

The Determination of Mosquito Females by
Microscopic Preparations of the Head^{1/}

I. Taxonomic Characters and Description of the Genera

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ABSTRACT. We researched peculiarities in the structure of the head and its appendages in females of 75 species of the mosquito fauna of the USSR. We discovered characters which permit the genera, subgenera, and, in some cases, the species of mosquitoes to be distinguished by microscopic preparations of the head. A concise description of the taxonomic characters is presented here, as well as a description of the genera.

In determining mosquito species, the coloration of various parts of the body, which depends mainly on the color of the scales covering the insect's body, plays an important role. Morphological characters are used primarily in determining the genus as well as the species of mosquitoes on the basis of the structure of the genitalia. Females usually predominate in collections of mosquitoes for understandable reasons. In mass collections the scales easily fall off, and a significant number of the females collected turn out to be indeterminable.

The question arises concerning the possibility of determining females by morphological characters which are distinguishable under the microscope, just as bloodsucking biting midges (*Ceratopogonidae*) and mosquitoes (*Phlebotominae*) are also determined by microscopic preparations. There are indications in the literature that morphological specific differences between mosquito females exist. Attention was paid to the diversity in the structure of the oral cavity of *Anopheles*; in many species it has a row of denticles of various shapes (Christophers, 1933). It is also known that the sizes and structure of the palps of females are not alike in different species. In the work of Natvig (1948), schematic drawings of the palps of females of some Northern European species are provided. The differences in structure of the genitalia of females of various genera found in Japan have also been described (La Casse and Yamaguti, 1955). Since that time, the structure of the internal sclerites of the genitalia of mosquito females, principally of the genus *Aedes*, has been researched in detail (Mohrig, 1967, 1969; Ryazantseva, 1970, 1972).

In our opinion, it would in practice be most convenient to determine mosquito females whose scales are no longer intact by microscopic preparations

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of the head. The processing of such preparations is not complicated. No dissection is necessary. The head is detached from the fresh or dried specimens, treated with a 10% caustic alkali, washed well with water, dehydrated, cleared, and placed in Canadian balsam. It is desirable that the dorsal side be turned upwards. The preparations are examined at a magnification of 80-200 times. It is not necessary to use a greater magnification.

Taking into account the complexity of the structure of the head and its appendages, it can be hoped that the researcher will succeed in finding the appropriate distinctive characters, if he takes into consideration the peculiarities in the structure of not just any single organ (palps, oral cavity), but of the various parts of that bodily section. We undertook our research in that direction. We examined more than 500 preparations of the head of 75 mosquito species of seven genera of the Soviet Union's fauna. In this communication, we present a concise description of the taxonomic characters and a description of the genera. A detailed description of the structure of the head does not enter into our task.

TAXONOMIC CHARACTERS

Dorsally between the eyes there is an elongated area, the frons^{3/}, usually there are no fine hairs and scales on it (Fig. 1). It can be narrow (*Culex*, *Mansonia*, *Orthopodomyia*, subgenus *Culicella*) or relatively wider (*Anopheles*, *Aedes*, subgenus *Culiseta*). Towards the rear, the frons widens into the occiput. The width of the frons can be determined by comparing its diameter with the diameter of a facet of the eye. If the width of the frons at its anterior end does not exceed the diameter of a facet, it may be designated as narrow; if it is 2-3 times wider than a facet, it is of medium width. The frons has a greater width in a few species of *Aedes* and especially in *Anopheles* which belong to the subgenus *Cellia*.^{2/}

Many fine hairs and scales are missing on the preparations. The sites where the scales were attached to the frons, designated by very small circles or ovals, are distinguished from the sites where the hairs were attached, which have larger circles or ovals depending on the size of the hairs. On the frons there can be a longitudinal suture like a dark line, which is pronounced in most *Aedes* and in almost all species of the subgenus *Ochlerotatus* in particular. The longitudinal suture on the frons is lacking in *Mansonia*, most species of *Culex* and *Culiseta*, and in some *Aedes*. This character has important taxonomic significance, but in individual cases it is highly variable. In some species (*Aedes vexans* Mg., *Culiseta alaskaensis* Ludl.), the longitudinal suture on the frons exists in some specimens but not in others.

