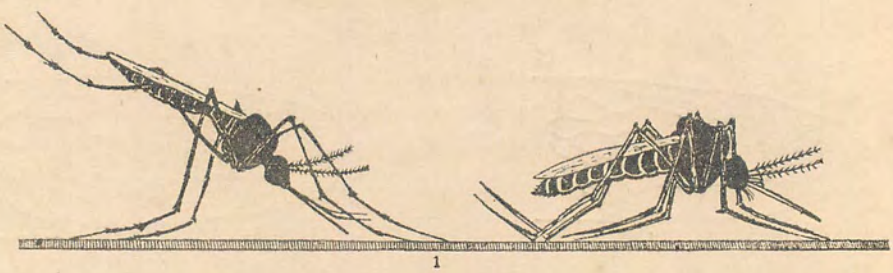

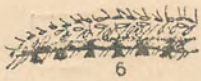

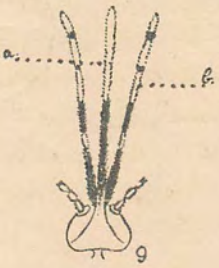

To Illustrate Dr. Barnes' paper on the Anophelene Mosquitoes of Siam.

ANOPHELENE MOSQUITOES WITH SPECIAL REFERENCE TO THE SPECIES FOUND IN SIAM.*

By M. E. Barnes, M.D.

With Plate 7.

[Read before the Natural History Society of Siam, Oct. 17, 1922.]

There are few subjects which have proved of more absorbing interest and which have stimulated more painstaking study among workers in tropical countries than have anophelene mosquitoes. Investigators in the most important tropical and sub-tropical countries have vied with each other in wading in rice-fields, plunging into swamps and bogs, examining collections of water in every conceivable place, to determine the facts concerning the life history of anophelenes. By the most painstaking labour men have devised and conducted delicate laboratory experiments; have dissected mosquitoes; have even measured the capacity of the mosquito stomach and calculated the number of human red blood cells which it can contain; have studied the habits of flight of various species; have learned what the larvae feed upon; have studied the natural enemies of mosquitoes; and have tested their susceptibility to various toxic substances. As the result of all of these investigations, we have to-day a growing accumulation of definite knowledge concerning this important group of the insect enemies of man. The application of this knowledge has already made possible the realization of such triumphs in applied sanitation as the world has never before witnessed.

My own study of these interesting insects during the past six years has resulted in the capture and certain identification of seventeen species of anophelenes, besides a few others the identification of which is as yet uncertain. The identifications have all been passed upon by Col. A. Alcock, Professor of Medical Entomology in the University of London, to whom I am greatly indebted for this service. Every species mentioned in this paper (with the exception of *Anopheles culicifacies)* was caught while attacking me in my

*The observations upon which this paper is based were made with the support and under the auspices of The International Health Board of the Rockefeller Foundation.

house, either in Bangkok or in the vicinity of Chiengmai. Some of them have been caught also by other individuals in other parts of Siam. *Anopheles culicifacies* is well known as a biting mosquito in other countries. The specimens of this species were caught resting in my house, but none have been caught while attacking. With reference to the species mentioned in the list below, therefore, it may be stated that each of them will enter a house in search of its victim.

There is considerable difference of opinion among entomologists as to the generic classifications as well as to the validity of certain specific names. Thus, the same species may be described under a variety of names by workers in various parts of the world. The tendency, however, is toward a simplification of classification instead of the hopeless multiplying of genera and sub-genera which has heretofore prevailed. Thus, in the latest edition of his book "Entomology for Medical Officers" (1920), Col. Alcock classifies all anophelenes under the single genus *Anopheles*. This procedure is adopted in presenting the list of the anophelenes thus far found in Siam. Synonymous names are those given by Alcock in the book mentioned above.

The exact identification of anophelene mosquitoes involves a most careful study of minute details. For the benefit of those who have never carefully observed this insect group, I will first call attention to some of the peculiarities upon which the differentiation of the various groups is based, and will then discuss other interesting and less technical phases of the subject.

DIFFERENTIATION OF ANOPHELENES FROM OTHER MOSQUITOES.

