ILLUSTRATED KEYS TO SPECIES OF CULEX (CULEX) ASSOCIATED WITH JAPANESE ENCEPHALITIS IN SOUTHEAST ASIA (DIPTERA: CULICIDAE)

R. REUBEN¹, S. C. TEWARI¹, J. HIRIYAN¹ AND J. AKIYAMA²

ABSTRACT. Illustrated keys are presented for identification of female mosquitoes of 19 genera occurring in Southeast Asia, five subgenera of Culex, and 16 commonly encountered species of Culex (Culex), nine of which have been incriminated as vectors of Japanese encephalitis. Because members of the Culex vishnui subgroup, which includes some important vectors, are notoriously difficult to identify as adults, separate adult and larval keys are provided. Notes and distribution on individual species are included.

INTRODUCTION

Japanese encephalitis (JE) has become an important public health problem in Southeast Asia during the last two decades and has extended its range, including new areas in India and Nepal (Umenai et al. 1985). There is an urgent need for entomologists working in operational control programs to be able to identify the vectors of this flaviviral disease in order to target their efforts efficiently. Although several genera of mosquitoes have been found infected with JE virus, the vast majority of the isolations have come from species of Culex (Culex) Linnaeus, several of which are recognized as major vectors in the region (Pant 1979). Apart from Cx. quinquefasciatus Say, which is strongly anthropophilic, these species feed extensively on cattle, pigs, and birds, but only occasionally on man, and are thus important in maintaining reservoirs of the virus in nature. Pigs and birds are recognized amplifying and maintenance hosts of JE virus (Pant 1979).

Identification of Culex S.S. can be difficult in Southeast Asia. Since the publication of Barraud’s (1934) volume in the Fauna of British India series, two important monographs have been published: a revision of the genus Culex in Thailand by Bram (1967) and a revision of the subgenus Culex in the Oriental Region by Sirivanakarn (1976). These are not widely available and are perhaps too detailed for the field entomologist. In this paper we provide simple illustrated keys to the genera of mosquitoes, the subgenera of Culex, and the common Southeast Asian species of Culex S.S. Using these keys, field entomologists can distinguish vector species from the many culicines which resemble Culex S.S., notably species of the subgenera Lutzia Theobald, Culciomyia Theobald, and Lophoceraomyia Theobald.

There are 42 species of Culex S.S. in Southeast Asia (Sirivanakarn 1976). However, this key is restricted to 16 common species, mostly of known or potential importance in the natural cycle of JE (Carey et al. 1968, Sirivanakarn 1976, Amerasinghe et al. 1988). The remaining 26 species would not normally be encountered in routine collections, but users of this key should be alert to the occasional aberrant specimen that may be a species not included here and should refer to Sirivanakarn’s (1976) key for correct identification. The key can be used in all countries of the region shown in Table 1.

Females of the Cx. vishnui subgroup are difficult to identify as adult females. However, because of the importance of members of the subgroup in the epidemiology of JE, it is frequently necessary to identify adult females for virus isolation attempts and other

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Table 1. Distribution of common species of Culex (Culex) in Southeast Asia.

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<th>Species</th>
<th>Bangladesh</th>
<th>Cambodia</th>
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studies. In places where studies have already been carried out, three species, Cx. tritaeniorhynchus Giles, Cx. pseudovishnui Colless, and Cx. vishnui Theobald, commonly occur and can be satisfactorily separated using our key. However, the other three species of the subgroup, Cx. alienus Colless, Cx. perplexus Leicester, and Cx. whitei Barraud, may be locally common and therefore, when commencing work in a new area, it is desirable to check the species composition. Because larval characters are diagnostic, we have provided both larval and adult keys for the six species of this subgroup.

