

Acknowledgments: The authors wish to acknowledge the advice and counsel of Dr T. Ramachandra Rao and Dr C. R. Anderson concerning the planning and conduct of this research.

REFERENCES

- Anonymous.** 1964. Kyasanur Forest Disease, 1957-1964. *Indian Council Med. Res.*, Delhi. 30 p.
- Barnett, S. F.** 1961. The control of ticks on livestock. *Food Agr. Organ. UN FAO, Agr. Studies No. 54*, 115 p.
- Hoogstraal, H.** 1966. Ticks in relation to human diseases caused by viruses. *Ann. Rev. Ent.* **11**: 261-308.
- McDuffie, W. C. & C. N. Smith.** 1955. Recommended current treatments for tick control. *Pub. Health Rep. US* **70**(3): 327-30.
- Mount, G. A., J. M. Hirst, J. G. McWilliams, C. S. Lofgren & S. A. White.** 1968. Insecticides for control of the lone star tick tested in the laboratory and as high and ultra-low-volume sprays in wooded areas. *J. Econ. Ent.* **61**(4): 1005-07.
- Singh, K. R. P., K. M. Parvi & C. R. Anderson.** 1964. Transmission of Kyasanur Forest Disease virus by *Haemaphysalis turturis*, *Haemaphysalis papuana kinneari* and *Haemaphysalis minuta*. *Indian J. Med. Res.* **52**(6): 566-73.
- Trapido, H., M. K. Goverdham, P. K. Rajagopalan & M. J. Rebello.** 1964a. Ticks ectoparasitic on monkeys in the Kyasanur Forest Disease area of Shimoga District, Mysore State, India. *Amer. J. Trop. Med. Hyg.* **13**(5): 763-72.
- Trapido, H., M. G. R. Varma, P. K. Rajagopalan, K. R. P. Singh & M. J. Rebello.** 1964b. A guide to the identification of all stages of the *Haemaphysalis* ticks of South India. *Bull. Ent. Res.* **55**(2): 249-70.
- Varma, M. G. R., H. Trapido & P. K. Rajagopalan.** 1957. Studies on ticks as possible vectors of Kyasanur Forest Disease. *Proc. 9th Pacif. Sci. Congr. Pacific Sci. Assoc.* 88-90.
- Varma, M. G. R., H. E. Webb & K. M. Parvi.** 1960. Studies on the transmission of Kyasanur Forest Disease virus by *Haemaphysalis spinigera* Neumann. *Trans. Roy. Soc. Trop. Med. Hyg.* **54**: 509-16.
- USDA, Agr. Res. Serv. and Forest Serv.** 1967. Suggested guide for the use of insecticides to control insects affecting crops, livestock, households, stored products, forests, and forest products. *Agr. Handbook No. 331*. 273 p.
- Work, T. H. & H. Trapido.** 1957. Summary of preliminary report of investigations of the virus research centre on an epidemic disease affecting forest villagers and wild monkeys of Shimoga District, Mysore. *Indian J. Med. Sci.* **11**: 340-41.

SOUTH EAST ASIA MOSQUITO PROJECT

DEPARTMENT OF ENTOMOLOGY

SMITHSONIAN INSTITUTION

J. Med. Ent. Vol. 6, no. 3: 251-256

UNITED STATES NATIONAL MUSEUM

30 August 1969

WASHINGTON, D. C. 20560

**CULISETA MARCHETTEI, A NEW SPECIES OF THE SUBGENUS
CLIMACURA HOWARD, DYAR, AND KNAB FROM MALAYSIA,
WITH NOTES ON ITS BIOLOGY (DIPTERA: CULICIDAE)¹**

By **Richard Garcia,^{2,3} John Jeffery² and Albert Rudnick²**

Abstract: The ♂, ♀, pupa, 4th instar larva, and egg of *Culiseta marchettei*, a new species from Malaysia, are described and compared with other species of the subgenus *Climacura*. Notes on collections, habitat, and rearing are presented.

The first record of the genus *Culiseta* Felt, 1904 in southeastern Asia, represented by the species described herein, was reported by Garcia et al. (1968). The medical importance of the genus in relation to arboviruses and aspects of the biology of the Malay-

sian species were also discussed in that report.

