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OVIPOSITION AND INCUBATION IN *BOOPHILUS DECOLORATUS* (KOCH, 1844) (ACARINA: IXODIDAE)*

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ABSTRACT

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Studies on the duration of the oviposition and incubation periods of *Boophilus decoloratus* (Koch, 1844) revealed that these non-parasitic periods are temperature dependent, increase in temperature causing shortening of the periods. Humidity had no effect on the duration of either the oviposition or the incubation periods. The relation between the mass of engorged female ticks and that of the eggs they produced was found to be linear. The viability of eggs produced during the first 13 days of oviposition (at 26 °C) was greater than that of eggs laid subsequently. The critical temperature for *B. decoloratus* eggs was found to be 42 °C and they were shown to be unable to take up water vapour from damp atmospheres. Both temperature and humidity affected the hatch of eggs. A simple model of the relations between the physical factors of the environment and the various biological phenomena studied has been given.

Résumé

LA PONTE ET L'INCUBATION CHEZ LA TIQUE, BOOPHILUS DECOLORATUS

Des études portant sur la durée de la ponte et celle de la période d'incubation de la tique Boophilus decoloratus *(Koch, 1844) ont révélé que ces périodes non-parasitaires dépendent de la température, l'élévation de celle-ci déterminant un raccourcissement de ces périodes. L'humidité n'a aucun effet sur la durée de ces périodes. Le rapport entre le poids des tiques femelles après l'ingestion complète du sang et celui de leurs oeufs est linéaire. La viabilité des oeufs pondus pendant les 13 premiers jours de la ponte (à 26 °C) dépasse celle des oeufs pondus plus tard. La température critique des oeufs se situe à 42 °C. Les oeufs sont incapables d'absorbes de la vapeur d'une atmosphère humide. L'éclosion des oeufs est influencée par la température et l'humidité. L'auteur présente un modèle simple exprimant le rapport entre les caractères physiques du milieu et les différents phénomènes biologiques qui ont été étudiés.*

INTRODUCTION

Detailed information concerning the life cycle of *Boophilus decoloratus* (Koch, 1844) has only recently become available. The author and his co-workers have published accounts of the anatomy and parasitic life cycle of this species (Arthur & Londt, 1973), its preoviposition period (Londt, 1974), the embryonic development of the larvae (Londt, 1975), gonad development and gametogenesis (Londt & Spickett, 1976), and the adult male's fertilization capacity (Londt, 1976). In addition the distribution of larvae in cattle pastures and their climbing behaviour on grass stems and water balance were discussed by Londt & Whitehead, 1972. The oviposition and incubation periods, however, have not yet been described. Knowledge of the duration of these periods is important when the possibilities of "Pasture Spelling" as a means of controlling this one-host species are considered or when an attempt is made to design acaricide bioassay techniques involving engorged adult female ticks.

OVIPOSITION PERIOD

The relation between engorged female mass and the mass of eggs produced

Twenty-one semi-engorged female ticks, selected for their wide range in size, were pulled off a laboratory infested Guernsey calf, mass measured and allowed to oviposit in tubes held in an incubator (26 °C; 95% R.H.). The relation between female mass and that of the eggs they produced (Fig. 1) is linear and similar to that demonstrated for a number of other tick species, including *Amblyomma americanum* (Gladney & Drummond, 1970); *A. maculatum* (Drummond & Whetstone, 1970); *A. hebraeum* (Norval, 1974); *Anocentor nitens* (Drummond, Whetstone, Ernst & Gladney, 1969); *Boophilus microplus* (Sutherst, 1971); *Hyalomma aegyptium* (Sweatman, 1968); *H. anatolicum* (Snow & Arthur, 1966) and *Rhipicephalus sanguineus* (Sweatman, 1967). The minimum mass a female must attain before eggs are produced was about 17 mg. This implies that, as unengorged females have a mean mass of 1,4 mg (Arthur & Londt, 1973), a blood meal of approximately 16 mg is required for egg production.

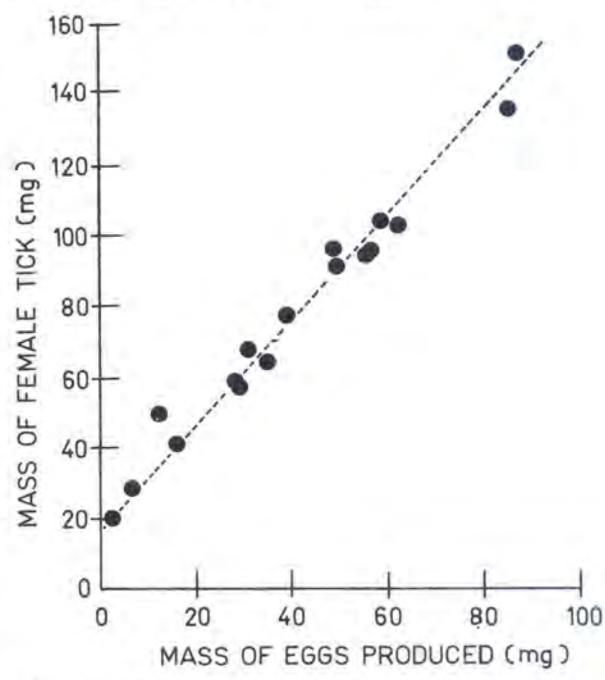

FIG. 1 The relation between the mass of *Boophilus decoloratus* female ticks and that of the eggs they produce

The influence of constant temperature and humidity conditions on the pattern of oviposition and duration of oviposition period

The influence of constant temperature and humidity conditions on the oviposition period of *B. decoloratus* females was studied, 6 different temperatures and 3

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different humidity levels being used (Table 1). The results of mass measuring daily collections of eggs from all the females studied are shown in Fig. 2 a–e.