MATERIALS AND METHODS

Keys were prepared after examining specimens from the collections of the National Institute of Virology, Pune, and the museum of the Centre for Research in Medical Entomology (CRME), Madurai. Scientists of the CRME have used these keys in studies of the epidemiology of JE in ricefield ecosystems. The distribution of the 16 common species of Culex S.S. in the ricelands of Southeast Asia is shown in Table 1. The morphological terms and abbreviations used in the keys are illustrated in Figs. 1 and 2. Some of the illustrations were taken from the monographs of Harrison and Scanlon (1975:Fig. 3E), Mattingly (1971:Figs. 5, 15b), Sirivanakarn (1976: Figs. 17, 20, 35, 40), and Tanaka et al. (1979: Figs. 176, 179, 184, 194). The terminology used follows Harbach and Knight (1980), and abbreviations of generic and subgeneric names are from Reinert (1975).

NOTES ON SPECIES

Cx. bitaeniorhynchus subgroup. This is a group of large mosquitoes (wing 3.8–5.2 mm), with the anterior 0.7 of the scutum covered with pale beige, yellow, golden, or dark brown scales, in distinct contrast to the posterior 0.3 of the scutum. Two species are common throughout the region, Cx. bitaeniorhynchus Giles and Cx. infusa Theobald. There have been two isolations of JE virus in nature from the former in India (Rodrigues 1988), and the species feeds extensively on birds and pigs as well as on man (Christopher and Reuben 1971). It could, therefore, play an important role locally. Little is known about the vectorial potential of Cx. infusa, which has been confused with Cx. bitaeniorhynchus in the past. Both have wings heavily speckled with pale and dark scales, but the abdominal terga of Cx. bitaeniorhynchus are marked with apical pale bands only, whereas
**Fig. 1. Morphology of female mosquito.**

*Cx. infusa* has both apical and basal bands and apicolateral pale patches.

A single isolation of JE virus has been made in India from a third species, *Cx. epidesmus* Theobald (Banerjee et al. 1979), which appears to be seasonally common in parts of the northern state, Uttar Pradesh (R.S. Soman, personal communication), where JE epidemics occur. It possesses a distinctive apical pale area on the wing, and the apical abdominal terga are largely golden-yellow.

*Culex fuscocephala* Theobald. This species is an efficient vector of JE in Thailand (Gould et al. 1974). Isolations from wild-caught mosquitoes have been made from that country and in India (Reuben et al. 1988) and Sri Lanka (Amerasinghe et al. 1988). *Culex fuscocephala* may be confused with *Cx. quinquefasciatus* because of the absence of a pale band on the proboscis of both species, but *Cx. fuscocephala* is easily distinguished by the alternating pale and dark markings on the pleuron and absence of pale banding on the abdomen.

*Culex gelidus* Theobald. This species is considered to be one of the most important vectors of JE in Sri Lanka, Thailand, Malaysia, Vietnam, and Sarawak (Gould et al. 1962, Macdonald et al. 1967, Nguyen et al. 1974, Gingrich et al. 1992, Peiris et al. 1993). Rel-
Fig. 2. Morphology of mosquito wing.

At relatively few isolates have been made in India (Reuben et al. 1988). It can be distinguished from all species except Cx. whitmorei (Giles) by the pure white scaling on the vertex and the anterior portion of the scutum. The absence of pale scales on the fore- and midfemora separates it from Cx. whitmorei.

**Culex mimeticus subgroup.** This is a group of species with spotted wings. It is of no known medical importance, but species of the subgroup are likely to be encountered in foothill villages.

**Culex perexiguus Theobald** (formerly Cx. univittatus Theobald). This species extends as far as Myanmar (= Burma) from the eastern Mediterranean and Ethiopian regions, where it is a vector of West Nile virus (Habach 1988). It is not a vector of JE, but it is widespread, though not common. In our experience, it needs to be carefully distinguished from Cx. quinquefasciatus and Cx. pseudovishnui, which it superficially resembles. It differs from the former by the presence of a patch of pale scales on the postspiracular area and from the latter by the absence of a pale band on the proboscis. In addition, there is a median longitudinal pale stripe on the midfemur.

**Culex quinquefasciatus.** A few isolations of JE have been made from several countries, including India and Vietnam, but the species is regarded as a poor vector and not of importance in the natural cycle of disease (Sirivanakarn 1976). The characteristic features are the absence of a pale band on the proboscis and the presence of basal pale bands on the terga.