^{2/} Until the catalog of mosquitoes (Stone et al, 1959) was published, the given subgenus was usually designated as *Myzomyia* Blanch. In the catalog, the name *Cellia* Theob. was given priority at publication time (for 18 days). In our opinion, there is no basis for similar cases of restoring names which have hardly been used for several years, especially since it is not always possible to determine the date of publication exactly. But insofar as almost all the authors have accepted the recommendation of the catalog, we consider it expedient to use the name *Cellia*.

^{3/} The interocular space of Knight, Mosq. Syst. Newsletter 2:23-33. 1970.
[Editor's note.]

On the ventral side of the head the eyes are also separated by a space which is sometimes very narrow, not more than the diameter of a facet (*Anopheles*, most *Culex*). More often this space is wider; in some *Aedes* it is 7-8 times greater than the diameter of a facet.

The space on the dorsal side of the head behind the eyes (the occiput) is covered with scales and hairs of varying sizes. In rare cases, large hairs on the occiput are present in only one row behind the eyes (subgenus *Stegomyia*). The large hairs are usually scattered over the occiput in a greater or lesser amount. Many hairs and scales are located on the rear surface of the head, but it is difficult to see them, since this part is placed more or less at a slant or vertically on the preparations.

The shape of the clypeus has some taxonomic significance. Its length is usually approximately equal to its width at the base; the anterior margin of the clypeus is more often bulging; sometimes it is almost straight. More rarely the length of the clypeus is obviously less than the width. In very rare cases the clypeus has a bundle of scales; the point where this bundle is attached looks like a dot- in *Aedes (Stegomyia) aegypti* L.

The dark outlines of the sclerotous wall of the pharynx (the "esophageal pump") are visible through the integument of the head. The pharynx (Fig. 2) consists of a narrow anterior part and a wider posterior part. Lightly colored large tracheal trunks adjoin the posterior part along the sides. In some mosquitoes, the posterior part of the pharynx is swollen and is not inferior in length to the anterior part (*Aedes, Culiseta*). In other cases, the faintly enlarged posterior section of the pharynx is shorter than the anterior part (most *Culex*). Towards the rear, the pharynx widens gradually; therefore the relationship of the length of both sections cannot be precisely measured. At the front of the pharynx lies the oval cavity or cibarial pump ("pharynx"). The oral cavity in *Culex* and in species of the subgenus *Cellia* contains a row or group of stick-like or scaly denticles. In this case the oral cavity is designated as "armed".

The structure of the appendages of the head (proboscis, palps, antennae) presents a great deal of variety. The length of the proboscis, varying greatly within each species, can serve to a great degree as an index of the dimensions of the insect body, although there is no strict proportionality here. Small mosquitoes have a proboscis up to 2 millimeters long; in medium-sized mosquitoes the proboscis is 2-3 mm; in large mosquitoes it is greater than 3 mm. We measure the length of the proboscis from the anterior margin of the clypeus to the apex of the labellae. In some mosquito species there is a light ring in the middle part of the proboscis which is rather distinctly visible on the preparations.

The palps in most mosquito species have five segments. Natvig (1948) discusses the question of the number of palpal segments; we agree with his conclusions. The first segment is small and free of scales (exceptions are *An. hyrcanus* Pall. and *An. lindesayi* Fil.). The second segment is also small, but it has a small bundle of scales and relatively long hairs. The third segment is longer and thicker. The fourth segment, which makes up roughly

half of the entire length of the palp in Culicinae, is stick-shaped or thickened, sometimes pin-shaped; the segment is thickly covered with scales and hairs. Numerous sensillae of a peculiar sort are located on this segment; they look like small light circles. The fifth rudimentary segment is usually very small in most species and is mainly spherical or egg-shaped. In many species (most *Culex*, some *Aedes*), the fifth segment is lacking. In individual cases it exists or is lacking in specimens of a single species, for example in *Aedes cinereus* Mg. and *A. vexans* Mg.