TABLE 1 Combinations of temperature, relative humidity and saturation deficit used to study the influence of these parameters on oviposition and incubation periods in *Boophilus decoloratus*

Temperature (°C)	Relative humidity (% R.H.)	Saturation deficit (mm Hg)
10	90	0,91
10	70	2,74
10	50	4,57
15	90	1,26
15	70	3,80
15	50	6,33
20	90	1,73
20	70	5,21
20	50	8,68
26	90	2,55
26	70	7,53
26	50	12,51
32	90	3,64
32	70	10,68
32	50	17,72
38	90	8,06
38	70	14,89
38	50	24,71

No oviposition took place at 10 °C. The oviposition patterns for females laying at 38 °C, 32 °C and 26 °C were very similar. Egg production usually reached a peak on the 3rd day, although in a few instances this peak occurred either on the 2nd or the 4th day after laying began (Fig. 2 a–c). Egg production then decreased fairly rapidly and finally ceased about Day 13 (range: 7–16 days). At 20 °C and 15 °C respectively no clearly defined peaks of egg production were found (Fig. 2 d–e), although it was still evident that the majority of eggs were produced during the first half of the oviposition period. The relation between temperature and the duration of the oviposition period, clearly illustrated in Fig. 3, shows that the duration of oviposition remains fairly constant at temperatures of 26 °C and higher, while lower temperatures have the effect of extending the oviposition period from 18,3 days at 20 °C to 32,2 days at 15 °C.

The relation between saturation deficit and the duration of the oviposition period (Fig. 4) is more difficult to interpret. The data plotted in Fig. 4 suggest that the oviposition period is possibly extended by a decrease in saturation deficit (i.e. an increase in humidity). This relation is not real, however, for the same reasons given by Londt (1974) when discussing the effects of saturation deficit on the preoviposition period of *B. decoloratus*. It is obvious that there is an overriding effect of temperature and that the influence of humidity is minimal. This suggestion is supported by the information contained in Fig. 2 a–e, where egg production curves plotted for each humidity level are very similar at each temperature level.

To assess the effects of temperature and humidity on the total egg output of female ticks, the mass of eggs per milligramme of engorged female mass was calculated for each tick to eliminate any effects of female size or degree of engorgement. The relation between temperature and egg output is shown in Fig. 5. There appears to be a tendency for ticks to produce more eggs at high temperatures than at low. However, as the variation about the means was great, this relationship is questionable. The relation between saturation deficit and egg output (Fig. 6) is again difficult to interpret. It appears to be affected by temperature, so it is suggested that humidity does not influence total egg output by female *B. decoloratus* ticks.

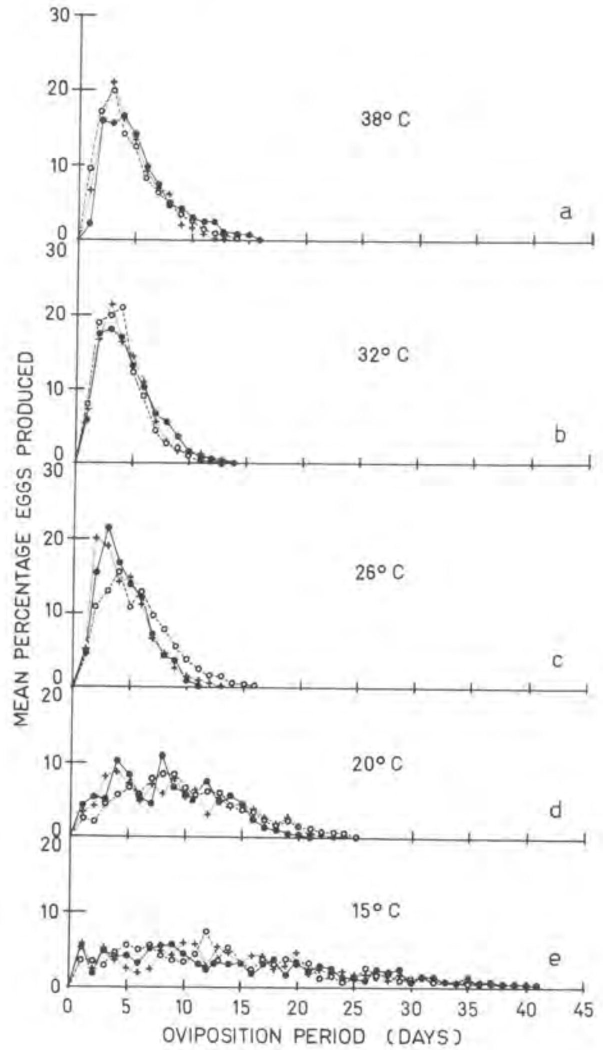

FIG. 2 a–e The influence of constant temperature and humidity on the oviposition pattern of *Boophilus decoloratus*. Data collected at 90% R.H. ●——●; 70% R.H. O----O; 50% R.H. +·····+

The influence of naturally fluctuating temperatures and humidities on oviposition

Two batches of fully fed *B. decoloratus* females were confined in open-ended plastic tubes by means of fine gauze and placed together with a thermo-hygrograph in a Stephenson's screen standing on the ground in a cattle pasture on the farm, Upper Gletwyn, near Grahamstown. *B. decoloratus* larvae had previously been collected in this pasture. These ticks were examined daily, their eggs removed and mass measured. The daily fluctuations in egg production in both batches of females are given (Fig. 7) and the duration of the oviposition period of each female shown (Table 2).

