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MOSQUITOS AND MALARIA.

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MOSQUITOS AND MALARIA

A STUDY OF OUR KNOWLEDGE ON THE SUBJECT
AT THE BEGINNING OF THE YEAR
1900

WITH AN ACCOUNT OF THE

NATURAL HISTORY OF SOME MOSQUITOS

BY

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WITH FIVE PLATES OF ILLUSTRATIONS

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1900.

To

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DIRECTOR-IN-CHIEF,

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A TRIBUTE

OF RESPECT AND ADMIRATION FOR ONE WHO HAS, BY THE LABOUR OF YEARS, BEEN THE MEANS OF ROBBING TWO OF THE MOST DEADLY OF TROPICAL DISEASES OF HALF THEIR TERRORS, BY HIS DISCOVERY OF VACCINES FOR CHOLERA AND PLAGUE.

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MOSQUITOS AND MALARIA.

MOSQUITOS AND MALARIA.

CHAPTER I.

During the summer of 1899 it was reported that a syndicate had been formed to buy up part of the Italian Campagna. The Campagna is a large tract of country in Italy, some 1,400 square miles in extent, with the City of Rome occupying practically the centre. The part of this once fertile and populous district which the syndicate proposed to acquire lies in the triangular piece of land bounded by the river Tiber to the north and west, and the Aino to the south and east, and is now so malarial that the land is almost valueless, cultivation is reduced to a minimum, and the scanty population is sickly in the extreme.

Why had this tract of land suddenly become saleable? Because a most important discovery in the causation of malarial fever had been made.

This discovery was the fact that mosquitos were the active agents, and probably the only agents, in carrying the fever from man to man, a theory that had been suggested as far back as the time of Lancisi, a Roman physician, but until now has never been proved.

This was not all the discovery. It was found that only a particular sort of mosquito could carry the fever, and that this sort could easily be recognized in the larval state, and was said to breed only in certain pools and marshes. Hence it became evident that within the near future it might be possible to exterminate the fever-carrying mosquitos, and make a district that was previously uninhabitable quite healthy.

The whole subject is of such interest and importance to every one in India, that we propose to give a sketch of the steps which led up to this discovery, and the possibilities that it opens out before us.

The Malaria Parasite.

The cause of malaria is a microscopic amœboid parasite which lives inside the red corpuscles. It does not live free in the blood stream like some other blood parasites.

"This parasite, discovered by Dr. Laveran in 1880, is the "most important in tropical pathology; not only does it give "rise to grave and fatal fevers, but in consequence of its "debilitating influences, it undermines the health of millions, "and directly and indirectly it is the principal cause of death "in the tropics and sub-tropics"—(Manson).

Zoologically it is placed among the Sporozoa, and is closely allied to the Coccidia. Many other vertebrata are affected by similar, though specifically distinct, parasites, which have been shown, by the observations of R. Pfeiffer and of Simond, to possess a double cycle of life, one form adapted for the multiplication of the parasite in the original host, or the intra-corporeal, the other form adapted for its transmission through a fresh host, or the extra-corporeal part of the cycle.

There are several varieties of the sporozoon special to man. Each variety passes through certain definite and well known stages in its development in the blood corpuscle, and gives rise, during its intra-corporeal life, each to its own particular type of fever, quartan or three-day fever, tertian or two-day fever, and so on.

But to follow out its extra-corporeal life for long baffled the ablest and most careful investigators, until the abovementioned discovery, made recently in India, put the scientific world upon the right track.

The mature parasite occurs in two forms : (*a*) a crescent-shaped body with pigment, and (*b*) a more or less spherical pigmented body. Both these mature forms are intra-corpuscular, although in some cases it may appear otherwise, for the whole of the protoplasm has been assimilated and nothing remains visible but the limiting membrane. The crescent bodies, which are only discoverable after the fever has lasted some days, may, in spite of large and continued doses of quinine, remain in the blood for weeks after the attack without giving rise to fever or other symptoms. They degenerate and disappear without multiplying, and their significance until recently was a subject of controversy. They were considered as sterile forms.

On the other hand, the spherical forms having reached maturity and completed a process of segmentation, escape from the including corpuscle, which breaks down, and the enclosed segments or spores become free in the blood stream. A proportion of these spores attach themselves to other blood corpuscles, which, in some unknown way they contrive to enter, and thus give rise to another attack of fever. By the action of quinine these spores are killed and subsequent attacks prevented.

But if the quinine is withheld and a drop of the patient's blood containing the mature parasites be placed upon a slide and watched under the microscope, they will sometimes be seen to throw out long arms or filaments of protoplasm, which wave about energetically and are called "flagella."

Great importance was attached to these flagella. The fact that the flagellated body did not come into existence until the blood had left the vessels, and was outside the human body, suggested that its function also was outside the body ;

and because the flagella were formed from parasites of large size and apparently mature, it was thought, and as we shall see, rightly, that the flagella were flagellated spores, or in other words, the flagellated body constituted the first phase of the extra-corporeal life of the parasite.

Moreover, the fact that the parasite was, whilst in the circulation enclosed in a blood corpuscle, and therefore incapable of leaving the body by its own efforts, suggested that it might be removed from the circulation by some blood-sucking insect; and Dr. Patrick Manson, in his Goulstonian lectures in 1894, expounded the now famous theory, based also on certain recondite changes in the parasite, that this organism requires a suctorial insect for its further development.

Then it was discovered, that if a mosquito sucked blood containing these spherical bodies, they soon began to throw out flagella, these flagella subsequently breaking away and becoming free in the stomach of the insect.

This gave a fresh impetus to the investigation of the mystery, and the next step was the discovery by Bastianelli and Bignami* that the crescent-shaped bodies, if ingested by mosquitos, turned into spherical bodies, some of which remained as they were, whilst others threw out flagella which broke away, became endowed with locomotive powers, and penetrated those bodies which had not flagellated. So that the crescent bodies now had the significance of sexual forms. What more was required? The chain of evidence seemed complete, and warranted the following hypothesis, put forward by Dr. Manson, the highest authority on the subject at the time.

He says ("Tropical Diseases"): "I conjecture that the "flagellum enters some cell of the mosquito, and therein, like "any gregarine or coccidium, becomes parasitic, growing and "sporulating afterwards, just as the non-flagellated plasmodium

* Annali di Medicina Navale, 1898.

" enters, grows, and sporulates in the human blood corpuscle. " The plasmodium, I hold, is an intra-cellular parasite, both " outside as well as inside the human body, and that when " outside the human body it is parasitic in the mosquito.

" I have further conjectured that the continuation and mul- " tiplication of the plasmodium outside the human body is " secured by the passage of the mosquito-bred plasmodial spore " into the larvæ of the same insect. The mosquito generally " dies in water beside the eggs she has deposited. When the " eggs are hatched the young larvæ commonly devour the body " of their parent, and consequently her parasites. Moreover, " being very voracious, they eat any organic matter they come " across. We can easily understand from this in what way " parasites from the mosquito may get access to the mosquito's " offspring. On the infected larvæ becoming mature insects, " the plasmodia they have swallowed continue, I conjecture, to " develope. These insects, in their turn, infect their larvæ and " so on. Continuation and multiplication of the plasmodium " outside the human body is in this way secured. Man, " I conjecture, may become infected by drinking water " contaminated by mosquitos ; or, and much more frequently, " by inhaling the dust of the mud of dried up mosquito- " haunted pools ; or in some similar way."

This hypothesis was accepted, and for years little more was heard of mosquitos and malaria, every careful person living in a malarious country believing that the best way to keep clear of malarial fever was to avoid drinking bad water and to boil all that he did drink.

The reader may be reminded that it has long been recognised that many parasitic animals require for their complete development to live in two hosts. The common tape-worm, *Tænia Solium*, passes its earlier stage in swine, and then goes on, when the swine's flesh containing it is eaten, to its adult stage in human beings. *Trichina Spiralis* has a similar history.

There are many examples in the insect world also of insects acting as extra-corporeal, or definitive hosts : for example, the blood worm *Filaria*, the cause of Elephantiasis, has been proved to divide its life between man and mosquitos. According to Manson it is conveyed out of the blood system by the mosquito. It then passes directly from the stomach into the tissues of the insect, which is then supposed to lay its eggs and die on the surface of the water, liberating the young filaria which again gets access to man when he drinks the water.

We also know of two important diseases in cattle, namely, Texas cattle fever, and tsetse fly disease, which are inoculated, the former by the bite of a cattle tick, *Boöphilus Bovis*, the latter by the bite of the tsetse fly, *Glossina Morsitans.*

It will be noticed that in these two instances the disease is conveyed directly by the bite of the insect, and does not require the intervention of water as is supposed in the case of Filariasis, and as was till recently (*vide* Manson's hypothesis) in the case of malaria.

It is very probable that before long it will be discovered that the mosquito is also the active agent in conveying Filariasis from man to man, and that the intervention of water is not required to complete the life cycle of the *Filaria.*

CHAPTER II.

Major Ross's Discoveries.

In a report to the Director General of the Indian Medical Service, dated 19th September 1897, Surgeon-Major R. Ross, I. M. S., described some peculiar pigmented cells observed by him in August 1897, in the stomach wall of two mosquitos, fed upon a patient suffering from malaria, and whose blood contained crescents.

Believing, as he did, that this pigment was malarial pigment which, owing to certain characteristics, is not easily mistaken for other pigment, he at once anticipated that he had discovered a further stage in the life history of the parasite of malaria in these insects.

An abstract of the report was published (*British Medical Journal*, December 18th, 1897) to which was appended some remarks by Dr. Manson, Dr. Bland Sutton, and Dr. Thin on the original specimens, which Ross had sent them for examination.

While Dr. Manson thought that the cells might be the extra-corporeal phase of the parasite of malaria, he, on the other hand, considered it possible for them to be either normal cells of the mosquito, or some parasite in it quite independent of malaria, and he indicated to Major Ross a series of experiments that it would be advisable to carry out.

In September 1897, Ross saw the pigment cells again whilst making the following experiments:—

1. Out of a large number of mosquitos of many species examined unfed, none contained pigmented cells.

2. Out of a large number of mosquitos fed on healthy persons, none contained pigmented cells.

3. Out of a very large number of brindled mosquitos fed on patients with crescents, none contained pigmented cells.

4. Out of a large number of grey mosquitos fed on patients with crescents, none contained pigmented cells.

5. Out of four dappled-winged mosquitos fed on a case with crescents, pigmented cells were found in three.

6. Out of a considerable number of dappled-winged mosquitos fed on healthy persons, none contained pigmented cells.

7. One grey mosquito caught feeding on a case of mild tertian malarial fever, contained pigmented cells.

8. Out of 34 brown-brindled mosquitos fed on a case of triple quartan malarial fever, none contained pigmented cells.

These experiments pointed to the following conclusions, *viz.*, that the pigmented cells were not normal in the mosquito (1.): nor did any mosquito become infected after sucking healthy blood, (2.6.); that it was not all mosquitos that became infected after sucking malarial blood, (3.4.8.); but that only the dappled-winged and one grey mosquito became infected after sucking malarial blood, (5.7.)

The fresh light which these new observations threw upon the subject was deemed of such importance that in February 1898, the Government of India placed Major Ross on special duty to prosecute his researches, and gave him the use of a laboratory in Calcutta for the purpose.

February was not the malarious season of the year, and suitable cases of malaria were not easy to procure. Ross, therefore, decided to carry out his experiments with bird instead of human malaria, his idea being that, as the two diseases were practically identical, he might expect to find his pigment cells in those mosquitos fed on birds with bird-malaria, if his theory that these cells were an extra-corporeal phase of the parasite, was right; and that, if having found them, he was able to infect other birds, previously healthy, with bird-malaria, then his theory would be very much nearer to being proved.

Labbé*, Koch, and others have made an exhaustive study of the two commonest forms of malaria parasites in birds, one called the "proteosoma," found commonly in larks and sparrows; the other the "halteridium," chiefly in pigeons and crows.

In the adult stage the proteosoma can be distinguished from the halteridium by the fact that the latter does not alter the position of the nucleus of the red blood corpuscle, while the former pushes it to one side. (Koch). The red corpuscles in birds, it must not be forgotten, are nucleated.

For an account of the development of the bird-malaria parasites, the reader is referred to the work of Koch and Kossel, and also to the various papers of MacCallum in America.

A vast number of careful experiments were carried out in the Calcutta laboratory. They are described in detail in Ross's second report, dated May 21st, 1898. It is only necessary here to give a summary of his results.

Having learnt from his previous experiments (5-7) that in all probability only one or two species of mosquitos were capable of carrying human malaria, he conjectured that similarly only certain species would be able to carry bird-malaria. His next experiments were therefore directed to the discovery of the particular species. After many failures, with all the species of mosquitos available, he found that the common one, which he

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* Parasites endoglobulaires du San des Vertébrés, archives de Zoologie Expérimentale et Générale, 1894.

called the "grey mosquito" (species then unidentified), was the only one which contained the pigment cells after having been fed on birds whose blood contained that form of bird-malaria parasite which is called the proteosoma.

In order to prove this more conclusively, the following experiment was carried out :—

Out of 245 grey mosquitos fed on birds with proteosoma, 178 or 72 per cent. contained pigment cells.

A large number of experiments are detailed in Ross's report, requiring the dissection and examination of many hundreds of mosquitos. The following may be mentioned :—

Grey mosquitos were fed—

(*a*) On larks, sparrows, and a crow with mature proteosomata.

(*b*) On larks and a sparrow with immature proteosomata only.

(*c*) On crows, pigeons, and other birds with halteridia only.

(*d*) On a man with crescents.

(*e*) On a case of immature tertian parasites.

(*f*) On healthy sparrows.

Only the insects fed on group (*a*) contained pigment cells. To still further show the restricted association between proteosomata in the birds, and the pigment cells in the grey mosquito, the following experiment, one of many, may be mentioned.

Thirty mosquitos caught in the larval stage, all in the same drain and hatched out in the same breeding bottle, were fed as follows :—

(*a*) Ten on a sparrow whose blood contained numerous proteosomata. When examined, these ten contained 1,009 pigment cells, or an average of 101 each.

(*b*) Ten on a sparrow with a moderate number of proteosomata contained 292 pigment cells, or an average of 29 each.

(c) Ten on a sparrow with no proteosomata contained no pigment cells.

It is probable that the grey mosquito mentioned in his early experiment No. 7 had fed upon a bird infected with proteosoma or halteridium previous to its being fed upon the case of mild tertian.

Now as to the technique of these experiments. In his report Ross says: Mosquitos are obtained by catching the larvæ in the pots of water, drains or puddles in which they live. They are then placed in wide-mouthed glass vessels, covered with muslin, until they hatch out into the perfect insect.

The vessel is then opened under an ordinary mosquito net under which has also been placed the subject for experiment—a bird in a cage. In the morning the mosquitos, gorged with blood, are caught in test tubes, in which they are kept, and when wanted are killed by chloroform or tobacco smoke. The tubes should contain a few drops of water for the inmates to drink and lay their eggs in, and should be changed for clean tubes every morning.

Grey mosquitos fed at night remain gorged and asleep all next day and night. Towards the end of the second day the stomach begins to "clear," that is, the meal has been partly digested and partly evacuated, leaving the organ partially empty. The eggs are usually laid on the third day, after which the insect generally dies if it has not been fed again. To keep them alive for twelve days or so, they must be fed every other day.

A mosquito, Ross says, is dissected for examination of the stomach in this manner: A needle is passed through the thorax, the legs and wings are pulled off. Holding the needle in the left hand the insect is then lowered into a drop of water, salt solution, or weak formalin placed on a glass slide. The last two segments are partially separated and held down upon the slide by another needle. The body is then moved away in such a manner that, if done skilfully, the alimentary and generative organs will remain on the slide attached to the last two segments of the tail.

The alimentary canal consists of an œsophagus, stomach, and a short straight intestine leading to the anus, containing six heart-shaped glands near the rectum. Five long tubes, the malpighian tubes, open into the stomach at its junction with the intestine.