**Culex sitiens Wiedemann and Cx. annulirostris Skuse.** Separation of females of these two species is difficult. Both have hindfemora heavily speckled with dark and pale scales anteriorly. However, neither has any known medical importance in Southeast Asia (Sirivanakarn 1976). Culex sitiens is a common pest mosquito in coastal areas. Culex annulirostris is an Australasian and Pacific species that in our region only occurs on some islands of Indonesia and adjacent to Papua New Guinea. It has been found to be naturally infected with JE virus in Guam (Pant 1979)
and with Sindbis and Murray Valley encephalitis viruses in Australia (Doherty et al. 1963).

**Culex vishnui subgroup.** This group contains three important JE vectors. *Culex tritaeniorhynchus* is extremely common and widespread. It has been incriminated as a major vector in India, Sri Lanka, Thailand (Rodrigues 1984, Leake et al. 1986, Amerasinghe et al. 1988, Gingrich et al. 1992), Sarawak (Macdonald et al. 1967), and outside the region in Japan, Korea, and Taiwan (Pant 1979). It can be easily identified by the dark brown scaling on the vertex and scutum, the accessory pale patches basal to the pale band on the ventral surface of the proboscis, and the narrow apical dark ring on the hindfemur. *Culex vishnui* is also extremely common and is known to be a vector in India, Thailand, and Taiwan (Carey et al. 1968, Cates and Detels 1969, Leake et al. 1986). *Culex pseudovishnui* has been implicated in southern India and Sri Lanka (George et al. 1987, Peiris et al. 1993). The role of the latter two species in the transmission of JE virus in other countries of Southeast Asia has not been established, partly because of the difficulty of telling them apart as adult females. *Culex vishnui* is a very variable species, with heavy speckling of pale scales on wings and femora in many specimens from southern India to none in parts of Southeast Asia. The scutal scaling varies from pale or golden to beige and light brown. There is a poorly contrasted dark stripe on the anterior dorsal surface of the hindfemur. *Culex pseudovishnui* typically has pale to golden scaling on the scutum, often forming a definite pattern, and the dark stripe on the anterior surface of the hindfemur is generally sharply contrasted with the pale areas. However, there is an overlapping of characters in a proportion of specimens. The larvae of these species are distinctive, and identifications should always be confirmed by individual rearings before starting studies in new areas. In India it was possible to identify correctly all except 12% of the adult catch (Reuben 1969).

**Culex whitmorei.** Several isolations of JE have been made from this species in India and Sri Lanka (Carey et al. 1968, Peiris et al. 1993), where it may play a role as a secondary vector. This species is similar to *Cx. gelidus*, having pure white scales on the vertex and scutum, but the pale scaling extends posteriorly to the prescutellar space and scutellum, and there is a speckling of pale scales on the fore- and midfemora.

### KEY TO THE GENERA OF MOSQUITOES IN SOUTHEAST ASIA (Females)

1. Proboscis rigid, apical half more slender and bent downward; posterior margin of wing emarginate just beyond CuA (Figs. 3, 4) ................................................................. *Toxorhynchites*  
   – Proboscis and wing otherwise ................................................................. 2

![Fig. 3.](image)

![Fig. 4.](image)
2(1). Scutellum evenly rounded; palpus about equal to the length of proboscis (Figs. 5, 6); abdomen with sterna and usually terga largely or wholly devoid of scales ........................................... Anopheles
   Scutellum trilobed (Fig. 1); palpus short; abdominal sterna and terga with dense, uniform covering of scales ........................................... 3

3(2). Tip of proboscis swollen, upturned, and hairy (Fig. 7) ........................................... Malaya
   Tip of proboscis sometimes swollen, otherwise unmodified ........................................... 4

4(3). Scutum with double median longitudinal stripe of broad, flat, usually white or silvery scales; prespiracular setae present; postspiracular setae absent ........................................... Topomyia
   Without this combination of characters ......................................................... 5

