedes 6 and 7, resulted in lesions at the points where the insects bit. These lesions were small and disappeared within a few days. Refeedings of Aedes 1, 3, and 8, which took place 9, 5, and 3 days respectively after the primary feeding, gave rise to typical fowl-pox lesions. The many negative results obtained in these experiments
were probably due to the fact that 4 of the 6 chickens, 63, 66, 67 and 69, employed were subsequently found to be either partially or completely immune to fowl-pox. Sufficient positive results were obtained, however, to show that Aedes mosquitoes are capable of inoculating healthy susceptible chickens with the virus of fowl-pox many days after they have fed on infected combs of diseased birds.

**Transmission of Fowl-Pox by Mosquitoes under Natural Conditions.**

The results of the work reported above show that Culex and Aedes mosquitoes are capable of transmitting fowl-pox. The tests, however, were made under experimental, not natural, conditions. It seemed advisable, therefore, to conduct at least 1 experiment in which the conditions approximated those occurring in nature.

**Experiment 18.**—Oct. 15, 2 healthy chickens (Nos. 61 and 62) and 1 (No. 57) with fresh fowl-pox lesions on both sides of the comb and on both wattles were placed in a mosquito-proof cage (Fig. 12). Five recently hatched Culex mosquitoes were also introduced into the cage. Oct. 22, 7 days after the experiment was started, 2 small lesions were observed on the lower part of the left wattle of Chicken 61. Oct. 26, lesions on Chicken 61 were still increasing in size (Fig. 11). One of the lesions was removed and fixed in Zenker's fluid. Stained sections (Fig. 13) showed a typical early fowl-pox lesion with many Bollinger bodies in the injured epithelial cells. The other lesion which was not excised went through the evolution usually observed in fowl-pox.

Chicken 62 never developed any lesions. This may have been due to the fact that it was immune to fowl-pox before the experiment was begun. In any event, it was tested, Nov. 10, and found to be immune at that time. It is unfortunate that tests for immunity cannot be made before the fowls are used for experimental purposes. Chicken 61, that showed the 2 typical lesions, later developed an immunity, which was probably acquired through the infection contracted during the experiment.

The control for the experiment was conducted in a manner similar to that employed in the test, with the exception that mosquitoes were excluded from the cage. 2 healthy chickens (Nos. 77 and 79) were placed in a cage with a fowl (No. 73) that had fresh fowl-pox lesions on the comb. The healthy chickens frequently pecked at the warty growths on the comb of the infected fowl. In spite of this close contact with the infectious agent, the healthy birds developed no lesions during the 12-day period of observation. They were later shown to be susceptible to fowl-pox.

The results of Experiment 18 clearly indicate that mosquitoes, under natural conditions, can transmit fowl-pox from diseased to healthy susceptible chickens.
Nature of the Lesions Produced by Insect Inoculation.

To present a study of the pathology of fowl-pox is not the purpose of this paper. It seems desirable, however, to describe the lesions produced by insect inoculation.

At the points on the comb or wattles of susceptible chickens where infected mosquitoes had fed or attempted to feed, minute nodules usually appeared within 5 to 9 days. Such lesions rapidly increased in size and assumed a grayish, glistening, translucent appearance. Frequently within 2 or 3 days the nodules were partially transformed into vesicles from which, by means of a capillary pipette, small amounts of whitish fluid rich in virus could be obtained (Figs. 1, 10, 11). If undisturbed the lesions continued to increase in size and gradually became large, yellowish, warty growths (Figs. 3 and 7). Finally regression and healing supervened and the wart-like scabs fell off, leaving superficial white scars. In non-immune fowls with 2 to 8 lesions, the duration of the disease was 3 to 6 weeks. In what appeared to be partially immune chickens, however, the incubation period was short, only 4 days at times, and the evolution of the disease was rapid, frequently requiring less than 2 weeks (see Experiment 9).

In certain instances, the stained sections of young lesions presented a marked hyperplasia of the epithelial cells with little, if any, reaction in the corium (Fig. 13a). In the case of 1 fowl, however, a lesion, approximately of the same age as the ones above described showed, in addition to the hyperplasia, vesicles in the epithelial layer and a marked cellular reaction in the corium (Fig. 14a). An explanation of this difference in the amount of reaction in the corium is not possible at present. Possibly this difference can be accounted for upon the ground that the depth of the insertion of the insects' proboscis varied considerably with each feeding and attempted feeding or that the reactivity of the fowls, without respect to specific resistance or immunity, was not the same in every case. Our impression, however, is that the marked early reaction in the corium occurred in a partially immune fowl (Experiment 9). This idea is at least in agreement with some observations recently reported by Andrewes (11) in regard to the histology of Virus III lesions in partially immune rabbits. From our studies it is clear that in some early fowl-pox lesions produced by the bites of infected mosquitoes a marked involvement of the epithelial cells may occur before any definite evidence of a reaction is discernible in the corium.
TRANSMISSION OF FOWL-POX BY MOSQUITOES

DISCUSSION.

