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Two *Armigeres* species of Manipur, the easternmost Northeast India

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Abstract

In the endeavour for diversity assessment of mosquitoes of Manipur, an attempt has been made to identify vector mosquitoes. In quest for proper investigation of the vector mosquitoes two *Armigeres* spp are reported. They are *Armigeres (Armigeres) obturbans* (Walk.), 1860 and *Armigeres (Armigeres) aureolineatus* (Leicesyer), 1908. They are systematically analysed in the present study. The two species are all identical externally from larvae to adult individual through pupal stages. But microscopic features are quite different in these species. Main differences could be found in the male hypopygium particularly the number of teeth along stylar. In *Armigeres obturbans* the number of teeth is 17 to 19 while in *Armigeres aureolineatus* it is only 15 to 16. The two species are from Jiribam and Moreh where the annual temperature is much higher than other parts of Manipur. It will be quite safe to argue that the species might inhabit the area with higher temperature. *Armigeres aureolineatus* is a new report from the state of Manipur as the available literatures. The species *Armigeres obturbans* could not be synonymized with *Armigeres subalbatus* as reported from Manipur. The ascertaining of the two species with the molecular work will the future perspective and exploration more species of the genus will have enriched the diversity of the state and India as whole.

Keywords: Manipur. *Armigeres obturbans*, *Armigeres aureolineatus*, Moreh, Jiribam, annual temperature

1. Introduction

Out of the total 3, 540 species recognised mosquitoes in the world ^[1] which are divided into two subfamilies and 112 genera, 393 species divided among 49 genera and 41 subgenera are reported from India and subfamily Anophelinae contains 61 species in one genus followed by subfamily Culicinae with 332 species in 11 tribes and 48 genera ^[2]. All these data support the fact that India ranked fifth in terms of mosquito biodiversity after Brazil, Indonesia, Malaysia and Thailand ^[3]. Mosquitoes are usually popular for their nuisance and vector of many diseases despite their role in ecology more precisely in food chains. They are divided into carriers and non-carrier. In India, 31 species are currently recognized for transmitting various mosquito-borne pathogens ^[2]. Among the carriers *Armigeres* are also important. The mosquito *Armigeres subalbatus* is a vector of filarial worms with medical importance, *Brugia pahangi* ^[4] and with veterinary importance, *Dirofilaria repens* ^[5]. *Ar. subalbatus* has also been found infected with Japanese Encephalitis Virus, JEV ^[6] and has been incriminated as a JEV vector in settings without rice fields ^[7]. Regarding blood feeding, it has been reported that this mosquito was able to experimentally feed on humans, mice, chickens and reptiles, but not amphibians ^[8], thus reinforcing the view that this mosquito can transmit zoonotic pathogens ^[9]. In India 20 species of *Armigeres* from two subgenera *Armigeres* and *Leicesteria* ^[2] were recorded and from Manipur 12-11 *Armigeres* and 1 *Leicesteria* ^[10] from different parts of Manipur were recorded. But systematic studies of available species had not been put up. The present study focuses on the morphological systematic studies of two species of *Armigeres* from Manipur and their distinctive features were analysed for the first time.

2. Materials and Methods

Larval stages of mosquito were collected from two breeding ground of Jiribam keithel (Bazar)- 24°48'00"/E 93°7'12" and Moreh (Tengnoupal District)- N 24°21'06"/ E 94°20'32", Imphal West during 24th April and 24th June, 2018. Both the places are humid tropical characterized

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but short winter and long summer with heavy rainfall. The immature larval stages were reared with appropriate foods in beakers till the emergence of adults and identification of the species were done from larvae, pupae and adults male individuals. The identification keys followed in present studies included: Barraud [11], Website of Walter Reed Biosystematics Units [12].