The first point to take up is the differentiation of anophelene mosquitoes from other mosquitoes. This is based upon one or more of the following peculiarities:—

1. *Attitude while at rest.* (Fig. 1)

With the exception of one or two species, the anophelenes rest with the head, thorax and abdomen extended in a straight line and at an angle with the surface upon which they rest. *Anopheles culicifacies* is the only exception to this rule thus far found in

Siam. When at rest, it rests like a *Culex*, i. e. with the abdomen approaching the surface upon which the insect rests.

2. *Spotted wings.* (Fig. 2)

With only a few exceptions, anophelenes have spotted wings, the spots being formed by definite collections of scales along the wing veins. Non-anophelene mosquitoes in some cases have spotted wings, but other characteristics distinguish them.

3. *Simple curved scutellum.* (Fig. 4)

Anophelenes universally have the scutellum in the form of a simple curve, instead of being tri-lobed as in other mosquitoes.

4. *Abdominal scales.*

In non-anophelene mosquitoes, the abdominal segments are as a rule thickly clothed with scales. In anophelenes the scales on the abdominal segments, if present at all, are as a rule few and scattered. When at all numerous they are usually limited to the terminal segments. The only marked exception to this is the single specimen described by Theobald under the name *Aldrichia*. This specimen had anophelene characteristics in the thorax and wings, but the abdomen was thickly clothed with scales as in *Culex*. No other such specimen has ever been found since.

5. *Length of the palps in the females.*

The proboscis and palps of anophelene females are of very nearly the same length. In non-anophelene mosquitoes the palps of the females as a rule are much shorter than the proboscis.

Generic Classifications.

The next point of interest is the method of differentiating the various genera into which anophelenes have been divided. As the number of valid species thus far described in various parts of the world is well over 100, this is no easy task. Careful study, however, shows that there are four important parts of the body which show sufficient constant differences to enable a classification of genera or sub-genera to be made. In giving the list of anophelenes found in Siam, all are grouped into a single genus *Anopheles*. In making the preliminary identification of the specimens, however, I used the classification adopted by James and Liston in their book "The

Anophelene Mosquitoes of India" 2nd edition. Their classification is most useful in identifying the mosquitoes of Siam, provided that one remembers the synonymous names for certain of their species. These authors pay special attention to differences shown on the following parts of the body of anophelenes:—

1. The abdomen. 2. The thorax. 3. The prothoracic lobes 4. The wing scales.

Some of the differences in these respects exhibited by anophelenes commonly found in Siam will be briefly mentioned.

1. *The Abdomen.*

a. Having no scales whatsoever:—*Anopheles, Myzomyia.*

b. Having scales only on the last two or three segments but not arranged in tufts:—*Nyssorhynchus, Nyssomyzomyia.*

c. Having a tuft of scales projecting ventrally from the seventh abdominal segment:—*Myzorhynchus.* (Fig. 5).

d. Having tufts of scales projecting ventrally from six abdominal segments:—*Christophersia.* (Fig. 6). This genus is united by other entomologists with *Cellia* in which the tufts in dried specimens project laterally instead of ventrally. In the specimens of this genus thus far examined in Siam, I have never found the tufts projecting laterally.

2. *The thorax.*

a. Having the thorax clothed with hairs, or long hair-like scales or both (Fig. 3):—*Anopheles, Myzomyia, Nyssomyzomyia, Myzorhynchus.*

b. Having the thorax clothed with short, broad, true scales (Fig. 4):—*Nyssorhynchus, Christophersia* (= *Cellia*).

3. *The prothoracic lobes.*

By carefully turning the insect under a high magnifying lens, these lobes may be seen projecting from the anterior aspect of the thorax.

a. With hairs only. No scales:—*Anopheles, Myzomyia, Nyssorhynchus, Nyssomyzomyia.*

b. With tufts of scales:—*Myzorhynchus, Christophersia* (= *Cellia*) (Fig. 4.).