Morphological characters of the 4th instar larva as outlined by Edwards (1932), Dobrotworsky (1954) and Belkin (1962) clearly place the Malaysian species in the subgenus *Climacura* Howard, Dyar and Knab, 1915.

The subgenus *Climacura* includes 4 described species with a peculiar, disjunct geographic distribution: *Culiseta melanura* (Coquillett, 1902), the type species from the eastern United States; *C. tonnoiri* (Edwards, 1925) and *C. novaezealandiae* Pillai, 1966 from New Zealand; and *C. antipodea* Dobrotworsky, 1962 from southeastern Australia. The presence of *C. marchettei* in southeastern Asia further broadens the distribution and adds a tropical habitat to an otherwise temperate zone subgenus.

Specimens of all stages used for the description and illustrations presented below were reared in the laboratory from eggs deposited by a single wild-caught female and were compared with wild-caught specimens. The terminology used follows that of

¹Supported in part by the University of California International Center for Medical Research and Training with Research Grant TW-00144 from the Office of International Research, National Institutes of Health, U. S. Public Health Service and by the U. S. Army Medical Research and Development Command, Department of the Army, under Contract No. DA 49 193 MD 2931.

²George Williams Hooper Foundation, University of California Medical Center, San Francisco, California 94122, U.S.A. and University of Malaya Faculty of Medicine, Kuala Lumpur, Malaysia.

³Present address: Division of Biological Control, University of California, Gill Tract, Albany, California, 94706, U.S.A.

Belkin (1962) except for the pupal chaetotaxy which follows Barr & Myers (1962).

Culiseta (Climacura) marchettei n. sp.

Types: *Holotype:* ♂ with associated larval and pupal skins (PR8-18-2) reared from gravid ♀ (PR8-18-0) collected in fresh water swamp, near Tanjong Rabok, Kuala Langat Forest Reserve, Selangor, Malaysia, 9 August 1968, Ahmad bin Abdul Hamid & K. T. Tee. *Allotype:* ♀ with associated larval and pupal skins (PR8-18-9), same data as holotype. *Paratypes:* 4 ♂♂ (PR8-18-3, 4, 6, 8) and 2 ♀♀ (PR8-18-5, 10) with associated larval and pupal skins, 10 larvae (PR8-18), and parent ♀ (PR8-18-0), same data as holotype. Type material deposited in the U.S. National Museum, Washington, D.C. This species was named in honor of Nyven J. Marchette in recognition of his contributions in the field of arthropod-borne diseases in Malaysia.

♀. *Wing*, about 3.2 (3.0–3.4) mm; *proboscis*, 2.2 (2.1–2.3) mm; *forefemur*, 1.5 (1.4–1.7) mm; *abdomen*, about 2 mm. *Head:* erect scales of vertex blackish; decumbent scales pale and narrow except for broad pale patch along posterior orbital margin; clypeus bare; labium slender with brownish scales except for a few lighter scales ventrally, a few subbasal bristles; palpus 1/4 to 1/5 length of proboscis with deep brownish scales, segments 2 and 3 with a few long bristles, 4 with numerous short and a few long bristles; antennal torus brownish without scales.

Thorax: mesonotal and scutal integument brownish, pleuron dull greyish brown with lighter areas along sutural lines; mesonotal scales narrow and dark, more numerous over anterior surface; scutellum with narrow dark scales; pleural scaling scanty; *apn* without scales; *ppn* with fine black hair-like scales; *ppl* apparently without scales; *psp* without scales; *stp* with several broad creamy translucent scales more or less in a row along lower posterior margin; *mep* with single hair-like pale scale; bristles dark with few exceptions; 3–4 *sp* bristles; *stp* with upper and posterior bristles, also a row of fine pale bristles extending along lower posterior margin; *apn* with a few long and numerous shorter bristles; *ppn* with 1 or 2 strong bristles; *mep* with apparently 1 strong lower bristle, upper and anterior bristles present, middle and posterior bristles absent.

