# STUDIES ON ULTRASTRUCTURES OF EXTERNAL GENTALIA IN SPECIES ANOPHELES (CELLIA) PULCHERRIMUS THEOBALD AND ANOPHELES (CELLIA) ANNULARIS VAN DER WULP

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### ABSTRACT

External genitalic attributes are highly species specific in different species of genus *Anopheles* Meigen. These features have already been studied by eminent taxonomists. However, nobody has studied ultra structures present on male genitalia of species *Anopheles (Cellia) pulcherrimus* Theobald and male and female genitalia *Anopheles (Cellia) annularis* Van der Wulp. These structures have been studied and illustrated in detail in present communication.

**Keywords:** Scanning Electron Microscope, Anopheles Pulcherrimus, Anopheles Annularis, Male and Female Genitalia

# INTRODUCTION

The plains of Punjab are low lying, wet and often inundated. Malaria in Punjab is seasonal and unstable, with epidemics recurring at approximately eight year intervals (Christophers, 1911; Yacob and Swaroop, 1945, 1946). Both species i.e. *Anopheles (Cellia) pulcherrimus* Theobald and *Anopheles (Cellia) annularis* Van der Wulp breeds in groundwater pools, tanks, ponds, ricefields and stream with green vegetation. The former species is not a vector, whereas the later has been reported as secondary vector of malaria in India (Nagpal and Sharma, 1995). Both the species can be identified morphologically and male and female genitalia are also species specific. The female genitalia of anophelines have been little studied taxonomically.

During the recent collection-cum-survey tours both the above said species were procured from different localities of Punjab and found to be very common. Scanning Electron Microscopic (SEM) studies have been conducted on the genitalic structures of these species for the first time. Many additional and new characters have come to light which can be incorporated in the detailed diagnosis of these species. These new attributes may further help to resolve the problem of species complex in these species.

### MATERIALS AND METHODS

In the course of mosquito survey, adults of both the species were collected from different localities of Punjab state. Specimens were collected by using test tube & torch method and oral aspirators. For SEM studies, method given by Lee and Craig (1983) has been followed. For genitalic attributes, last three segments of both male and female specimens were dissected with the help of forceps. These were first boiled in 10% KOH for 10-15 minutes, washed with water several times, air-dried and mounted. After mounting, these were sputter-coated with gold and scanned with JSM-6610LV scanning electron microscope. 5-7 specimens were used for each investigation.

Taxonomic keys developed by Puri (1931), Robert and Ross (1943), Nagpal and Sharma (1995), Amersinghe *et al.*, (2002) for identification of collected anopheline specimens was used. The terminology for various morphological characters and abbreviations has been adopted given by Harbach and Knight (1980, 1981).

### **RESULTS AND DISCUSSION**

General Structure of Male Genitalia: The arrangement and number of parabasal and other differentiated spines on the coxites are characteristics for different subgenera of *Anopheles*. In subgenera *Cellia*, two (occasionally one or three) large parabasal spines arising from eminences found, in subgenera

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# **Research** Article

*Anopheles* there are 4-5 smaller spines not on raised area and internal spine of subgenera *Anopheles* is usually absent (Reid, 1968). The shape of dististyle is represented by an oblong, conical, sickle-shaped bent formation. Study under SEM demonstrated that it is uniformly bent rather than bent apically, as it was mentioned by some authors (Mohrig, 1969).

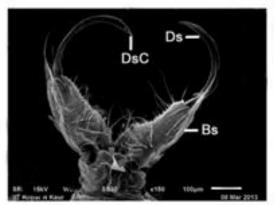


Figure 1:Male genitalia (whole view)