5(4). Upper calypter bare; vein IA reaching wing margin at or before level of base of crossvein mcu (Fig. 8) ......................................................... 6
   Upper calypter fringed; vein IA reaching wing margin well beyond level of base of crossvein mcu (Fig. 9) ......................................................... 7
6(5). Outstanding scales on outer half of wing with emarginate tips (Fig. 10) ........................... Hodgesia
- Wing scales normal (Fig. 8) ........................................... Uranotaenia

7(5). Prespiracular area with setae and sometimes scales (Fig. 11) ...................................... 8
- Prespiracular area bare (Fig. 12) ........................................... 9
8(7). Remigium and base of subcosta on ventral side without setae; at most 1, 2 small prealar setae (Fig. 13) ......................................................... \textit{Tripteroides} \\
- Remigium and base of subcosta on ventral side with setae; many prealar setae (Fig. 14) ........ \textit{Culiseta}

9(7). Antenna short, thick, and tapering; mid- and hindfemora with large tufts of suberect scales at apex (Figs. 15, 16) ................................................................. \textit{Aedoeomyia} \\
- Antenna and femora otherwise ................................................. 10

10(9). Mesopostnotum with a group of small setae; scutum covered with flat scales, usually with bright metallic reflections (Fig. 17) ......................................................... \textit{Heizmannia} \\
- Mesopostnotum and scutum otherwise ........................................ 11
11(10). Fore- and midtarsomere 1 distinctly longer than tarsomeres 2-5 together (Fig. 18) .... Orthopodomyia
   - Tarsomeres otherwise ................................................................. 12

Fig. 18.

12(11). Postspiracular setae present, or foreungues toothed, or both (Figs. 19, 20) .................... 13
   - Postspiracular setae absent; ungues simple (Figs. 21, 22) ................................ 16

Fig. 19. Fig. 20. Fig. 21.

13(12). Wing with broad, often asymmetrical scales; dark and pale scales intermixed (Fig. 23) .... Mansonia
   - Wing with normal scales ........................................................................ 14

Fig. 23.

14(13). Proboscis curved and laterally compressed (Fig. 24) ................................................... Armigeres
   - Proboscis slender and usually straight, never laterally compressed (Fig. 25) ............. 15
15(14). Upper calypter bare, alula with broad scales; overlapping silvery scales on interocular space; post-pronotum without scales (Figs. 26–28) .......................................................... Udaya
- Without this combination of characters .......................................................... Aedes

16(12). Antenna with flagellomere 1 about 3 times length of flagellomere 2; alula fringed with narrow scales (Figs. 29, 30); palpus less than 0.2 length of proboscis; scutellum with narrow scales only; wing with anterior fork cell as long as or longer than vein R_{2+3}; small or very small species .............. Ficalbia
- Without this combination of characters .......................................................... 17
17(16). Alula bare or with flat, decumbent scales (Fig. 31); proboscis usually swollen .......... *Mimomyia*
   - Alula with narrow fringe scales (Fig. 30) .............................................. 18

Fig. 31.

18(17). Hindtarsi with ungues very small and inconspicuous; pulvilli present (Fig. 32) ........... *Culex*
   - Hindtarsi with ungues not very small; pulvilli absent (Fig. 33); yellow or yellowish brown mosquitoes ................................................................. *Coquillettidia*

Fig. 32.

Fig. 33.

**KEY TO THE SUBGENERA OF CULEX IN SOUTHEAST ASIA (Females)**

1. Four or more strong lower mesepimeral setae present (Fig. 34); large species ........... *Lutzia*
   - Lower mesepimeral setae absent or 1,2 weak ones (Fig. 35); small to moderate species ............ 2

Fig. 34.

