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IS A. MELAS THEO. A DISTINCT SPECIES?¹

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Research carried out so far on the specific relationship between A. gambiae and its brackish water "form" found in West Africa, "A. melas", has given only contradictory results. Morphological differences are far from being constant, the value of the physiological test may be doubted, following recent experiments by Fox in Liberia and, finally, the results of interbreeding between the two "forms" have not solved the problem.

Cytogenetics has shown its value in the study of the anopheline complexes of Europe and South America and it is to this science which we turned in an endeavour to make a contribution to this problem.

Thanks to the welcome given us by the Government of Nigeria, we were able to work there in March 1959 with an "A. melas" strain maintained in the Federal Malaria Service insectarium, as well as with "A. melas" bred from eggs laid in the laboratory by wild females captured in dwellings near Lagos.

The average temperature of the larvae tanks was 27.5-28°C. A study of the chromosomes in salivary glands extracted from the larvae at the end of the third and at the beginning of the fourth stage, was made using the "squashing" method already repeatedly described, after fixation and then staining with carmine in acetic acid.

¹ This preliminary report forms part of a series of researches into the cytogenetics of the anopheline vectors of malaria. It was presented in November 1959 at the Technical Meeting on Malaria Eradication, held at Brazzaville.

Cytogenetic analysis

One fact should again be mentioned; namely, the difficulty of reading chromosomes of "A. melas" (or A. gambiae) as compared with the extreme ease with which the chromosomes of A. maculipennis, for example, can be studied.

Thus out of 480 slides prepared, only 45 or 9.4 per cent. could be analysed. Since each salivary gland consists of 18-24 cells, each with a chromosome set, there should, after all, have been an average of 2000 chromosomes for study. In actual fact, only 660 could be analysed, distributed as follows:

X	67
II G	128
II D	145
III G	171
III D	149

Temperature and feeding are two factors which strongly influence the readability of the chromosomes, as is shown by experiments made on gambiae (which we shall describe elsewhere). By prolonging the period of larval life, low temperatures and under-feeding appreciably increase the percentage of usable slides.

The results of cytogenetic analysis of "A. melas" are as follows:

(a) the free distal extremities of the chromosomes are identical with those of A. gambiae; the succession of bands along the length of the chromosome is the same;

(b) a large number of heterozygous inversions were noted, similar to the inversions previously described in A. gambiae from Lagos or Sokoto; there seems to be no doubt that these are due to the relatively high temperature (27.5-28°C);

(c) on the other hand, the presence should be mentioned of:

(i) a deficiency affecting half of sector 28 and sector 29 of chromosome III D (2 per cent. of the specimens examined, i.e. 3 out of 149);

(ii) a large heterozygous inversion in the form of a loop affecting part of sector 38 and sectors 39-42 of chromosomes III G (7 per cent. of the tests, i.e. 11 out of 171). This inversion corresponds to the 39-41 inversion previously described in the case of A. gambiae but covers a larger zone.

Conclusions

There seems to be no doubt that "A. melas" should be considered as a variety of A. gambiae and not as a separate species.

The cytogenetic analysis carried out at Lagos should only be regarded, however, as a preliminary study. It is in fact necessary to:

- (a) define the nature of the chromosome III D deficiency which may be completely independent of any relationship between gambiae and melas, and due to the presence of an abiotic factor in the strain studied;
- (b) determine whether the heterozygous inversion of III G may be considered characteristic of the melas variety;
- (c) study cytogenetically the descendants of crossings of gambiae and melas. The two varieties are sympatric, and successful crossing in the insectarium probably requires merely the working out of a good technique;
- (d) analyse the effects of breeding the variety melas in soft water on the heterozygous inversions;
- (e) finally, study the chromosomes of the variety found in the brackish water of the Indian Ocean (Tanganyika, Mauritius and Zanzibar).