The stomach wall consists, beginning from the inside, firstly of several layers of large cells which contain nucleus and nucleolus; then a delicate homogeneous membrane, which Ross calls " the outer homogeneous coat "; then the muscular coat, consisting of bands of muscular fibres crossing each other at right angles and obliquely ; and lastly the ramifications of the air vessels externally.

For a further description of the anatomy, Ross's report must be consulted.

He then proceeds to describe the development of the pigmented cell. He says : " Taking an insect fed, we will say, during the previous night upon a healthy sparrow, we dissect it in the manner described above."

" The stomach full of blood, with the œsophagus, intestine, malpighian tubes and ovaries attached, lies before us on a glass slide."

" With the point of a needle we open the side of the organ, and the contained blood runs out freely into the water of dissection."

" If now a cover glass is applied, we can easily examine with the highest powers the stomach tissues or their contents, or if we wish we can wash away the latter by irrigation with water or weak stains, and study the tissues alone."

In the case of the mosquito fed on a healthy sparrow, we shall find that the contents consist of red and white corpuscles crowded together, and a little remaining serum densely coloured with exuded hœmoglobin. On examining the tissues we find no trace of the characteristic pigment of proteosoma, or halteridium.

Taking, however, an insect fed at the same time on blood containing mature proteosomata, we shall find in the contents, besides the condition of blood just described, not only traces of these parasites, but entire ones still enclosed in the corpuscles, or lying free in the yellow serum. In some of these the pigment is still oscillating.

On the second day the blood in the cavity of the stomach of the insect fed on healthy blood is still further disintegrated. The red corpuscles and leucocytes have, as a rule, disappeared, and a fairly homogeneous reddish-black mass of multitudes of granules with brownian movement remains, or the stomach may by this time have poured its contents into the intestines, which in its turn may have evacuated them, in which case the insect is ready to feed again.

But if the insect is one which has fed on blood containing mature proteosomata we shall still find some traces of these parasites in the granular mass of disintegrated blood in the stomach in the shape of (*a*) small clusters of pigment, (*b*) scattered pigment granules, or (*c*) pigment contained in a yellowish hyaline cell, the remains of the entire dead parasite.

In the tissues something abnormal is at once noted. The external muscular and homogeneous coats of the stomach are found to be studded with small clusters of pigment identical in appearance with the characteristic pigment of proteosoma. These clusters of pigment constitute the youngest form of the pigment cells, but their cellular outline has not yet become apparent. They lie invariably either in the homogeneous coat, or still further outwards between the strands of muscular fibre, in the manner with which we are familiar in the case of *Trichina* embryos. They have never as yet been found lying in or amongst the innermost layer of cells, or in the cavity of the stomach itself.

During the morning of the second day after feeding, when the pigment cells first appear, there is considerable variety amongst them. The earliest form is a mere cluster of about 20 grains of pigment; a little later these 20 grains are seen to be enclosed by a faint oval outline; in another and probably a later form still we find interspersed amongst the pigment distinct vacuoles and refractive granules. These varieties have three things in common, their pigment, their oval outline, and approximately their size. Often all the pigment cells in one mosquito belong to the same variety. As the day advances the cells become larger, the vacuoles and granules become more numerous, the pigment tends to collect round one or two of the larger vacuoles, and the outline of the cells becomes more distinct.

It will be remembered that cells similar to these were found on the second day in mosquitos fed exclusively on human blood containing crescents. (Experiment 5.)

On the third day, he says, the pigment cells are larger and their outline is rounder; they are now more uniform in appearance and belong distinctly to two varieties only, one containing granules and some large vacuoles, the other clear and hyaline with some faint vacuoles towards the centre. In the former the granules are more numerous and closely packed together than on the previous day, the vacuoles are larger, and the pigment, the particles of which are less than on the previous day, tends to accumulate round one or two of the larger vacuoles in both varieties.

In some mosquitos nearly all the cells belong either to one, or the other variety, but in others the two varieties are found side by side. As the third day advances the pigment tends to decrease in amount, and in some of the largest cells a few bright refractive points begin to appear, scattered through the granular plasma.

On the fourth day, if the insect has survived without re-feeding, the pigment cells will scarcely exceed in size those observed on the third day, but if it has been fed again they are much larger and the granular and hyaline varieties can still be clearly differentiated. The pigment may still oscillate within a small area, but is now greatly reduced in amount. In some the pigment has disappeared, while the bright refractive points referred to have increased in number and size. In the granular variety the number of granules and vacuoles increases with the growth. At this stage the cells protrude from the external wall of the stomach like warts on a finger, not into the cavity of the stomach, but into the general body-cavity, the cœlum.

If the mosquito has been re-fed on the original bird, on the second day after first feeding, or on one infected with mature proteosomata, a fresh generation of young pigment cells, similar to those observed on the second day, will now be seen.

From this point, then, the parasitic nature of these bodies became obvious, and Ross assumed that they were coccidia, and therefore henceforth called them proteosoma-coccidia.

On the fifth day, he says, the coccidia continue to grow and protrude into the cœlum, but the difference between the granular and the hyaline cells has become less marked. The pigment has disappeared, and each coccidium now consists of a capsule containing a more or less granular and vacuolated substance studded with numerous small bright points.

On the sixth day the coccidium may have reached a growth of 60 or even 70 μ, easily visible with a magnifying glass.

These changes can be hastened or retarded by raising or lowering the temperature. Instead of one week they can be prolonged to two weeks or more if necessary.

(Some excellent diagrams, showing the appearance of the coccidia at different stages, are given in the report.)

Beyond this point Ross observed no further growth in the coccidia, although he examined coccidium infected mosquitos kept alive till the twelfth day.

If mosquitos of this stage, he says, be dissected in plain water, the coccidia will be seen bursting in all directions, pouring out their contents slowly into the surrounding fluid, and leaving behind a collapsed and wrinkled capsule still attached to the stomach wall. In some of these collapsed capsules can be seen a number of black sausage-shaped bodies. He describes also a peculiar striated appearance which he noticed in many mature coccidia between the 6th and 12th days, the striations appearing to radiate from one or two of the bright points previously mentioned as appearing on the third day.

Further investigation, he says, is needed as to the significance of the striation, and of the black bodies, but he ventures the following hypothesis.

Since no further growth of the coccidia has been noticed, we may expect that they have become ripe for sporulation, and since they are in a closed cavity, namely, the body-cavity of the host, there appears to be no means by which they can escape from that host during its life, to undergo sporulation in external nature, just as there appeared to be no way in which the malarial parasite could escape from the human body for the same purpose. It would appear, therefore, that sporulation must occur either (*a*) in the living insect after twelve days or (*b*) sometime after the insect's death. The first would point to a completion of the life cycle by a direct infection of men and birds by the coccidium spores in the mosquito ; the second to a more circuitous infection, perhaps by a second generation living free in water, which would coincide with Manson's suggestion that malaria may be conveyed by drinking water which has been contaminated by mosquitos.

That the young forms of coccidia of the second day are directly derived from the ingested proteosomata is proved beyond doubt by the following facts :—

(1) That they contain the characteristic pigment of proteosoma.

(2) That they are to be found in a large percentage of grey mosquitos fed on blood containing proteosomata, and are never observed in the insect fed otherwise.

(3) That they occur always on the second day after the insect is fed on blood containing mature proteosomata, and only on the second day after such feedings, not before.

No sign of movement has been observed in the coccidia. The older coccidia have never been seen in grey mosquitos except when fed on birds infected with proteosoma.

Whether the coccidia are intracellular in their younger stages, that is, originally contained in a cell of the mosquito, remains doubtful, and this point will probably not be cleared up until the gap in the continuity of development of the coccidium from the flagellum, or from the mature parasite which has been penetrated by a flagellum soon after reaching the stomach of the mosquito, has been bridged over.*

In concluding the report, he says, the cultivability of proteosoma becomes then a fact of great practical importance, implying, as it does, the ultimate solution of the problem, which has so long puzzled investigators, of the life history of the parasites of malaria outside the human body.

It remains, however, to discover—

(*a.*) The sporulation of the proteosoma-coccidia and further stages.

(*b.*) The appropriate second hosts for the different species of malaria parasites.

We shall see presently how near he himself was to the discovery of (*a*).

* MacCallum—On the Flagellate forms of the Malarial Parasite, *Lancet*, November 13th, 1897.

CHAPTER III.

Further Discoveries by Ross.

In October 1898, Major Ross handed to the Director-General a third report, dealing with some further researches of the utmost importance on the development of proteosoma-coccidia.

He takes up the subject again at the point at which his former report left it, namely, the appearance in the mature proteosoma-coccidium after the seventh day of what he now considers to be reproductive elements, namely, (*a*) a large number of delicate thread-like bodies (the striations mentioned as radiating from certain refractile particles), and (*b*) a smaller number of large black sausage-shaped bodies, which he now considers to be spores.

The minute thread-like bodies, he says, taper to each extremity, and contain vacuoles, and chromatin granules, in their middle. They are closely packed in the capsule of the mother coccidium. No certain indications of movement in them have been noticed, but as they are invisible in water, and only became visible when stained, or when shrivelled by the salt of the solution in which they are examined, it may be doubted if they have been seen alive. This, probably, is the reason why they were mentioned in his previous report only as striations.

The large black spores are cylindrical, closed at the ends, and are straight, curved, sigmoid or variously twisted.

"I have never," he says, "observed both the thread-like elements and the black spores in the same coccidium."

Over-shadowed by the interest attached to the thread-like bodies, the large black spores seem to him to be of little importance.

On the eighth or ninth day, shortly after the coccidia have matured, and produced their reproductive elements, they burst *in situ* in the living insect, and pour those elements into the general cavity of the body. They are swept away by the so-called blood or circulating juices, and distributed throughout the tissues of the mosquito. If we kill a mosquito 10 days after it has been fed on a sparrow with proteosoma, we shall most certainly find large numbers of the thread-like bodies in the juices of the head and thorax, and also some of the black spores in almost all the muscular and connective tissues of the body.

While examining these thread-like elements in the blood of mosquitos, he says: "I observed that they were frequently to be found collected within the cells of some gland in the thorax, the nature of which was unknown to me at the time. This eventually proved to be the salivary or poison gland of the mosquito."

Closely similar to the well-known salivary gland of other insects, this organ is situated in the neck, or anterior part of the thorax of the mosquito, and consists of a number of lobes. Each lobe is made up of numerous large cells clustered round a central duct. The main effluent duct runs up the under-surface of the head, enters the base of the central unpaired stylet of the proboscis (Pl. III, fig. 3), traverses the whole length of it and opens at its extremity in such a manner that the secretion of the gland must be poured into the tissues in the manner of a hypodermic needle. The use of the secretion is probably to produce an influx of blood to the spot.

It is in this gland that the thread-like reproductive elements of the proteosoma-coccidium are found accumulated in large numbers, either floating singly in the cavity of the grape-like cells, or crowded together in them in hundreds. It is impossible to mistake their identity or, Ross thinks, to err in the interpretation of these observations. The existence of this gland affords the minute thread-like bodies, which clearly have the significance now of reproductive elements, the opportunity, so to speak, of returning to a warm-blooded host, where, if that host be a bird of an amenable species, they doubtless originate an infection of proteosoma, thus completing the life cycle of the parasite.

All that was now required to obtain complete proof of this theory was actually to infect healthy birds in the manner suggested. This was accomplished. Four sparrows and one weaver bird, whose blood on several examinations had been found to be entirely free from proteosoma, were subjected nightly to the bites of numerous grey mosquitos fed more than a week previously on a sparrow whose blood contained proteosomata. Ten days afterwards these birds were found to have become infected with swarms of the parasites. All of them died very soon, and the liver and spleen in each one were found to be profusely charged with the characteristic black pigment of proteosoma.

Out of 28 healthy sparrows experimented on in this way, 22 or 79 per cent. became infected in from five to eight days. In all the birds in which the parasites appeared after the experiment, the invasion presented such constant and unmistakable characters that there was absolutely no room for doubting that the infection was due to the mosquitos. And owing to the fact that the proteosoma, the halteridium and other malaria parasites of birds, so closely resemble the hæmamœba, or malaria parasite of man, there seemed no reason to doubt that man is infected in the same way, but it remained to be proved.

"The parasites of this group," says Koch, "show such remarkable analogies in the processes of their development that the laws which govern the development of one variety may, *mutatis mutandis*, safely be assumed to hold good in that of the others. For instance, if halteridium (of pigeons and crows) forms "vermiform bodies" at one stage of its growth, or proteosoma (of sparrows and larks) similar "thread-like bodies" at the same stage, it is tolerably certain that the human malaria parasite will be shown to do likewise."

CHAPTER IV.

Dr. C. W. Daniels' Report.

In scientific work, before any new observation is accredited, it must be confirmed and accepted by men of recognised scientific standing.

Therefore, in December 1898, the Royal Society, in conjunction with the Colonial Office, sent out Dr. C. W. Daniels, a member of the Malaria Committee of the Society, to Calcutta, for the purpose of confirming the results of Major Ross's experiments.

Dr. Daniels' report will be found in the Proceedings of the Royal Society, Vol. 64, and also in the *Journal of Tropical Medicine* for July 1899.

According to this report Dr. Daniels seems to have examined all Dr. Ross's preparations, to have carried out many of his experiments afresh, initiating other ones where there seemed a weak link in the chain of evidence, and to have confirmed the whole of his work in all its details.

The black tubular spore-like bodies (the sausage-shaped bodies of Ross) seem to have particularly arrested his attention.

He says: " Mention has been made of the differentiation of " the coccidia (previous to the formation of the germinal " threads) according to the appearance of their contents, into " clear and granular; the evolution of the latter into the " coccidia containing germinal threads can be traced day by " day. This differentiation was clearly visible in my series.

"In a minority of the coccidia, and in most infected mos-
"quitos, when the germinal threads are mature, certain black "tubular bodies are to be found in cysts with otherwise clear "contents. Most of the infected mosquitos contained coccidia "with these bodies, though in a few insects all the cysts "contained germinal threads only. In some cysts the black "spores were numerous and occupied the entire cyst; in other "cysts there were only a few.

"In most instances germinal threads were not found in the "black spore bearing cysts, but there were a few such cysts in "which it was doubtful whether germinal threads were present "or not, or whether the appearance arose from overlying "threads which had escaped from a neighbouring capsule.

"These black spores are very resistant. I have seen some "which have been kept in water for months by Ross, and which "had undergone no visible change. They withstand irrigation "with *liquor potassæ*.

"When the cysts are ruptured, the black spores are to "be found all over the body of the mosquito, but not included "in cells. They do not seem to accumulate in any particular "organ.

"The most plausible view of the nature of these black "spores seems to be that they are 'resting spores' and that "through them, by another cycle, the proteosoma can be "propagated in conditions unfavourable for direct propagation "by mosquito insertion into a warm-blooded animal.

"If this be the case three courses suggest themselves :—

"(*a*) From the black spores may arise bodies capable of non- "parasitic life, which at certain stages of their exis- "tence, and in certain conditions, on introduction into "a warm-blooded host by inhalation, through drinking "water, or even by injection by a mosquito or other "blood-sucker, in transferring them from the medium "in which they live, may resume parasitic habits.

" (b) That they may be ingested by mosquito larvæ, and in " them undergo such development as will result in " the formation of germinal threads in the adult mos- " quito, which, in turn, may be injected into the ap- " propriate bird.

" (c) That they may, if swallowed or inhaled by an appro- " priate warm-blooded host, so develope as to reach " the circulation and pass into the sporulating phase.

" Such experiments as have been made on this subject are " inconclusive and it is obvious that until the nature of these " black spores is determined we cannot exclude, even for " proteosoma of sparrows, the possibility of any one of the many " alternate channels of infection. Intervention of the mosquito " intermediate host may be only an occasional requirement.

" Still less are we justified in concluding that malaria in man " can only be acquired through, and directly from, the mos- " quito; or in devoting our attention exclusively to that " channel."

