

## ULTRASTRUCTURE OF MAXILLARY TEETH AND SENSILLA OF THREE ANOPHELINE SPECIES OF MOSQUITOES (INSECTA: DIPTERA: CULECIDAE)

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### Abstract

The maxillary stylets and maxillary palps of three species of mosquitoes belonging to genus *Anopheles*, viz., *Anopheles culicifacies* Giles, *Anopheles fluviatilis* James and *Anopheles subpictus* Grassi have been studied using scanning electron microscope. The studies reveal the presence of lateral teeth and absence of mesial teeth on the maxillary stylets. The number, shape and size of lateral teeth differ in different species. The maxillary palps have been found to bear various capitate pegs which also shows differences in their shape and size. These discriminatory structures are, however, too minute to be studied under ordinary light microscope. The scanning of their surfaces under electron microscope brings to light the true structure of the teeth and the pegs and makes it possible to compare its structure in different species and genera. The studies so far made on the ultrastructure of maxillary teeth and maxillary sensilla largely pertain to the species available outside India.

### Key words

*Anopheles culicifacies*, *A. fluviatilis*, *A. subpictus*, ultrastructure, scanning electron microscopy, maxillary palps, maxillary stylets, Punjab

### Introduction

Mosquito-borne diseases are responsible for major loss of human life in different parts of the globe. It is believed that malaria alone has killed more people than all the wars man has fought so far. Mosquitoes play a crucial role in our environment and being an integral part of our biodiversity, clarity in taxonomy is emphasised.

The existence of sibling species has made the task of epidemiologists and entomologists very difficult. In addition to this, a major proportion of *Anopheles* species shows morphological variations which poses great difficulty in their proper identification and speedy separation. Therefore, in order to establish the existing morphological criteria and searching hidden taxonomic characters, the studies have been conducted on maxillary stylets and palps of three species of mosquitoes belonging to genus *Anopheles*. During the present investigation it was found that the number arrangement, shape and size of lateral teeth and sensilla are of great taxonomic value in these

three species.

Hence, keeping in view the importance of scanning electron microscopy, the work has been taken up on such aspects of mosquitoes which reveal additional and more reliable discriminatory characters for the identification of different species.

### Material and Methods

The three species belonging to genus *Anopheles*, viz., *A. subpictus* Grassi, *A. culicifacies* Giles and *A. fluviatilis* James, were collected from various resting places such as cattle sheds, rice fields, water coolers, walls of houses and other man-made structures in Punjab.

For scanning electron microscopic studies, the specimens were first fixed in 3% glutaraldehyde, dehydrated in graded series of alcohol, air-dried mounted on specimen stubs, sputtered with gold and scanned under JSM 6100 scanning electron

microscope. The terminology used is adopted from Lee (1974), Lee and Craig (1983), and, McIver and Chariton (1970). The standard error has been calculated by using the following formula.

$$S_M = \frac{\delta}{\sqrt{N}}$$

$$\delta = \frac{\sqrt{\sum x^2}}{(N-1)}$$

## Results and Discussion

Different types of structures and sensilla on the maxillary stylet and palps in three species of mosquitoes belonging to genus *Anopheles* are described below.

### Maxillary stylet

(Figs. 1-4). Our observations show that the two maxillary stylets are flattened structures and are located near the base of the proboscis but over most of the length of the proboscis they lie side by side and occupy a ventrolateral position at rest. The inner swollen edges of these stylets surround the midrib of the hypopharynx while their outer parts are flattened and overlap other stylets. The inner edge of each stylet is armed with posteriorly directed teeth in its distal part which are usually referred to as lateral teeth. The number, shape as well as size of these teeth are taxonomically significant and show variations in different species of *Anopheles* as shown in Table 1 .

An examination of lateral teeth on the lacinia of different species shows the presence of lateral teeth and absence of mesial teeth in all the three species of genus *Anopheles* included in this study. The number of lateral teeth varies between twelve to sixteen. The lacinial teeth have been studied in a number of species under different genera. Lee and Craig (1983) observed lacinial teeth in three species of genus *Anopheles*, viz., *A. faurauti*, *A. merus* and *A. stephensi*, the number of which ranges from 13-19.

The studies by Hudson (1970) and Lee (1974) show that the mesial set of teeth are missing in *Wyeomyia smiths* (Coquillett) as well as in *Aedes simulans* (Newstead & Carter). However, according to Lee and Craig (1983) who have studied a large number of species, the mesial teeth are present in all the species of *Aedes* and two species of *Armigeres* studied by them. According to them, the mesial teeth are present in the representative species of five other genera but absent in genera *Anopheles*, *Culex* and four other genera.

It follows from the above that the genera *Aedes* and *Armigeres* always have both sets of teeth on the lacinia, whereas *Culex* and *Anopheles* always lack the mesial teeth.