3. Results

The general features of the *Armigeres* of the two species from two extremes of Manipur viz., Jiri Keithel and Moreh Hill (Fig. 1), were quite similar and no external features could differentiate the two. The features from larval studies revealed that the average of 10 specimens in the *Armigeres obturbans* was 12 mm while that of the *Armigeres aureolineatus* was only 9 mm (Fig. 2). The mentum and premandibles are quite similar. Mantums of the species are crown shape with 8 lateral teeth in *Ar. obturbans* and only 6 in *Ar. aureolineatus* (fig. 2 A and B) with the identical premandibles in these species. The trumpet structure was roughly L shaped quite identical in two species only visible differences were tracheation. In *Ar. obturbans* the tracheation was limited to the basal while it was

extended upward to near the tips of the trumpet (Fig. 3 A and B). Another prominent differences were the number of the seta present on the 9-VII-VIII which were strongly developed and plumose type. The number of seta on VIII was 13 in the species, the seta on VII was 7 (seven) in but only 6 (six) in *Ar. aureolineatus*. The number of setae on 9-VI was 3 (three) in the *Ar. obturbans* and only two in *Ar. aureolineatus* which were stouter and longer than of *Ar. obturbans* (Fig. 3 C and D) in males. The adults were quite decorated particularly the ventral abdomens. The male *Ar. obturbans* showed somewhat white blunt laterally while they were quite pointed in *Ar. aureolineatus* (Fig.4 A and D). The male gonostylus with distal comb-like process was the characteristics of the genus *Armigeres*. The comb occupies almost 50% of the length of gonostylus in *Ar. aureolineatus* but it is little less than (47%) in *Ar. obturbans*. the comb also different in these species-17 - 19 in latter while it was 15-16 in former (Fig. 4 G and H). The species delimitation is done on the basis of the comb numbers. The gonocoxite were quite identical in these species (Fig. 4 B and E). The phallosomal teeth spines were sharper in *Ar. aureolineatus* than slightly blunt, *Ar. obturbans* and also ridges were longer in *Ar. obturbans* (Fig. 4 c and F).