Dr. Daniels, with the cautiousness of a scientist, wished to draw attention to the fact that, although Major Ross's discoveries have put observers on the high road to proving that the mosquito conveys malarial fever from man to man, they do not prove that it is the only way that the fever can be carried.

Dr. Daniels wrote his report a year ago. Since then the fact that mosquitos can infect human beings with malaria seems to have been proved to the satisfaction of all authorities, but our knowledge of the function of the black spores remains where it stood at the time of Dr. Daniels' report, as far as we are aware, in spite of careful study by Koch and others.

For this reason, and also because they are so resistant to external influences, because they do not occur in every coccidium infected mosquito, because they occur in coccidia without the thread-like bodies and which are otherwise hyaline, because drinking water containing them has failed repeatedly to produce malaria, because all investigations have failed to discover them in the larvæ, or in anopheles, born from larvæ fed upon insects containing them, and for many other reasons deduced from our more complete knowledge of mosquitos, enabling us to explain almost all the many facts and beliefs of ages past in connection with malaria, it is now generally believed that they are some degenerative or abortive form of cell. Nevertheless it should be borne in mind that these black bodies still may turn out to be "rest-spores," or to have some function.

CHAPTER V.

THE WORK OF THE ITALIANS AND OTHERS.

Whilst Ross was working in Calcutta with proteosoma, the probable connection of mosquitos and malaria was being studied by the distinguished Italian malariologist Bignami. His first experiments in which a number of mosquitos of several species were brought from a malarious district and set free in a suitable room in Rome, in which a man, who had never previously had malaria, slept, were made in 1894. The result was negative, as also were the results of several similar experiments during the succeeding three years.

In the summer of 1898, Grassi, a well known Italian naturalist, made an exhaustive study of the mosquitos that prevailed in the different parts of Italy, and he found that in all the malarial zones, mosquitos belonging to a particular genus, called by zoologists *anopheles*, were to be found, and in every spot where they were found malaria was prevalent. This fact in conjunction with Ross's papers, showing that a special mosquito was needed as the host of proteosoma, at once suggested to Bignami, the course which he had to take.

The same man, who had voluntarily submitted himself for experiment before, and whose history Bignami publishes to show that he had never been exposed to the chances of malarial infection, again submitted himself to experiments. Guided by Grassi's information, certain mosquitos were brought from Maccarese, where fever prevailed, and were identified by Grassi as belonging to the genus *anopheles* and identical with the species which he had found in all the malarial zones. These were set free in a suitable room in Rome in which the man lived and slept.

Of course, although these mosquitos proved ultimately to be the human malarial bearing species, it could not be known whether they had previously fed on a person whose blood contained the mature parasite, but it was thought more than likely, for the neighbourhood was a very bad one.

For some time after the minimum period, the experiment did not succeed, but ultimately it was successful, and the subject of the experiment began to suffer from symptoms of fever. His blood was examined and the parasites of what is called summer-autumn fever, identical with the type of fever which prevailed at Maccarese, were found. Thus Bignami first succeeded in getting positive evidence of the direct inoculation of malaria by mosquito bites.

In the district of Rome in which the experiment was carried out, there had not been a case of malarial fever caught in that district within the memory of medical men, the centre of Rome being absolutely free from fever.

This result has been amply confirmed by further experiments by Bignami and Bastianelli, and it has now been conclusively shown that anopheles may be made to bite a man suffering from malarial fever, and if then kept at a proper temperature (30°C.) for the requisite number of days, to allow the sporozoites to be present in the veneno-salivary gland, and then made to bite a healthy man who is free from malarial infection, this man will, after the regular incubation period, develope malaria. And it is important to note that the parasite so conveyed from man to man retains its specific type. It further results from their experiments that a very few bites are sufficient to convey an attack of fever, probably a single one sufficing. What is important is the species of mosquito, and the stage of development of the parasite when it is ingested by the mosquito.

They also have shown that the development of the parasite of human malaria in the body of anopheles is indentical with the development of proteosoma in the " grey mosquito."

It seemed at one time quite possible that each of the main types of the human malaria parasite, each of which gives rise to a distinct clinical type of fever, might be found to require a particular species of anopheles to propagate it, but recent observations by the Italians partly disprove this. Ross also found that in certain barracks at Sierra Leone, three types of malarial fever were to be found among the troops, but only one species of anopheles could be found in the buildings. Hence it was inferred that this one species conveyed all three types, and as a matter of fact the zygotes (the coccidia, or pigment cells) of both quartan and tertian fever, were identified in the insects.

He says (*B. M. J.*, Sep. 99, p. 746) that the insects were caught engorged with blood, whilst resting on the walls, were kept two or three days, then examined, and the young zygotes of quartan and tertian fever could be recognised at once on inspection of their pigment; that of the quartan zygotes being coarse, of a dark-brown colour and comparatively scanty; that of tertian being very copious, fine and of a light-brown colour.

It was not possible to tell by inspection, from which species of parasite the zygoblasts (thread-like bodies) had been derived.

Kilburn and Smith have shown that the hæmatozoon which causes Texas fever of cattle is transmitted from the tick, which is its temporary extra-corporeal host to a second generation of ticks, by which it is again conveyed to cattle. The resemblance between the hæmatozoon of Texas fever and the human malaria parasite is so great, that the proof that the one was conveyed by an insect was considered almost a proof that human malaria must be similarly conveyed—even before it had been demonstrated that it is so, and rendered it necessary to investigate whether in anopheles also the parasite is transmitted from parent to offspring. The Italian observers, therefore, made a diligent examination of this question. They cultivated the larvæ of anopheles taken from near houses that were infected with malaria, and when the new generation of winged insects was developed, they allowed them to bite healthy people. These experiments have so far turned out negative.

At the hospital of Santo Spirito, a person who had never had malaria exposed himself voluntarily to the bites of new-born anopheles. He was bitten for 20 days by numerous insects of this species, which were developed in a room in the hospital. He nevertheless continued to enjoy good health.

In the laboratory of comparative anatomy, Professor Grassi and five persons belonging to the laboratory exposed themselves to the bites of numerous anopheles developed in the laboratory from the larvæ or nymphæ taken at many parts of the Campagna, from places near dwellings in which malaria had run riot. These persons were bitten daily. Not one of them became infected with malaria.

So far all these experiments have been negative, and up to the present time only those anopheles that had bitten human beings suffering from malaria have been shown to be capable of propagating the disease.

After an attack of malarial fever the parasites, usually in cases in which the treatment has been faulty, may retire from the general circulation and, it is believed, pass what is called their "latent phase" in the spleen, bone marrow and some other of the deeper organs of the body. They may remain quiescent for many months until some devitalising influence, such as a chill, or prolonged exposure to the sun, brings them once more into the peripheral circulation, where they sporulate afresh giving rise to a recurrence of the fever without the intervention of any mosquitos.

Malarial fevers are such a curse outside the walls of Rome, that the Italians have every possible reason for leaving no stone unturned which may possibly lead them to a solution of the difficult problem of how to lessen the scourge. Many remarkable and interesting experiments are detailed in the publications of Grassi, Celli, Bastianelli, Bignami, Casagrandi, Santori, and others during the past eighteen months.

From the foregoing further proof of the mosquito theory of malaria seems unnecessary here.

Professor A. Celli, of Rome, summarises the subject in the following concise way:—

"Man and mosquitos are without doubt the sources of "malarial infection which circulates, so to speak, from man to "mosquito and from mosquito to man, and so on. In this circu-"lation of the malarial contagion, the presence of malarial man "is indispensable, inasmuch as down to the present time the "hereditary transmission of malaria from mosquito to mosquito "has not been demonstrated experimentally, or morphologically, "nor have resisting parasitic forms been found in the environ-"ment external to the body of the mosquitos. Malaria is "therefore a typical contagious disease.

"Where there is malaria, mosquitos abound, but malaria "does not exist in every place where mosquitos abound, which "is explained by the fact that in malarious regions particular "species of mosquitos live. Those capable of lodging the "specific parasites and of infecting man, appertain to the genus "anopheles.

"All the four species (A. *claviger* or *maculipennis*, A. "*bifurcatus*, A. *superpictus*, and A. *pseudopictus*) of this genus "which exist in Italy are injurious. It is not probable that "species of mosquitos belonging to the more common genus "Culex, or other blood-sucking insects, which are found in "malarious places, can also transmit malaria.

"And since the specific germs external to man live not in "the soil, but in the body of the mosquito, it follows that "malaria can no longer be cited as a classic example of a "disease of the soil. The same may be said of water. It may "be, and in fact is, the habitat of the eggs, larvæ and nymphæ "of the mosquitos which eventually become malarigenous. "Therefore soil and water pass into the number of indirect "epidemic causes, or those which we term predisposing." (*B. M. J.*, February 10th, 1900).

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CHAPTER VI.

The Natural History of some Mosquitos.

We now come to the study of the mosquitos themselves.

For a description of how to collect them for the cabinet, and the apparatus which is necessary, those interested may refer to a pamphlet on the subject issued by the Natural History Department of the British Museum, South Kensington. It is also published in the *Journal of Tropical Medicine* for January 1899.

The collection and classification of the many different species of mosquitos, in various parts of the world, is by no means sufficient from a medical point of view, and too much attention, we think, is being paid to it to the exclusion of more useful investigations.

The mosquito, most of us know, undergoes four stages of development. The first stage, that of the eggs, is passed on the water ; the second, or larva stage, is passed in the water ; the third, the pupa or nympha stage, is also passed in the water ; and the fourth stage, that of the adult insect, is passed chiefly on the wing. Beyond this the natural history of mosquitos is, to a large extent, unknown, and our chief aim in publishing this work is to stimulate the interest and to enlist the energies of naturalists, medical men and others in India.

What is required is a more intimate knowledge of the life-history of the mosquito, and of our enemy Anopheles in particular, in all four of its stages. The distribution of the different species; when and where they lay their eggs; the conditions of temperature, &c., which modify the length of time of hatching of those eggs; the amount of cold or of dessication that they can withstand; the distinctive characters of the larvæ of each species; the food they each feed upon; the character of the localities in which they are found; the distance from human habitations; the number of broods in the year of each species; the duration of life of the mature insects; the time of day or night at which they feed,—are some of the many points which still require investigation.

Classification.

We have as yet no certain knowledge of the classification and identification of the Indian CULICIDÆ, but the European can be divided, according to Ficalbi, into three genera, based on the following characters:—

The first, *Anopheles* (from the Greek, meaning hurtful, strange to say) in both sexes, has the palpi about the same length as the proboscis (Pl. III.); the second, *Culex* (to which Ross's "Grey Mosquito" belongs), the palpi about the same length as the proboscis in the male only, in the female much shorter (Pl. I.); the third, *Aedes*, the palpi in both sexes much shorter than the proboscis.

It matters little what classification is used, for we are chiefly interested in the two genera Culex and Anopheles, seeing that these two genera comprise almost the whole of the family of Culicidæ. Culex is of world-wide distribution and Anopheles has to be differentiated from it.

The palpi, upon which entomologists have based the above classification, are organs growing from the front of the head and seem, in most cases, to have much the same function as the antennæ, or feelers.

Collectors and others will do well to provide themselves when it appears with a copy of the work, of Major G. M. Giles, I.M.S., a manual of over 200 species of mosquitos.

Genus Anopheles.

Fortunately Anopheles, as first pointed out by the members of the Sierra Leone Mosquito expedition (1899), can easily be recognised and distinguished at a glance from mosquitos belonging to the much more common genus Culex, by its attitude whilst resting on the wall (Pl. IV., fig. 1).

In this situation it looks, at a short distance, not unlike a thorn sticking in the wall, pointing upwards at an angle of 45° or 50°.

The proboscis, with the head, points in the same direction as the axis of the body. It is thick and noticeable, owing to the palpi being held close to it (Pl. III., figs. 1, 2). The legs are long and exceedingly slender, the body is slender, and the wings are small and light coloured, except for four conspicuous brown spots on their anterior margins (Pl. IV., figs. 2, 3, 5).

Its appearance has been likened to a miniature humming-bird moth.

It is a fragile mosquito, but very active and difficult to see on the wing, and can easily pass through the meshes of ordinary mosquito-netting. Add to this its alleged muteness and its painless bite, and it is easy to understand how it is that a careless observer may assert that a certain district, although known to be malarious, is free from mosquitos.

The different species vary a good deal in colour and markings. Some are of a light fawn colour, some a dark-greenish black ; some species have two spots on the wing, whilst others have three, and one at least has none.

From its position on the wall alone, native assistants can easily be taught to recognise Anopheles.

Genus Culex.

On the other hand, Culex, the common mosquito, rests upon the wall with its body either parallel to it, or pointing in towards it at the tail end, as it were (Pl. II., fig. 1). It has a distinctly hump-backed appearance. Its proboscis is slightly curved and carried at a considerable angle with the body; and its head is at a lower level than the thorax. They vary greatly in markings, some species having the legs and proboscis ringed black and white, in others they are plain. In some the wings are beautifully marked, but in the commoner species they are plain brown, grey, or greenish-black. They usually rest with the body well away from the wall, but after feeding may crouch flat against it. The hindermost pair of legs is generally to be seen curved up over its back. This is not so we observe with Anopheles, which rests stilt-like, unless disturbed, when it at once puts them out behind.

Ova of Anopheles.

If the debris, floating on the surface of the water, in which some species of Anopheles is breeding, be examined with a lens, the tiny black rod-shaped eggs will be seen, either single, attached to floating objects, or collected together in characteristic little patterns (Pl. IV., fig. 6). Often they are attached to some fixture at the edge of the water. They require about twenty-four hours or more to hatch out, the time varying according to the temperature.

Ova of Culex.

No lens is required to find the eggs of Culex, at least of the commoner species, for they are massed together vertically in black boatshaped collections of a hundred or more, each mass being an eighth of an inch, or even nearly a quarter of an inch in length (Pl. II., figs. 7, 8). They require about the same time as those of Anopheles to hatch out, all the eggs in one boat doing so nearly simultaneously.

Larvæ of Anopheles.

The larvæ of Culex and Anopheles also have some very obvious differences between them. The larva of Anopheles floats horizontally at the surface of the water like a tiny piece of stick (Pl. V., fig. 4). This position seems never to vary. If a specimen be placed in a drop of water upon a glass slide, and examined under the microscope, using the low power, or even a good hand-lens, it will be seen that the breathing tubes, or tracheæ, which run one on each side of the middle line, down the length of the body, and can be seen throbbing, end in two round apertures, in the last segment (Pl. V., figs. 2, 3). Hence the necessity for coming as near the surface of the water as possible to breathe.

We have found it convenient to add a little acid to the water on the slide to quiet the struggles of the specimen.

Looking down upon it in a tumbler, it will be noted that the head is black, and is narrower than the thorax (Pl. V., fig. 1). Looking at it from below the level of the water in the tumbler, it will be seen that the head is carried in the same plane as the body, just as the proboscis in the adult insect is carried. When at the edge of the tumbler it nearly always has its head towards the centre (Pl. IV., fig. 11), and if disturbed swims across the surface backwards with a series of vigorous jerks, until, if too much disturbed, it can be pushed about with a penholder as if it were dead.

As a rule the larvæ of Anopheles are always to be found at the surface. They feed chiefly at the surface, but if the puddle in which they are living is only a few inches deep, they can be watched slowly sinking one by one to the bottom to feed amongst the mud or stones, and then after a minute or less jerking themselves backwards up to the surface again by alternately curving the tail round to the head first on one side then on the other. Sometimes, depending possibly upon the clearness of the water, they will disappear instantly from the surface in a body when disturbed or when a dark shadow falls upon them, so quickly that it is difficult to catch them. In these cases they are likely to be mistaken for the larvæ of Culex.

Owing to their bodies being nearly transparent, they vary in colour according to the food upon which they are feeding. If it is algæ alone they are dark, but if the water is muddy and algæ scarce, they are very light coloured except the head which seems to be always black.

Larvæ of Culex.

The distinctive feature of the larva of Culex is that it hangs head downwards, from the surface of the water, breathing through a prolongation from its tail (Pl. V., fig. 8).