Fig. 35.
2(1). Pleuron with distinct scale patches on at least upper and lower mesokatepisternum and anterior mesepimeron (Fig. 36) ................................................................. Culex
- Pleuron without distinct scale patches (Fig. 37) ................................................................. 3

3(2). Acrostichal setae well developed on scutum (Fig. 38); small, dark species; usually without pale bands on abdominal terga .................................................................................. Eumelanomyia
- Acrostichal setae not developed except at extreme anterior end (Fig. 39) .......................... 4

4(3). Scaling on scutum sparse, rough in appearance; usually small, dark species ................... Lophoceraomyia
- Scaling on scutum very dense, smooth in appearance; vertex with broad scales forming a border to eyes (Fig. 40); moderate sized species ........................................................................ Culiciomyia

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3 Females of Lophoceraomyia generally are collected in association with males, which are distinguished by possessing modified tufts of scales and setae on one or more flagellomeres of the antenna. Their presence is useful in confirming the determination of females.
KEY TO THE COMMON SPECIES OF *CULEX* (*CULEX*) IN SOUTHEAST ASIA (Females)

1. Proboscis and tarsi without pale rings; 1 or 2 lower mesepimeral setae present (Fig. 41) .................. 2
   - Proboscis and tarsi with pale rings (Figs. 42, 43); lower mesepimeral setae absent ....................... 4

2(1). Abdominal terga without pale bands, occasionally a few indistinct bands on posterior segments; pleuron with striking pattern of dark and pale stripes (Fig. 44) ........................................... *fuscocephala*
   - Abdominal terga with basal pale bands; pleuron without striking pattern of dark and pale stripes (Figs. 45, 46) ................................................................. 3
3(2). Postspiracular area with pale scale patch (Fig. 47) ........................................... *perexiguus*
- Postspiracular area without pale scale patch (Fig. 48) ........................................... *quinquefasciatus*
4(1). Wing with pale spots on at least 2 areas of costa and 1 area of other veins (Fig. 49) . . . Mimeticus Subgroup
- Wing without distinct pale spots ................................................................. 5

5(4). Abdominal terga II–VIII largely yellowish or golden; pale yellow scales on apical portions of wing
veins C, R₁, R₂, R₃, and R₄,₅ forming apical pale area (Figs. 50, 51) ...................... epidesmus
- Abdominal terga II–VIII with dark and pale bands, or completely dark; wing tip without definite
pale area ............................................................................................................ 6

6(5). Wing heavily speckled with pale scales; abdominal terga II–VI with apical bands and/or apicolateral
pale patches and basal pale bands .................................................................... 7
- Wing without speckling of pale scales; abdominal terga II–VI with basal pale bands .............. 8

7(6). Abdominal terga II–VII with evenly broad apical pale bands and without apicolateral pale patches;
femora, tibiae, and wings heavily speckled with pale scales (Figs. 52, 53) ............ bitaeniorhynchus
- Abdominal terga II–IV largely dark, or with narrow apical bands, apicolateral yellowish patches,
and median basal pale bands or spots; terga V–VII with apical and basal bands, basal bands narrower
(Fig. 54); legs and wings lightly or moderately speckled ....................................... infula
8(6). Anterior 0.7 of scutum covered with pure white scales ............................................. 9
   - Anterior 0.7 of scutum covered with beige, yellow, golden, or dark scales .................. 10
9(8). Anterior surface of fore- and midfemora without speckling of pale scales; scales on prescutellar space, behind wing base, and on scutellum entirely dark; pale band on proboscis narrow, less than length of basal dark area (Figs. 55–57) ................................................................. gelidus
   - Anterior surface of fore- and midfemora speckled with pale scales; scales on prescutellar space, behind wing base, and on scutellum mainly pale; pale band on proboscis broad, as long as or longer than basal dark area (Figs. 58–60) ............................................................ whitmorei
10(8). Anterior surface of fore- and midfemora speckled with several pale scales, at least on apicodorsal surface ................................................................. 11
- Anterior surface of fore- and midfemora entirely dark .................................................. 12

11(10). Wing scales mainly dark; scutal integument dark; speckling of pale scales on femora contrasting sharply with dark-scaled area (Fig. 61) ........................................... sitiens, annulirostris
- Wing with few to several scattered pale scales; scutal integument light brown; speckling of pale scales on femora not contrasting sharply with dark-scaled area (Fig. 62) (see key to Vishnui Subgroup) vishnui

