BOOPHILUS MICROPLUS (CANESTRINI) (ACARIDA : IXODIDAE) ON THE BOVINE HOST II. DISTRIBUTION OF STAGES DURING DEVELOPMENT

BY

Gordon F. BENNETT [1]

ABSTRACT

Distribution of the larvae, nymphs and adults of a population of *Boophilus microplus* was determined over 21 body areas of the host. The data indicated that certain areas were more favoured (or tick survival in these areas was better) by the ticks than others, each stage having ' preferred ' zones, the older ticks proportionally more numerous on the posterior portions of the host. The pattern of distribution of the tick populations on susceptible, resistant, and resistant calves restrained from self-grooming, were similar.

ABSTRAKT

Die Verteilung der Larven, Nymphen und Erwachsenen einer Bevölkerung von *Boophilus microplus* wurde auf 21 Körperpartien des Wirtstieres beobachtet. Die Resultate ergaben, dass bestimmte Körperpartien von den Zecken bevorzugt wurden (oder die Zecken überlebten besser auf diesen Körperteilen). Jedes Stadium wiederum hatte bevorzugte Körperteile. Die älteren Zecken waren proportionell zahlreicher auf dem Hinterteil des Wirts vertreten. Die Art der Verteilung der Zecken-Bevölkerung auf anfälligen, widerstandsfähigen, und widerstandsfähigen Kälbern, die am Selbst-Säubern verhindert wurden, war ähnlich.

INTRODUCTION

LEGG (1930) and WILKINSON (1955) noted pronounced irregularities in the distribution of a cattle tick population on its host. ROBERTS (1968 *b*) stated that removal of all ticks from cattle following a constant, prolonged, daily regime of infestation should show whether certain stages of the tick predominate on certain body areas of the host, and whether such distribution is influenced by the susceptibility of the particular animal to infestation by *Boophilus microplus*. This study was designed to test Robert's hypothesis, and to indicate if the distribution of the stages of a tick population differs on susceptible, resistant, and resistant animals restrained from self-grooming. The technique used herein, differing from that of ROBERTS (1968 *b*), was to remove all ticks from the *living* host, noting their distribution as they were removed.

1. Department of Biology, Memorial University of Newfoundland, St. John's, Newfoundland, Canada.

Materials and methods

The animals used in this study were two pure-bred Jersey (four months old) and three Jersey-Sahiwal crossbred (six months old) steers, obtained from the Division of Animal Genetics, CSIRO, at Badgery's Creek, New South Wales. These animals were infested with 1 000 larval ticks daily after the rationale of Roberts (1968 *a*) and maintained in roofed stalls. These animals, which had no previous experience with ticks, were allowed to stabilize to a relatively constant yield of mature female ticks (Roberts, 1968 *a*). Two of the Jersey-Sahiwal crossbreds (Animals 3 and 4) were placed in anti-grooming harnesses (Bennett, 1969) for four weeks prior to examination ; both pure-bred Jerseys (Animals 1 and 2) and the other Jersey-Sahiwal crossbred (Animal 5) were unrestricted. The five animals were infested for a minimum of 60 days before all ticks were picked off. The average daily yield of mature female ticks for the 40 days prior to removal of all ticks was used to determine the susceptibility of the experimental calves. The susceptibility rating refers to the survival and maturation of larvae to the adult stage and is equivalent to Snowball's (1956) " survival percentage ". On this basis, Animal 1 was considered to be susceptible, animals 2, 3 and 4 as moderately susceptible and animal 5 as moderately resistant.