If examined on a glass slide, under the low power of the microscope, as before, it will be seen that the breathing tubes are prolonged from the surface of the body, near the tail, into a chamber of considerable length, in which they are dilated in a fussiform manner, and terminate at the end of it in small stigmatic orifices (Pl. V., figs. 13, 14). The dilatations contain a small amount of residual air and act as buoys. This prolongation from the tail, containing the ends of the breathing tubes, seems to be the most reliable characteristic of the larvæ of the many species in this genus.

The head of the larva of Culex is massive, and as broad or broader than the thoracic part of the body. It is bent slightly beneath the body just as the head is in the adult insect (Pl. V., figs. 11, 12).

It is very active in the water, and if disturbed riggles at once to the bottom, often remaining there for a considerable length of time if the disturbance is continued. When feeding it seems continually on the move, riggling from one place to another with a series of figure of eight riggles, very different to the motion of the larva of Anopheles. It will sometimes go to a depth of several feet. Looking down from above the larvæ of at least one species of Culex appear to be floating like Anopheles, flat upon the surface, but if put in a test tube, and examined from below the level of the water, the long breathing tube, near the tail, will at once be seen (Pl. V., figs. 9, 10).

The larvæ of the different species of Culex vary greatly in colour and habits. Some are very dark, some are light and some are of a beautiful transparent green.

The Nymphæ.

There is one stage in the life-history of both Anopheles and Culex during which they are almost identical in appearance. This is the nympha or pupa stage (Pl. II., fig. 10, Pl. IV., fig. 8,). Having reached its full size in from 8 to 20 days or more, after leaving the egg, the larva will be seen, if watched carefully, to become somewhat irritable, swimming fitfully about the tumbler in which it is kept. Then quite suddenly a slit appears in the back, and after some kicks and riggles the nymph emerges, leaving the shed epidermic cast, which retains the form of the larva to sink to the bottom of the water.

It is now comma-shaped and seems to have nothing in common with its former self. It is even more active than the larva, jerking itself down to the bottom of the vessel by its lobster-like tail, much in the same way as a shrimp moves, and then floating up to the surface again to breathe. Instead of breathing like the larva, through its tail, it now does so by means of two shining earlike projections from the sides of the thorax containing the ends of the tracheæ. An examination with a lens will reveal the cases for the antennæ, wings, legs, &c., of the adult insect.

The nymph remains in this state for one, two, or even three days or more if the temperature is low, becoming darker and darker in colour, then, whilst at the surface of the water, the thoracic part of the pupa case splits longitudinally, the new-born pest emerges, and is seen standing over it on the surface.

After an hour or so, under favourable conditions, its legs harden, its wings unfold, and, if not imprisoned, it flies away "to join its companions in their mazey dance," or if a female, to work its sweet will upon the nearest warm-blooded animal.

Habitat of Culex Larvæ.

The larvæ of Culex will be found thriving in warm countries, in almost every pot, tub, cistern, broken bottle, pond, or ditch, or in fact any place where a little water lodges. They are not particular about their diet, feeding voraciously on anything, but seem to prefer animal matter and sewage. They soon attack the body of a drowned mosquito, even before it is quite dead, and we have watched the older ones devour the younger and sickly ones on several occasions. Culex larvæ are the worst offenders in this respect. But if the water in a tumbler in which some Anopheles larvæ have been kept without food for some days be stirred up, the little black heads of the departed ones will appear at the surface as circumstantial evidence of the cannibalistic propensities of those remaining.

The writer recently discovered in a building in Bombay a number of fire-buckets each containing apparently sweet water, and in each of these were living scores of Culex larvæ in many stages of growth. What did they live upon? Mr. Hankine,* probably not knowing of their cannibal habits, has suggested that they live on bacteria and tend to purify the water, which may or may not be so.

Habitat of Anopheles Larvæ.

Anopheles is a puddle, or even a stream-breeding mosquito. It selects small, slow runnels containing algæ, or green floculent water-weed, or small puddles, or even stagnant collections of water containing green algæ. Never, as far as our experience goes, does it select sewage or foul water to breed in as Culex does. In sewage and foul water algæ do not grow. The larvæ of Anopheles feed voraciously on this green floculent water-weed, and seldom, if at all, on animal matter, and therefore what suits the algæ suits the larvæ. Places likely to be dried up or scoured out by rain will not suit them, nor places in which the water runs too fast or which contain fish.

does not breed in sewage or foul water.

feed on algae

* Bacteriologist to North-West Provinces Government.

Small pools or puddles on level ground, which cannot dry up, because they are fed from some small spring oozing out of higher ground, are said to be best suited for the growth of algæ and therefore also for the breeding of Anopheles.

The locality, we imagine, varies greatly according to the climate and temperature. Grassi has asserted (*B. M. J.*, Feb. 10, p. 323) that one of the most constant conditions which determines the presence of the larvæ of Anopheles is the existence on the water of Lemna (Duck weed). This does not agree with our experience in India. Very rarely have we found these larvæ in tanks in which the water is covered with Lemna. But our investigations on this point are not yet complete. Whether it is, as we suspect, that Lemna is, to a certain extent, antagonistic to the growth of the algæ, or whether it is that the larvæ have not sufficient surface space, owing to the incredible rapidity of growth of the Lemna, which covers large surfaces several times in the year, we have not determined.

Dr. S. P. James, I.M.S., writing from Quilon, says: "On "looking in the large collections of water which covered the "rice-fields round my bungalow, I was surprised to find "Anopheles larvæ in great numbers. In this part of Travan- "core they are common in the water of rice-fields, and also in "the pools of water which lie between the rows of earth on "fields where the native vegetables are planted in rows. Many "of these rice-fields contain minnows."

There are many instances on record where the formation of rice-fields has caused the re-appearance of malaria in places where it had become extinct.

General.

The sex of all mosquitos may at once be distinguished by their antennæ, which in the male are conspicuously feathery, or plumose, forming tufts in front of the head (Pl. I., figs. 1, 2).

In the female they are plain, with the exception of some fine hairs at the base of each joint (Pl. I., fig. 3). It is said that only the female Culex sings. This is a point upon which further study is needed. It is possible that all species are not alike in this respect. We have satisfied ourselves that both the male and female of at least one species of Culex does so. It is asserted that both sexes of Anopheles are silent, and that the bite of the female is less painful than that of Culex, people frequently not being aware of their presence, or of having been bitten by them.

The male and female of all mosquitos each possess a proboscis, but only the females bite and suck blood. In some species, for example, *Corethra*, the proboscis is so short that it is almost invisible to the naked eye. These gnats, we think, do not suck blood.

Mosquitos are fecundated on the wing shortly after feeding. On the second day after feeding the ovaries appear as two thick yellow masses loosely attached to the alimentary apparatus and to the terminal segments of the body (Pl. II., fig. 5). On the third day the eggs are usually laid and the insect frequently dies if it has not been fed again on the previous night.

Mosquitos are believed to return to the spot in which they were born to lay their eggs. Hence the places probably remain permanent breeding haunts.

According to Grassi, Anopheles bites only at sunset and sunrise. Ross says (*B. M. J.*, Sept. 30, 1900, p. 869), they bite at night, sleeping on the walls during the day-time, where they can be easily caught by placing a test tube or a bottle slowly over them. The tube is then stoppered with cotton-wool. In forty-eight hours after feeding, the stomach becomes empty, then the eggs are laid and the insect is ready to feed again. This can be effected by placing the mouth of the tube on the skin. Presently Anopheles "lays hold" and gorges itself even in the day-time. It even voids blood while sucking. Grassi corroborates this, and says that Culex, in this respect, is extremely refractory. He also says that blood is digested in Anopheles about twenty hours sooner than in Culex which, if a fact, is extraordinary and may be correlated in some obscure way with the development of the malaria parasite.

Dr. Bancroft says, that if mosquito larvæ (speaking probably of Culex) are fed partly on animal matter they grow faster and to a larger size.

We have several times seen the larvæ of Anopheles die within the hour when transferred from the tumbler in which they had lived to the tumbler in which Culex had, and was being hatched out in the water of a drain.

Our own method of observation is to keep the larvæ in a series of tumblers with fine mosquito netting over them, changing the water from time to time, or adding more from the tank, or puddle, from which they have been brought, and if possible, containing whatever they have been feeding upon. When they reach the nymph stage, they are transferred with a few drops of water to one of a series of inch, or inch and-a-quarter glass test-tubes kept in a stand. Each tube is labelled, and inside is placed a strip of rough white paper about the width of the tube for the mature insect to show itself upon when it emerges, as it does, in one, two, or perhaps three days, from the pupa case which remains floating at the surface. The top of the tube is plugged with cotton-wool. These pupa-cases often serve as tell-tale evidence of the existence of a breeding haunt.

The nymphæ of course do not require food, but to feed the mosquitos a section of ripe banana is placed each day, or every other day, in the mouth of the test tube instead of the cotton wool. Both males and females suck the juice of the fruit.

According to Professor Celli it would seem that Anopheles, during the warm season, is capable of producing five generations in one season, 40 days being the time occupied from *ovum* to *imago*.

It is not safe to observe mosquitos under ordinary mosquito netting, arranged above the tumblers in which they have developed, for the females, particularly of Anopheles, can pass through the meshes with the greatest ease wherever there is an angle, a crease or a corner in the netting for them to push their legs against. The males can also pass through, but the females are the most determined, and we have watched 50 or more pass through into an outer netting in the course of a morning. Where the net is stretched tight we have not seen them get through. The material known as "white leno" is said to be more serviceable.

Will mosquitos suck blood from a corpse? This is a question of some interest, and affords food for reflection upon the coagulation of blood, and upon the duration of life of blood parasites after the death of their host. On one occasion in Nigeria we proceeded to a hut in order to make a *post-mortem* examination upon the body of a white non-commissioned officer who had died from Black-water Fever three and-a-half hours previous to our arrival. On lifting the corner of a mackintosh sheet which had been laid over the body, we were surprised to see a cloud of mosquitos rise from beneath it. The sheet was replaced leaving as before the shoulder and neck slightly uncovered, and in a few minutes a number of large brown Anopheles were probing the corpse in their characteristic attitude.

Sucking corpse

How much desiccation can the eggs and the larvæ of Anopheles withstand? Ross says:—(*B. M. J.*, Oct. 14th, 1899.) "If the puddle dries up without complete desiccation of the "mud at the bottom, the larvæ survive and spring into activity "again. When, however, the rain holds off for 4 days, not "a single larva, large or small, can be detected when rain fills "up the puddle once more. In twenty-four hours, however, "a fresh crop of newly hatched larvæ may be found.

"Whether or no the eggs have withstood the desiccation, "or a fresh lot of eggs has been laid, has not been ascer-

"tained. Either of the following hypotheses would explain "the continuance of the species in a locality. (*a*) The eggs "can withstand prolonged desiccation, or (*b*) the adults can "hibernate throughout the dry season."

Bignami, Grassi and others have proved that mosquitos may live in captivity as long as 8 weeks, and there is little doubt that, if conditions require it, they can, in the tropics and sub-tropics, remain alive in dark corners of houses for long periods.

But what becomes of them in cold climates?

When in Norway it has always been a puzzle to us to find an explanation of how the millions of mosquitos, which are such a pest in most Norwegian valleys, and even worse at the Saëters high up in the hills, come there. We have found it impossible to fish in some of the valleys without a head-net and gloves.

Did the mosquitos themselves, or did the eggs, or the larvæ, or the pupæ, like the Lemmings survive the long winter months beneath the many feet of snow?

TABLE OF COMPARISON.

CULEX.	ANOPHELES.
Mosquito.	
1. Incapable of conveying malaria to man (?)	1. Conveys malaria to man.
2. Palpi in male about same length as proboscis. Sometimes longer and feathered (Pl. I., figs. 1. 2).	2. Palpi in male same length as proboscis and sometimes clubbed (Pl. III., fig. 1).
3 Palpi in female much shorter than proboscis.	3. Palpi in female same length as proboscis. Not clubbed.
4. Rests on wall with body parallel to, or pointing in towards it posteriorly.	4. Rests on the wall with body at angle of 45° from the wall.
5. Head below thorax, and looks hunch-backed.	5. Head, proboscis and body in same straight line, and looks like a thorn sticking in the wall.
6. Proboscis curved and carried at an angle with the body.	6. Proboscis nearly straight, and held in same direction as axis of body.
7. Sometimes rests with body flat against the wall.	7. Body always well away from the wall.
8. Grey, brown or greenish black.	8. Very light or dark brown.
9. Wings in most species not marked.	9. Wings in most species with 4 or 3 dark spots on the external (anterior) margins.
10. Sings (either one or both sexes).	10. Is mute (?)
11. Bite painful.	11. Bite painless (?)
Ova.	
1. Float free in black boat-shaped masses.	1. Float singly or in little patterns.

Culex.	Anopheles
Larva.	
1. Floats head downwards.	1. Floats horizontally at surface.
2. Breathing tubes open at extremity of a prolongation from the tail. (Pl. V, fig. 14).	2. Breathing tubes open near the tail. No prolongation. (Pl. V, fig. 13).
3. Wriggles to bottom of water when disturbed with figure of eight riggle.	3. Jerks backwards along surface of water, but may go instantly to bottom when disturbed.
4. Head carried slightly beneath the thorax as in mosquito.	4. Head carried in the same plane as the body as in mosquito.
5. Head same colour as body.	5. Head black.
Nympha.	
1. Generally larger than anopheles.	
Habitat of Larvæ.	
1. Any pot, tub, cistern, drain, ditch or swamp.	1. Puddles or ponds containing green algæ, &c.

The above table will not hold good in every case, but for primary purpose of comparison it will be found useful by those who have not yet made a study of the subject.

CHAPTER VII.

Methods of Destruction of Mosquitos.

Let us summarise what we have learnt so far : firstly, that certain mosquitos convey malaria from man to man ; secondly, that those mosquitos belong only, as far as we know at present, to the genus of Anopheles ; thirdly, that the larvæ of Anopheles all have certain peculiarities by which they can, with a little skill, be recognised at sight. This being so, it is clear that the next thing we must inquire into is, whether it is not possible to declare war against Anopheles, and endeavour to exterminate him, or at least to greatly lessen his numbers by attacking him in his larva stage, which evidently is his most vulnerable state.

It is well for Culex and other genera that we have as yet no indictment against them, and fortunate for us, for it would be almost an impossible task to deal, once a week, with every tub, pot, ditch, puddle, or well, which contained larvæ.

With the knowledge that the larva of Anopheles can be easily recognised and breeds in certain places containing algæ, and also if, as we suggest later on, its breeding places are proved to be always close to the haunts of man or to human habitations, then it would seem that our task is at least within the bounds of possibility.

Having discovered the breeding places, the first method which suggests itself is to fill them up with a few cart-loads of earth, If their area is large, then drainage would seem to be the only successful plan. Many examples of drainage and cultivation, having reduced malaria to a minimum, can be cited, for example, Lincolnshire in our country. On the other hand there are localities where malaria remains in spite of both cultivation and drainage. Drainage is of course expensive, and few towns in the tropics could afford to drain a large malarious area, and what is more, dry cultivation, although naturally the best against mosquitos, and consequently against malaria, is unfortunately not remunerative.

We must therefore consider drainage as a crude measure and not feasible in the majority of cases. Flooding, another crude measure, might be more successful in a few cases.

Larvicidal Substances.

If the spot cannot be filled in or drained, we next turn our attention to some larvicidal substance, or substance that will kill the larvæ in the water. They have to come up to the surface of the water to breathe. This being so, it does not require a great stretch of imagination to see that, if we can find a substance which will float evenly over the whole surface of the water, the imprisoned larvæ, being unable to breathe, will soon die. Such a substance is petroleum, or kerosene. A very small quantity of this will cover a considerable area, provided the surface is not influenced by wind or running water. Any other oily substance will act equally well, provided it is light and diffusible, but the disadvantage of them all is that they foul the water for drinking purposes, unless it is drawn from a tap below the level of the surface. Not less than one ounce of kerosine for each ten square feet of surface should be used.