Legs: dorsal, anterior and ventral surfaces of forefemur dark-scaled, posterior surface pale-scaled; dorso-anterior surface of midfemur and hindfemur dark-scaled, ventral and about 2/3 posterior surfaces pale-scaled, remainder dark-scaled; foretibia and midtibia dark-scaled except for faint line of paler scales along posteroventral surface; hindtibia dark-scaled except for patch of whitish scales at apex forming an indistinct ring, a few pale scales at base; foretarsi dark-scaled except for some lighter scales on ventral surface of segments 2–4, more extensive on 5; midtarsi with fewer lighter scales; hindtarsal segments 1–4 banded basally with white scales and a few white scales apically, segment 5 essentially white-scaled except for a few brownish scales; all claws simple.

Wing: membrane essentially clear, scales uniformly dark, long slender plume scales primarily along midportion veins M and RS; cell R_2 about 5 times as long as vein R_{2+3} , crossveins without scales.

Haltere: integument of base and stem light, knob dark with dark scales.

Abdomen: tergite I with bristles only; tergite II with deep brown scales; tergite III brown-scaled dorsally and basal patch of white scales laterally; tergite IV essentially as III except for

extension of lateral white scales dorsally; tergite V extension of lateral white scales forms complete narrow basal band dorsally; tergite VI and VII with basal white bands about 1/4 and 1/2 length of segments respectively; tergite VIII mostly pale-scaled; sternites II–VI with translucent pale scales; sternite VII pale-scaled except for a few darker scales apically, integument light basally and dark brown apically producing a banded appearance; sternite VIII pale-scaled, integument dark.

♂. Similar to ♀ with following exceptions. *Palpus:* longer than proboscis by more than 1/2 of segment 5; segment 2 about 3 to 4 × as long as 1, segment 3 about 1-1/2 to 2 × as long as 2 and over 2 × the length of 4 and 5 combined; distal portion of segment 3 and segments 4 and 5 with long hairs; segments 4 and 5 with bristles distally and 5 with bristles along lateral margin; segments 2–3 pale-scaled laterally, brown-scaled dorsally; segment 4 largely brown-scaled laterally and dorsally or sometimes pale-scaled on proximal 1/2 laterally; segment 5 with only dark brown scales. *Legs:* claw of foretarsus and midtarsus with teeth, hindtarsus simple. *Wing:* cell R_2 about 3 × as long as vein R_{2+3} . *Abdomen:* tergite III with lateral patches of white scales extending dorsally; tergite IV with narrow basal white band about 1/5 length of segment; tergites V–VII with broader bands increasing from about 1/3 to 1/2 length of segments.

MALE GENITALIA (FIG. 1b). As figured. *Segment VIII:* tergite pale-scaled, integument dark; sternite brown-scaled except for narrow band of pale scales. *Segment IX:* tergal lobe bearing 3 to 5 bristles. *Sidepiece:* long and conical, more or less uniformly covered with medium to small bristles; basal lobe distinct, conical with 1 or 2 strong apical bristles and medium to small basal bristles. *Claspette:* absent. *Clasper:* simple; with a broad, blunt spoon-shaped spiniform. *Phallosome:* aedeagus more or less cylindrical, lateral walls strongly sclerotized and terminating apically with 3 to 4 prominent teeth. *Proctiger:* very large and prominent; basolateral sclerotization broad; paraproct very heavily sclerotized, projecting in a moderate dorsal curve terminating with 3 to 4 very strong spines; dorsal surface bearing 3 to 4 cercal setae.

PUPA (FIG. 1 a, c). *Abdomen*, about 3.0 mm; *trumpet*, about 0.35 mm long and 0.15 mm wide; *paddle*, about 0.75 mm long and 0.45 mm wide. Chaetotaxy as figured. *Cephalothorax:* generally lightly pigmented, wing case slightly darker, hairs lightly pigmented and fine; hairs 9-C and 7-C single or branched; 5-C forked at about midpoint with 4 branches; 10-C moderately strong with 8–10 branches; 11-C moderately strong with 4–6 branches; 12-C forked at about its midpoint. *Abdomen:* hairs 1, 4–III strong with 5 or 6 and 4 or 5 branches respectively; hairs 1, 5–IV strong, with 3 or 4 and 4 or 5 branches respectively; hairs 1, 5–V strong with 3 or 4 and 4 or 5 branches respectively; hair 9–VIII with 8–12 branches. *Paddle:* midrib moderately strong and pigmented for about 7/8 its length, remainder weak and undifferentiated from integument; apical 1/2 fringed with rows of spicules as illustrated, outer spicules stronger than inner ones; inner and outer paddle hairs often reversed in position, 1 single and short, the other longer with 2 or 3 branches.