The antennae consist of 15 segments. The first is small, ring-shaped, and without scales (exception - *Culiseta longiareolata* Mcq.). The second segment is large, spherical, and covered with scales and hairs primarily on the interior side; the scales may also be present on some other segments. The subsequent segments have a cylindrical shape; the third is relatively long; the fourth is significantly shorter. At the base of the segments, beginning with the fourth, whorls of long hairs are arranged. In addition, numerous shorter hairs of varying size and structure are arranged on the segments of the antenna. The third segment is covered with the very smallest hairs - microtrichia. They are clearly visible at a magnification of about 200X. In some species of *Culex* and of the subgenus *Culiseta*, the microtrichia are developed on the 4th and 5th segments as well, and in *Urano-taenia unguiculata* Edw., they are developed up to and including the 8th segment.

Many characters, distinguishable on microscopic preparations of the head, have diagnostic significance: the form and chaetotaxy of the frons; the width of the space between the eyes on the ventral side; the hairs of the occiput; the shape of the pharynx and the correlation between the length of its anterior and posterior sections; the "armament" of the oral cavity; the shape of the clypeus; the structure of the palps and antennae; the peculiarities of the hairs covering the head; etc. The majority of characters mentioned are characterized by a great degree of variability, which must be taken into account when determining mosquito species.

Quantitative characters also have great significance - not so much the absolute dimensions of the proboscis, palps, and antennae (whose lengths vary greatly within a single species), but rather the correlation of these dimensions. The length of the palps compared to the length of the proboscis is an especially important and relatively stable character. In order not to complicate the procedure of examining the preparations, we limited ourselves to four measurements: the length of the proboscis; the total length of the palp from the base of the first segment to the apex of the last (if both palps were preserved on the preparation, we used the average length); the length of the 4th (the longest) segment of the palp; and the total length of the 5th and 6th segments of the antenna. The total length of these latter segments was taken because they are more "typical", whereas the preceding segments are shortened. Also, the 5th and 6th segments usually lie next to the 4th segment of the palp on the preparation, which allows for the possibility of roughly comparing their lengths without resorting to measuring them.

The results of the measurements permit the following indices to be compiled: the relationship of the total length of the palps to the length of the proboscis (P/P), in Culicinae it is 0.12-0.45, and the relationship of the length of the 4th segment of the palps to the total length of the 5th and 6th segments of the antenna (P/A), in Culicinae it varies from 0.48 to 1.46.

As an example we cite the average quantitative indices for three species of *Aedes*; the first - a small mosquito with very short palps and relatively long antennae; the other two - rather large mosquitoes, but one is "short-palped" while the other is "long-palped."

Species	Length				Indices	
	Proboscis (in mm)	Palps (microns)	4th seg- ment of palps (microns)	5th & 6th segments of antenna (microns)	P/P	P/A
<i>A. cinereus</i> Mg.	2.05	.371	189	311	0.18	0.61
<i>A. punctor</i> Kirby	3.25	.621	304	317	0.19	0.95
<i>A. flavescens</i> Müll.	3.11	.884	516	386	0.28	1.34

GENERAL DESCRIPTION OF THE GENERA

1. *Anopheles* Mg. The determination of this genus is not difficult; as is generally known, the palps are approximately equal in length to the proboscis. The frons is moderately wide (subgenus *Anopheles*) or very wide (*Cellia*). The index of P/A in *Anopheles* is significantly less than three; in *Cellia* it can be greater than three. Attention should be paid to the antennae, on which there are conical sensillae in the form of small, sharply outlined light circles. The oral cavity is not armed (i.e., it has no denticles) (*Anopheles*) or else it is provided with an arcuate row of spine-shaped formations (*Cellia*).