Tar has been recommended as being efficacious, and in our opinion is the best of all the cheap larvicidal substances. If petroleum is used, it so soon evaporates that the place must be treated every three days or so. But if a small quantity of tar or an old tar-barrel is placed in the pond or puddle, the water remains for weeks coated with the oily products of the tar which kill the larvæ and prevent the mature insects from laying their eggs there.

According to Professor Celli and Dr. Casagrandi (*B. M. J.*, September 9, 1899, page 683), who have spent much time in the investigation of this matter :—" Among the mineral sub-
" stances which kill the larvæ potassium permanganate, even
" 5 per cent., acts slowly in clear water, and in marshy water
" it is practically inert, having to oxidise all the organic sub-
" stances. For the same reason its action is destroyed in
" sulphurous waters. Lime, copper, iron sulphates and ammonia,
" even in very large proportions act rather slowly. Sulphurous
" water is one of the most active substances ; corrosive sub-
" limate, as strong as 1 per cent., kills the larvæ slowly and
" does not kill the nymphæ.

" Among the vegetable substances potent poisons to the larvæ
" are the leaves of strong tobacco, and some of the insecticide
" powders of commerce, which are obtained from the unex-
" panded flowers of chrysanthemums. The commercial extract
" of tobacco, a saturated aqueous infusion of quassia, of solanum
" nigrum, or of daphne quidium, are also efficacious. Some
" of the aniline dyes are likewise efficacious.

" Of all the substances water saturated with sulphurous
" oxide, or potassium permanganate mixed with hydrochloric
" acid act the most rapidly."

" Saltwater, which at 5 to 10 per cent. kills the larvæ in
" 15 hours, in concentrated solutions, kills them in 30 minutes,
" and the nymphæ in an hour.

" Caustic lime, ammonia, and chloride and carbonate of lime
" only act in very high proportions. Of the more powerful
" bactericidal substances formalin and lysol are not very
" efficacious, and corrosive sublimate is even less so.

"Young larvæ are much more easily destroyed than the old. "With a rise of temperature the time for the destruction of the "larvæ and nymphæ by different substances is shortened. "The first to lose its larvicidal action is petroleum, which soon "evaporates and no longer covers the surface of the water. "It does not kill the fish or lower forms of life. The insecti-"cide powders are poisonous to some worms, molluscs and "fishes. The staining substances, if concentrated solutions "are used, destroy all animals in the water and their action "is of considerable duration."

Culicicidal Substances.

According to the same observers, the substances which kill the mosquito itself may be divided into odours, fumes and gases. Among the odours which caused death were turpentine, iodoform, menthol, nutmeg, camphor and garlic. Among fumes, tobacco, chrysanthemum flowers, fresh eucalyptus leaves, quassi wood, pyrethrum, and also simple wood smoke; among the gases the most practical and efficacious was sulphurous oxide. Mosquitos also detest the smell of many of the essential oils, of which the oil of lavender is said to be the best. Soaps made with the extracts of tobacco or turpentine are useful. The latter is said to prevent mosquitos from biting a person in the open air for one or two hours.

Three teaspoonsful of powdered cinneraria, mixed with a little nitre and ignited, is sufficient to kill all the mosquitos in an ordinary sized room. A sulphur candle is also of service.

A cyanide jar we find is useful for killing specimens for examination.

A better knowledge of the habits of mosquitos, that is, of the place and time of breeding, may possibly render their destruction easier.

M. Grellet (*B. M. J.* December 9, 1899) has recently communicated to the French Academy of Medicine some interesting observations on the influence exercised by lime on a malarious soil. He says that up to the year 1840 malaria was rife among the inhabitants of Châtillon—sur—Loing, after that date it disappeared completely. Between 1824 and 1840 lime was applied to the soil for purely agricultural purposes, and by the time the whole plateau had been thus treated malaria had disappeared. No difference had been made in the agricultural methods employed, nor drainage, nor sanitary works carried out. He mentions other instances of the sort. The value of a process which at once checks malaria and improves the soil is obvious.

Vast sums have been spent by nations and private individuals for preserving the vine from oidium and phylloxera, and as a recent estimate makes the number of people who die each year directly and indirectly from malarial fever in India alone as high as 500,000, it is to be hoped that something will be done to lessen the amount of malaria in this country.

CHAPTER VIII.

The Ætiology of Malaria.

Seasons.

Even with the small amount of knowledge we already possess of the life-history of Anopheles, and the many factors, such as temperature, rainfall, food, &c., which influence its reproduction, we begin to realise how it is that malaria has such a close connection with the seasons—so close that malarial fevers are named after the seasons.

Professor Celli says (*B. M. J.*, Feb. 10, 1900) that "in order to "follow the annual course in Italy of malaria, it is necessary "to distinguish carefully the primary forms from the recurrents "or relapses, and also the three principal forms of malarial "fevers—the spring and quartan, tertian, and summer tertian.

"It is then seen that the true malarial season—that is, the "season of primary infections—is the second six months of the "year. Its sequels, with the recurrent fevers are prolonged "through all the first six months of the following year, gradu-"ally declining from January to June. The fevers, therefore, "of the first half of the year are generally recurrents of the in-"fections contracted in the second half of the preceding year.

"By means of these recurrents the seed of contagion is "maintained. In the summer season it is transmitted by the "agency of mosquitos, and opens the subsequent epidemic "year.

"The epidemic year of malaria, therefore, extends from the "July of one year to the June of the following year. It is "remarkable that the quartan is the last to appear and also the "last to recur ; the mild tertian and the severe have a similar "but not identical annual course, the second in summer and "autumn notably prevailing over the first, hence the name "Æstivo-autumnal Tertian.

"This tertian is the first to cease recurring, and consequently "mild tertian and quartan prevail in the spring and have, "therefore, *a fortiori* been called spring fevers.

"The life of Anopheles is in Italy in direct and intimate "relation with the annual epidemic course of malarial fevers. "The new generations begin to bite in the second half "of June and in the first half of July. Towards the end of "June the first infected mosquitos are observed, and their "number increases in July and August."

The following table of Professor Celli's (*B. M. J.*, Feb. 10, p. 324) illustrates his meaning more clearly. The *italics* are ours :—

	Æstivo-autumnal, (*Severe Summer Tertian.*)	Benign Tertian.	Quartan (*Spring.*)
March 15th to June 30th ..	3	6	12
July...	10	3	0
August	26	7	1
September	18	3	1
October	10	2	3
November	2	1	2
December	0	0	0

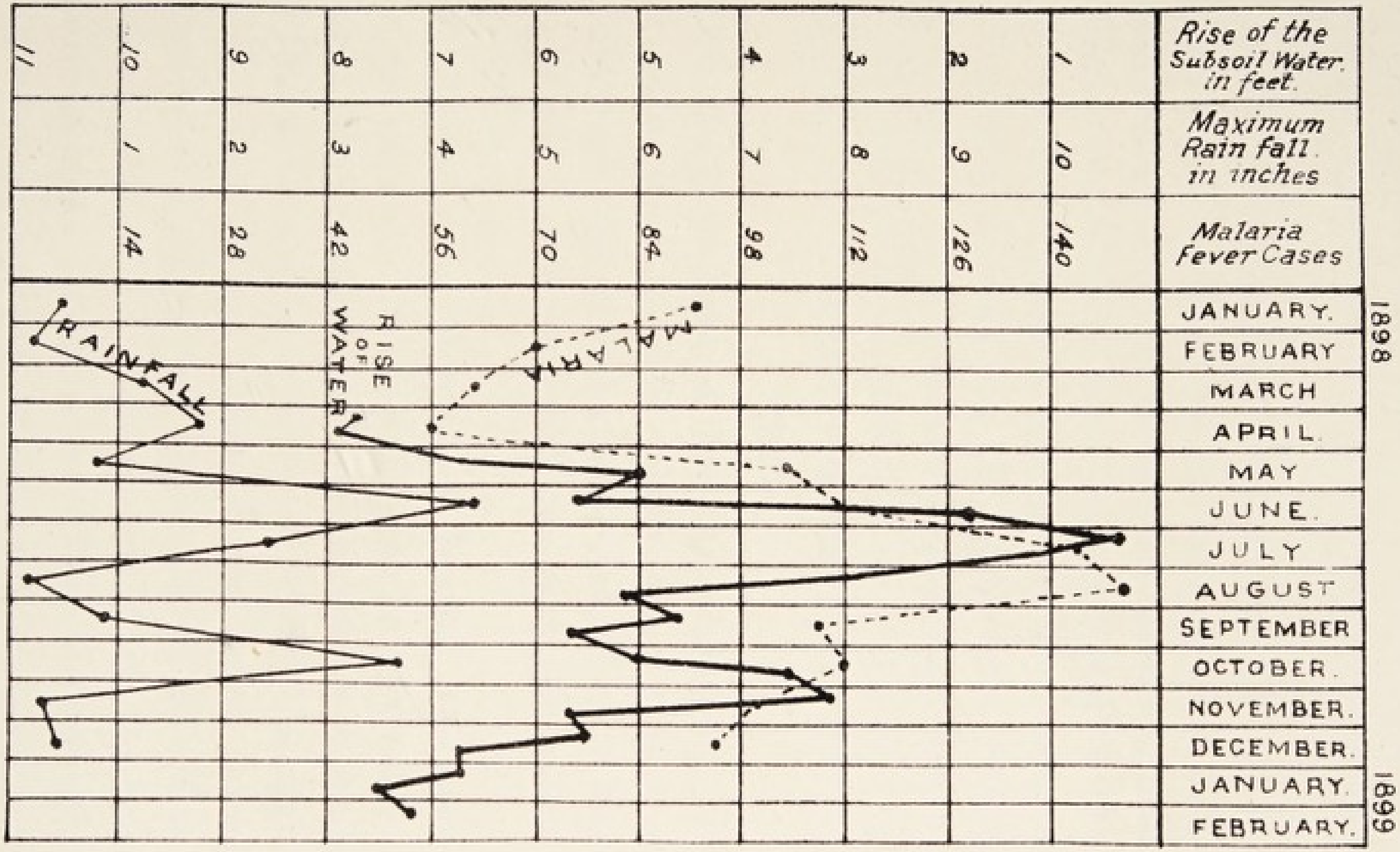
It will be seen that from July 1st onwards to the summer and autumn, the type of disease changed completely from what it had been from March to June.

At Grosseto, in Tuscany, the dangerous malarial season of first attacks is said to last 3 months only, beginning in the last week of June. Three weeks after the temperature has risen to 27° C, the sudden outbreak of malaria commences. Celli says: "Certainly the temperature in-"fluences the sudden appearance of the fevers in July, and their "prolongation in the autumn and sometimes to the beginning "of winter if the cold weather sets in late." (*B. M. J.*, Feb. 10, page 303).

Dr. Strachan, Chief Medical Officer at Lagos, West Africa, writes (*Journal of Tropical Medicine*, February 1900):—"I "have for many years noted that the greatest number of the "cases of malaria occur shortly after the cessation of the rains, "and but few during the rains, unless the latter had been in-"terrupted by short periods of dry hot days. It seemed, "therefore, to me that saturation of the soil with water, or, in "other words, rise of the sub-soil water, might eventually be "found to have a casual relationship to malarial fever.

"It is interesting to note on the accompanying chart, which "is the outcome of a series of observations in respect to the "curves of rainfall, rise of ground-water, and malaria at "Lagos, during 1899, that the relationship of the malarial "curve to that of the rainfall, confirms the opinion expressed "above, an opinion formed originally in Jamaica. It will be "seen that the height of the malarial curve follows very close-"ly that of the ground water, and at a distance of several days "the crest of the rain curve. Owing to the absence of suffi-"cient elevation above the sea-level, the ground water here "rises to the surface and remains there for some time. Keep-"ing in mind the mosquito theory, therefore, it is not difficult "to find an explanation in a place like Lagos, where a virulent "form of malaria is common and where Anopheles is found in "myriads."

Some medical men of large experience in the tropics have maintained that the greatest number of cases occur during the rains. This may be so, but is certainly not our experience in Africa with regard to first infections.



Rise of the Subsoil Water in feet.
Maximum Rain fall in inches
Malaria Fever Cases
1 10 140
2 9 126
3 8 112
4 7 98
5 6 84
6 5 70
7 4 56
8 3 42
9 2 28
10 1 14
11
1898
JANUARY.
FEBRUARY
MARCH
APRIL.
MAY
JUNE.
JULY
AUGUST
SEPTEMBER
OCTOBER.
NOVEMBER.
DECEMBER.
1899
JANUARY.
FEBRUARY.
MALARIA
RISE OF WATER
RAINFALL

A series of observations extending over at least one year, from several places in which malaria is prevalent, similar to those of Dr. Strachan's, would be of the greatest interest, particularly if in conjunction with observations on the hatching out of Anopheles.

It will be found that the height of the fever curve, that is, the time of year at which the greatest number of primary infections occur, is different in different countries, and in fact in different places in the same country; and it will be also found that these differences have a very close connection with the temperature, rainfall, &c. We know that the temperature, rainfall, &c., directly influences the hatching out of Anopheles, but it remains for us to prove that in all places the hatching out of Anopheles coincides with the fever curve.

SOIL.

It has often been reported that the *disturbance of soil* has given rise to an out-break of malarial fever, and it is a fact that, indirectly, the disturbance of soil, if on a large scale, may give rise to such an epidemic, particularly during the warm season.

Dr. Manson says (Manual of Tropical Diseases):—"The "Medical History of Hong Kong may be cited as an illustra-"tion. At the commencement of the occupation of this island by "the British, for a short time, it was healthy enough. Then "on its cession being completed, and when barracks and "houses were being built and roads laid out, it became excess-"ively unhealthy, the soldiers dying by the hundreds of "pernicious fevers. In time the sickness and mortality grad-"ually decreased, and now, so far as malaria is concerned, the "city of Victoria is healthy. But even at the present day "wherever in the outskirts, in the course of construction of "houses, roads, forts, and similar works, soil is turned up, fever "often, of a most pernicious type, is nearly sure to break out "among those engaged in the work."

Scores of other instances may be mentioned, and many theories have been advanced to explain them. But now a vivid picture of what no doubt happens in these cases rises up before us.

We have to begin with one essential of an epidemic, a large collection of men, say European soldiers, or coolies making a railway. They are housed in barracks or open sheds and no mosquito nets are provided. Mosquitos are plentiful, including Anopheles, the more so owing to the unusual supply of food, and possibly a pond or puddle close by not previously used as a breeding place, and one that has been excavated to obtain earth for an embankment is, from the same cause, swarming with larvæ.

If malaria is prevalent in the neighbourhood, the thing is simple, but, if not, one of the men has a recurrent attack, or a new comer has been bitten by an infected Anopheles on his journey, and at 4 p.m. on April 30th, we will say, he commences to pass through the stages of a typical attack. He is bitten repeatedly that afternoon and evening by both Culex and Anopheles, but Culex we know is not susceptible, and the Anopheles do not become infected, because the crop of malaria parasites in the corpuscles of that man's blood are not yet mature and therefore cannot flagellate.

At midnight a few mature forms might be found, and from then onwards through the morning (May 1st) any Anopheles which draws his blood becomes infected. What then happens?

On May 2nd or 3rd we find that the external wall of the stomach of that mosquito, if examined, is found to be studded with the earliest form of pigment cells (coccidia).

These go on developing until, on May 8th or 9th or later, according to the temperature, the adult coccidia (zygotes) burst, and pour into the cœlum of the insect their minute reproductive elements, the threadlike bodies (zygoblasts). A few hours afterwards they are to be found in most of the tissues, including the veneno-salivary gland.

The man has probably had a second rise of temperature which, we will suppose, was at the same time of day as before, on May 2nd. If the same Anopheles, which has probably been feeding every other night, happens to have bitten the man again on the morning of the 3rd, when his second crop of parasites is mature, then we should find, on examination, another crop of coccidia in its stomach wall, which would mature in their turn, and reach the salivary gland about May 11th or later.