LARVA (FIG. 2). *Head*, about 0.8 mm long and 1.1 mm wide; *antenna*, about 0.7 mm long; *siphon*, about 1.1 mm long. Chaetotaxy as figured. *Head:* moderately pigmented, integument with rather evenly distributed fine pits; mental plate with 6–7 teeth on each side of a stronger median tooth; hair 1-C moderately developed, curving mesad and tapering to a fine point; 6-C long, single and simple; 7-C usually with 5–8 branches; 13-C with 6–7 branches, longer and more strongly developed than 11-C. *Antenna:* shorter than head capsule, uniform in width from base to hair 1-A and slightly narrower distally; hair 1-A on distal 1/7 of segment; spicules slender over proximal portion, much broader over distal portion.

Thorax: lightly pigmented with fine uniform sculpturing;

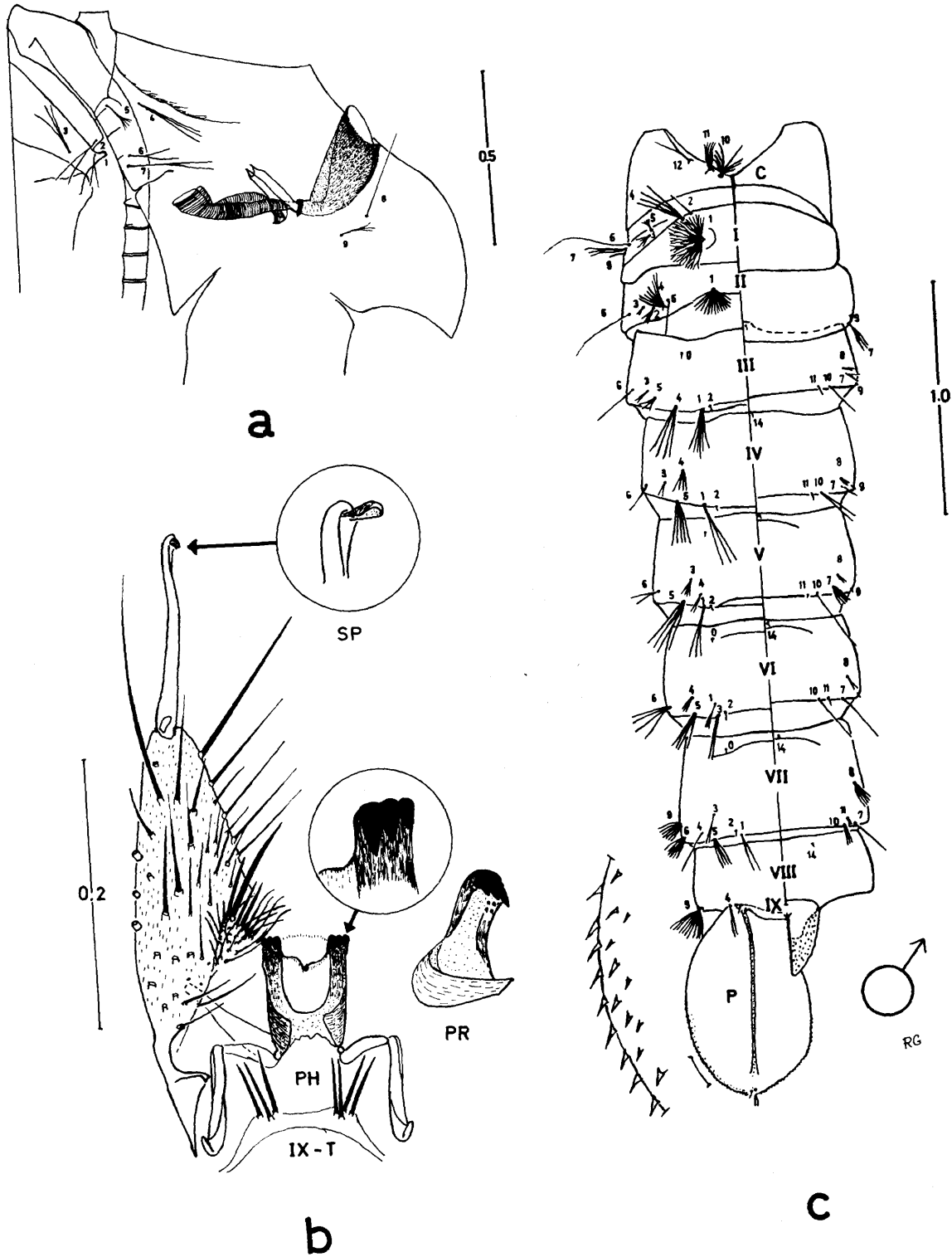


FIG. 1. *Culiseta marchettei* n. sp.: (a) pupa, cephalothorax; (b) ♂ genitalia; (c) pupa, abdomen and metanotum.