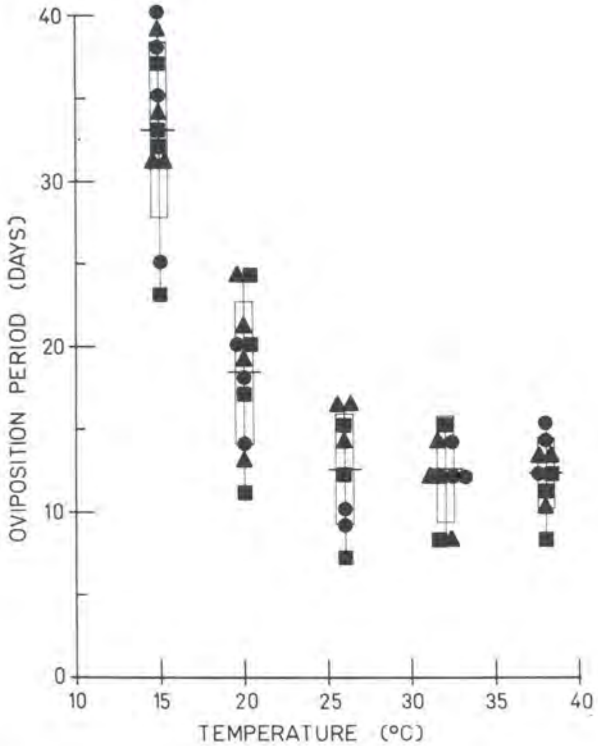

FIG. 3 The relation between temperature and the oviposition period duration of *Boophilus decoloratus*. Data collected at 90% R.H. ●; 70% R.H. ▲; 50% R.H. ■. The horizontal lines indicate means; open boxes indicate one standard deviation about the mean and vertical lines the range at each temperature level

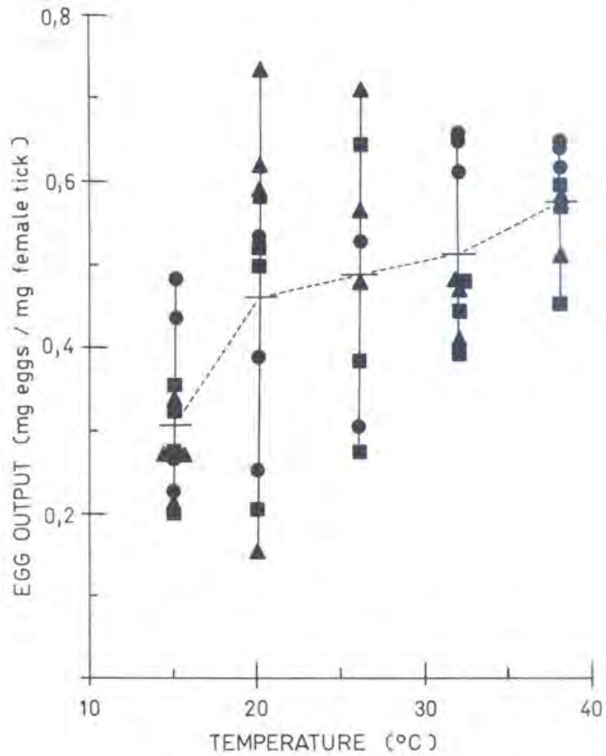

FIG. 5 The relation between temperature and egg output by *Boophilus decoloratus*. Data collected at 90% R.H. ●; 70% R.H. ▲; 50% R.H. ■. Horizontal lines indicate means and vertical lines range at each temperature

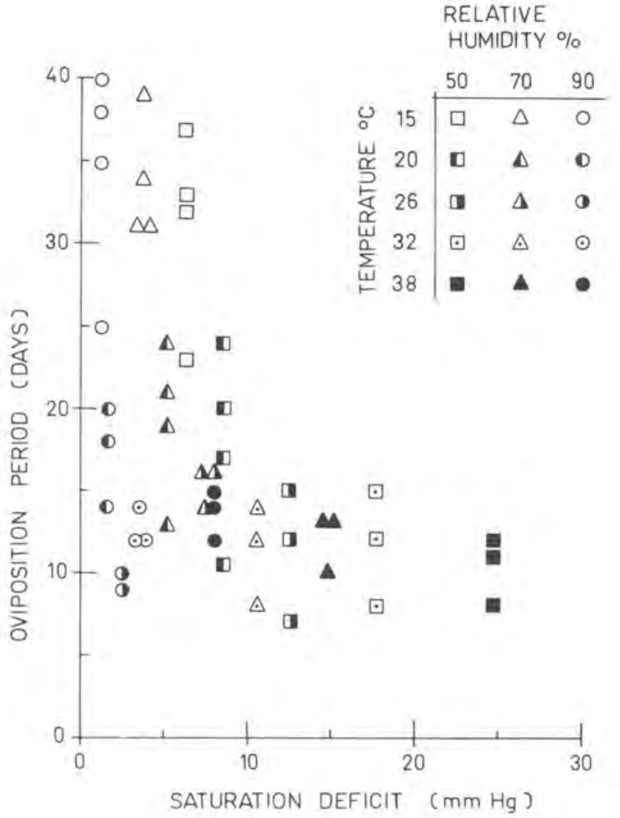

FIG. 4 The relation between saturation deficit and the oviposition period duration of *Boophilus decoloratus*