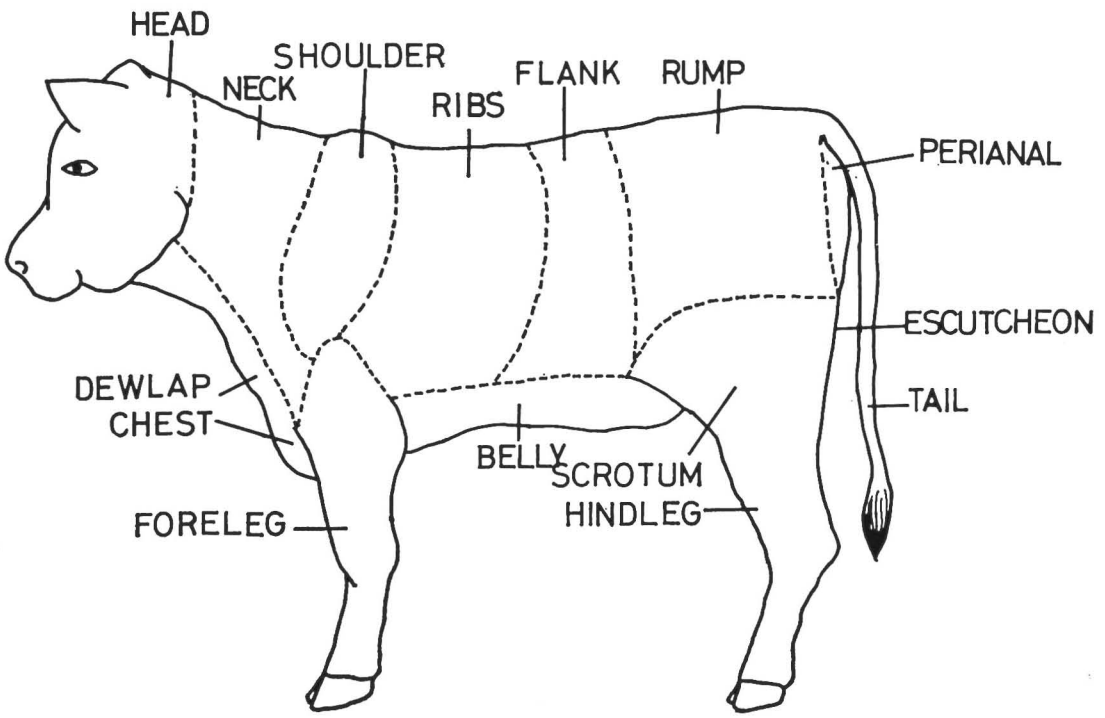


Fig. 1 : Body regions used in study of tick distribution.

To determine the distribution of the various stages of the tick population on the host, all ticks were removed from the living host by picking them off with forceps. To facilitate the removal of the ticks, the hair of the host was clipped close to the skin and the body marked, with indelible pencil, into 21 arbitrarily chosen zones (Fig. 1), viz. — head, dewlap, brisket, belly,