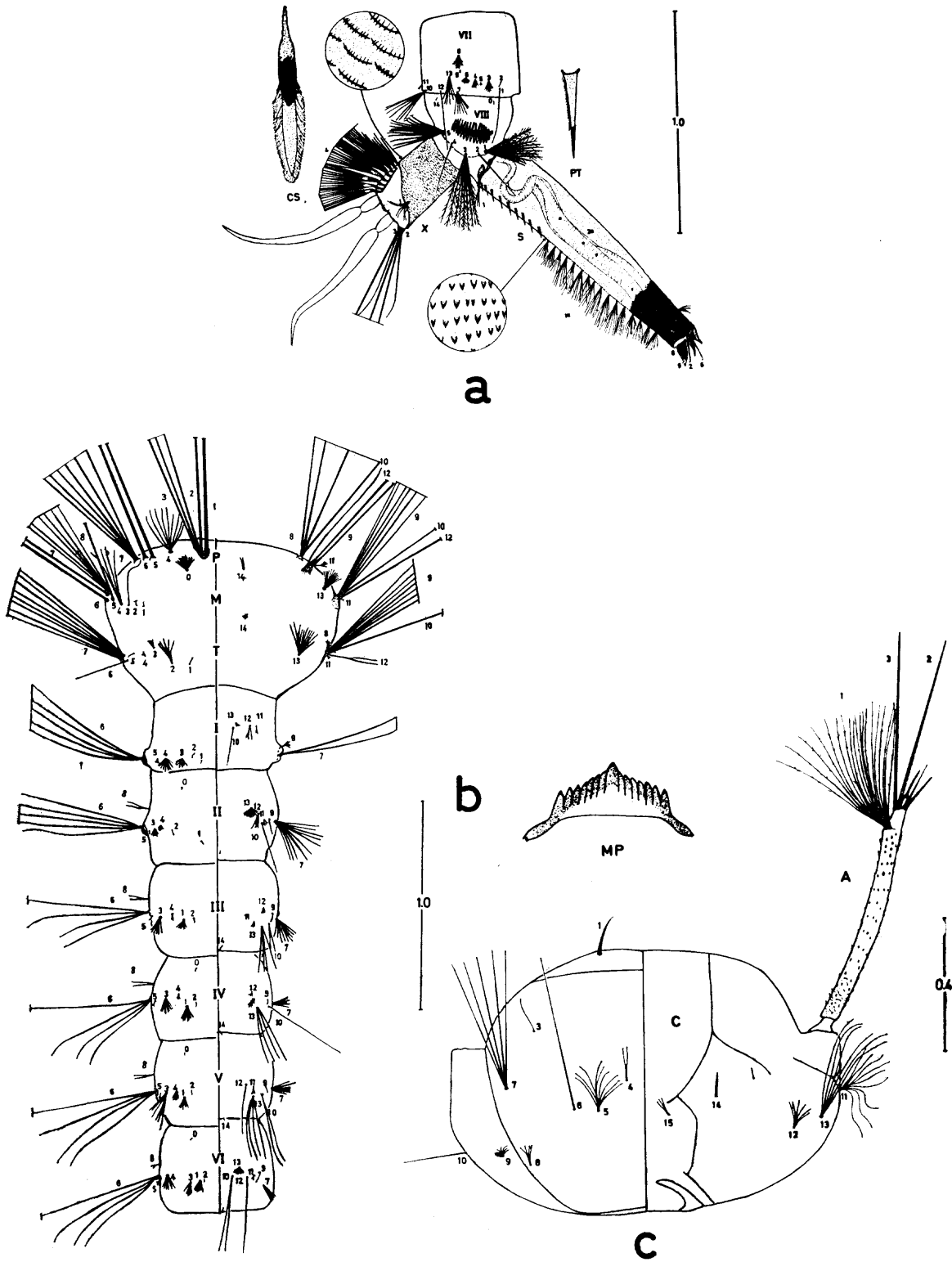


FIG. 2. *Culiseta marchettei* n. sp., 4th instar larva: (a) terminal segments; (b) thorax and abdomen; (c) head.

hair 4-P with 5-9 branches; hairs 7, 8-P, 8, 9-M and 7, 9-T with fine lateral branchlets.

Abdomen: hairs 6-I, II with 4 or 5 branches with fine lateral branchlets; hairs 6-III-VI with 4 or 5 branches. **Segment VII:** hair 10-VII long, 6-8 branched; hair 12-VII long, single, simple; hair 13-VII moderately developed with 6-7 branches. **Segment VIII:** comb, in a single row, 13 to 20 spindle shaped, finely-fringed scales, 1-3 scales at end of row usually weakly developed, center of scales darkly-pigmented and slightly swollen; hair 3-VIII with 9-10 branches. **Siphon:** index 4.0-5.7, moderately pigmented over basal 3/4 except basal black ring, apical 1/4 black, acus moderately pigmented, sides nearly parallel over basal 3/4 then tapering to about 3/5 of its width, integument with small bulbous spicules in subparallel rows; 9-11 pecten teeth in a straight row about 1/3 length of siphon, slightly wider spaced distally, each simple acute spine with a fine fringe at margin of basal 2/3; hair 1-S small, at extreme base, usually with 3-5 branches; 1a-S well developed, represented by a single row of 13-18 hairs usually 6-branched, extending over about distal 2/3 of siphon, middle hairs