Medicine. — *Correlation between intestinal and salivary infection in Anopheles maculipennis.* By N. H. SWELLENGREBEL and A. DE BUCK. (Zoological Laboratory, Department of Tropical Hygiene, Royal Colonial Institute, Amsterdam.) (Communicated by Prof. W. A. SCHÜFFNER.)

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Since August 1929 we have regularly infected *A. maculipennis* with the parasite of benign tertian malaria for the purpose of treatment of general paralysis. All matters relating to this last point will be published separately by one of us (DE B.) in collaboration with Dr. KORTEWEG.

Here we only wish to deal with the subject mentioned in the title of this note.

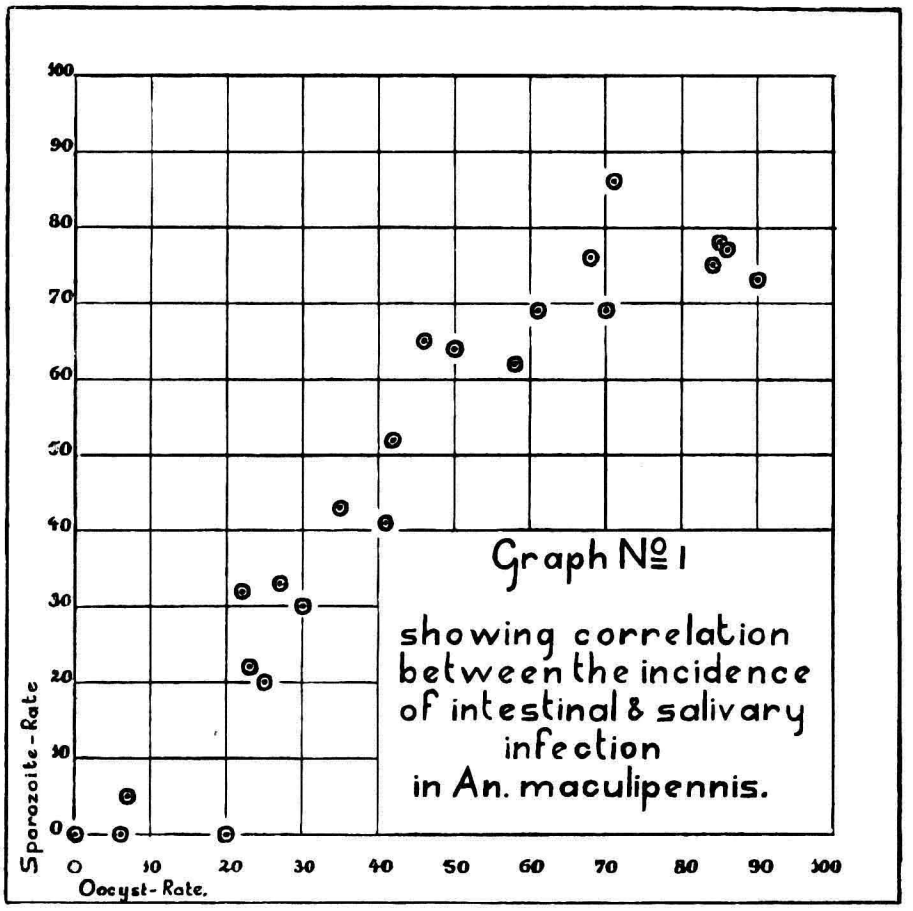
At present there exists a general tendency to discredit the results of investigations on the rate of malarial infection in nature of various Anopheline species, when based on dissection of the gut only. It is asserted that it is not rare to find intestinal infection, not followed by salivary infection. Consequently the rate of the former cannot be considered a reliable index of the importance of a particular Anopheline species as a malaria vector in nature.

Among our batches of infected *A. maculipennis* two had an Oocystrate of 20 % and 6 %, whereas salivary infection was wholly absent. Still the mosquitoes had lived long enough (24 and 36 days) and at a sufficiently high temperature (22° C., like all our batches) for the sporozoites to make their appearance in the salivary glands. This does not rigidly prove that the oocysts degenerated before reaching maturity, but it looks very much like it. So far as this goes our findings countenance the critical attitude mentioned above.

But observations of this kind cannot justify the assertion that the oocyst-rate is unreliable in establishing the epidemiological significance of a given species of Anopheles. This assertion could only be maintained by showing the oocyst-rate and sporozoite-rate to vary independently from one another i.e. by proving the absence of any close correlation between the two.

A number of 23 batches, out of a total of 45 we infected during this year, afforded us an opportunity to verify this point. The average number of Anopheles in each batch was 59 to start with and 28 by the time sporozoites invaded the salivary glands. The mean oocyst-rate (not including mosquitoes dying during the first 6 days after the infecting

meals [1])), for all the batches was 45 %, the mean sporozoite-rate (not including mosquitoes dying at a time sporozoites cannot be expected to



have made their way to the glands) 47 %. The close correlation between the two is plainly shown in the accompanying graph and is more accurately expressed by the formula :

$$r = + 0{,}939 \pm 0{,}024.$$

This unambiguous result should remove all doubt as to the accuracy of the oocyst-rate as a measure to test the suitability of *Anopheles maculipennis* to transmit malaria. And as this doubt mainly originated by investigations on this particular species, we see no reason why it should continue with regard to other species.

It may be objected that the results of experimental infections cannot be applied to conditions obtaining in nature, because there the examination includes mosquitoes which imbibed an infecting meal, but which had not

[1]) The infection of our mosquitoes was carried out as a rule by feeding them on a gamete carrier 3—4 times within a period of 5—6 days.

yet arrived at a stage when oocysts or sporozoites may be expected to have become visible. To meet this objection we have also established the oocyst- and sporozoite-rate of all mosquitoes examined, including the ones which could not be expected to show infections of either kind. The result is different as both oocyst- and sporozoite-rate have decreased, but they remain none the less approximately equal: 39 % and 42 % and the correlation existing between them, although inferior to the first, viz.:

$$r = + 0{,}735 \pm 0{,}092.$$

is still a high one.

Still one may argue: if there exists even the faintest doubt, why not prefer the sporozoite-rate to the oocyst-rate? The accuracy of the latter is at least equal to the former. There is no doubt Anopheles is harmful only when its glands are infected and it is only this degree of harmfulness we are concerned with.

But is the sporozoite-rate the equal in accuracy of the oocyst-rate? We maintain it is not. In various investigations on natural infection of Anopheles, the oocyst-rate almost invariably surpasses the sporozoite-rate, the relation between the two ranging from $1\frac{1}{2} : 1$ to $6 : 1$; in *A. maculipennis* this relation is $5 : 1$.

This may be the true state of things for a particular batch of mosquitoes which happened to be infected simultaneously and to be caught and examined at a time only a few oocysts had attained to maturity. But the reverse may happen as well, when the batch is caught at a time oocysts have disappeared from the stomach wall, and in the long run both rates should become about equal in nature as well as in the experiment. That they do not is explained by the fact that is so much easier to overlook a slight salivary infection than to miss the oocysts which gave rise to it.

Conclusions.

1. In *Anopheles maculipennis* the incidence of intestinal infection allows of a more accurate estimate of the actual number of sporozoite carriers than does the incidence of salivary infection.

2. At present there is no reason why this conclusion should not apply to other species likewise.

3. For practical purpose, in establishing the rate of material infection in nature precedence should be given to the examination of the gut if circumstances do not allow of a more complete investigation.