## Determination of Physiological Age for Female Glossina morsitans

RECENTLY, considerable attention has been paid to a method of determining the physiological age of female insects of economic importance by the appearance of the ovarioles after ovulation. This appearance of the ovarioles after ovulation. method, which was originally worked out by Soviet entomologists<sup>1,2</sup> for *Anopheles*, and has since been extended to other blood sucking Diptera, depends upon the recognition of the number of follicular relics ('corpora lutea') appearing as dilatations of the follicular tubes. Each dilatation represents the relic of a single egg follicle and is evidence of a gonotrophic cycle. If the duration of the cycle is known it is possible to calculate the calendar age of the female, a factor of great practical application in determining the epidemiological importance of populations of insect vectors or the degree of success of control programmes. In the laboratory, Bertram and Samarawickrema<sup>3</sup> recognized Mansonioides uniformis which had completed five gonotrophic cycles by the presence of five dilatations in the follicular tubes. A study of the ovarioles of Glossina morsitans after ovulation has been made, and it is suggested that a similar method of determining the physiological age of female tsetse flies should be possible.

Each ovary of G. morsitans consists of two polytrophic ovarioles, each at a different stage of development<sup>4</sup>. Only one egg matures at a time, and a sequence of ovulation occurs so that the first and third eggs are ovulated from the right ovary and the second and fourth eggs from the left ovary. Subsequent eggs are



Fig. 1. Ovariole of *Glossina morsilans*, immediately after ovula-tion of egg. g, Germarium; rnc, remains of the nurse cells; eft, expanded follicular tube



Fig. 2. Ovariole of Glossina morsitans, six days after ovulation. g, Germarium; f2, first signs of second egg follicle developing in the germarium; f1, remains of first follicle

produced from each ovariole in a continuation of the same sequence.

Immediately after ovulation the follicular tube of the ovariole, which has been greatly stretched to accommodate the mature egg, is an open expanded sac which retains the remains of the nurse cells and the follicular epithelium (the follicular relic) (Fig. 1). In the next few days the follicular tube begins to contract and the follicular relic is passed out of the ovariole. A greater part of the expanded follicular tube then becomes detached, possibly because the germarium is pushed towards the anterior end of the ovary by the growth of the other ovariole. After four days all that remains of the 'dilatation' is a small piece of tissue remaining on the posterior end of the follicular tube which resembles the 'funicle stumps' of parous Anopheles gambiae described by Lewis<sup>5</sup>, and is evidence of ovulation (Fig. 2). As a complete dilatation as described for M.  $uniformis^3$  is not formed, serial dilatations are never found in Glossina morsitans, and ovarioles which have ovulated twice or more cannot be distinguished from those which have only ovulated once. The sequence of ovulation, however, enables the number of eggs produced by a female to be determined up to the production of the fourth egg; after this it is impossible to tell how many eggs have been produced.

At 26° C., the fourth egg is ovulated (from the left ovary) 40 days after the emergence of the fly and therefore, at this temperature, flies may be aged up to at least the fortieth day by the number and position of the funicle stumps and the relative sizes of the four egg follicles. After the ovulation of the first egg (eighth or ninth day at 26° C.) eggs are ovulated about every 10 days, and the uterus contains either an egg or a developing larva. Each of the latter 10day periods, therefore, may be subdivided by the stage of the offspring in utero. It is suggested that a combination of these observations could be used to determine the age of wild-caught tsetse flies.

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