longer than end hairs; 2-S well developed with apical hook, inserted at apex of siphon; 2a-S represented by a single row of very small hairs with 3 or 4 branches except for a rather strong distal hair with 4 or 5 branches on subdorsal surface. **Anal Segment:** saddle complete, uniformly pigmented except for darkening at basal margin and apical dorsal surface, integument with a series of curved lines of several fine spicules in subparallel rows; hair 1-X small, 5-7 branched; hair 2-X usually 3-, sometimes 4-, branched; ventral brush with 7-9 pairs of hairs, basal 1 or 2 hairs usually on saddle.

EGG. About 0.6 mm long and 0.12 mm wide at midpoint; elongate-oval, posterior end narrower; gray with darker posterior end; deposited in raspberry-shaped clusters of up to 149 eggs.

Specimens examined: 125 adult females; 139 adult males; reared series from single parent ♀ consisting of 4 4th instar larval skins, 4 pupal skins, 10 4th instar larvae, 3 adult ♀♀, and 5 adult ♂♂; 2 4th instar larvae reared from another parent ♀; and 6 egg clusters.

Distribution: *West Malaysia*, all from fresh-water peat-swamp forests. 105 ♀♀ and 130 ♂♂ near Kampong Tanjong Rabok, Kuala Langat Forest Reserve, Selangor. 6 ♀♀ and 5 ♂♂ about 3 km W of Pacific Tin mine, Batang Berjuntai, Selangor. 14 ♀♀ and 4 ♂♂ on highway to Pekan, 32.5 km from Kuantan, Pahang.