12(10). Erect scales on vertex and scales on anterior 0.7 of scutum entirely deep brown; proboscis often with accessory pale patches on ventral surface, proximal to median pale ring; hindfemur pale with distinct, narrow dark ring distally (Fig. 63) (see key to Vishnui Subgroup) ... tritaeniorhynchus
- Erect scales on vertex usually pale in center and dark laterally; scutum largely pale beige, yellow, or golden-scaled; proboscis without accessory pale patch; hindfemur with pale stripe on anterior surface contrasting well with dark areas (Fig. 64) (see key to Vishnui Subgroup) ................. pseudovishnui

KEY TO THE SPECIES OF THE VISHNUI SUBGROUP IN SOUTHEAST ASIA (Females)

1. Wing entirely dark-scaled ................................................................. 2
- Wing with few to several scattered pale scales, at least on base of vein C .......................... 3

2(1). Proboscis longer than forefemur; prescutellar space entirely pale-scaled (Fig. 65); integument of scutum light brown ................................................................. whitei
- Proboscis shorter than or as long as forefemur; prescutellar space usually with dark scales (Fig. 66); integument of scutum dark brown to blackish ....................................... perplexus
3(1). Proboscis usually with accessory pale patches or stripe on ventral surface; hindfemur pale with an apical dark ring (Figs. 63, 67); erect scales on vertex all dark ........................................... *tritaeniorhynchus*
  
- Proboscis without accessory pale patches or stripe; hindfemur with dark and pale areas, with or without speckling, but not with apical dark ring; erect scales on vertex mixed pale (cream, pale yellow, or beige) and dark, rarely all dark ........................................... 4

4(3). Anterior surface of hindfemur with pale stripe contrasting well with dark-scaled area (Fig. 64) ........ 5
  
- Anterior surface of hindfemur with pale stripe not contrasting with dark-scaled area (Fig. 62) ........ *vishnui*

5(4). Prescutellar area entirely covered with pale scales (Fig. 65) ........................................... *pseudovishnui*
  
- Prescutellar area mainly with dark scales (Fig. 66) ........................................... *alienus*

(Fourth-stage larvae)

1. Individual comb scales apically rounded (fan shaped), fringed with subequal spicules; siphonal tufts weak, with 2–5 branches (Figs. 68, 69) ........................................... *tritaeniorhynchus*
  
- Individual comb scales with a well-developed median apical spine and short, subequal lateral spicules (Fig. 70); siphon tufts variable ........................................... 2
2(1). Pleural area of thorax with spicules; about 22 comb scales; siphonal subventral tufts strong, with 6 or more branches (Figs. 70–72) ......................................................... vishnui
- Pleural area of thorax without spicules; subventral tufts of siphon with 5 or less branches ........... 3

![Fig. 70.]

3(2). About 40 comb scales; subventral tufts of siphon usually bifid, weak, short, length equal to or less than the siphonal width at their point of attachment (Fig. 73) ......................................................... whitei
- About 25 or fewer comb scales; subventral tufts strong, greater than siphonal width at their point of attachment ................................................................. 4

![Fig. 71.]

4(3). Proximal subventral tufts of siphon long, strong, greater than 3 times width of siphon at their point of attachment (Fig. 74) ................................................................. alienus
- Proximal subventral tufts less than 2 times width of siphon at their point of attachment ............ 5

![Fig. 72.]

![Fig. 73.]

![Fig. 74.]
5(4). Comb scales 4–8 (rarely up to 19), large, strong, spiniform, approximately in a single row; abdominal seta 6-V,VI with 3 branches; siphon lightly to strongly curved (Figs. 75–77) ....... *pseudovishnui*

- Comb scales 12–20, short, arranged in 2,3 irregular rows; abdominal seta 6-V,VI bifid; siphon slender, straight (Figs. 78–80) ................................................. *perplexus*
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