### Maxillary palps

(Figs. 5-8). The maxillary palps are well developed parts of the maxillae. The segments of the maxillary palps are covered over with four different kinds of structures. These are categorized as microtrichia, scales, sensilia chaetica and capitate pegs (McIver & Chariton, 1970).

The microtrichia are short, non- innervated cuticular projections and densely cover all the segments of the maxillary palps (Fig. 5). These are of three subtypes, viz., long straight, short curved and straight with swollen bases. The last type of microtrichia are missing in some of these species. The scales are present on the basal four segments of palpus with higher concentration on segment four. Each scale is broad and elongated and consists of a pedicel and a squama. The sensilia chaetica are innervated structures arising from sockets and are pointed at their apices.

These sensilia are found on all the five segments but their number is more on the fourth segment. The capitate pegs, which are also innervated structures, occur mainly on the distal half of the ventral surface of segment four of the male anophelines and on second to fourth in female anophelines. The capitate pegs show variations in their size and shape in different species.

The details of the length and shape of these sensilia in different species are given in Table 2.

Table 1. Various parameters of maxillary teeth in three species of genus *Anopheles*\*

Species	Number of teeth	Size (µm)	Distance between adjacent teeth (µm)	Shape
<i>Anopheles culicifacies</i>	16	0.3-1.8	1.4-6.2	1-12 blunt, 13-15 somewhat reduced, 16 reduced
<i>Anopheles fluviatilis</i>	12	0.5-2.3	2.5-8.8	All blunt
<i>Anopheles subpictus</i>	14	1-3	2-8.5	1-11 blunt, 12-14 somewhat reduced