Remarks: Belkin (1962) points out the remarkable similarities between *C. melanura* of the eastern United States and *C. tonnoiri* of New Zealand. The 4th instar larva and certain characters of the adults of *C. marchettei* also show some striking similarities to these species and in particular to *C. melanura*. In the larva these include the comb scales in a single row, the small basal hair 1-S with 3-5 branches on the siphon, a row of well developed hairs 1a-S, the dorsolateral hairs 2a-S, the well developed apically-hooked hair 2-S, and the stronger and larger hair 13-C compared to 11-C. The adults of *C. marchettei* also have poorly developed pleural scaling, a small number of spiracular bristles, and a relatively short

vein R_{2+3} in the female. Vein R_{2+3} in the male is much longer.

The larva of *C. marchettei* can be distinguished from all other members of the subgenus *Climacura* except *C. melanura* by the presence of the strongly developed 1a-S hairs. A number of characters separate it from *C. melanura*: the shape of the siphon, pecten teeth, and comb scales and the well developed subapical dorsolateral hair 2a-S as well as other differences in the chaetotaxy. The adults of *C. marchettei* can easily be distinguished from other *Climacura* species by the distinct banding of the abdominal tergites and the basal white bands on the hind tarsi.

The male genitalia of *C. marchettei* are very distinct. The very strong heavily sclerotized ventro-dorsally projecting teeth of the aedeagus separate it from all other *Climacura* species. This structure viewed laterally in a fresh specimen bears a resemblance to the dark sclerotized spines of the proctiger.

Biology: Adults of *C. marchettei* have been collected in 3 fresh-water peat-swamp forests. These swamps are characterized by: relatively tall straight trees, some with flanging buttresses, dense shade, moderate undergrowth and brown clear water produced by the leaching of pigments from the swamp vegetation. The swamp floor is usually porous and uneven due to the intermingling roots, fallen trees and other debris. There is visible standing water in the depressions during the rainy seasons, while during the drier periods the water level may drop 1/3 m or more below the level of the forest floor.

The 2 collecting areas in the state of Selangor are located a few km inland from the west coast and are separated longitudinally by about 60 km. The 3rd locality near Pekan in the state of Pahang is near the east coast, approximately 190 km from the other sites. These 3 areas represent the only fresh-water swamps in which intensive mosquito collections have been conducted by our laboratory. Although other habitats, such as mangrove swamps, lower and upper dipterocarp rain forests, secondary forests, rubber plantations, domestic animal shelters, and other agricultural areas have been collected intensively for several years, individuals of this species have not been recovered. These results suggest that *C. marchettei* is restricted to fresh-water peat-swamp forests.

The principal method of collection of *C. marchettei* was sweeping around tree buttresses, low vegetation, and crevices in the root system of swamp trees employing battery-operated hand vacuum devices. TABLE 1 demonstrates the results of collections in the Tanjong Rabok swamp forest. The numbers

TABLE 1. *Culiseta marchettei* adults collected by sweeping in fresh-water peat-swamp forest, Tanjong Rabok, Selangor (30 May-18 November, 1968).*

| MONTH | No. ♂♂ | No. ♀♀ | NUMBER/ MAN-HOUR** |
|-----------|--------|--------|-----------------------|
| May | 27 | 19 | 8.0 |
| June | 29 | 21 | 2.5 |
| August | 3 | 2 | 1.3 |
| September | 0 | 3 | 1.2 |
| October | 7 | 11 | 7.7 |
| November | 63 | 42 | 7.8 |

*Mosquitoes were collected by a portable vacuum device from tree buttresses, low vegetation, crevices and ground debris.

**Calculated by dividing number of collecting hours into number of mosquitoes recovered.

recovered per man hour are relatively low compared to some of the other species collected by this method. The higher recoveries during May, October, and November indicate fluctuations in the populations of *C. marchettei* which may be related to higher levels of swamp water during the rainy periods. However, the collecting periods during August and September were of short duration and the data may not be valid for comparison.

Several intensive searches for larval breeding sites in the swamps have been unsuccessful.

During late October and early November, adult specimens were collected on 4 occasions from animal bait traps. One female *C. marchettei* was recovered from a trap placed at an elevation of 13.7 m in the lower canopy and baited with a pig-tail monkey (*Macaca nemestrina*). At an elevation of only 1 m a trap baited with a chicken yielded 1 female and 2 traps each baited with a chicken and dry ice yielded 3 and 4 females. Of the 9 specimens recovered, 2, from the chicken-baited traps, were freshly engorged with blood suggesting that *C. marchettei* may prefer avian hosts.