2. *Uranotaenia* Arrib. The palps are very short and have four segments, but the boundaries of the segments are so obscure that the palps seem to have only two or three segments. On the upper side of the head, the parts of the eyes which are turned toward the middle line of the body are cuneiformly tapered. The frons is therefore, strictly speaking, not pronounced (Fig. 1). Microtrichia are developed on the third through the eighth segments of the antenna.

3. *Orthopodomyia* Theob. This genus is characterized by relatively long palps with five segments; the index of P/P is about 0.4 - highest in the Culicinae. The palps are very thin; the third segment is not thicker than the parts of the antenna which lie next to it. The frons is long and narrow with hairs and scales.
4. *Mansonia* Blanch. The palps have five segments. The scales of the palps and the proboscis are wide. The frons is long and very narrow in the anterior half; in the posterior half it has hairs and scales. A representative character of *M. richiardi* is the wide, light ring in the middle part of the proboscis.
5. *Aedes* Mg. The palps in most species have five segments; in some species, however, the fifth segment is lacking. The frons is moderately wide; more seldom it is wide. There is a longitudinal suture in most species, particularly in almost all *Ochlerotatus*. The posterior section of the pharynx is greatly swollen and is longer than the anterior section; sometimes both sections are approximately the same length. Species of the subgenus *Stegomyia* are distinguished by the arrangement of the hairs on the occiput: a row of 5-7 very large hairs is located behind the eyes on each side; there are no other hairs behind them. Very long hairs on the antenna are characteristic for the subgenus *Finlaya*; thus, on the fifth segment of the antenna, the length of the hairs of the whorls is at least twice as great as the length of the given segment. In species of the subgenus *Ochlerotatus*, the hairs of the antennae are relatively shorter, the palps have five segments, and the longitudinal suture on the frons is well developed (with the exception of 1 or 2 species). Mosquitoes of the *cantans* group are characterized by relatively long palps (P/P = 0.24-0.30), as well as by a rather large fifth segment of the palp, which is usually characteristically cone-shaped. In mosquitoes of the *communis* group, the P/P ratio is 0.17-0.23; very rarely it is 0.24.
6. *Culex* L. The palps have four segments; in rare cases, and even then not in all specimens of the given species (*C. hortensis* Fic., *C. orientalis* Edw.), there is also a small fifth segment. The length of the palps is usually not great (P/P = 0.17-0.24 and in mosquitoes of the subgenus *Lutzia* it is only 0.26-0.28). The antennae are relatively long; the index of P/A in the subgenera *Culex* and *Neoculex* is less than one (in *Barradius* it is only 0.5-0.6), but in *Lutzia* it is greater than one. The posterior section of the pharynx is slightly enlarged, and it is appreciably inferior in length to the anterior section (Fig. 2); more seldom both sections are of approximately the same length. The oral cavity has an arcuate row of stick-shaped structures or else it has a patch of scaly denticles (the "armament" of the oral cavity can be successfully examined only on well lighted preparations). The eyes on the upper side are approximated along a significant distance; they sometimes almost adjoin; the frons, as a rule, does not have a longitudinal suture. More seldom the frons is somewhat wider (*Barradius*, *Neoculex*).
7. *Culiseta* Felt. Species of the subgenus *Culiseta* are, on the basis of head structure, close to *Ochlerotatus*, but they are distinguished by the elongated form of the frons, which is narrow in the anterior half and greatly widened in the posterior half. Species of the subgenus *Culicella* are,

on the basis of head structure, closer to *Culex* - the frons is very narrow and long. On the lower side of the head, the eyes in *Culicella* are approximated; in *Culiseta* they are further apart. In both subgenera the oral cavity is unarmed (i.e., without denticles); the palps, as a rule, have five segments. *C. (Allotheobaldia) longiareolata* Mcq. stands alone: this species is characterized by the presence of a bundle of scales on the first segment of the antenna and by relatively longer palps.