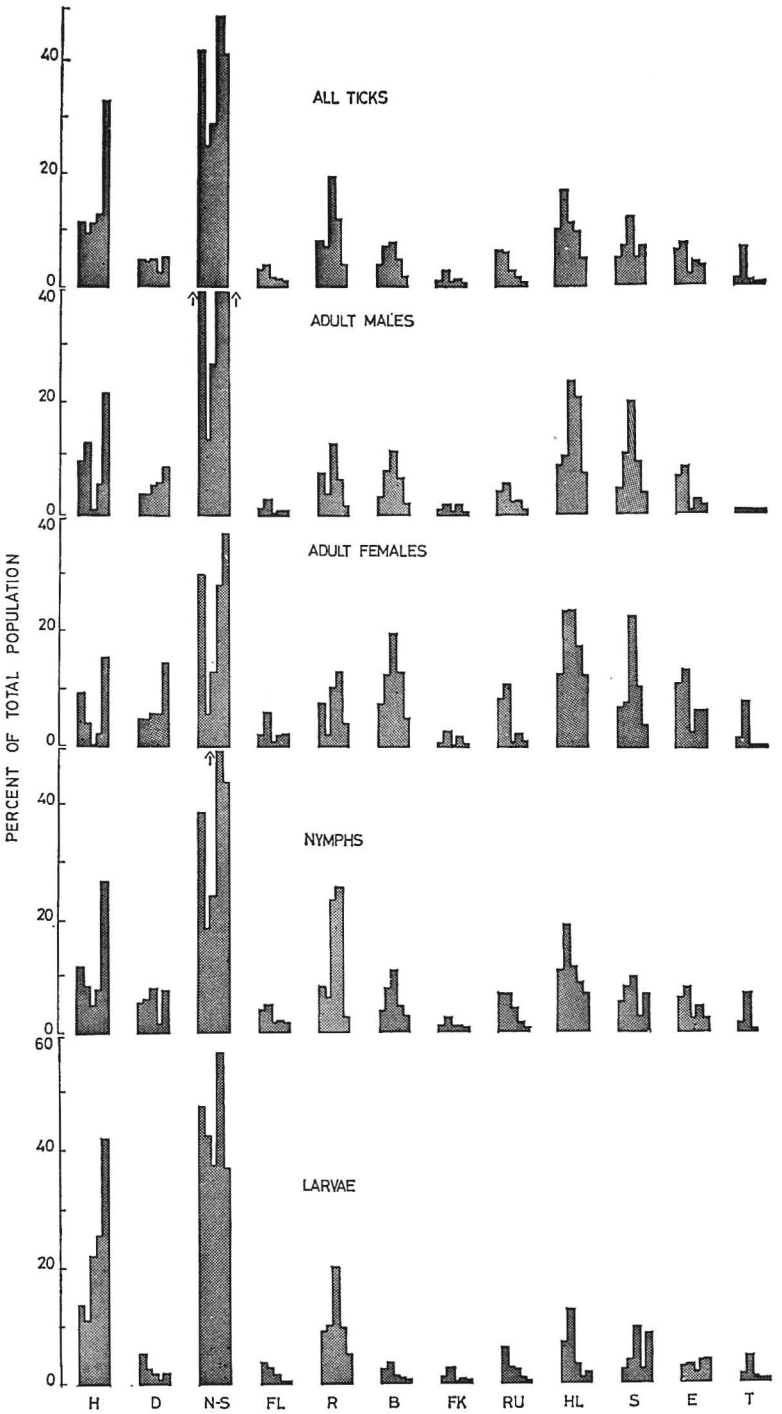


FIG. 2 : Distribution of stages of tick population on host.

scrotum, escutcheon, tail, right and left foreleg, neck, shoulder, rib cage, flank, rump and left and right hind leg. The data are presented as the combined results for the right and left sides and the dewlap-brisket (= chest), reducing the 21 zones to 12 in the analyses. An attempt was made to remove all ticks in these areas on the first day of examination. To ensure that ticks had not been overlooked, especially the smaller larval and nymphal stages, each animal was examined on a further two occasions, and the ticks found classified as to the age they would have been on the first day of examination. The animals were returned to their stalls and ticks maturing throughout the following 29 days were recorded as an additional control of the efficiency of the examinations.

The physical labour of removing the ticks was carried out by a minimum of three persons at any one examination, usually four or five on the first examination which extended over a period of four to seven hours. Fortunately, the calves used in this study were exceptionally docile, permitting detailed and prolonged examination of the eyelids, ears, escutcheon, scrotum, etc. The animals often lay down during the examination and slept with their heads on the lap of one of the examiners, thus providing the opportunity for a particularly close scrutiny for ticks on this region. This docility permitted the examination and removal of ticks without the use of narcotizing or anaesthetic agents.

Ticks collected from each zone were preserved by freezing until they were examined and classified. Frozen specimens were found to be superior to those kept in the usual preservatives as they retained the colour characteristics of the living ticks and did not materially change in mass or linear dimensions. Sex ratios were based on the engorged, resting nymphs ; female nymphs were readily identified by both their larger size and prevailing orange colour. Infestation with larvae was terminated 24 hours preceding examination ; thus all ticks in the study were 24 hours of age or older.

Results

Removal of all ticks from an animal required a minimum of 36 manhours ; for most animals, 50 man-hours was required spread over three examinations. The first examination resulted in the removal of 70 % or more of the total ticks but two further examinations did not yield all the remaining ticks ; up to 3.3 % of the population was recovered during the ensuing 29 days in the form of engorged adult females — presumably a similar proportion of adult males were present but not recovered. Small larvae and nymphs were often overlooked on the first examination and were found later during examinations when their increase in size made them easier to locate.