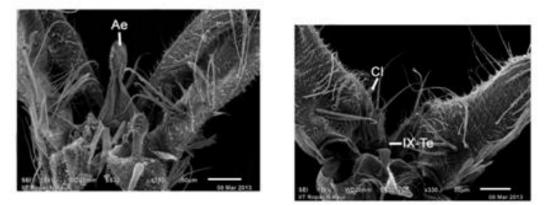


Figure 2 and 3: Phallosome

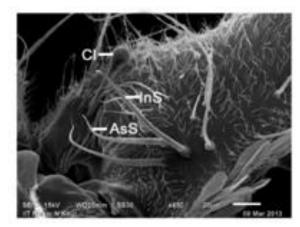


Figure 4: Different types of spines on basistyle

Genitalia of Anopheles (Cellia) pulcherrimus Theobald

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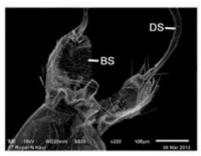


Figure 5: Male genitalia

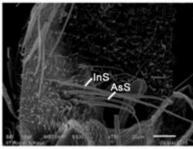


Figure 7: Different types of spines on basistyle

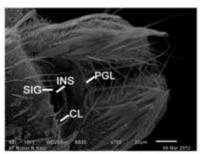


Figure 9: Female terminalia

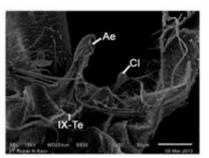


Figure 6: Phallosome (Magnified)

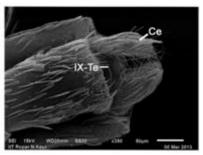


Figure 8: Female genitalia

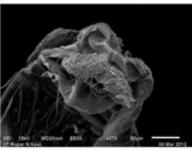


Figure 10: Dorso-caudal view

Genitalia of Anopheles (Cellia) annularis Van der Wulp

# Anopheles (Cellia) Pulcherrimus Theobald

**Male Genitalia** (Figure 1-4): Shape of dististyle is same as it was defined by Mohrig (1969) and claw present at tip. Basistyle are covered by several kinds of setae and with numerous ling and short microtrichae. One internal spine which is very long and thin as compared to all other spines. Three accessory spines and one parabasal spine with parabasal lobe are present, all are almost same shape and size. Claspettes with two pair of long and thin spines on ventral lobe which is slightly V-shape and covered with minute scales. Shape of ventral lobe was different in all species studied, so it is species specific character and clasepettes are very prominent with spoon-shape (lower stem like part is thin with rounded upper portion). Aedeagus looks like pyramid.

# Anopheles (Cellia) Annularis Van der Wulp

**Male Genitalia (Figure 5-7):** Structure of this species is almost same as *Anopheles pulcherimus* except some differences. Dististyle with thin and short spine near tip. Claspettes without spines on both ventral and dorsal lobe, shape of vental lobe is almost U-shape and claspettes are prominent with spatula-shape (lower part is not very thin as in above said species) and aedeagus pyramid like as in *Anopheles pulcherrimus*.

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Female Genitalia: Female genitalia of *Anopheles pulcherrimus* has not been studied in present communication, only *Anopheles annularis* studied in detail.

**Dorsal View of Female Genitalia (Figure 8-10):** Female genitalia having cerci, Postgenital lobe (PGL), IX-tergum, insula, cowl and sigma. A pair of cerci covered with several types of long setae from inner side and with short setae on upper side. Postgenital clearly visible bearing pair of long and thin seta arising from its tip. Shape of IX-tergum very species specific and in the present species it is almost band like, upcurved with dentate structure i.e. IX- tergum with numerous teeth like structure at margins which is not found in any other species studied by us. Insula slightly upaward in middle; Cowl slightly downward at end and sigma is very fine structure, taper at apex and both slightly joint at apex.

### Conclusion

As highlighted in the introduction new taxonomic attributes like shape of dististyle and basistyle, number of internal and accessory setae, shape of claspettes, shape of aedeagus, shape of IX-tergum, shape of insula and cowl have been studied in detail. These features can be used to revise the characterization of species *An. pulcherrimus* and *An. annularis*. This will not only update the status of these taxa but also these structures will be taxonomically significant for identification of these species.

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### Abbreviations

Ae (Aedeagus), AsS (Accessory spine), Basistyle (Bs), Ce (Cerci), CL (Cowl), Cl (Claspettes), Ds (Dististyle), INS (Insula), InS (Internal spine), PGL (Post Genital Lobe), SIG (Sigma), IX-Te (IX-Tergum).

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