

Culex (Culex) pipiens and *Culex (Cx.) torrentium* (Diptera: Culicidae) in
England: Notes on their Taxonomy and Biology

P. G. Jupp
Arbovirus Research Unit
National Institute for Virology
Private Bag X4
Sandringham 2131
South Africa

ABSTRACT. The sympatric sibling species *Culex torrentium* Martini and *Cx. pipiens* L were collected in Surrey and *Cx. pipiens* in Huntingdonshire, southern England. The 2 species were studied taxonomically mainly to find a diagnostic character for separating the females and some biological observations were also made.

Males could only be separated by previously described genitalia differences, while in the female study of the pre-alar scales showed that a diagnosis could be made according to the number of these scales.

At Silwood Park in Surrey *Cx. torrentium* comprised a high proportion (62.1%) of the total collection of both species, while *Cx. pipiens* occurred alone in a collection of hibernating mosquitoes from Huntingdonshire.

Hibernating *Cx. pipiens* taken by air to South Africa were reactivated by an increase in temperature and day length (provided by artificial illumination) at 17 days and at 5 months after collection, so that they took bloodmeals and oviposited.

INTRODUCTION

In the 1977 summer specimens of *Culex pipiens* L. were collected in England as part of a broader taxonomic study on members of the *Culex pipiens* complex from several countries. Large numbers of *Culex torrentium* Martini occurred in these collections which made it necessary to be able to separate the 2 species.

Apart from Mattingly's (1951) brief report on the discovery of *Cx. torrentium* in England, Service (1968) is the only worker who has investigated *Cx. torrentium* and its relationship to *Cx. pipiens*. He did studies on both taxonomy and biology and his findings on the taxonomic separation of the 2 species, with terminology after Knight (1971) and Knight & Laffoon (1971), were as follows:

- (1) Eggs and larvae cannot be distinguished.

- (2) Pupae can be separated to some extent: when seta 4 on abdomen segment VIII is single on one or both sides the pupa can be confidently ascribed to *Cx. torrentium* but specimens which have 2 branches on both sides cannot be identified with certainty.
- (3) The genitalia are completely diagnostic for adult males owing to differences in the structure of the phallosome (see also Mattingly, 1951). In *Cx. torrentium* the basal lateral projection of the paraprocts is very well developed whereas in *Cx. pipiens* it is vestigial. In addition the dorsal arm of the inner division and the outer division of the lateral aedeagal plate have differently shaped tips.
- (4) The possession of pre-alar scales is not diagnostic for males of *Cx. torrentium* as 10% of a sample of this species was found to lack these scales on both sides of the thorax, making them indistinguishable from *Cx. pipiens*. No pre-alar scales were found in males of *Cx. pipiens*. Owing to these findings in males Service cautioned against relying on pre-alar scales for identification of females.

The present paper reports taxonomic and some biological studies carried out on the 2 sympatric sibling species. A search was made for morphological characters by which larvae, adult females and adult males of the 2 taxa could be separated. A means of identifying the female mosquito was the principal objective.

MATERIALS AND METHODS

The collections were all made in the grounds of the Field Station of the Imperial College of Science and Technology at Silwood Park near Ascot in Surrey, southern England, except for one which was made at Monks Wood in Huntingdonshire. Egg rafts, larvae and pupae were collected at Silwood Park. At Monks Wood a sample of adult *Cx. pipiens* females was collected by mechanical aspirator from the walls of an underground room where this species had just entered into hibernation.

Each field-collected larva or pupa was reared in isolation so that each emergent adult could be preserved along with its associated pelt or pelts for later taxonomic study. Each field-collected egg raft was allowed to hatch and a sample of the resulting larvae reared so that associated larvae, larval pelts, pupal pelts and male and female adults all originating from the same individual egg raft could be preserved. This procedure was carried out for a number of egg rafts so that different batches of associated immature-adult specimens could be obtained i.e. different sibling groups. Thus a sibling group was the progeny reared from one egg raft deposited by one female mosquito. A number of such different sibling groups were also reared from rafts subsequently deposited by the mosquitoes collected at Monks Wood.

For the taxonomic studies, with the exception of certain instances stated in the 'Results' section, one larva, one adult male and one adult female were used from each of the sibling groups which had been reared. The male was identified from the structure of its genitalia, so that the identity of the associated larva and female mosquito was known. Using specimens identified in this way an evaluation was made of 4 morphological characters for separating the 2 species. These were as follows:

- (1) The number of branches borne by the siphonal setae la-s and lb-s in the larva.
- (2) The value for the cross-vein index in the wing of the adult female, that is the ratio of the combined lengths of the 2 cross-veins to their distance apart.
- (3) The value for the maxillary palp hair index in the adult male, that is the ratio of the length of the shaft of the palp bearing hairs to its total length (segments 2 + 3).
- (4) The presence and number of pre-alar scales in the female mosquito.