The distribution of the three developmental stages of the tick on the five hosts is shown (Figs. 2 and 3), and the data indicates that certain areas of the host — namely forelegs, flanks, rump and tail — did not support large populations of ticks. One other area, the chest (dewlap-brisket), was not particularly favoured, even on the most susceptible animal (animal 1). The tick population was not equally distributed on both sides of the host. Three animals (Table I) had significantly (P_{01}) higher numbers of ticks on one side, while the remaining two had equal numbers on each side. In considering only the maturing female ticks, however, there was no significant difference in the number of ticks on the two sides of four of the animals.

Larvae

The distribution of larvae over the host was generally similar for all five animals. Larvae predominated on the neck-shoulder region and head, decreasing in numbers towards the posterior

TABLE I. — Comparison of the total ticks (all stages) and maturing female ticks (17-18 days of age) on the right and left sides of a host *.

Animal	Total Ticks		Maturing Ticks	
	Right	Left	Right	Left
1	2 149	2 312	15	10
2	1 048	1 102	11	9
3	468	333	7	6
4	490	738	12	17
5	416	408	3	8
Total	4 571	4 893	48	50

* Numbers in this table exclude ticks on the head, belly, scrotum, dewlap, escutcheon and tail.

regions. Larvae were more numerous on the scrotum, escutcheon and hind leg (mainly inner thighs) than on the flank, rump and belly and this suggests that some areas are ' preferred ' by the larvae. Within the broadly defined zones, there was some evidence of preference for ' microhabitats '. On the head, for example, the majority of the ticks occurred on the ears, generally on the inner margin of the apex. On the neck-shoulder region, the majority of the larvae were on the mid-dorsal line, especially concentrated at the apex of the shoulder blades. On animal 5, the larvae on the head and neck-shoulder regions represented over 75 % of the total larvae and on the other animals, over 50 % of the larvae occurred here. This high proportion undoubtedly represents the fact that infestation was by a neck collar method and these areas are adjacent to the source of infestation. The higher proportion on animal 5 may result from the fact that these areas represented the only two areas which could not be effectively self-groomed.

Nymphs

The distribution of nymphs was similar to that noted for larvae (Figs. 2 and 3) with the neck-shoulder region harbouring the largest single population of this stage. However, nymphs were more widely distributed over the body and proportionally more occurred on the posterior regions of the host. On some animals the head region, although supporting a large population of larvae, supported relatively few nymphs. The escutcheon region seemed favourable for nymphs. Distribution of nymphs suggested that the population was shifting from the anterior to the posterior.

The ease of sexing engorged nymphs allowed the sex ratio to be calculated more accurately than using the adults, as the loss of wandering adult males from the host was always possible. The sex ratio of the ticks varied among the five animals, from almost equal males to females (as postulated by HITCHCOCK, 1955) to a ratio of 1 male : 1.4 female on animal 2. Four of the animals showed a preponderance of females. The sex ratio also varied markedly on various portions of the same host. On all animals, the posterior regions had a higher female to male ratio whereas on the neck-shoulder region, males usually predominated (Fig. 3). The evidence suggests that (a) the female nymphs prefer the posterior regions and seek them out or (b) there is a differential survival of the sexes on the various regions of the host.

