ARTICLES

The Taxonomic Status of <u>Culex</u> (<u>Culex</u>) <u>univittatus</u> Theobald (Diptera: Culicidae) in South Africa*

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I have investigated populations of the mosquito <u>Culex</u> (<u>Culex</u>) <u>univittatus</u> Theobald from the Transvaal and Natal provinces of South Africa. Two forms of the mosquito occur in this area: a highveld (H) form which is found on the high inland plateau, the so-called "highveld" region with a temperate climate, and a coastal (C) form which is found in the subtropical lowlands including the coastal plain of eastern and northeastern Natal. The H form is the same as the type form of <u>C</u>. <u>univittatus</u>, whereas the C form appears to be undescribed. The latter has some morphological similarities to <u>C</u>. <u>univittatus var neavei</u> Edwards but is not the same. I have made a comprehensive study of the two forms in an attempt to decide their true taxonomic status. At present I am preparing the results of this work for publication and the following is an outline of my findings.

Morphological Findings

Material from Ndum in northeastern Natal and from Olifantsvlei near Johannesburg, Transvaal, was compared, representing the C and H forms of the mosquito respectively. Differences were not detected in larvae from the two places but in adult mosquitoes four characters showed significant morphological differences between the two forms. These characters were the appearance of the hind and mid-femora in both sexes and the nature of two of the appendages borne by the coxite in male terminalia. A quantitative examination of these characters was carried out on large samples of mosquitoes from the two localities.

Hind femur

For the hind femur a "femoral index" (R) was calculated. This was the proportion of the length of femur occupied by the dorso-anterior black stripe and was expressed as a percentage. It was found that this

* The studies and observations on which this paper are based were financed jointly by the South African Institute for Medical Research, the Poliomyelitis Research Foundation and the South African Council for Scientific and Industrial Research. index always had a value of 75% or greater in H females and males whereas in C mosquitoes the value was most often less than 75%. However, the ranges of R for the two forms overlapped in both sexes; in the females, where this overlap was slightly less than in males, 10 mosquitoes out of a sample of 94 of the C form possessed an R value of 75% or higher. This overlap precludes the femoral index from being suitable as a primary practical taxonomic character for separation of the two forms. However, comparison of the means of the R values found for the two samples, using the "t" test, showed that the two populations were significantly different (P <.001) in respect of both sexes.

Mid femur

The mid femur was inspected for the presence of an anterior white stripe. All H females possessed this character while out of a sample of 94 C females it only occurred in one individual. In the male samples the stripe was present in 86% of the H sample but was absent in the C form. This character difference is suitable therefore for distinguishing the females but not the males of the two forms.

The leaflet

The leaflet borne by the sub-apical lobe of the male coxite was found to be narrow in the H form, as shown in fig. 1, or broad in the C form as in fig. 2. In order to express this difference quantitatively a leaflet index (R,) was calculated which was the ratio of the greatest width (s) to the greatest length (1) expressed as percentage. For 56 leaflets measured from C males, the range of R_1 was 50-67%, whereas for 53 measured from H males the range was 33-50%. This overlap was due to 3 H and 3 C males having an index of 50%. Comparison of the means of the R_1 values found for the two samples, using the "t" test, showed a significant difference (P <.001). Because of this and since the two types of leaflet are readily distinguished without measurement it is considered that this character can be used as a practical means for separating males of the two forms.

Seta 'f'

This appendage, also borne by the sub-apical lobe of the male coxite, was found to possess an unswollen tip in the H form, as shown in fig. 1, or a swollen tip in the C form as in fig. 2. In the samples of male terminalia examined this difference appeared to be a consistent one. However, if this appendage is not mounted on its side in permanent preparations its club-shape in the C form may not be fully visible as the swelling appears to be predominantly dorso-ventral. This character should therefore be regarded as a subsidiary one for distinguishing the two forms.

Frequency distribution curves for femoral and leaflet indices

These were plotted to see whether these two continuous characters showed a normal distribution and were thus determined by polygenes. Typically normal curves were found in the case of the R, index for both mosquito forms and for the R index in H males and females. However, the distribution of R in the C form did not follow a typically normal curve the curve for both males and females was basically a normal one but with three peaks present. The explanation for this is probably that control in this case is largely polygenic but with 3 oligogenes exerting their influence.

Scatter diagram of femoral and leaflet indices

The knowledge that the R and R₁ indices were normally distributed permitted the construction of a scatter diagram for male mosquitoes from Ndumu and Olifantsvlei. Values for the two indices measured on the same individuals were used for this diagram which is shown in fig. 3. It can be seen that plots for the C and H forms formed two separate areas on the diagram, which confirms the morphological distinctness of the two populations.