\* Five observations for each species



Figure 1. Maxillary stylet in *Anopheles culicifacies* Giles

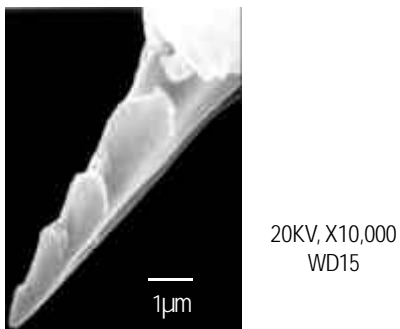


Figure 2. Maxillary teeth in *Anopheles culicifacies* Giles

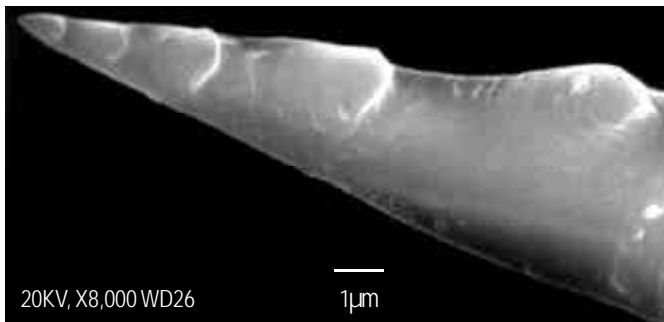


Figure 3. Maxillary teeth in *Anopheles fluviatilis* James

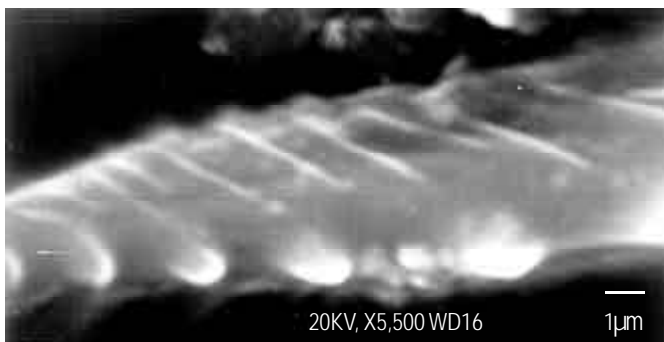


Figure 4. Maxillary teeth in *Anopheles subpictus* Grassi

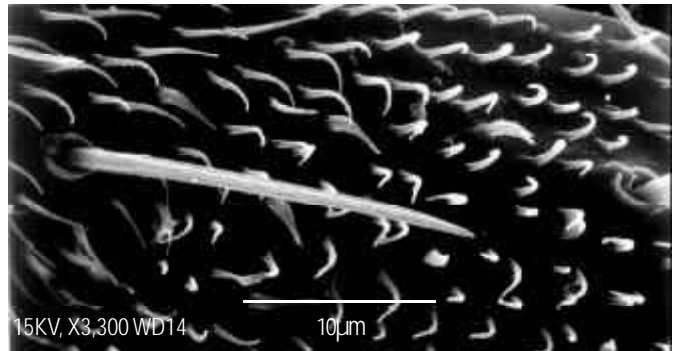


Figure 5. Sensilla chaetica and microtrichia on maxillary palps

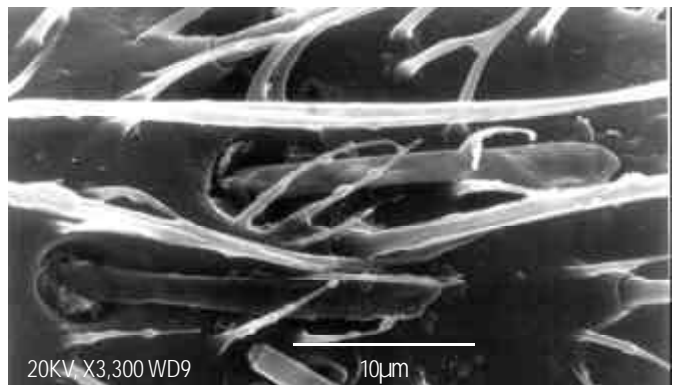


Figure 6. Capitata peg in *Anopheles culicifacies* Giles

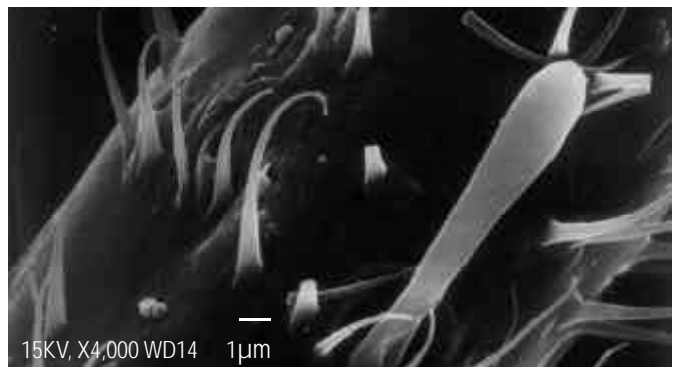


Figure 7. Capitata peg in *Anopheles fluviatilis* James

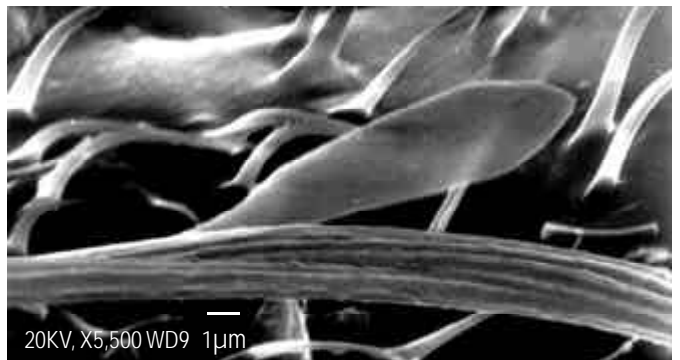


Figure 8. Capitata peg in *Anopheles subpictus* Grassi

**Table 2. Disposition of capitata pegs on the maxillary palps of three anopheline species\* of mosquitoes.**

Species	Shape	Mean length	Standard error
<i>Anopheles culicifacies</i>	Roughly club-shaped	20.7µm	1.0
	Club-shaped		
<i>Anopheles fluviatilis</i>	Club-shaped	15.6µm	0.8
<i>Anopheles subpictus</i>	Spatula-shaped	14.7µm	0.8

\* Five observations for each species.

Previous reports also reveal the presence of the same four types of structures on the maxillary palps as observed in the present study.

However, McIver and Siemicki (1975) have also reported one additional sensillum, the campaniform sensillum, one each on maxillary palps of three species of *Anopheles*. The campaniform sensillum has not been observed in species studied by the authors. The exact size of the sensillum has not been mentioned by McIver and Siemicki (1975). It is possible that either the sensillum is too minute to be observed because of the dense covering of scales and sensilia chaetica or it is diffused and hardly visible on the outer surface. In addition to anopheline species work has been carried out on four species of *Culex* and two species of *Aedes* by McIver and Chariton (1970), and McIver (1971, 1972) who have reported the same four types of structures observed by the authors but none of them have mentioned the shape and size of capitata pegs present on second to fourth segment of the maxillary palps.

The length of capitata pegs in the species studied falls within the range of 14-20.7µm as against the length range of 20-25µm in other species of this genus (McIver & Siemicki, 1975). Hence,

the size and shape of capitata pegs can be used in the separation of species.

It follows from the above observation that the shape of capitata pegs differs in different species of *Anopheles*.

Therefore, the scanning of maxillary structures prove to be of value in the taxonomy of mosquitoes. Even the species of a complex or sibling species can be discriminated on the basis of these magnified maxillary structures.

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