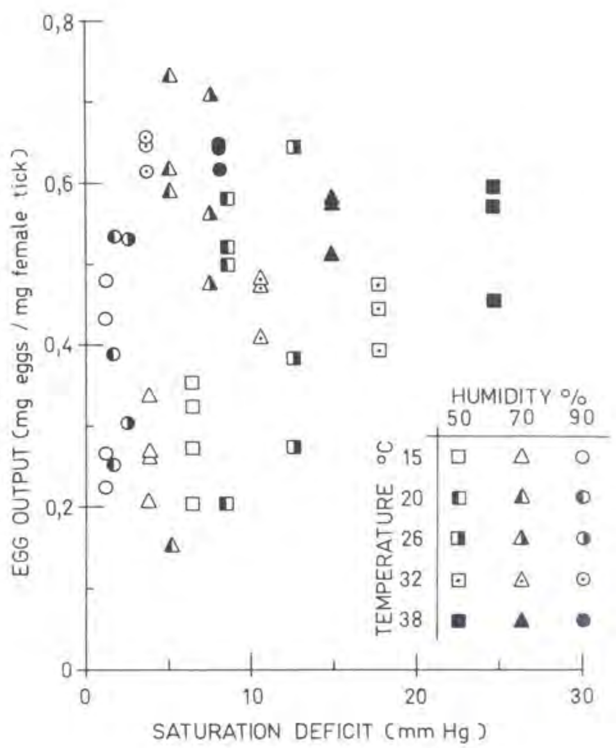

FIG. 6 The relation between saturation deficit and egg output by *Boophilus decoloratus*

TABLE 2 The oviposition periods, in days, of *Boophilus decoloratus* females laying under naturally fluctuating macroclimatic conditions

No. of female	Duration of oviposition period (days)	
	Batch 1	Batch 2
1	38	37
2	36	25
3	34	37
4	—	41
5	—	41
6	—	29
7	—	37
8	—	32
9	—	36
Mean	36	35

The mean mass of eggs produced fluctuated from day to day, the fluctuations being directly related to temperature changes expressed as daily hour degrees above a baseline of 10 °C (Fig. 7). To assess the possible effects that fluctuations of relative humidity and saturation deficit had on oviposition, the number of relative humidity units (i.e. hour % R.H. units above a baseline of 50% R.H.) and the number of saturation deficit units (i.e. hour mm Hg units above a baseline of zero) were calculated for each 24-hour period and plotted together with the fluctuations in hour degrees (Fig. 8). The fluctuations of relative humidity units (hour % R.H. units) were the converse of temperature and saturation deficit units, as would be expected, otherwise all 3 physical parameters followed the same pattern. As oviposition studies under controlled laboratory conditions showed that egg production is not dependent on humidity, it can be accepted that the fluctuations in oviposition illustrated in Fig. 7 are the direct consequence of temperature fluctuations.

INCUBATION PERIOD

The viability of eggs produced on different days of the oviposition period

Boophilus microplus eggs laid towards the end of the oviposition period yielded a lower percentage hatch than those produced during the first few days (Hitchcock, 1955). It was therefore decided to determine the viability of eggs laid on successive days throughout the oviposition period of *B. decoloratus*. Eggs were collected daily from 10 fully engorged female ticks and placed in separate glass vials in an incubator (26 °C; 95% R.H.) to hatch. When hatching had ceased, the percentage hatch of each egg batch was determined and then plotted against the day on which the eggs were produced (Fig. 9). Except for Day 8, for which a mean hatch of 82% was recorded, eggs laid over the first 13 days of the oviposition period yielded hatches of over 95%. The percentage hatch of eggs produced after Day 13 decreased progressively; of those laid on Day 19 (the last day of the oviposition period) just under 10% hatched. The reasons for this decrease in hatch have not been established but the decrease could be due to any of the following factors:

1. A possible decrease in the supply of nutrients to the eggs during their formation in the ovaries.
2. A possible decrease in the water content of the eggs during their formation.
3. A possible decrease in the efficiency of Gene's organ in the waterproofing of eggs.
4. A possible breakdown of the fertilization mechanism.