Geographical Relationships

Initially the discovery of the H form at Olifantsvlei and the C form at Ndumu suggested that the two forms were allopatric with the occurrence of the former at high altitude on the highveld plateau and the latter at low altitude in the coastal lowlands. In view of this, collections of <u>univittatus</u> were made at further localities in the two areas to investigate this allopatry as well as at localities with altitudes intermediate between that of the highveld and the coastal lowlands where it was thought the ranges of the two forms might overlap. Mosquitoes collected at the various localities were classified into the two forms on the basis of the four character differences already described.

Figure 4 shows the distribution of the C, H and possible intermediate forms of univittatus in the Transvaal and Natal according to all the population samples collected and identified from 16 localities. It is clear from the maps that the C and H forms are allopatric, the C form occurring in the coastal lowlands of eastern and northeastern Natal at altitudes up to 500 feet while the H form occurs in the highveld region of the Transvaal above an altitude of 3,000 feet. At three localities of intermediate altitude, namely Ingwavuma and Ubombo situated on the Lebombo mountain range and at a locality 17 miles east of Pongola west of the Lebombo mountains, heterogenous populations were found. This indicated that these localities were within the region of overlap between the ranges of the two forms of the mosquito. Examination of the female mosquitoes from these three localities showed the presence of H and C forms and this prompted the construction of scatter diagrams for the R - R_1 ratios on male individuals coming from the same localities to see whether this would reveal the presence of intermediate forms. Too few males were available from Ingwavuma to produce a worthwhile diagram for this locality but plotting the R - R_1 ratios for the samples from the other two places on the same scatter diagram as used for the Ndumu-Olifantsvlei samples revealed the occurrence of possible hybrids (intermediates) along with the H and C forms (see fig. 3). This finding suggested that some hydridization may occur at areas where the two allopatric forms overlap, although the paucity of intermediates indicates that it is probably only an occasional phenomenon.

Differences in Host Preferences

The relative aviophilic and anthropophilic tendencies of the two forms of <u>univittatus</u> were investigated by the use of avian-baited traps and man-baited catches at Ndumu and Olifantsvlei respectively. The results showed that both forms are highly aviophilic and suggested that the C form of the mosquito is more highly anthropophilic than the H form. These findings tend on the whole to agree with the identification of blood meals made by the precipitin test on mosquitoes collected at Ndumu and Olifantsvlei respectively (Paterson et al. 1964, Anderson 1967).

Laboratory Colonization and Crossing Experiments

Colonization

Colonies of both forms of univittatus were established in the laboratory so that crossing of the two forms could be attempted. Special treatment was found necessary for successful colonization which, because of the eurygamous nature of the mosquitoes, included the use of large "walk-in" cages and artificial lighting simulating dawn and dusk periods. For each form a principal colony was first established in a walk-in cage and from this a stenogamous sub-colony in 14 in. square cages was eventually obtained.

Certain differences in behavior between colonies of the two forms were observed, the most important of which related to mating. In both the principal and sub-colonies of the H form mating activity began after a particular light intensity had been reached during the simulated dawn and dusk periods. At this time the males swarmed and mating on the wing took place. However, this activity was never observed in either of the C colonies - it might have occurred during periods of complete darkness or when the light intensity was too low to permit observation but obviously the colonies of the two forms had a different mating behavior. Such a difference could act as an isolating mechanism causing reproductive isolation between populations of the two forms.

Crossing experiments

In view of the difference in mating behavior outlined above the results obtained in the attempted crosses between the two forms were not entirely unexpected. Reciprocal crossing experiments were carried out with both the principal colonies and sub-colonies of the two forms using large numbers of mosquitoes but not a single fertiel female mosquito was detected. It appears that there was complete reproductive isolation between the H and C colonies. It may have been possible to obtain a few hybrid progeny if further crossing experiments had been conducted in which females of, for example, the C form had been placed in a cage with 75% C males and 25% H males. In this way there might have been a few accidental matings between heterogenous forms. The converse experiment could have been done using H females.

Conclusions

The evidence indicates that in South Africa what has previously been considered as <u>Culex</u> univittatus consists of two species. The degree of morphological and ethological difference shown by the H and C forms is sufficient to make their separation possible on taxonomic grounds and observations on mating behavior of laboratory colonies of the two forms and attempts at cross-mating suggested that a behavioral isolating mechanism exists which would preclude any significant cross-mating in The forms are allopatric in the areas studied, each being adapted nature. to different environments, the H form to the temperate inland plateau and the C form to the warmer coastal lowlands, which fact is indicative of physiological differences between each form. Where their ranges meet at intermediate altitudes the two forms co-exist and it appears that there

is not interbreeding except for the possible production of occasional hybrids. The H form is similar to the type form, while the C form is apparently an undescribed species.

References Cited

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Sub-apical lobe of right coxite from inner side to show leaflet and seta "f"; fig. 1 - highveld form, fig. 2 - coastal form.



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Fig. 4. The distribution of the coastal, highveld and possible intermediate forms in the Transvaal and Natal from all localities sampled. Altitudes in feet are given in brackets; S.L. = sea level.