Characters (1) and (3) had previously proved diagnostic and (2) almost diagnostic for distinguishing *Cx. pipiens* and *Cx. quinquefasciatus* Say in South Africa (Jupp, 1978).

For clarity the methods used for handling the hibernating mosquitoes are included in the 'Results' section.

RESULTS

Taxonomy

The first 3 morphological characters evaluated proved valueless for separating the 2 species as their examination on small samples showed an overlap between the 2 taxa. However, study of the fourth character, the occurrence of pre-alar scales in females, showed that in 15 *Cx. torrentium* belonging to different sibling groups 1 or more pre-alar scales were present on both sides of the thorax. In a further 12 *Cx. torrentium*, which were siblings of 9 of the above mosquitoes, there were 2 specimens that had scales on only one side of the thorax - 3 scales and 1 scale respectively. However of 35 *Cx. pipiens* belonging to different sibling groups, 3 mosquitoes possessed a single pre-alar scale on one or both sides of the thorax. In view of this, all the available female siblings of these 3 mosquitoes were examined which revealed that the occurrence of pre-alar scales varied within each of the sibling groups as follows:

Number of siblings in sibling groups examined	Number individuals with:		
	1 scale on each side	1 scale on one side	No scales
2	1	0	1
6	1	3	2
6	0	2	4

These findings show that the presence or absence of pre-alar scales is not absolutely diagnostic for separating females of the 2 species, although the presence of 2 or more scales on one or both sides of the thorax would identify a specimen as *Cx. torrentium*. This criterion was used in identifying the emergent females reared from immatures collected in the field and in those cases where only 1 scale occurred, or where a specimen was damaged, the associated pupal pelt was mounted and examined. For each of these pelts a single-branched seta 4 on one or both sides of abdominal segment VIII confirmed that they were all *Cx. torrentium*.

Biology

At Silwood Park, larvae, pupae and egg rafts were collected from artificial containers of water, including metal water tanks, wooden barrels and concrete animal drinking troughs, and in one instance from the stagnant margins of a stream. A total of 206 emergent mosquitoes were identified. This figure was made up of 95 males and 86 females which had been reared individually from larvae or pupae collected in the field and of 25 sibling groups of mosquitoes successfully reared from egg rafts. Out of this total of 206 mosquitoes, 128 individuals or different sibling groups were identified as *Cx. torrentium* i.e. 62.1%. All of the 11 larval collections made contained *Cx. torrentium* and 7 of them also contained *Cx. pipiens*.

Seventy one individuals of a sample of about 250 *Culex* mosquitoes collected hibernating at Monks Wood on 30th August, were all shown to be *Cx. pipiens* from examination of males reared from egg rafts they subsequently deposited. Hence it is likely that the complete sample was only *Cx. pipiens*. The sample was placed in a 20 cm³ cage of cotton netting on the top of which was placed a thick wad of cotton wool soaked in 4% sucrose solution. Thereafter this wad was kept moist and renewed when necessary. The cage was enclosed in a card board box, just big enough for it, so that the relative humidity was 80-84% and the light excluded. The temperature was not controlled but the box was kept in a cool garden shed until flown to Johannesburg 14 days after the mosquitoes had been collected. On arrival in Johannesburg the box was stored in a cool garage and after 3 days the 3 mosquitoes were divided into 2 groups of about 100 insects each in 35 cm³ cages. The mosquitoes in the first cage were released from hibernation immediately by placing this cage in an environment which, although humid, (RH 68-70%) had a higher temperature of 26-27°C and artificial lighting to simulate a day length of 16 1/2 hours (an English summer's day) and a night of 7 1/2 hours. Seven days after placing this cage under these conditions 6 of the insects engorged on a pigeon offered to the mosquitoes. However, many others died from uncertain causes, possibly because of insecticide on the feathers of

the pigeon used for feeding. Over the next 3 weeks opportunity to feed on pigeons was provided 4 more times. This resulted in 25 insects taking blood-meals. The last 4 surviving mosquitoes, which remained unfed, were dissected and found to contain large (1 mosquito), medium-sized (1 mosquito) or small (2 mosquitoes) fat bodies.