4. *Wing scales.*

The wing scales present slight variations in the various species, but the determination of these differences requires careful measurements which are beyond the boundary of interest of the ordinary observer. For comparison it is usual to select the scales on the third longitudinal vein, or those found near the bifurcations of the second and fourth longitudinal veins.

a. With scales of the usual expanding type, i. e. being broadest in the middle and tapering gradually toward the apex :— *Nyssomyzomyia, Nyssorhynchus, Myzorhynchus, Christophersia* (= *Cellia*) and *Anopheles.*

b. With narrow scales, broadest near the apex and tapering abruptly toward the apex and gradually towards the base:—*Myzomyia.* (Fig. 8).

By careful examination, most of the anophelenes of Siam may be classified into one of these genera. It is then easier to determine the species by reference to keys, and by observing other peculiarities which are noted below :—

1. *Palpal markings.*

In some species, the palps are of uniform color (e. g. *barbirostris*). In others there are bands of white scales of varying width and situation. These markings are fairly constant for the species. (Fig. 9).

2. *Proboscis markings.*

In *Anopheles punctulatus*, Donitz, the outer half of the proboscis is white. (Fig. 9). In *Anopheles aconitus*, Donitz, the outer half of the proboscis is yellow or yellowish-white. These are the only anophelenes thus far found in Siam which show peculiar marking of the proboscis.

3. *Leg markings.*

The extent of variation in the leg markings may be illustrated by the following species (omitting reference to the slight banding at the joints which occurs in some of the species) :—

a. Uniform colourings :—e. g. *A. funestus.*

b. Definite markings located in peculiar positions:—e. g. the white oval mark on the femur of the mid-leg in *A. fuliginosus*; the broad white band at the lower end of the tibia of the hind leg in *A. leucosphyrus.*

c. Speckling:—e.g. in *A. punctulatus*, and *A. ludlowi.*

d. Terminal tarsals white:—e. g. in *A. fuliginosus* and *A. jamesi.*

4. *Wing markings.*

The variations in the markings of the wings are quite definite, but they require very careful examination. An easily observed peculiar wing marking is the "T" mark on the wings of *A. rossii.* Another example which may be mentioned is the third longitudinal vein in *A. culicifacies* which is black-scaled in the greater part of its course. In A. *funestus* which it greatly resembles, the third longitudinal vein is white-scaled.

The wing fringe varies considerably in its markings in different species, and there are also important variations in the venation of the wings, but for these peculiarities, reference should be made to books on the subject.

In identifying anophelene mosquitoes, the study of the scales requires a good microscope. It is possible, however, to identify most of the species found in Siam by a careful examination with a good hand lens, and by reference to the following key which is modified from James and Liston in "The Anophelene Mosquitoes of India".

Life History of Anophelenes.

The life history of anophelene mosquitoes is of the greatest practical importance and much laborious investigation remains to be carried out before the peculiarities of habits of these various species are fully known. Such detailed knowledge is essential to the devising of adequate and practicable measures against anophelene mosquitoes.

Anophelenes lay their eggs in natural collections of water, and consequently they are not house-breeding mosquitoes. Only in times of great drought will they occasionally oviposit in water in artificial containers. With the exception of *Anopheles rossi*, to

KEY TO THE SPECIES.

WINGS SPOTTED:—

A. Palps unbanded.

Densely scaled thick palpi. Ventral tuft of scales projecting from 7th abdominal segment. Very large mosquitoes. .. .. .. *Anopheles barbirostris.*

B. Palps with five white bands.

Tufts of scales projecting ventrally or laterally from six abdominal segments. .. .. *Anopheles kochi.*

C. Palps with four white bands.

Tarsal joints banded, but none of the hind tarsal segments are white. Tuft of scales projecting ventrally from 7th abdominal segment (sometimes this is not prominent). Tips of wings are white. .. .. *Anopheles sinensis.*

Legs banded and much speckled. None of the hind tarsal segments are white. Outer half of the proboscis is white. .. .. *Anopheles punctulatus.*

Legs banded and much speckled. A broad white band at the tibio-tarsal joint on hind legs. *Anopheles leucosphyrus.*

Legs banded but not speckled. The last hind tarsal segment is pure white. .. .. *Anopheles karwari.*

D. Palps with three white bands.

(1) With one or more of the hind tarsal segments pure white.