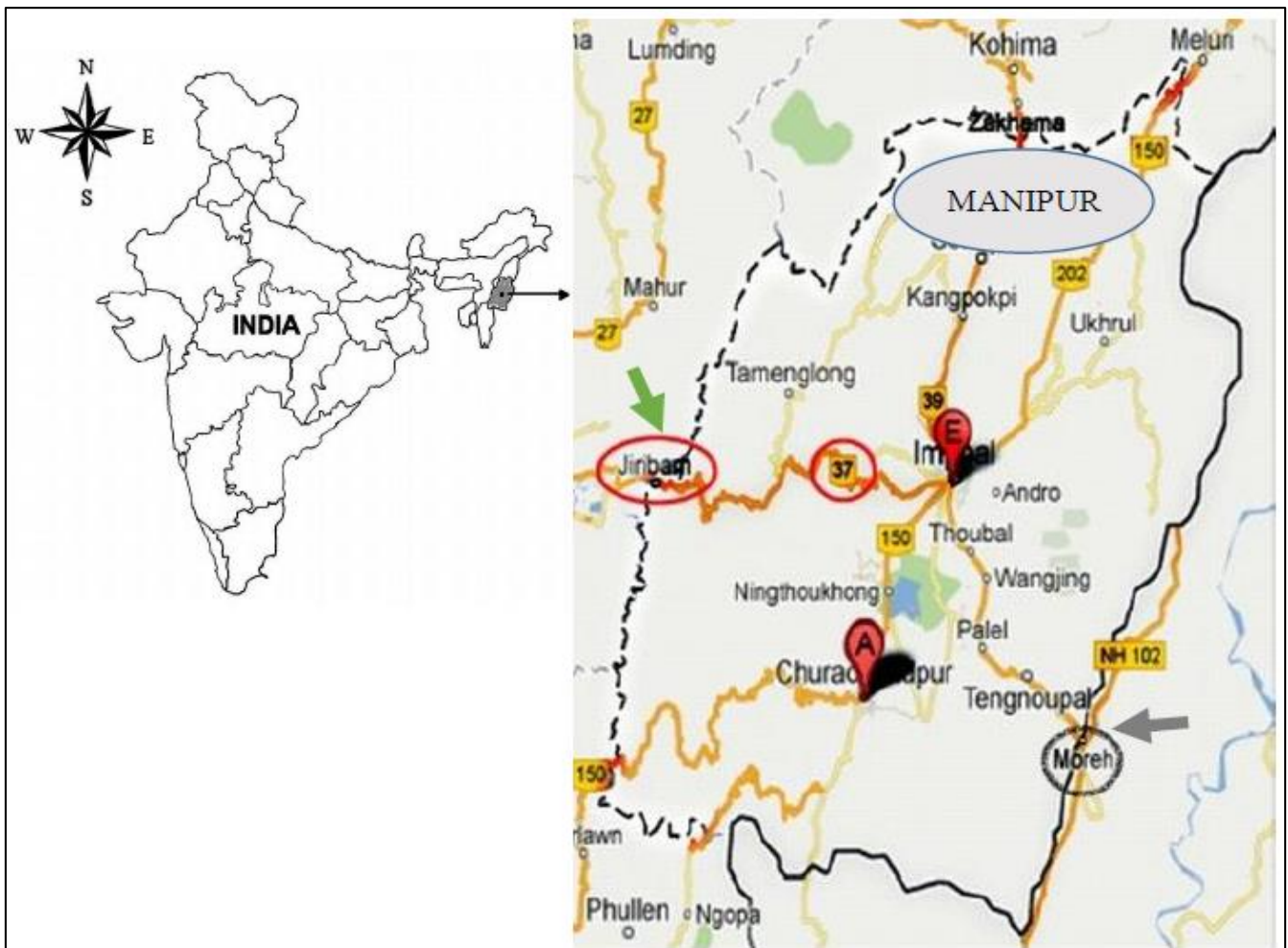


Fig 1: The location sites in Manipur in India. The arrowed locations are the study sites.

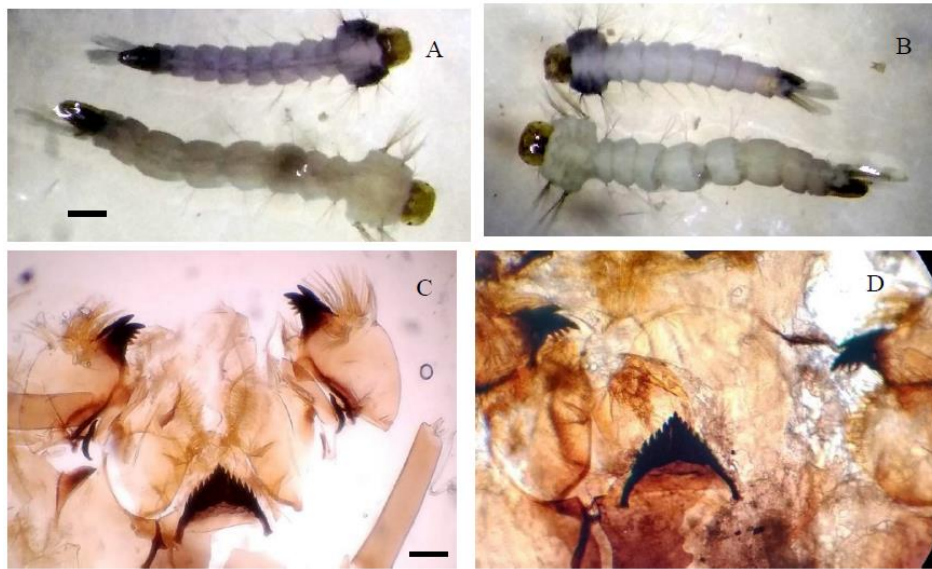


Fig 2: The larval mentum and pre mandible of the *Armigeres obturbans* (A) and *Armigeres aureolineatus* (B). Bar represents 1 mm.