In any case, on the night of May 9th and on every night, or on alternate nights, according as it fed, for some time afterwards, that Anopheles would infect any person whom it bit. If that person were restless, it might inject half a dozen doses of sporidia contained in the secretion of the salivary gland before sucking its fill.

On May 15th, 16th, or 17th, leaving 5 days as the minimum time for incubation, any of those men bitten would develop an attack of malaria, and each of those men, if bitten by Anopheles, when their parasites were mature, would initiate attacks in other men, and so on.

It is thus easy to understand how an epidemic is fostered. When we come to think that half a dozen Anopheles may have bitten the original man on May 1st, and the same ones and half-dozen more, on May 3rd, after his second rise of temperature, and that owing to bad treatment and want of quinine, he may have had several rises, being bitten each time, we can easily understand the widespread nature of that epidemic.

Bignami and Bastionelli (*Lancet*, January 13th, 1900, page 82) say :—" Wherever people congregated there also " Anopheles gathered. For instance, the harvesters slept in " the open until June 22nd, but on that date a large company " of them were housed in a big granary which served them as " a dormitory. On the 23rd, after a long search, a single " Anopheles was found in the granary ; on the morning of the " 24th eight were captured ; and on July 1st no less than 366."

They also (*B. M. J.*, same article) mention another instance where 42 corn-threshers, near Ostia, in the Campagna, commenced work on June 21st and continued till the beginning of August. Some of these men had had fever the previous year and were suffering from relapses when the work was begun. "Of the Anopheles collected in the huts of seven "of these people on July 1st, two out of seven were found to "be infected (with the coccidia of malaria); of those collected "on the 9th and 10th, two out of 17; of those captured on the "17th, 15 out of 32, or about 40 per cent.; and on July 20th "the number of infected Anopheles had risen to 11 in 17, or "about 64 per cent., some of them presenting ruptured capsules "and mature sporozoites. At the end of July and the beginning of August nearly all the men, who had slept in those "huts, were affected with Æstivo-autumnal or tertian fever.

"Evidently they had been infected by the mosquitos which "had invaded their huts and which had taken the parasites from those amongst the labourers who were suffering from relapses of fever. The progressively increasing number of Anopheles found infected, explains in "the clearest manner, it appears to us, the final outbreak of "the disease amongst nearly all the labourers."

It can now no longer be admitted that the simple stirring up of the soil in malarious places determines the liberation of germs of this infection, nor is it likely that breathing the dust of dried up marshes can have any effect in the production of malaria.

Water.

Hitherto it has been usual to attribute many attacks of malarial fever to the drinking of bad water. Now we know that the water of swamps and jungle-pools has much less danger in store for us than we imagined. Every one who has travelled in Africa, can call to mind occasions on which he has been obliged to drink foul, and sometimes stagnant swamp water, and the subsequent jubilation at escaping the expected attack of fever.

One Italian experimentalist has recorded the fact that for a long period he drank quantities of water sent to him from various malarial localities, some samples containing larvæ, while others contained the bodies of mosquitos, but he escaped with nothing worse than a mild dose of dysentery.

Nevertheless it cannot be said as yet that the possibility of infection by drinking water, by inhalation, or by food, has been absolutely disproved, but there seems to be no evidence whatever in favour of any of these channels.

Geographical.

"The geographical range of malaria is very great, it "extends in the Northern hemisphere from the Arctic Circle "to the Equator, and in the Southern probably as widely. "It is not uniformly distributed throughout this vast area, "but occurs in limited endemic foci which tend, speaking "generally, to be more numerous and larger as the Equator is "approached." (Manson.)

It now becomes important for every observer to help in mapping out the geographical distribution of Anopheles, in order to see whether it agrees in all cases with the range of malaria.

A study of the mosquitos of South America, for example, would be particularly interesting, for the reason that South of the Brazils malaria is, we believe, unknown or very rare. Nevertheless, this part of South America is supplied with mosquitos perhaps more generously than any other place in the world. On the Rivers Plate and Parana, mosquitos can be seen in clouds in the day-time as locusts are often seen.

Our diary tells a painful tale of woe occasioned by a trip we took some years ago along the banks of the Parana above the city of Rosario for the purposes of ornithology. For the greater part of the day we walked through miles of a tall umbelliferous plant which grew in profusion everywhere. From this plant as we advanced, rose clouds of small brown mosquitos which settled all over us, making a gorey mess of hands and face. Their bite seemed to cause very little pain or inconvenience at the time. Twelve hours afterwards, however, we were in bed with a temperature of 102° F. vowing vengeance on all mosquitos, with eyes closed by œdematous eyelids, and lips, fingers and ears double their normal size. A splitting headache and attacks of vomiting completed our discomfort. But no malaria followed. Before leaving the country we saw many other cases of similar acute poisoning from mosquito bites but no malarial fever.

Many of the South Pacific Islands are said to be entirely free from malaria.

Malaria is not contracted in the desert. "In the Sahara there is no malaria unless in the oases; in many of these it is rife, in Biskra for example." (Manson.)

From the natural history, as well as from the medical point of view, a study of the mosquitos of the West Indies would be of interest. In the island of San Domingo they are remarkable for their conspicuous and even brilliant markings. A very fatal form of malaria, called billious remittent fever, the *fevre billosa* of Spanish winters, is prevalent in several of the islands at certain times of the year; the native doctors continually mistake it for yellow fever.

We retain a vivid recollection of certain mosquitos in San Domingo. On the few drier and wooded parts of a big morass of many miles in extent, at the mouth of the river Yuna in that island, is to be found growing a good sized tree, belonging, possibly to the Magnolias. The natives call it the "pitch tree," because they produce from its fruit a black substance which they use as pitch. Beneath this tree nothing seemed to grow, the ground being covered with the large dead leaves of the tree. If we or the natives wandered carelessly into one of the open spaces thus formed beneath these trees, we were instantly attacked by the biggest, blackest and most vicious mosquitos it has ever been our lot to encounter. We had to make an immediate retreat, in fact to rush through the thickest undergrowth that could be found, or the attacking party would follow us like a little swarm of hornets.

There are said to be several non-malarious areas in India; for example, Jasalmir State. It will be interesting to hear whether Anopheles is to be found in this State.

Altitude.

Mountainous districts are usually free from malaria, because if there is any slope, the water-courses and puddles are scoured out by the rain, often to the bare rocks, and what water remains soon drains away. In such places algæ, the chief food of the malarial mosquito, cannot grow, and therefore in them Anopheles cannot re-produce its species.

Altitude has no influence whatever upon the production of malaria, except in lowering the temperature below that required for the hatching of mosquitos, and for the development of the coccidia. The temperature in a narrow, ill-ventilated, imperfectly drained valley, at an elevation of several thousand feet, may be so high that it becomes just as malarial as places in the plains below.

Swamps and Morasses.

Why is malaria always contracted near water? Because mosquitos require water for the reproduction of their species.

Why is it associated with swamps and morasses? Because these are almost a necessity for pools containing algæ.

This brings us to some interesting points upon which further knowledge is needed. The Sierra Leone Expedition, last year, reported (*B. M. J.*, October 14th, 1899, page 1033) that "in not a single instance have the breeding pools of Anopheles been found far from human habitations. The females must pass frequently between the pools in which they lay their eggs and the houses where they obtain their food. It must, therefore, be convenient for the one to be as near the other as possible. If we find Anopheles in a house we may assume that the breeding place is close by."

Morasses and swamps, usually swarm with mosquitos, but we believe that in most, if not in all cases, they will be found to be Culex, the females of which suck the blood of cattle which are herded on the few dry spots, wild animals, birds, and man if he is present.

Where do these mosquitos breed? It is believed that, owing to the danger incurred from small fish, they cannot live in the water of big morasses, but it is a fact that they can, for at certain times of the year, when the river, with which these big morasses is generally connected, is low, the water percolates so slowly through many miles of the densely packed rushes that grow in these situations that in parts it becomes actually stagnant, and there mosquitos breed in abundance. Through these reeds fish probably cannot penetrate. We have several times identified the larvæ of Culex in such situations on the upper waters, of the River Niger, whilst being forced through the rushes in a canoe, many miles from any human habitation. But we have never identified the larvæ of Anopheles in similar situations. They are less conspicuous than those of Culex and may have been overlooked. Anopheles, it will be found, is the domestic mosquito, breeding only near human habitations.

If the morass is uninhabited, then, in all probability, Anopheles will not be found in it, if later it becomes inhabited Anopheles will soon appear, and its breeding spots will be found to be within a hundred or two hundred yards of the habitations.

Sarrasin, in his recent interesting work on the Celebes, states that malaria is not contracted in the jungles, but only near villages. Our own experience in Africa is similar. A country in which malaria is prevalent, but in which Anopheles does not occur has yet to be found. If that country is found, then the statement that Anopheles is the only genus of mosquitoes which can convey the fever, is disproved. And a country in which Anopheles occurs but in which there is no malarial fever has yet to be found.

The oft repeated travellers' tales about fever in uninhabited countries have still to be substantiated.

If a primary attack was proved to have been contracted more than 8 days after entering an absolutely uninhabited country, it would clearly point to the fact that monkeys, apes, or some other animal inhabiting the country, was capable of suffering from human malaria, and we should assume that it had been conveyed to the patient by Anopheles.

Inoculation.

Koch has shown (*B. M. J.*, Feb. 10, 1900, page 326) by experiment that the higher apes cannot be infected with human malaria by injections of blood drawn from patients suffering from malarial fever, but he has not shown that no wild animals can suffer from human malaria conveyed by the agency of mosquitos.

It is doubtful, judging from our knowledge of the double cycle of life of the Sporozoa, whether man or animals can be infected with malaria by injections of malarial blood. It is possible that the injection of a large quantity of blood containing active sporulating parasites might produce an attack. The results of experiments, which we are unfortunately not able to quote, seem to show that it can.

Inasmuch as our knowledge of the form which the parasite takes during its periods of latency is as yet limited, it might be argued that, if some form of the parasite can return after several months to the peripheral circulation and re-sporulate, is it not possible that the crescent bodies or blood from the spleen or liver of a malarial patient, if injected into the blood of an animal or human being, could produce an attack? Further experiment only will show.

Winds.

No relation can be shown between winds and malaria, on the contrary they tend to clear the air. Mosquitos cannot fly in a wind.

Nor can any relation be shown between mists or fogs and malaria, except that they are associated with what we ambiguously call chills, often the immediate cause of a recurrent attack. The once dreaded miasmas of marshes and mangrove swamps have now few fears for us.

A puddle of water under a bedroom window may be infinitely more dangerous than a square mile of swamp or rice-fields a mile away. (Ross).

" It is certain, that 1,000 or 1,500 yards of water between a " ship and a malarious coast will usually suffice to secure im-" munity to the crew. A similar distance from a malarious " source is probably quite as effective. One village may " be sickly, whilst a neighbouring one may be healthy. " (Manson).

Ship Malaria.

Several instances are on record of outbreaks of what was reputed to be malaria on ship-board on the open sea. As it is not likely that the disease can be conveyed in drinking water we now refuse to accept the diagnosis of malaria.

But it may be quite possible that Anopheles, having once gained access to a dark and ill-ventilated cabin, may remain there a considerable period after the ship has put to sea, and if a case of malaria occurs in that cabin, it is conceivable that the insect, having fed upon it, might convey the infection to others.

IMMUNITY.

For the acquirement of artificial immunity, most is at present to be hoped from methylene blue (Celli, *B. M. J.*, February 10th, 1900, p. 305).

Koch in his second report (*B. M. J.*, February 10th, 1900, p. 325) to the German Colonial Office, on the work of the Malaria Expedition to the Dutch Indies, conducted under his direction, mentions some interesting discoveries with regard to the subject of immunity.

It is well-known that malarial attacks are more severe, more common, and much more dangerous in young children than in adults, and Koch's experience in East Africa strengthened his knowledge of this fact. Therefore, in order to judge of the peculiar malarial conditions of certain regions, he examined a large number of children in places which appeared especially favourable for malaria.

In the first village he found the adults apparently healthy, and they stated that they suffered little or not at all from fever ; but out of 86 children whose blood was examined, parasites were found in 8 (9·2 per cent.). Of the children under one year 16 per cent., and of those above that age 4 per cent., were malarial.

In the next place, out of 141 children examined, 18 (12 per cent.) were malarial. Of those under one year 15·5 per cent., and of those over one year 7 per cent. were malarial.

In a third place, 1,000 metres above the sea-level, 189 were examined, of these 43 (22·8 per cent.) were found with malaria parasites. Of those under one year 41 per cent., and above one year 14·6 per cent. were malarial.

"The rapid diminution in the frequency of malaria with "increasing age can be explained," he thinks, "only in one way, "that the men in these regions, where they are constantly "exposed to the infection, go through it in early youth, and "those who do not succumb to it acquire a greater or less "complete immunity." It has often been noticed in Africa and other malarious countries, that those soldiers or recruits who are drawn from Europe or non-malarious districts hardly ever escape infection, and suffer more severely than those from a malarious district, and that the latter show a certain power of resistance.

Koch thinks that these observations also "afford a plausible "explanation of the fact that children of Europeans in the "tropics, where malaria exists more or less everywhere, thrive "so ill; namely, that in regard to malaria they are obviously in "a much more disadvantageous position than native children "born of immune parents; when the latter become infected "with malaria, they have it in a milder form than the highly "susceptible European children. Finally the prophylaxis of "malaria, as far as it has to do with diseased men, may derive "the greatest advantage from this knowledge."

CHAPTER IX.

The Prophylaxis of Malaria.

It remains to point out the new lines on which the prophylaxis, or prevention of malarial fever, should be carried out. Our watchwords are: Sanitation; Quinine; Isolation; Education.

Sanitation.

Space does not permit of a full description of the various means of hydraulic sanitation, but to those who have read the foregoing pages closely, it will be evident that the sanitation for the prevention of this particular disease, becomes easy in proportion as our knowledge of the biology of the hurtful species of mosquitos advances.

In all malarious districts it is necessary to remove stagnant water, and to drain or fill up useless ponds and ditches in order that the larvæ may not be bred in them. Water-taps which, with the exercise of a little ingenuity on the part of the people, can be left running should be avoided. All systems with surface and open channels will be efficacious in proportion to the velocity of the water. Levelling the ground, or in towns paving it, so that the sub-soil water does not appear at the surface, is also most important. Paving

The planting of trees is injurious, as likely to harbour mosquitos, but in low situations, which are in danger of the sub-soil water coming to the surface, palms or quick-growing trees act beneficially in subtracting a large quantity of water and drying up the soil. Trees.

A house, camp or town will be healthy in a malarious district if it be on an elevation where the insects from the plains or a neighbouring marsh do not reach, and where there is sufficient wind or free current of air through all the buildings, rooms or tents. A house in a lower situation should be raised upon supports, the area of windows and doors should be large, the walls should be of plain deal or other smooth substance, and superfluous curtains, ornaments, and dark corners should be avoided.

In our experience a tent of the I. P. pattern has the great disadvantage that the wind cannot circulate freely through the upper parts of it, and the flaps for the "kanâts," are exceedingly well fitted for harbouring numbers of mosquitos through the day-time.

Quinine.

It is true that an ideal prophylaxis would be attained by removing the conditions which are necessary to the development of Anopheles in malarious areas. But seeing that malaria is now proved to be a contagious, and in fact an infectious disease, by the agency of certain mosquitos, "the careful treatment of the malarial patient constitutes one of the principal tasks of social hygiene."

Quinine, as is well-known, is the specific disinfectant for amœboid organisms, and consequently for the malarial parasites in their amœboid stage. Unfortunately, however, quinine does not act upon the mature forms of the parasite, which are, strictly speaking, not amœboid, the crescents for example.

It is, therefore, important that the quinine should be given early in order to prevent the parasites from reaching maturity; if they are allowed to do so, they become capable of remaining not only in the deeper organs, but in the peripheral circulation for weeks.

Isolation.