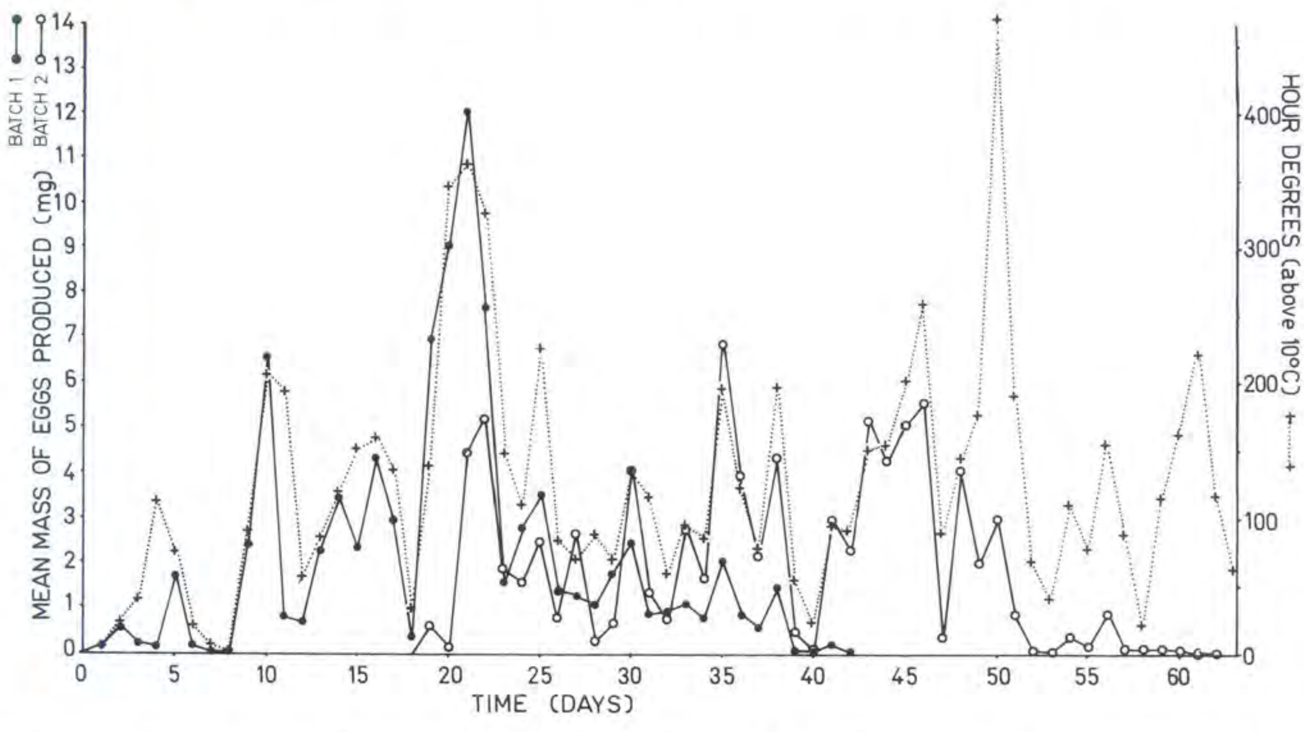

FIG. 7 The oviposition patterns of *Boophilus decoloratus* females (●——● = Batch 1; O——O = Batch 2), in relation to natural daily fluctuations in hour degrees (above 10 °C) (+·······+)

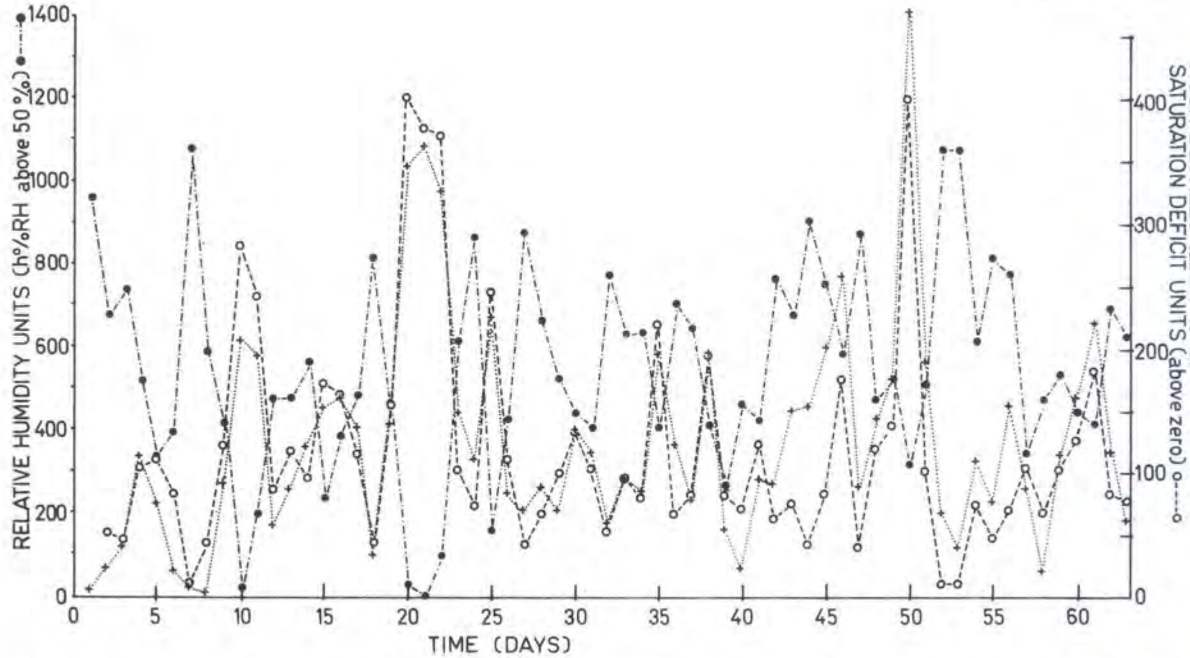

FIG. 8 The interactions between temperature (h °C above 10 °C, +·······+), relative humidity (h % R.H. above 50% R.H., ●-.-.-●) and saturation deficit (h mm Hg above zero, O----O) during studies on oviposition by *Boophilus decoloratus* under naturally fluctuating climatic conditions

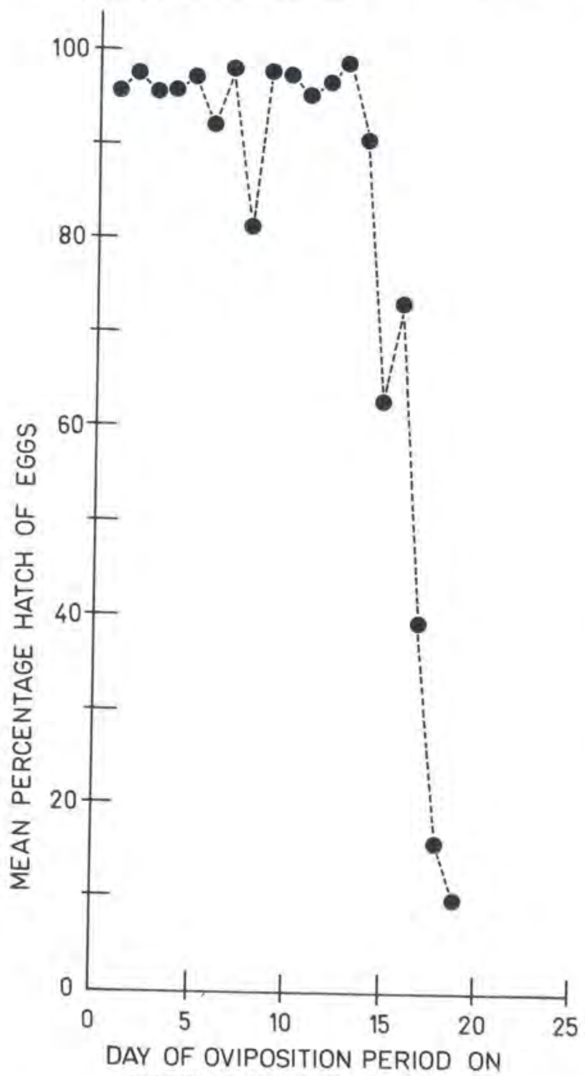

FIG. 9 The percentage hatch of *Boophilus decoloratus* eggs in relation to the day on which they were laid