The second cage was kept humid (RH 68-70%) and in the dark at 16°C, in conditions similar to those which prevailed in their natural hibernating site at Monks Wood. When this cage was finally placed under the conditions to terminate hibernation, it was found that only one mosquito had died after 5 months of hibernation in an artificial environment. Three days later a large proportion of the mosquitoes engorged on a pigeon and 40 insects subsequently deposited rafts, all of which hatched. The higher feeding rate by this group of mosquitoes was thought to be due to the fat bodies having been depleted in a higher proportion of individuals before the pigeon was offered to them.

DISCUSSION

Service (1968) showed that *Cx. torrentium* could not be distinguished in the larval stage from *Cx. pipiens* and that males of *Cx. torrentium* could not be distinguished by the pre-alar scale character but only according to the structure of the genitalia. The present findings do not alter this conclusion since the siphonal setae proved to be of no value for diagnosis in the larva and the maxillary palp hair index was likewise valueless in males. However, examination of female specimens in the present study indicated that although the 2 species could not be separated according to their wing venation, they could be differentiated by the pre-alar scale character. In the rather small samples studied it was shown that all females of *Cx. torrentium* except one specimen could be identified by the presence of 2 or more pre-alar scales on 1 or both sides of the thorax. Since siblings of this single exception could also be identified it is thought that the specimen in question may have lost 1 or more scales due to being rubbed. *Cx. pipiens*, although usually having no scales, occasionally had only a single scale on 1 or both sides. This method of diagnosis needs to be tested out on larger samples of female mosquitoes of known identity belonging to both species.

Cx. torrentium was more prevalent than *Cx. pipiens* at Silwood Park accounting for 62.1% of the total collections of both species. This is a much higher proportion than Service (1968) found in collections made at Poole, Dorset, from 1964-1966 (37.2%), Monks Wood (15.9%) and from a large area over the breadth of southern England (32.3%). The collections at Silwood Park suggest that a future survey in Britain might reveal that *Cx. torrentium* has displaced or partly displaced *Cx. pipiens* in certain areas.

No *Cx. torrentium* occurred among the 71 mosquitoes identified from the sample which was collected hibernating in an underground room at Monks Wood. Service (1968) also failed to find adults of this species hibernating in man-made shelters. Hibernating *Cx. pipiens* transferred to the laboratory in South Africa were reactivated at 2 different times after collection - after 17 days and after 5 months. Increased temperature and day-length,

the latter provided by artificial illumination, stimulated the mosquitoes to feed and develop their ovaries. This shows that *Cx. pipiens* can be transferred from one country to another while in a state of hibernation and subsequently reactivated. The same could well apply to other species of *Culex* from temperate countries.

ACKNOWLEDGMENTS

I wish to thank the Board of the Poliomyelitis Research Foundation for the award of a travel grant to visit Great Britain. I am also indebted to Prof. T.R.E. Southwood for allowing me to work at the Field Station of the Imperial College of Science and Technology and to Dr. P.F.L. Boreham who provided me with laboratory facilities there and assisted in many ways. Dr. M. W. Service of the Liverpool School of Hygiene and Tropical Medicine, Drs. P. F. Mattingly and G. W. White of the British Museum of Natural History and Mr. M. Yates of Monks Wood Experimental Station gave me valuable advice on the taxonomy and collection of *Cx. torrentium* and *Cx. pipiens* for which I am very grateful. Lastly I wish to thank the Secretary for Health, Dr. J. de Beer, for permission to publish.

REFERENCES

- Jupp, P.G. (1978). *Culex (Culex) pipiens pipiens* L and *Culex Cx. pipiens quinquefasciatus* Say in South Africa: morphological and reproductive evidence in favour of their status as two species. *Mosquito Systematics* 10:461-473.
- Mattingly, P.F. (1951). *Culex (Culex) torrentium* Martini a mosquito new to Great Britain. *Nature Lond.* 168:172.
- Service, M.W. (1968). The taxonomy and biology of two sympatric sibling species of *Culex*, *C. pipiens* and *C. torrentium* (Diptera: Culicidae). *J. Zool., Lond.* 156:313-323.
- Knight, K.L. (1971). A mosquito taxonomic glossary. VII. The pupa. *Mosquito Systematics Newsletter* 3:42-65.
- Knight, K. L. and Laffoon, J.L. (1971). A mosquito taxonomic glossary. V. Abdomen (except female genitalia) 3:8-24. VIII. The larval chaetotaxy 3:160-194.