Tarsal joints banded. Legs not speckled. Mid-legs with a large oval white spot on the femur. Hind legs-broad distal white band on 2nd tarsal segment. 3rd, 4th, 5th tarsal segments pure white. .. .. *Anopheles fuliginosus.*

Similar to above, but with speckled legs, and without the oval spot on femur of mid-leg. Hind legs-broad apical bands on 1st and 2nd tarsal segments, 3rd, 4th, 5th tarsal segments pure white. .. .. *Anopheles jamesi.*

As above, but with palps and legs speckled, and with distal white band on 2nd tarsal segment. 3rd, 4th, 5th tarsal segments pure white. .. *Anopheles maculipalpis.*

White patch on middle of 2nd hind tarsal segment. Broad white bands over distal joints of 2nd, 3rd, and 4th hind tarsal segments. 5th hind tarsal segment pure white. .. .. *Anopheles maculatus.*

Resembles *maculatus*, but has many scales on dorsal part of each abdominal segment. .. *Anopheles willmori.*

(2). Tarsal joints banded, but none of the hind tarsal segments are pure white.

Legs not speckled. On the wings is a "T" mark. .. .. *Anopheles rossii.*

Legs speckled. On the wings is a ".T" mark, with a full-stop in front of the T. .. *Anopheles ludlowi.*

Outer half of the proboscis is yellow or yellow-white. .. .. *Anopheles aconitus.*

(3). Legs uniformly coloured, without bands or white segments.

Six white patches on the wing-fringe. 3rd longitudinal vein is white-scaled. .. .. *Anopheles funestus.*

Three white patches on the wing-fringe. 3rd longitudinal vein is black-scaled. .. .. *Anopheles culicifacies.*

which pollution does not appear to be objectionable, anophelenes as a rule choose by preference fairly clean water. Each species shows a certain degree of selection in its breeding place. Usually, the waters selected contain vegetation, such as grasses.

Although various observers report that certain anophelenes breed in flooded rice-fields, I have seldom found any species except *A. barbirostris* and *sinensis* in the cultivated parts of the field. I have collected other species in the grassy ditches along the edge of such fields. Whether this is an accidental observation or whether the plowing of the soil and the growing rice render the waters unfavourable to the larvae, I am unable to say. In the flooded and grass-grown land lying fallow, I have collected the larvae of *A. fuliginosus, sinensis, barbirostris, aconitus,* and *rossii.* These same larvae may also be found in the grassy portions of small ditches and similar collections of water. If the water in these places is kept free of all vegetation so that fish have free access to every part of the pool or ditch, very few anophelene larvae will be found, provided the surface is free from floating matter. If small sticks and portions of leaves are floating on the surface, anophelene larvae may be found, even though the water is well stocked with fish. In such pools I have watched fish come up to a *sinensis* larva and even touch it. Under these conditions, the only chance the larva has to escape is to lie perfectly motionless. If it makes the slightest movement, the fish seizes it. If it lies perfectly motionless, the fish does not usually take it. The larvae of *A. fuliginosus* and *jamesi* may be found in moats and also in grassy swamps. Other larvae, such as *A. maculatus* prefer running streams, and I have collected *A. aconitus* larvae also in slow-moving streams. *A. ludlowi* prefers brackish water. Thus, on the island of Koh Phra, in a small salt-water marsh, large numbers of such larvae developed, and the mosquitoes carried malaria to the guards on the island. In India, larvae of some species have been found in wells in which the water was from 20 to 30 feet below the surface of the ground. Anophelene larvae have also been found in water collected in holes in trees, and in the cut joints of bamboo.

In Siam, the eggs of *A. rossii* hatch in from one to two days. The larvae mature in from seven to ten days, and the mosquitoes emerge from the pupal case in from one to two days. The complete cycle from egg to imago usually requires from 10 to 15 days, although in favourable conditions it may be complete in eight days. Anophelene larvae may be recognized by their short breathing tubes, and by the presence on their abdominal segments of peculiar palmate hairs. When they approach the surface of the water, they lie with their bodies horizontal to the surface. Culicine larvae have long breathing tubes, and when near the surface they hang with their heads down. The larvae feed upon algae, broken-down vegetable matter, and even upon other larvae.