Critical temperature of cuticular wax layer of eggs

The critical temperature is the temperature at which the protective wax molecules covering the cuticle are reorientated, with consequent loss of water from the tick. Lees & Beament (1948) determined the critical temperatures of the egg waxes of 6 tick species, as follows: *Ixodes ricinus* 35 °C; *I. canisuga* 42,5 °C; *Dermacentor andersoni* 43 °C; *Hyalomma savignyi* 44 °C; *Ornithodoros moubata* 45 °C and *O. delanoei acinus* 46 °C.

The critical temperature of *B. decoloratus* eggs was established by mass measuring egg batches before and after exposure for 1 hour, to different temperatures. The results obtained indicate a critical temperature of 42 °C (Fig. 10). Londt & Whitehead (1972) recorded temperatures of this order in field situations where vegetation cover was limited and where no *B. decoloratus* larvae were ever collected. It is possible that eggs produced by adult female ticks in situations where temperatures of 42 °C or over are experienced, would lose water very rapidly and die.

The uptake of water vapour by eggs

Londt (1975) demonstrated that *B. decoloratus* eggs lost water at all humidity levels below 100% R.H. and that successful development of the embryos was entirely dependent on the rate at which water was lost. Eggs losing more than about 35% of their initial mass through water evaporation did not complete embryonic development. In previous studies it was shown that *B. decoloratus* larvae were able to take up water vapour when the atmospheric humidity exceeded 70% R.H. and imbibe free water through their mouthparts (Londt & Whitehead, 1972). No information has been available to date regarding the uptake of water vapour by eggs. To assess the ability of *B. decoloratus* eggs to take up water vapour, 4 batches were mass measured and placed in an incubator (26 °C) in 4 different humidity chambers: 95% R.H. (1,30 mm Hg); 80% R.H. (5,04 mm Hg); 60% R.H. (10,02 mm Hg) and 40% R.H. (15,00 mm Hg). After 24 hours all the egg batches were again mass measured, transferred to the 95% R.H. chamber and thereafter mass measured at 24 hour intervals. The results (Fig. 11) show that, during the first 24 hours, eggs held at 95% R.H. increased slightly in mass whereas the rest all lost mass: the lower the humidity the greater the loss recorded. After their transfer to 95% R.H., none of the egg batches increased markedly in mass; so they are apparently unable to take up water vapour from damp atmospheres. This means that the egg stage of *B. decoloratus* is more sensitive to desiccation than the larval stage, a fact that may have important implications in future biological and ecological research on these ticks.

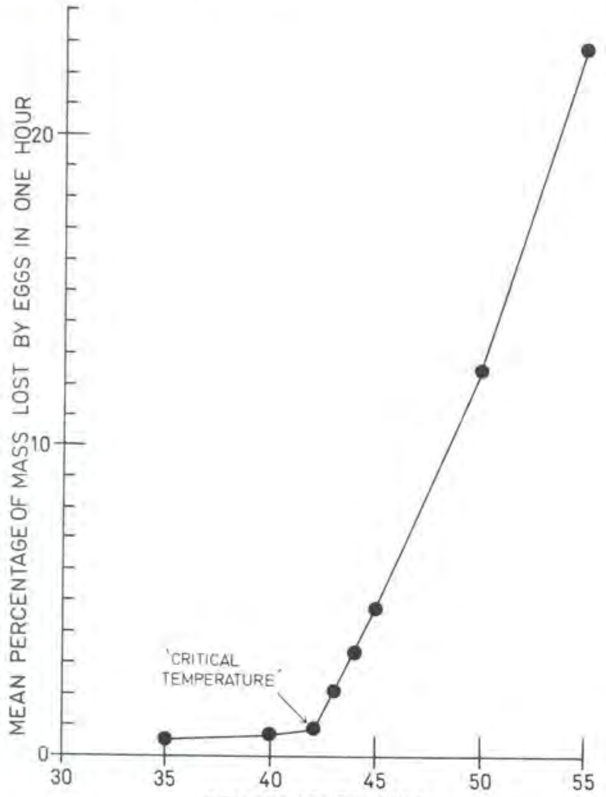

FIG. 10 The mean percentage mass lost by *Boophilus decoloratus* eggs exposed to constant temperature values between 35 °C and 55 °C for 1 hour. Critical temperature of 42 °C is indicated

The influence of constant temperature and humidity conditions on egg development and hatch

To assess the influence of different combinations of temperature and humidity on the duration of the incubation period of *B. decoloratus* eggs and the larval yield, batches of eggs were exposed to the same 18 combinations of temperature and humidity used in work on the oviposition period (Table 1). All vials were examined daily and the length of the incubation period recorded when the first larvae hatched. Three weeks later all larvae and eggs were counted and the percentage hatch determined.

The relation between temperature and incubation period duration is shown in Fig. 12 and that between saturation deficit and incubation period duration in Fig. 13. No hatch took place at 10 °C, 15 °C and 38 °C. At 20 °C only eggs held at 90% R.H. hatched while at 26 °C and 32 °C only eggs held at 90% R.H. and 70% R.H. hatched. The incubation period was extended by a decrease in temperature. Again it can be seen that temperature has an overriding effect and that humidity has no influence on the incubation period duration.