Attempts to rear this species in the laboratory from eggs deposited by wild-caught gravid females have been partially successful. Females were normally held in an airconditioned room with minimum and maximum average temperatures of 20°C and 25°C respectively. Oviposition usually occurred within 1-3 days and the 1st instar larvae emerged after about 5 days. At room temperatures ranging from 27°C to 30°C larvae held in open pans usually died in the early instars. In the airconditioned room, larvae in open pans occasionally reached the 4th instar but failed to pupate. The only successful rearing to the adult stage was in the airconditioned room in enamel pans covered with metal lids to eliminate light. These pans were disturbed

once daily for observation, feeding or removal of surface film. A small quantity of a commercially prepared chick starter feed was ground to a powder and mixed with distilled water. Several drops of the mixed suspension and a trace of liver extract were added to the pans of distilled water initially. Thereafter, a few drops of the food suspension were added when needed. From a cluster of 147 eggs, approximately 24 reached either the 4th instar, pupal or adult stages, and required a period of 39 days from the egg to the first emerged adult.

Acknowledgments: We wish to express our sincere appreciation to Dr S. Ramalingam and members of his staff, especially K. Ramakrishnan, in the Department of Parasitology of the University of Malaya for valued advice and assistance. We also wish to acknowledge with thanks the support of Professor J. Ralph Audy, Director of the Hooper Foundation, and Professor T. J. Danaraj, Dean of the Faculty of Medicine, University of Malaya.

REFERENCES

- Barr, R. B. & C. M. Myers. 1962. Pupae of the genus *Culiseta* Felt. I. The homology of larval and pupal setae (Diptera: Culicidae). *Ann. Ent. Soc. Amer.* **55**(1): 94-98.
- Belkin, J. N. 1962. The Mosquitoes of the South Pacific (Diptera, Culicidae). Univ. of Calif. Press, Berkeley, I: 608 p., II: 412 p.
- Coquillett, D. W. 1902. New forms of Culicidae from North America. *J. N. Y. Ent. Soc.* **10**(4): 191-94.
- Dobrotworsky, N. V. 1954. The genus *Theobaldia* (Diptera, Culicidae) in Victoria. *Proc. Linn. Soc. N. S. W.* **79**: 65-78.
1962. Notes on Australian mosquitoes (Diptera, Culicidae). VI. Five new Victorian species and a description of the larva of *Aedes milsoni* (Taylor). *Ibid.* **87**: 291-302.
- Dumbleton, L. J. 1965. Developmental stages and biology of *Culiseta tonnoiri* (Edwards) and a note on *Culex peruvigilans* Bergroth (Diptera: Culicidae). *New Zeal. J. Sci.* **8**: 137-43.
- Edwards, F. W. 1925. Mosquito notes. -V. *Bull. Ent. Res.* **15**: 257-70.
1932. Diptera Fam. Culicidae. Fasc. 194, 258 p., 5 pls. In Wytzman, P., ed., *Genera Insectorum*. Bruxelles.
- Felt, E. P. 1904. Mosquitoes or Culicidae of New York State. *N. Y. State Mus. Bull.* No. **79**: 241-400.
- Garcia, R., J. Jeffery & A. Rudnick. 1968. A first report of the genus *Culiseta* Felt in Malaysia (Diptera: Culicidae). *Med. J. Malaya* **23**(1): 29-31.
- Howard, L. O., H. G. Dyar, & F. Knab. 1915. The mosquitoes of North and Central America and the West Indies. *Carnegie Inst. Wash.*, Pub. **3**: 452.
- Pillai, J. S. 1966. *Culiseta novaezealandiae*, a new species of the subgenus *Climacura* Felt (Diptera: Culicidae: Culisetini), with notes on its ecology and development. *Trans. Roy. Soc. N. Z.*, Zool. **8**(11): 125-33.
1968. Notes on mosquitoes of New Zealand. II. The male terminalia of *Culiseta (Climacura) tonnoiri* and its ecology (Diptera: Culicidae: Culisetini). *J. Med. Ent.* **5**(3): 355-57.