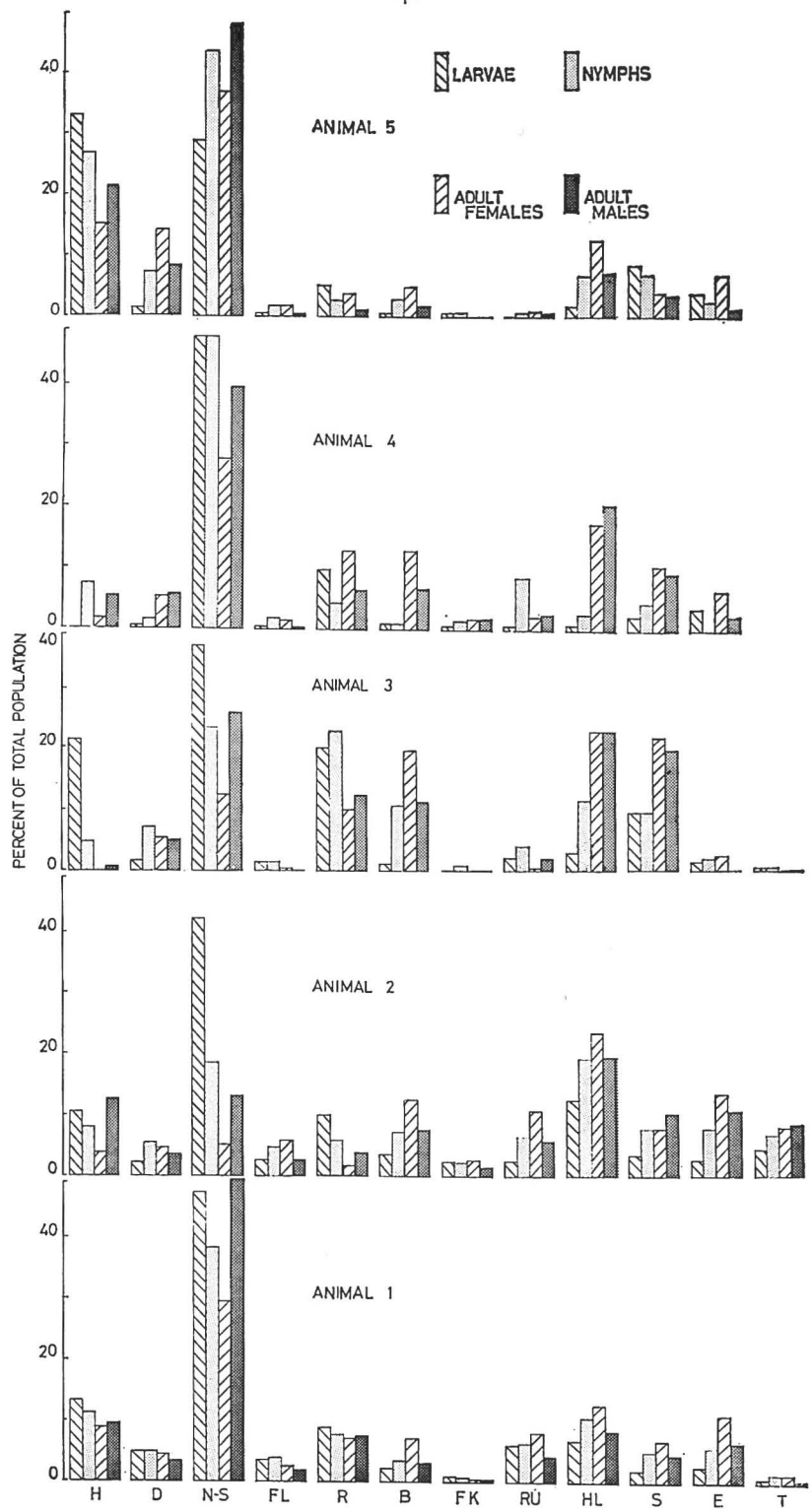


FIG. 3 : Distribution of tick population on different hosts.
Animal 1 — susceptible, Animals 2, 3 & 4 — moderately susceptible ; Animal 5 — moderately resistant.
Animals 3 & 4 in anti-grooming harnesses.

Adult females

Distribution of the adult females on the host differed considerably from that seen for the immature stages. While large numbers still occurred on the neck-shoulder region (Figs. 2 and 3), the proportion of the female population occurring on the head was sharply lower, especially on animal 3, where no females were found, althought 20 % of the larval population occurred there. A marked shift of the females to the posterior regions of the host were noted in all animals (Fig. 3). This shift of the tick population from the anterior to the posterior as the population matures is illustrated in Fig. 4 in which the posterior zone is defined as that part of the host caudad to the rib cage. Despite the general trend, however, there was considerable variability between the individual calves. It is noteworthy that despite variation in the level of resistance of the hosts, and the fact that two animals were artificially restrained from self-grooming, the distribution of ticks on all the animals was basically similar, suggesting that the ticks selected sites of attachment and that grooming by the host had relatively little effect.