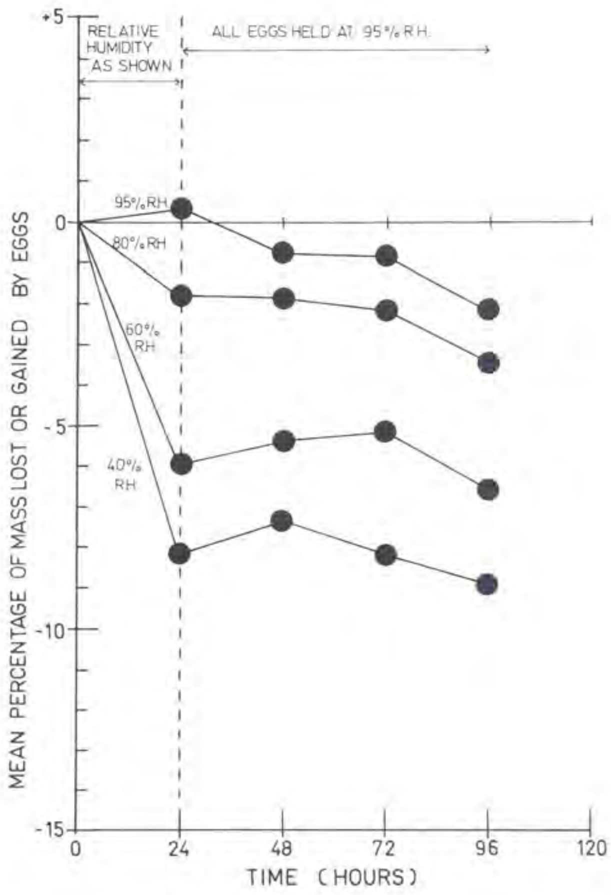

FIG. 11 The effects of placing *Boophilus decoloratus* eggs in a damp atmosphere (95% R.H.) after an initial 24 hour period in 1 of 4 different relative humidity levels. The temperature was 26 °C throughout

While considering the effects of temperature and saturation deficit on egg hatch (Fig. 14–15), it is evident that there is a definite saturation deficit dependence and an apparent, possibly indirect, temperature dependence. Temperature influences the duration of the incubation period and eggs held at low temperatures will have longer to lose water through evaporation before they hatch. At similar saturation deficit levels, eggs may develop into larvae at 32 °C whereas they may not be able to complete development at 20 °C because this would take much longer. It is possible, therefore, that the results shown in Fig. 14 and 15 reflect another limiting factor such as the water content of the eggs.

Discussion

The effects of temperature and humidity on various aspects of the non-parasitic life cycle of *B. decoloratus* reported in this and previous publications (Londt, 1974, 1975) may be summarized by means of a model (Fig. 16) resembling that described by Macleod (1962) for the dynamic community relations in which *I. ricinus* participates. Each disc represents a measurable parameter. A clockwise rotation of any disc means an increase in the effect of the parameter involved. It is important to note that the main 'drive' for the model is through the physical factors

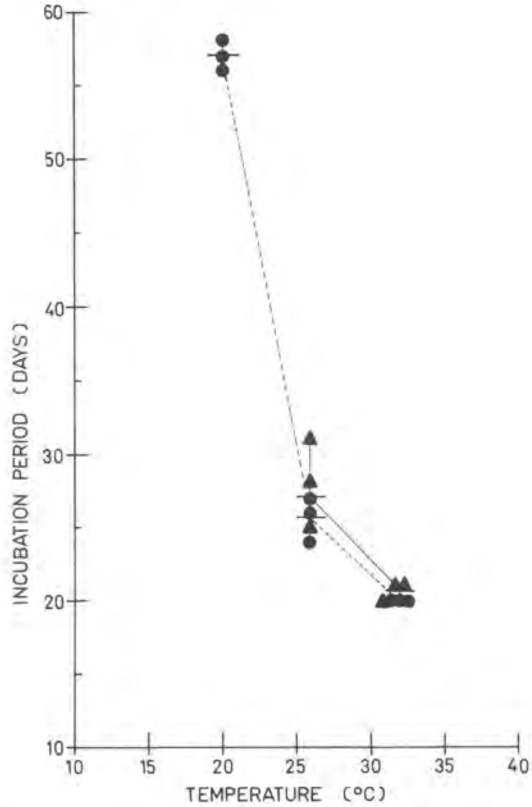

FIG. 12 The relation between temperature and incubation period duration of *Boophilus decoloratus* eggs. Data collected at 90% R.H. ● and 70% R.H. ▲. Eggs held at 50% R.H. failed to hatch. For convenience means at each temperature level have been joined

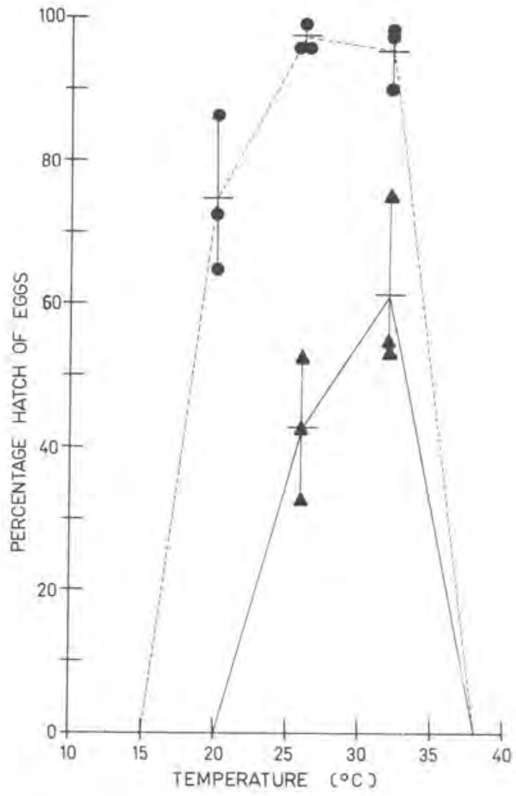

FIG. 14 The relation between temperature and the percentage hatch of *Boophilus decoloratus* eggs. Data collected at 90% R.H. ● and 70% R.H. ▲. For convenience means at each temperature level have been joined