Shortly after emerging from the pupal case, the female mosquitoes seek a blood meal. This meal may be secured from various animals, including man. Birds and fowls are favourite victims as the combs are rich in blood. The female mosquito alone is able to suck blood. After the meal, the females usually rest for a few hours to await the proper development of the ova, and then fly to their preferred sites to oviposit. About 100 ova are deposited at one time. It is not definitely known how many times a female oviposits during her life. Under laboratory conditions a female has been observed to oviposit as many as five times, but climatic conditions and the accessibility of food are variable factors which affect this act.

The length of life of anophelenes under laboratory conditions varies from a few days to as long as three months. Under natural conditions adult mosquitoes are preyed upon by many enemies, including nomadic spiders, dragon-flies, bats, etc., so that probably the average duration of life is much shorter.

Relation of Anophelenes to Malaria.

The study of anophelene mosquitoes has been greatly stimulated by the discovery of their relationship to malaria. Malaria is a disease which has been recognized from the remotest times. The clinical types which we recognize to-day were described by Hippocrates in the 5th century B. C. But the problem of the mode of transmission of malaria baffled physicians for thousands of years

until between 1895 and 1898 when Sir Ronald Ross, by a series of brilliant investigations, definitely demonstrated that malaria is a mosquito-borne disease. So far as is at present known, anophelene mosquitoes alone are involved in its transmission, and certain species of anophelenes appear to be much more efficient carriers of the disease than others.

As resistance to infection develops in a patient afflicted with malaria, sexual forms of the malarial parasite are produced. When a susceptible anophelene mosquito imbibes this blood, fertilization of the parasite takes place in the mosquito's stomach, and the parasite burrows into the wall of the stomach. After a period of from 10 to 14 days or more, the fully developed oocyst ruptures, and small sickle-shaped sporozoites are liberated in the body fluids of the mosquito. These sporozoites find their way to the salivary glands of the insect. Thereafter, whenever this particular mosquito sucks human blood, she is capable of infecting the victim with the organisms which produce malarial fever.

Of the seventeen species of anophelenes which have been mentioned in this paper, almost all except *A. rossii* are capable of developing malarial infection under laboratory conditions. Nine of these seventeen species are found in Bangkok, namely, *A. rossii*, *fuliginosus*, *sinensis*, *funestus*, *aconitus*, *minimus*, *punctulatus*, *barbirostris*, and occasionally *ludlowi*. Of 498 anophelenes caught in the evenings in my house in Bangkok, 55.8 per cent were *A. fuliginosus*, and 29.5 per cent were *A. sinensis*. Both of these species are efficient carriers of malaria in other lands, and presumably would be equally efficient in Siam. So far as anophelene mosquitoes are concerned, therefore, there is no reason why Bangkok should not have malarial cases in abundance. As a matter of fact, the hospitals constantly have cases of this disease originating in people who have never been outside of Bangkok. The number of these cases appears to be somewhat on the increase. A large city, such as Bangkok, which at present has comparatively few human carriers of the disease, even with a plentiful supply of anophelene mosquitoes is, however, much less liable to develop a high rate of

morbidity from malaria than is a village, because there is much less chance for large numbers of anophelenes to bite a human carrier of the disease. In some of the villages near Chiengmai, on the contrary, as high as 86 per cent of the children have enlarged spleens. This means that all of these children sooner or later are active carriers of the sexual forms of the parasites. It means further that enormous numbers of the anophelenes in these villages feed upon infected blood. In spite of all natural enemies, a considerable number of these insects is certain to survive for the ten days required for them to become infective. Thus, malarial infection is kept constantly active in these villages.

This direct connection between anophelene mosquitoes and such a serious disease as malaria, invests the study of these insects with great practical importance. Every observation, every investigation, every experiment, if carefully made, becomes of value. It is, moreover, a study in which all living in Siam have abundant access to the material, for none are immune from the attacks of these pests. Finally, it is a study in which there is a great field for new research. In spite of all that has hitherto been done, the definite information which we possess in regard to this important group is as yet very inadequate.

List of the Anophelenes of Siam. [1]

The anophelenes thus far found in Siam are named in the following list, together with the localities in which they have been collected. Where the collection was made by another observer, the name of the collector is given. Where no name is mentioned, the collection was made by the writer.