Hence it is at once apparent how important isolation becomes. A patient, in this condition, is a source of infection, and therefore a source of danger to others, and should at once be removed to a place where there are no malarial mosquitos, or isolated in a properly arranged mosquito net, or a room protected by mosquito netting.

It is needless to point out that an impervious mosquito net is the surest and safest preventive, and the best and readiest means of isolation. Numberless instances can be mentioned of persons living in notoriously malarious places having remained comparatively free from fever owing to the careful use of mosquito netting under all possible circumstances.

Professor Celli says all the employés of the Tivoli Railway have been personally examined by him, and all those who have been protected by mosquito nets have enjoyed an absolute immunity from malarial disease, those who have not been so protected have all suffered.

An experiment of Professor Grassi's is interesting in this connection. He obtained the use of two rooms in a cottage at Maccarese, one of the most malarious villages in the Campagna. The windows were covered with perforated zinc, and for eight days he left Rome every evening at 5-30, accompanied by a family from the healthiest district of Rome, consisting of father, mother, and 5 children, varying from 1 to 9 years of age, for the above rooms where they passed the night. The windows were left open, but the entrance of mosquitos was prevented by the zinc. They breathed the night air of this intensely malarious spot and drank the water that they found there. Professor Grassi took care that none of the family left the rooms between sunset and sunrise and at about 8 A. M. every morning, he and the family returned to Rome. Twelve days after they had permanently returned to the city, not one of them had been attacked with malaria.

Professor Grassi's results have been so encouraging that the Adriatic Railway, with 6,417 employés, has decided to enforce a rigid mosquito prophylaxis, including mosquito net protection, fumigation of houses, and the prophylactic use of quinine and methylene-blue. Other railways and commercial bodies in Italy are following suit.

Education.

The scattered literature of malaria consists of an amazing collection of coincidences. Laveran's discovery of the parasite at first only tended to enlarge the collection till the flow of hypothesis and conjecture, turned into the right channel by Manson, was changed for fact by the discoveries of Ross.

"Hitherto our efforts to obtain a more scientific frontier of "defence against this disease, have always been defeated by "our ignorance as to the exact manner in which the infection "is produced." (Celli.)

We are now no longer in ignorance, and it behoves medical men in India, not only to take an interest in the subject themselves, but to get others to do so, that the knowledge gained may be the sooner turned to practical use.

The author regrets, that owing to his having been transferred to other duties, he has not been able to treat the ætiology of malaria, from a mosquito stand-point, as fully as he intended; but he nevertheless ventures to hope that the work as it stands, the materials for which have been gathered under some difficulties during the intervals between his official duties, may be of service to many who are interested in "Mosquitos and Malaria," but who have not the time or the opportunities for referring to the many publications, both British and Italian, on the subject.

APPENDIX.

I have the permission of Mr. Haffkine to say that any specimens of mosquitos, larvæ, etc., sent for examination to the Plague Research Laboratory, Bombay, and marked 'Mosquitos,' will be attended to, in the event of my absence, by the staff of the Laboratory ; and that the receipt of all specimens, notes of occurrences, or other information bearing upon the subject, will be duly registered in a book kept for the purpose. The record compiled from this book will be ultimately published.

It is hoped that observers in all parts will contribute to this register ; and one of the first things to be aimed at will be the construction of a map showing the distribution of the fever-carrying mosquitos in India.

Mosquitos can be conveniently sent for identification in small boxes or bottles, or compressed between folds of dry blotting-paper. Cotton wool should not be used. The larvæ should be sent in a 2½ per cent. solution of formalin, but glycerine and water, boric-acid solution, or weak alcohol will do nearly equally well, or they may be sent alive in a corked tube containing some of the water in which they have lived if sufficient air is allowed them.

Small glass tubes, 1½ inches long, fitted with corks, and carried in a small box, tin-case or larger tube, are most useful for carrying home specimens or sending them by post.

The subject of "Mosquitos and Malaria" when more widely understood, must become of the greatest public importance in this country, as it now is in Italy, and soon will be in all our Colonies.

There are many points yet which remain doubtful and which require further study; and before any really useful rules can be formulated for the destruction of the various fever-carrying species, the details of their biology must be much more fully worked out than they are at present.

If, by this publication, I have been the means of interesting others in the work and thereby adding to our knowledge of those details, I shall have been amply repaid for the undertaking.

C. CHRISTY.

Bombay, April, 1900.

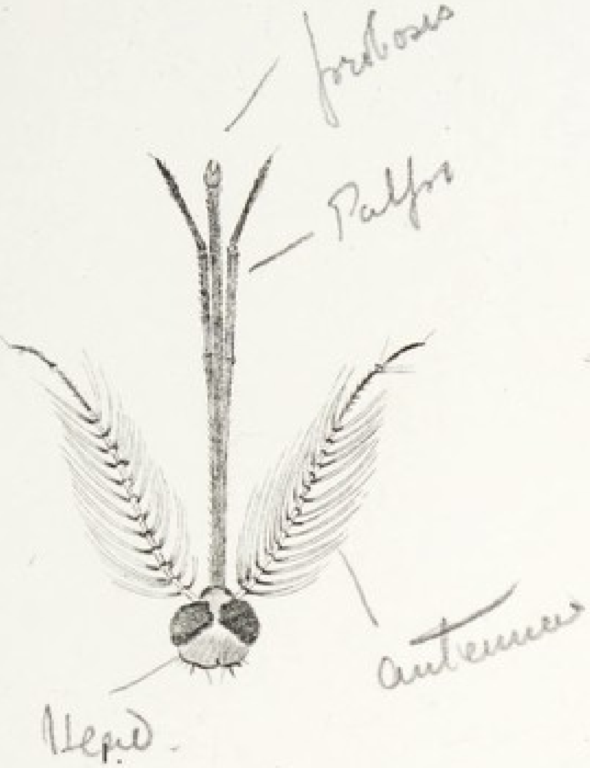
EXPLANATION OF PLATE I.

Fig. 1.—Head of male culex magnified. Showing palpi longer than the proboscis. Position of parts as in life.

Fig. 2.—Head of male culex magnified. Showing palpi longer than the proboscis and feathered like the antennæ.

Fig. 3.—Head of female culex magnified. Showing the very short palpi.

PLATE I (*Culex*)



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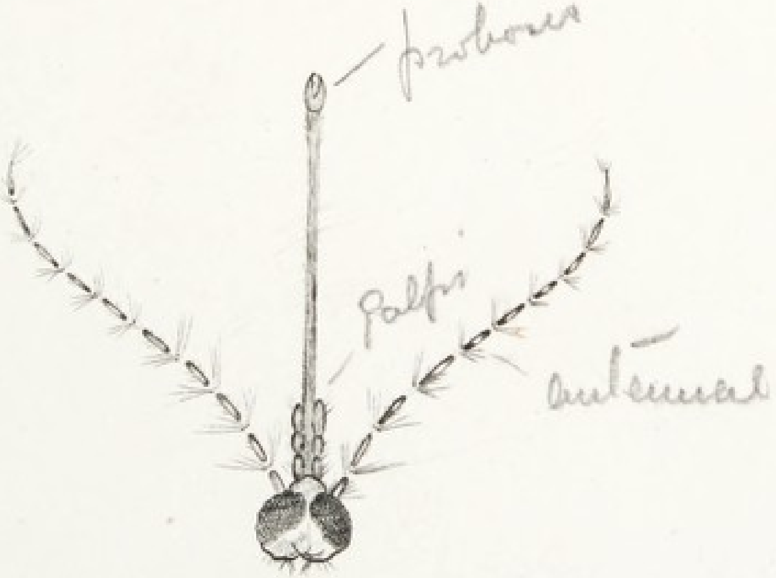


3.

L.N.TASKAR. *del.*

EXPLANATION OF PLATE II.

Fig. 1.—Attitudes assumed by culex when at rest on the wall. Natural size. Note posterior pair of legs extended above the body.

a. c. d. e. f. g. h. females. *b.* male.

Fig. 2.—Female culex. Natural size.

Fig. 3.—Male culex. Natural size.

Fig. 4.—Female culex, enlarged.

Fig. 5.—Female culex with eggs fully developed.

Fig. 6.—Male culex with palpi and antennæ feathered.

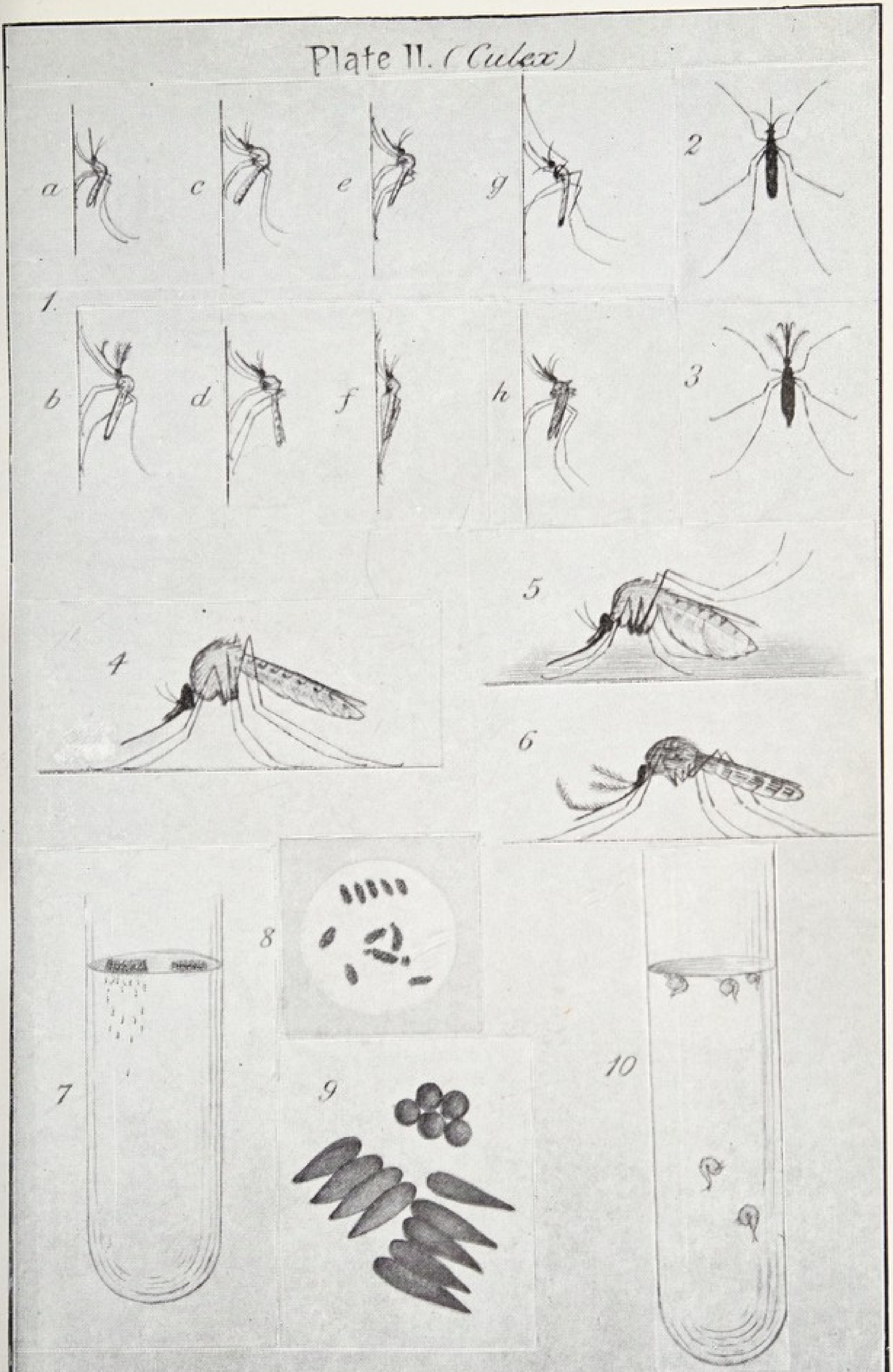
Fig. 7.—Eggs of culex in black boat-shaped masses. Natural size.

The left hand figure in the test-tube shows the larvæ just hatching out into the water.

Fig. 8.—Egg-boats seen from above. Natural size.

Fig. 9.—Eggs, enlarged. Side and end views.

Fig. 10.—The nymphæ. Natural size.



Govt Photozinco. Office, Poona 1900.

EXPLANATION OF PLATE III.

Fig. 1.—Head of male anopheles, magnified. Showing palpi about same length as proboscis and clubbed. Position of parts as in life.

Fig. 2.—Head of female anopheles, magnified. Showing palpi about same length as proboscis. Position of parts as in life.

Fig. 3.—Head of female anopheles magnified. Parts separated to show the large hypodermic-needle-like central stylet of the proboscis; 2 pairs of fine hair-like stylets which in life lie along the central stylet, (function of these stylets doubtful); the proboscis or sheath, open on its under surface, for the reception and protection of the central and the 2 pairs of hair-like stylets; the 2 palpi; and the two antennæ.

PLATE III (*Anopheles*)

1.

2.

3.

L N TASKAR *del.*

EXPLANATION OF PLATE IV.

Fig. 1.—Attitudes assumed by anopheles when at rest on the wall. Natural size. Note posterior pair of legs extended beneath the body.

a. b. c. males, *d. e.* females, *f.* female lifting palpi and antennæ whilst in the act of probing.

Fig. 2.—Female anopheles. Natural size.

Fig. 3.—Male anopheles. Natural size.

Fig. 4.—Male anopheles in the act of probing. Natural size.

Fig. 5.—Female anopheles, enlarged.

Fig. 6.—Eggs of anopheles floating in a test tube. Natural size.

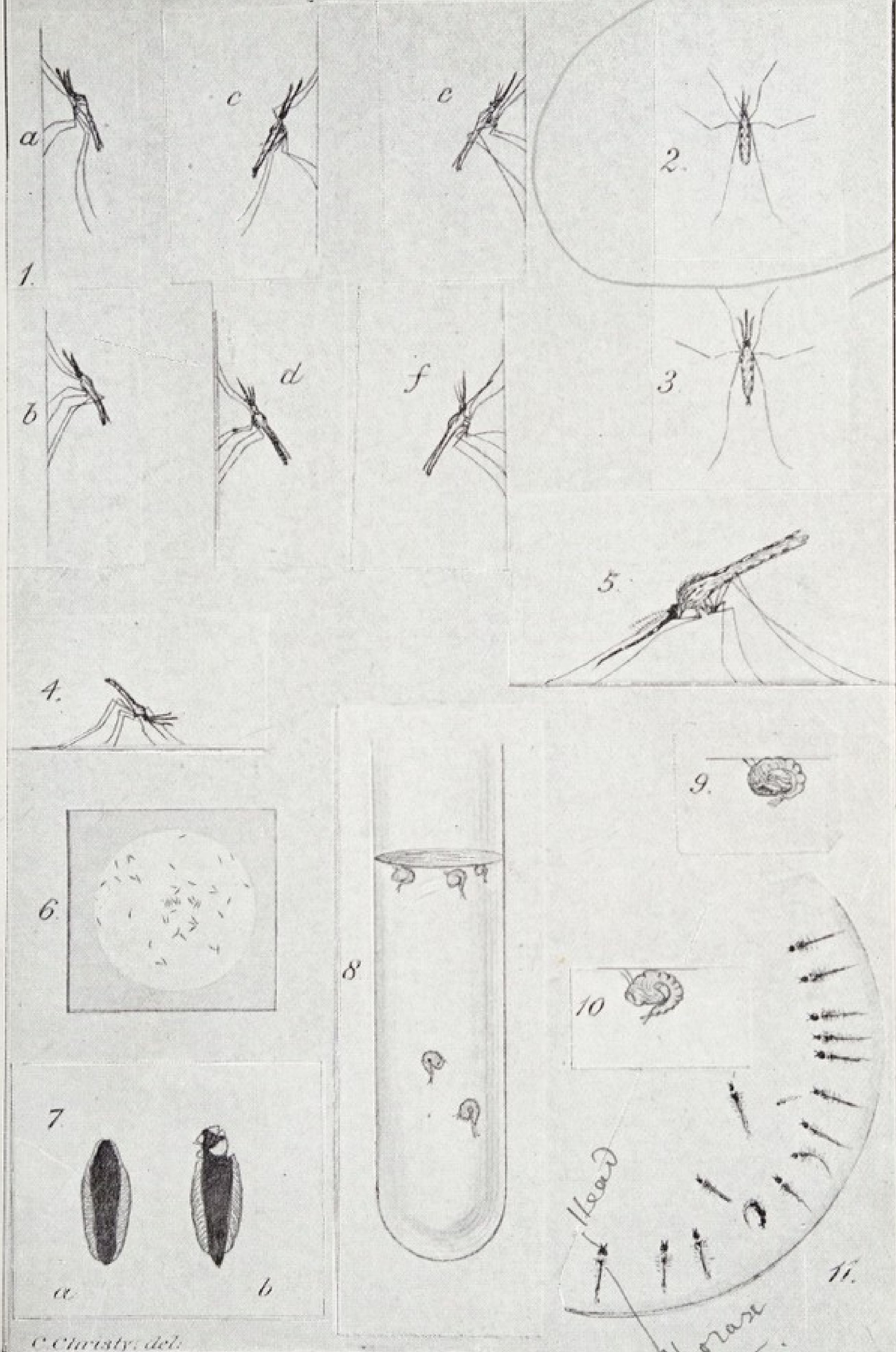
Fig. 7.—Eggs, much enlarged. *a.* unhatched, *b.* hatched-out shell.