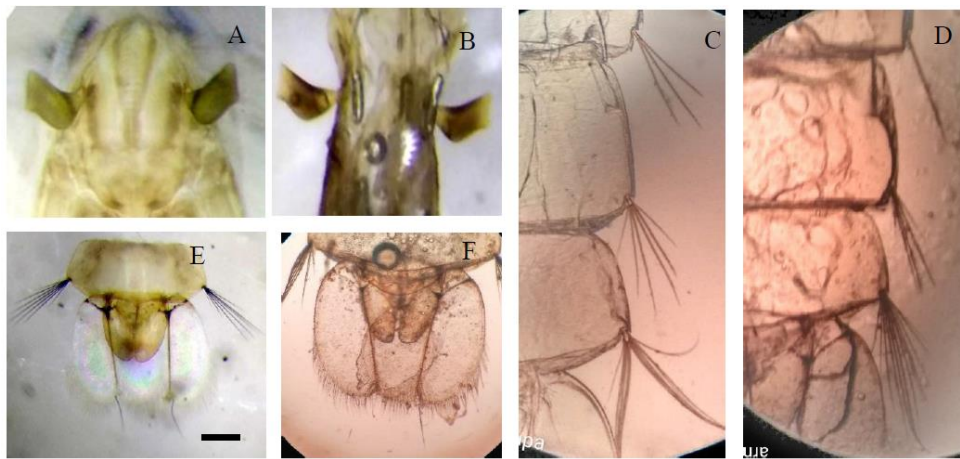


Fig 3: The pupal features of *Armigeres obturbans* (A, C and E) and *Armigeres aureolineatus* (B, D and F). Bar represents 1 mm.

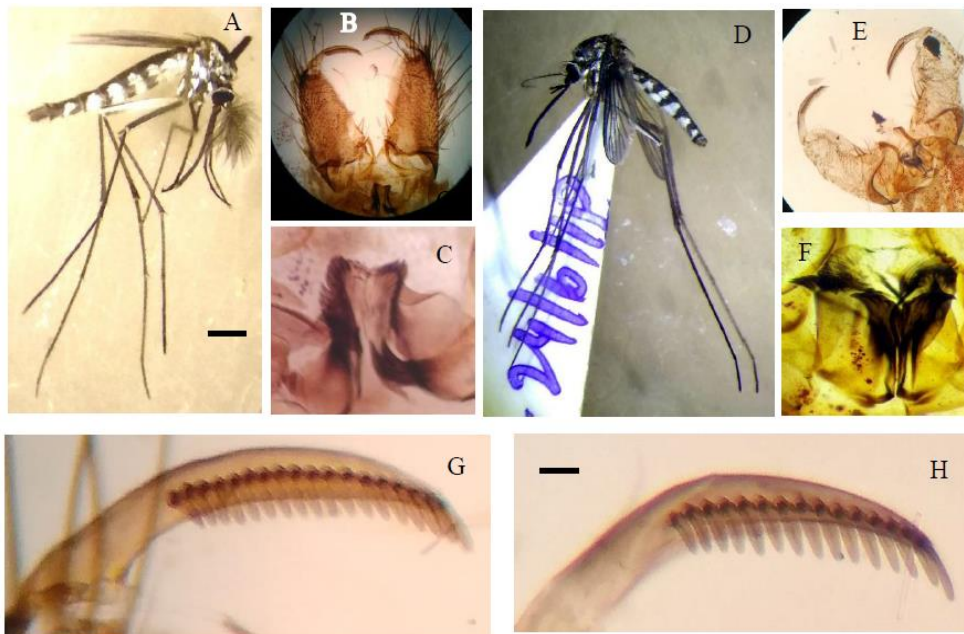


Fig 4: *Armigeres obturbans*: Male adult (A), hypopygium (B), phallosome (C), and *Armigeres aureolineatus* adult female (D), hypopygium (E), phallosome (F). the Gonostylar teeth of *Armigeres obturbans* (G) *Armigeres aureolineatus* (H). Bar represents 1 mm.