1. *Anopheles funestus*, Giles (= *Myzomyia listoni*, Liston).
   Chiengmai, April 9, 1920.
   Bangkok, Oct. 25, 1921.
2. *Anopheles rossii*, Giles.
   Bangkok, Dec. 1916.
   Chiengmai, March, 18, 1917.
   Lampang, June, 1920.

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1 For this list with more complete notes refer to the American Journal of Hygiene, (Baltimore) current volume.

Meklong, Nov. 1921.
Rajburi, Oct. 1921 (Dr. J. R. Redfield).
Prachinburi, July, 1922 (Lieut. Sawat Manmhai).
Trang, July, 1922 (Lieut. Tong Kham Raggabhai).
Prapatom, 1906, 1907 (Dr. P. G. Woolley—*cf.* Theobald Monograph Culicidae of the World, Vol. V, page 19).

3. *Anopheles aconitus*, Donitz (=*A. albirostris*).
Chiengmai, Aug. 24, 1920.
Bangkok, Oct., Nov., Dec. 1921.

4. *Anopheles ludlowi*, Theobald.
Bangkok, July, 1921.
Koh Phra, Nov. 16, 1921.
Koh Mak, Dec. 19, 1914; Koh Kut, Dec. 30, 1914 (C. Boden Kloss).[2]

5. *Anopheles culicifacies*, Giles.
Chiengmai, Aug. 22, 1920.

6. *Anopheles punctulatus*, Theobald (=*A. tessalata*).
Chiengmai, March 31, 1920.
Bangkok, Oct. 1921.

7. *Anopheles leucosphyrus*, Donitz (= *A. elegans*).
Hoi Chan Kean, Chiengmai, May 9, 1918.
Chiengmai, Nov. 5, 1913.

8. *Anopheles minimus*, Theobald.
Bangkok, 1921.

9. *Anopheles barbirostris*, Van der Wulp.
Bangkok, Nov. 26, 1914 (C. Boden Kloss).[2]
Bangkok, Dec. 1916.
Chiengmai, Feb. 1917.
Lampang, June, 1920.
Meklong, Nov. 1921.
Rajburi, Nov. 1920 (Dr. J. R. Redfield).
Prachinburi, July, 1922 (Lieut. Sawat Manmhai).
Trang, July, 1922 (Lieut. Tong Kham Raggabhai).
Prapatom, 1906, 1907 (Dr. P. G. Woolley. *cf.* Theobald Monograph Vol. V).

10. *Anopheles sinensis*, Wiedemann (= *A. Vanus*, Walker; *A. nigerrimus*, Giles; *A. pseudopictus*, Grassi; *Myzorhynchus minutus*, Theobald; *Myzorhynchus peditaeniatus*, Leicester; and perhaps *A. plumiger*, Donitz).

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2 Included in an unpublished paper "On a Small Collection of Mosquitoes from Siam" by A. T. Stanton, M.D. and F. W. Edwards, B.A., which is in the files of the Natural History Society of Siam.

Bangkok, Nov. 28, 1914 (C. Boden Kloss).[2]
Bangkok, Oct. 1921.
Prapatom, 1906, 1907 (Dr. P. G. Woolley, *cf.* Theobald Monograph, Vol. V).
Koh Chang, Dec. 6, 1914 (C. Boden Kloss).[2]

11. *Anopheles maculatus* var. *willmori*, James (=*Neocellia dudgeoni* and *indica*, Theobald).
Chiengmai, Royal Pages College, May 9, 1917.

12. *Anopheles maculatus*, Theobald (= *Nyssorhynchus pseudowillmori*, Theobald).
Chiengmai, Hoi Chan Kean, May 8, 1918.

13. *Anopheles karwari*, James.
Chiengmai, Hoi Chan Kean, May 8, 1918.

14. *Anopheles maculipalpis*, Giles (= *Nyssorhynchus indiensis*, Theobald).
Chiengmai, Hoi Chan Kean, May 12, 1917.

15. *Anopheles jamesi*, Theobald.
Chiengmai, Aug. 27, 1920.
Chiengmai, Nong Seng village. Aug. to Nov. 1920.