A more detailed description of the subgenera and species will comprise the contents of subsequent works. At the present time we are unable to claim that females of all mosquito species can be determined by preparations of the head. It is possible to determine some species with complete certainty -- others with varying degrees of reliability. The devised method may prove to be useful as a supplement of generally accepted methods, especially in determining specimens whose scales have fallen off.

The study of details of head structure, apart from its applied significance, can also be of theoretical interest. At the present time, a description of the structure of the male genitalia is obligatory in describing various taxons of mosquitoes. It is possible that in the future, the description of peculiarities of head structure in the female (by means of microscopic preparations) will also be recognized as an essential part of the description of genera and species of *Culicidae*.

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Fig. 1

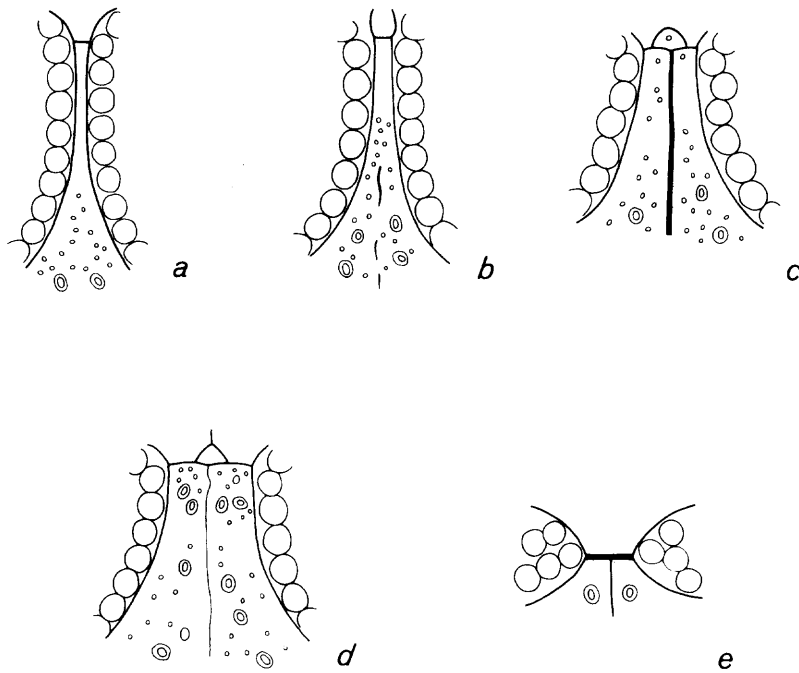


Fig. 1. The frons. a. *Culex pipiens* L.; b. *Culiseta alaskaensis* Ludl.; c. *Aedes punctor* Kirby; d. *Aedes caspius caspius* Pall.; e. *Uranotaenia unguiculata* Edw.

Fig. 2

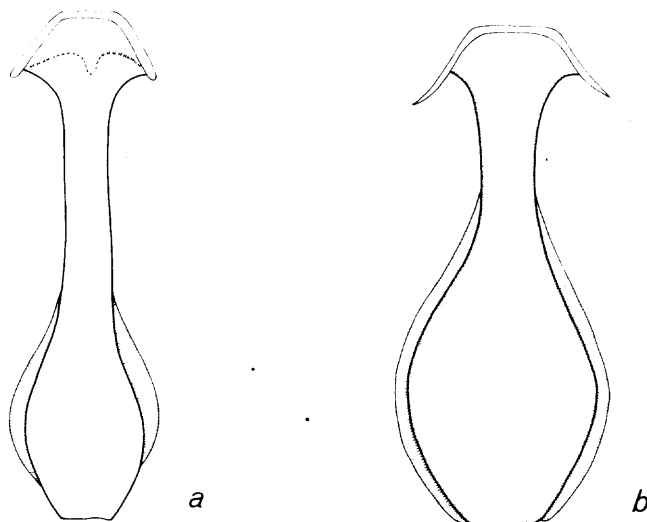


Fig. 2. The pharynx and oral cavity (diagram). a. *Culex pipiens* L.; b. *Aedes communis* Deg.