To incriminate an insect as an important vector of disease, it is essential to establish the fact that it bites readily and comes in close contact with the susceptible hosts. The intimate relation between fowls and mosquitoes has long been recognized. Furthermore, *Culex* mosquitoes feed readily on birds and are known to transmit bird malaria.

Under the conditions of the experiments reported in the present paper it is evident that *Culex* and *Aedes* mosquitoes are capable of transmitting fowl-pox from diseased to healthy susceptible chickens. Moreover, the indications are that these insects may play an important rôle in the rapid and thorough infestations of flocks under natural conditions. The question, however, as to whether this mode of transmission is the most significant one remains to be answered by experiments conducted in the field. Certainly there is no reason why other biting insects may not also be of importance (10).

While in most epidemics of fowl-pox the lesions occur on the chickens' comb and wattles, in occasional outbreaks the majority of the manifestations of the disease is found in the mouth and throat. Inasmuch as the mucous membranes are inaccessible to mosquitoes and since the eating of virus alone usually does not result in infection, the spread of this type of the disease is hard to relate either to mosquitoes or to the simple ingestion of virus. It is possible that another infection, *e.g.*, bacterial, may injure the mucous membranes, thus making them susceptible to fowl-pox. In this manner a combination of infectious agents may account for the unusual and frequently fatal form of the disease.

It has been shown that mosquitoes are capable of transmitting fowl-pox at various times during the first 14 days following the infective meal. Tests at intervals longer than 2 weeks were not made. Therefore, the total duration of infectiousness of the insects remains to be determined. In view of the fact that the virus is active in minute quantities and is highly resistant to drying, the results of our experiments can be explained entirely upon the grounds that the mosquitoes mechanically transmit the disease without the occurrence of any multiplication of the virus in the insects. Before definite conclusions
can be reached, however, this phase of the problem will require further investigation.

The fact that insects may play a definite part in the rapid spread of certain diseases by mechanically transferring the virus from diseased to healthy individuals has been recognized by other investigators (13–16). This applies particularly to the virus diseases of plants (17). Nevertheless, from the results of our experiments, it appears that the importance of this mode of dissemination of certain virus diseases (12) of animals has either been underestimated or not sufficiently investigated.

CONCLUSIONS.

_Culex_ and _Aedes_ mosquitoes are capable of transmitting fowl-pox from diseased to healthy susceptible chickens.

The mosquitoes remain infectious for at least 14 days following a meal on diseased fowls.

BIBLIOGRAPHY.


15. Duke, H. L., Further inquiries into the zoological status of the polymorphic mammalian trypanosomes of Africa, and the means by which they are spread in nature, Parasitology, 1923, xv, 258.


EXPLANATION OF PLATES.

PLATE 38.

Figs. 1 to 3. Photographs of Chicken 47, showing the development of 4 fowl-pox lesions caused by the bites of an infected Culex mosquito.

Figs. 4 and 5. Photographs of Chicken 56 that was inoculated with infectious material taken from the lesions on Chicken 47.

PLATE 39.

Figs. 6 and 7. Photographs of Chicken 51, showing the development of fowl-pox lesions induced by the bites of infected Culex mosquitoes. a is primary lesion; b are secondary nodules.

Fig. 8. Chicken 59 with lesions produced by the bites of infected Culex mosquitoes.

Fig. 9. Chicken 68 with lesions induced by the bites of infected Aedes mosquitoes.

Fig. 10. Chicken 70 with 2 young fowl-pox lesions on the wattle induced by the bite of Culex 10, 14 days after its infectious meal. Lesion a was removed for histological study (see Fig. 14).

Fig. 11. Chicken 61 with 2 young fowl-pox lesions on the wattle. The bird contracted the disease in a cage in which healthy and infected fowls were placed with Culex mosquitoes. Lesion a was removed for histological study (see Fig. 13).

PLATE 40.

Fig. 12. The cage in which the transmission of fowl-pox by mosquitoes under natural conditions was tested.

PLATE 41.

Fig. 13. a represents a section of a young fowl-pox lesion taken from Chicken 61. Note the hyperplasia of the epithelial cells and the absence of reaction in the corium. × 50. b represents a normal epithelial cell. × 1200. c, d, e represent epithelial cells with Bollinger bodies in their cytoplasm. × 1200.

Fig. 14. a represents a section of a young fowl-pox lesion removed from Chicken 70. Note the hyperplasia of epithelial cells, the vesicles in the epidermis, and the marked reaction in the corium. × 50. b, c, d, e represent epithelial cells with Bollinger bodies in their cytoplasm. × 1200.
(Kligler, Mackensie, and Rivers: Fowl-pox.)
(Kligler, Muckenfuss, and Rivers: Fowl-pox.)