Fig. 8.—The nymphæ or pupæ. Natural size.

Figs. 9-10.—Nymphs, enlarged.

Fig. 11.—The larvæ of anopheles floating in the water of a tumbler. Natural size.

Plate IV. (*Anopheles*)



C. Christy: del.

Govt Photozinco. Office, Poona 1900.

EXPLANATION OF PLATE V.

Fig. 1.—Anopheles larva drawn under $\frac{3}{4}$ inch lens.

Fig. 2.—Posterior end of anopheles larva, looking from above. Showing openings of tracheæ.

Fig. 3.—Posterior end of anopheles larva. Side view. Showing position of tracheæ.

Fig. 4.— Larvæ of anopheles in test-tube. Natural size.

Figs. 5-6.—Larvæ of anopheles in position of rest at surface of water. Magnified 6 diams.

Fig. 7.—Culex larva, enlarged (young).

Fig. 8.—Culex larvæ in test-tube. Natural size.

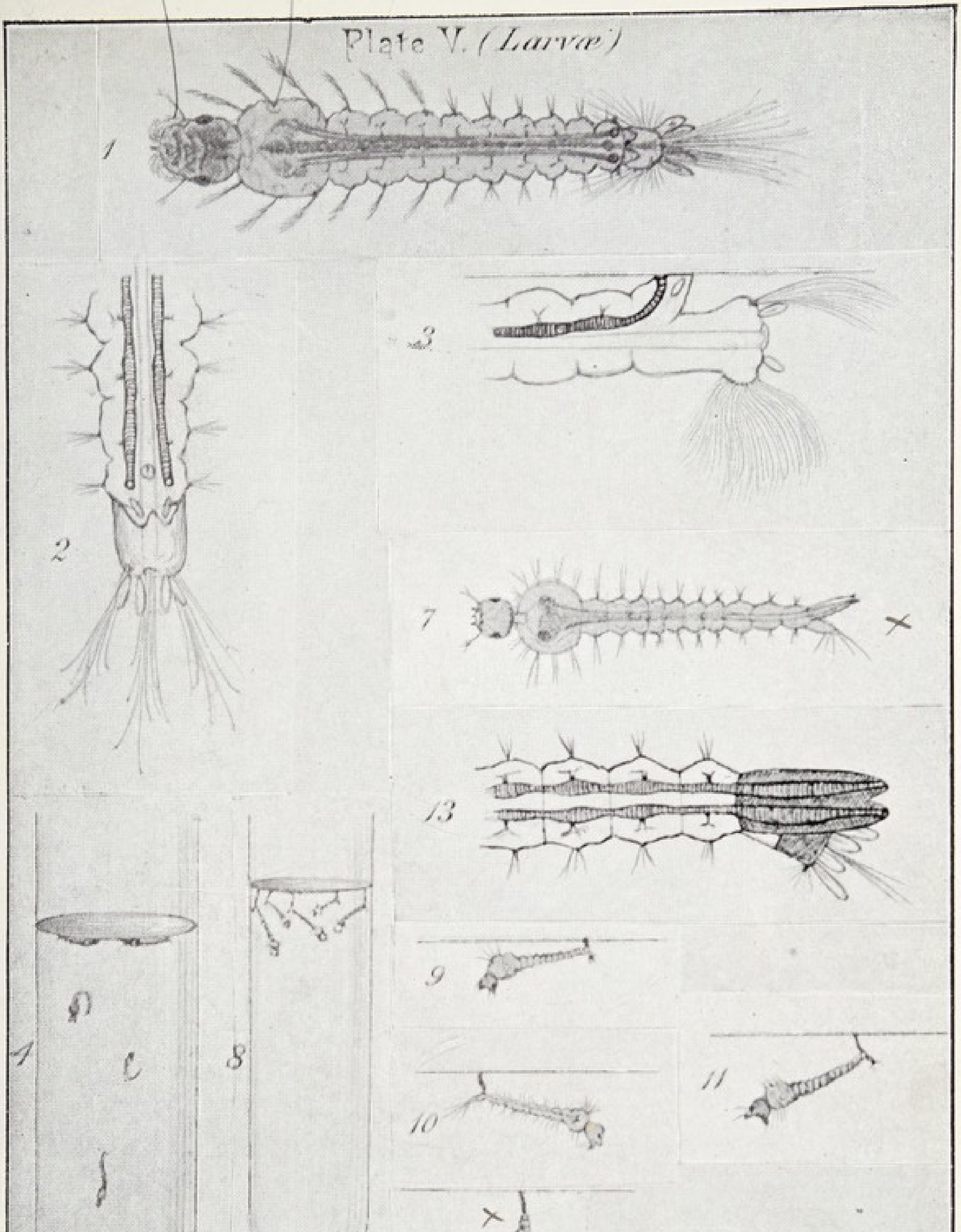
Figs. 9-10.—Culex larvæ, enlarged. Showing position of rest at surface assumed by same species.

Figs. 11-12.—Culex larvæ enlarged. Showing the usual position of rest at the surface assumed by most species.

Fig. 13.—Posterior end of culex larva looking from above. Showing the black buoy-like prolongation at the end of which the tracheæ open.

Fig. 14.—Posterior end of culex larva. Side view. Showing position of tracheæ and the fussiform dilatations.

Plate V. (*Larvæ*)



Govt Photozinco. Office, Poona 1900.

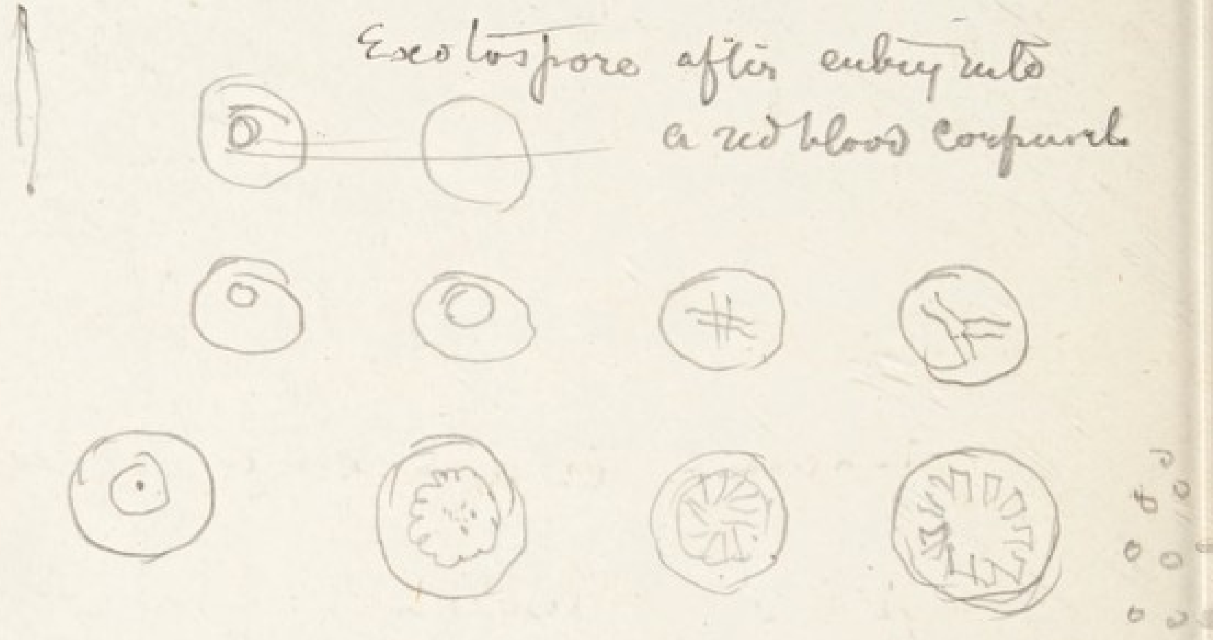
The Culex pipiens

Egg raft. The massed eggs float as a ~~raft~~ boat shaped raft on the surface of the water

The larva is generally seen at rest with the tip of the air tube of the abdomen at the surface of the water but when alarmed it strikes down into the water with a jerking movement

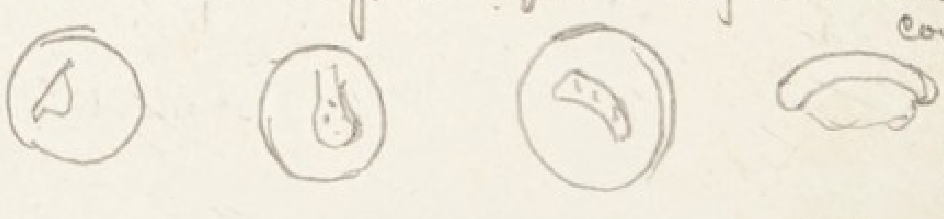
Pupa. The pupa rests with the two horn like air tubes on the thorax at the surface of the water but it can swim rapidly by the jerking of the abdomen which is provided at its extremity with two [illegible]

Escoto spore or germ of Malaria introduced into the blood of man by the prick of a mosquito



Crance

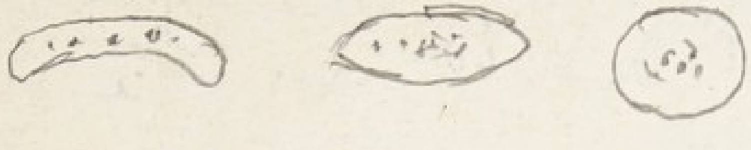
Enhaemospore after entering into blood corpuscle



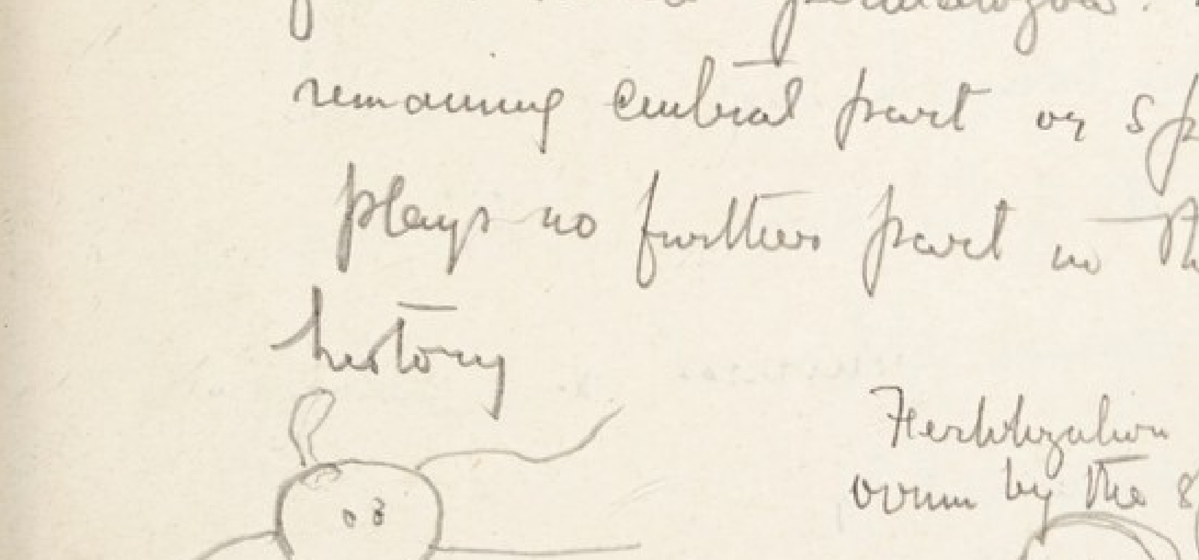
fully formed Cresc

Crescents appear to undergo no further development in man but if infected blood be sucked by a mosquito they undergo several changes.

The male enlarges and becomes spherical

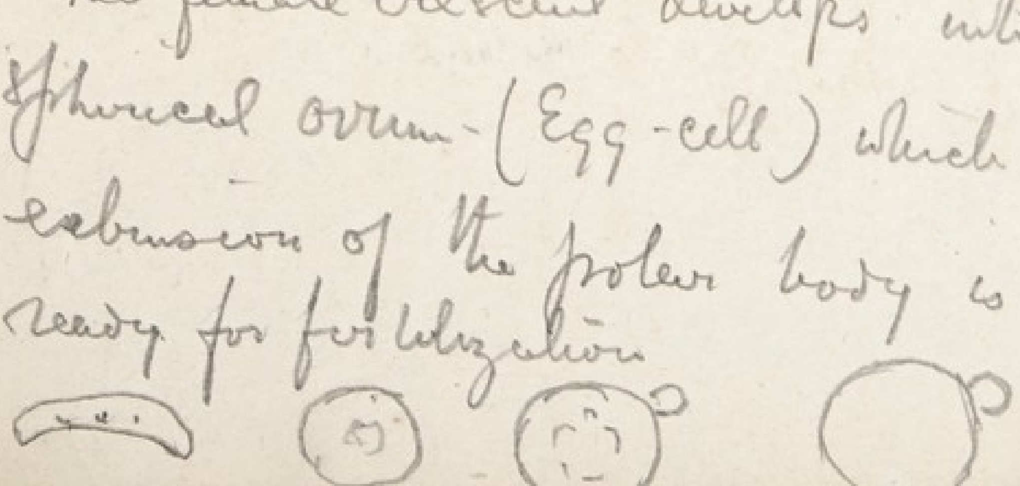


Several projections appear upon the surface of the male cell. These lengthen and are finally set free as mobile spermatozoa. The remaining central part or spermatophore plays no further part in the life history



Fertilization of the ovum by the spermatozoa

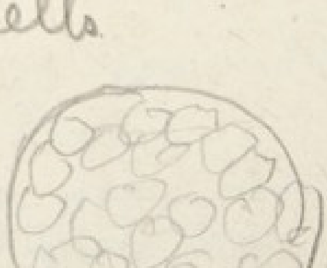
The female crescent develops into a spherical ovum-(Egg-cell) which after extrusion of the polar body is ready for fertilization



The fertilized egg or Zygote takes nourishment and becomes the active motile form vermicule

after boring through the stomach wall of the mosquito the vermicule becomes spherical and enlarges considerably

further stage the protoplasm broken up into spore mother cells



Sphere at its maximum of development the interior is occupied by countless needle shaped spores or germs which on the bursting of the sphere escape into the various organs of the body of the mosquito

Mobile spores (Exotospores) liberated by the bursting of the sphere. Such of these as find their way ~~into~~ to the salivary glands escape through the ducts of those glands and are transferred to the blood of man when the mosquito bites