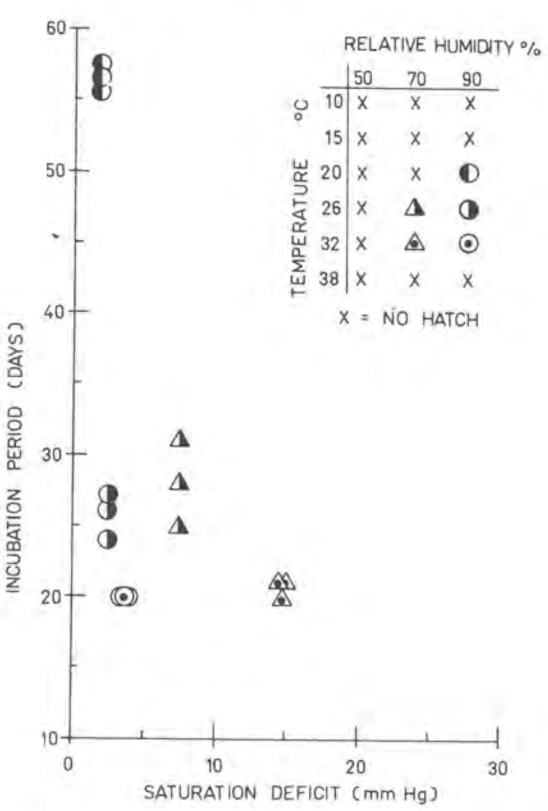

FIG. 13 The relation between saturation deficit and incubation period duration of *Boophilus decoloratus* eggs

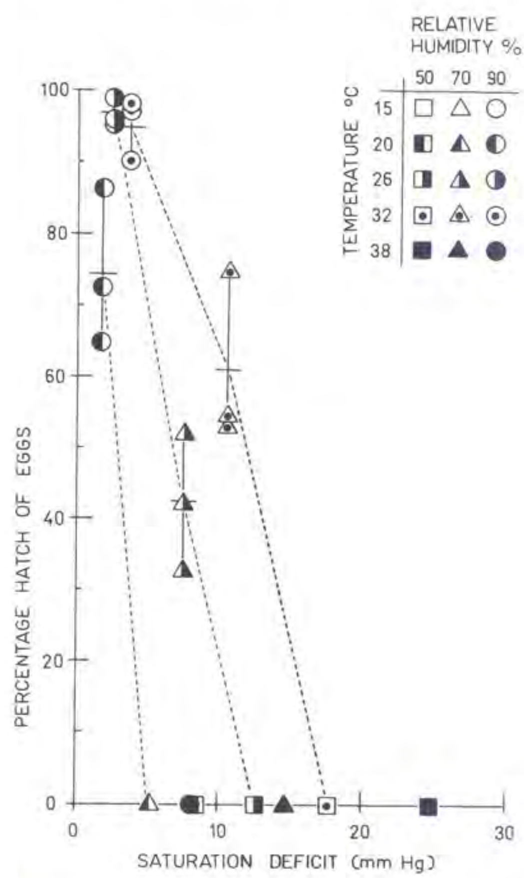

FIG. 15 The relation between saturation deficit and the percentage hatch of *Boophilus decoloratus* eggs. Means for data at each temperature level have been joined

of the environment. This means that a change in temperature results in a change in metabolic rate but that the reverse is impossible. Thus it can be seen that, with temperature increase, relative humidity decreases and saturation deficit increases. Temperature increase also causes a decrease in the durations of the non-parasitic phases (preoviposition, oviposition and incubation periods), presumably by increasing the metabolic rate of the ticks. A rise in temperature also appears to increase the total egg production by female ticks, presumably because the metabolic rate and efficiency of food utilization increase. A rise in saturation deficit, which could be caused by a rise in temperature, results in increased water loss from tick eggs; this directly affects the percentage hatch of the eggs.

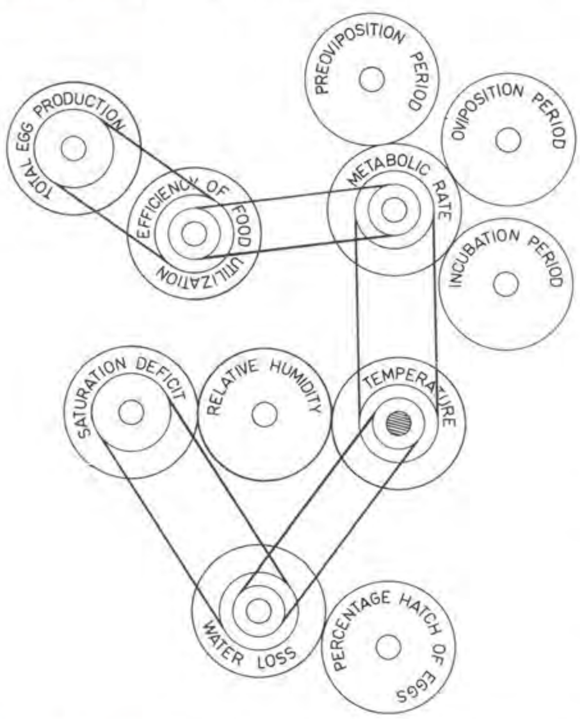

FIG. 16 A model of the interactions between the physica factors of the environment (temperature, relative humidity and saturation deficit) and various parameters investigated during a study of the biology of *Boophilus decoloratus*

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