4. Discussion

The two species reported here viz., *Armigeres aureolineatus* and *Armigeres obturbans* are quite distinct from each other on the basis of their larval, pupal and male adult features. The species delineation could be made from any stage of the insect. The study sites in the present studies are the area where the mean temperature of the regions are higher than the mean temperature of the state. So it is rather safe to conclude that the species are more or less adapted to higher temperature of the region from the studies. From Moreh, Rajput and Singh^[11] had reported *Armigeres obturbans*/*Armigeres subalbatus*, *Armigeres kuchingensis* Var. *durhami*/ *Aermigeres durhami*; *Armigeres theobaldi*. From Jiribam, Rajput and Singh^[11] had reported *Armigeres obturbans*/ *Armigeres subalbatus* and *Armigeres flavus*. Other species reported so far from the region other than *Ar. obturbans* might be from other parts of the region different from present location. Other important to discuss is the synonymizing of *Ar. obturbans* with *Armigeres subalbatus*^[11]. According to them *Armigeres obturbans* Walker, *J. Proc. Linn. Soc. Lond. Zool.* 4: 91; Barraud, 1934: 314-317; *Armigeres subalbatus* (Coquillett), La Casse and Yamaguti, 1950: 53; Knight and Stone, 1977:172. Right now we do not have any paper/literature to support or disprove the fact but recently Liu *et al.*^[13] discovered new virus from the *Armigeres obturbans* called Kadipiro Virus (KDP). KDV was first isolated from mosquitoes collected in Indonesia^[14]. KDV belongs to the genus Seadornavirus within the family Reoviridae^[15]. Five strains of this virus were isolated from *Culex tritaeniorhynchus*, *Anopheles sinensis* and *Armigeres obturbans* in Yunnan province in China in 2005. A 758 bp amplicon was obtained using KDV-specific polymerase chain reaction primers for the five strains^[16]. The nucleotide homology between the KDV Chinese isolates and KDV prototype (JKT-7075) was 90%, indicating that these five isolates are indeed KDV. This was the first report of KDV being isolated outside Indonesia^[17]. At present, there is no report that KDV is associated with human or livestock diseases. Qasim *et al.*^[18] also used *Armigeres obturbans* instead of *Ar. subalbatus*. Again in other studies *Ar. subalbatus* is highly cannibalistic^[19] and studies reported that this behavior help in reducing the density under unfavorable conditions. Here in our laboratory they consume only the cast skins by most of the larval stages. This could be due to differences in the species. From the above points it is evident that *Ar. obturbans* should not likely to be synonymized with *Ar. subalbatus*.

The present study reports the occurrence of *Armigeres aureolineatus* from Moreh hills is rather new as previously reported works. The similarities of larval features of this species with *Ar. obturbans* is stated by Barraud^[11] but only the differences in the length (Fig. 2). The adult hypopygium characteristics of the *Ar. aureolineatus* as stated by Barraud^[11]: *bl* with three strong spines. Style rather wide, with comb of 15 or 16 stout teeth along a little more than apical ½. All these features are in accordance with the present results except “a conspicuous tuft of hairs at apex coxite near base of style” which might be due to negligence in preparing the slides or absent at all. One or other the number of tooth is the delineating factor in this regards. The ascertaining validity of the two species in the present could be done through molecular studies in future.

5. Conclusions

The two species in the present studies are quite identical all features base on larvae, pupae and male adult features except some minute microscopic features. *Armigeres aureolineatus* is a new report from the state of Manipur as the available literatures. The species *Armigeres obturbans* could not be synonymized with *Armigeres subalbatus* as reported from Manipur. The ascertaining of the two species with the molecular work will the future perspective and exploration of more species of the genera will be enriched the diversity of the state and world as whole.

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