16. *Anopheles fuliginosus*, Giles (= *Nyssorhynchus nivipes*, Theobald; *A. leucopus*, Donitz; *Calveryina lineata*, Ludlow).,
Chiengmai, March 8, 1917.
Bangkok, Oct. 1, 1921.
Prachinburi, July 1922 (Lieut. Sawat Manmhai).

17. *Anopheles kochi*, Donitz (=*Christophersia halli*, James; *Cellia flava*, Ludlow).
Chiengmai, Aug. 13, 1920.

18. *Anopheles* (? *formosus*, Ludlow).
The specimens were badly rubbed so that this identification is uncertain.

Collecting and Preserving Mosquitoes.

1. *Collecting mosquitoes.*

A test tube is one of the most convenient devices for collecting mosquitoes, as its open mouth can be slipped over the mosquito, while it is attacking its victim, without disturbing it. Previous to use, a few drops of chloroform should be sprinkled upon a small pledget of cotton, which is then pushed to the bottom of the test tube. A piece of soft paper should be placed above the cotton to prevent the insects coming in contact with the chloroform. As soon

as the insects are dead, they should be removed from the chloroform tubes.

2. *Mounting.*

If it is desired to mount the specimens, No. 20 entomological pins are very useful for the purpose. The pin should be pushed through the thorax in such a way as to spread the wings. The point of the pin can then be inserted into a suitable mounting base, and the position of the legs adjusted by careful manipulation with a long needle.

3. *Drying.*

It is absolutely essential that the specimens be thoroughly dried before being mounted in collecting boxes or shipped for identification, as otherwise they will be destroyed by moulds. Two hours exposure to the sunlight is usually sufficient in this country.

4. *Permanent mounting.*

The box for permanent mounting should have a tight-fitting lid, and a suitable mounting medium on the bottom. For the latter purpose any of the following preparations will be found satisfactory :—

a. A thin sheet of cork, impregnated with beechwood creasote.

b. A layer of corrugated card-board such as is used in packing bottles for shipment. This must be impregnated with creasote.

c. A layer of paraffin. This is recommended as convenient and satisfactory. A sufficient quantity of paraffin to form a layer one-fourth of an inch in thickness is melted. To it is added approximately ten per cent of powdered naphthalene (moth balls) and ten per cent of beechwood creasote. The mixture is then poured into the box and allowed to solidify.

In each of these methods, the inner sides of the box should be painted with creasote at frequent intervals to prevent the entrance of small ants or mites, which otherwise are certain to destroy the collection.

d. For mounting type specimens the writer uses glass-shell vials 25 mm. high x 15 mm. diameter. The corks are first impregnated with creasote, and the pins holding the thoroughly-dried

specimens are inserted into the cork. The cork is then put in place, and the vial sealed with a few drops of melted paraffin. The labels are pasted upon the sides of the vial. The type specimen can be examined through the transparent vial, or if necessary, the cork can be removed to permit more careful examination.

5. *Labelling.*

The data concerning the specimens should be entered upon small card-board discs through which the mounting pin passes.

6. *Sending specimens for examination.*

A convenient method of sending specimens for examination is to enter the data upon small pieces of soft paper, e.g. cigarette paper. These papers are then rolled into small cylinders, the throughly dried specimens are dropped into these cylinders, and the ends closed by twisting the paper. These cylinders can then be loosely packed in a convenient mailing box or tube and sent by mail. If more than one specimen is enclosed in a cylinder, the specimens should be separated by small pieces of soft paper.

EXPLANATION OF PLATE 7.

1. Anophelene (left) and Culicine (right) attitude.
2. Spotted wing of anophelene.
3. Thorax clothed with hairs.
4. Thorax clothed with scales.
   a. Prothoracic lobe.
   b. Scutellum.
5. Abdominal scale tuft (in *Myzorhynchus*).
6. Abdominal scale tufts in (*Cellia*).
7. Common type of wing scale.
8. Wing scale as in *Myzomyia.*
9. Markings of Proboscis and Palps as in *A. punctulatus.*
   a. Proboscis.
   b. Palp.

(Drawings after James and Liston.)