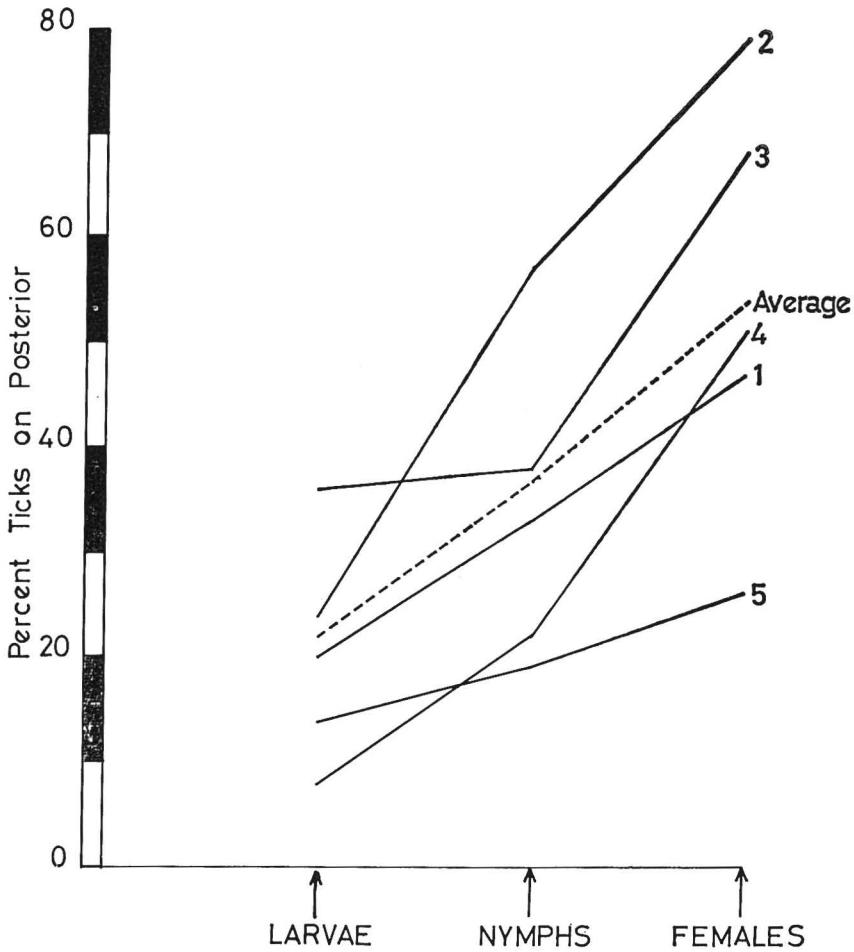


FIG. 4 : Shift of tick population during maturation of population.

Adult males

The distribution of adult males was broadly similar to that described for the adult females (Figs. 2 and 3). There was some indication of an anterior to posterior shift of the male population. While the presence of males could be correlated with the presence of females in some zones, the presence of males seemed to be correlated more with the distribution of nymphs (animal 3, head ; Fig. 3). Twenty percent of the adult males were found sharing the same attachment site as the engorged nymph. Surprisingly, however, these males were not invariably associated with female nymphs ; on animal 1, for example, of the 89 males associated with nymphs, 33 (37 %) were associated with male nymphs.

Discussion

The distribution of the tick population indicated that the forelegs, flanks, rump and tail supported comparatively few ticks. This distribution occurred on all five animals, despite the fact that two of the hosts were susceptible, two were moderately resistant but restrained from self-grooming and one was resistant but unrestrained. Presumably, therefore, the unattractiveness of those areas not utilized by the tick was due to some inherent factor common to these five areas in all five animals. Elucidation of the differences in the skin between various regions of the same host might be profitable for future research.

Distribution of the different stages of the tick on the same animal was not uniform (Figs 2 and 3). The greatest proportion of the larval population was found on the neck-shoulder and head regions. This may have been due to the animals being infested with larvae released from a vial placed in a collar around the neck. Assuming no movement of the population, the proportions of each stage on each area should be the same throughout development. However, it is clear that as the population matures, the proportions of each stage on each area vary. On some animals (Fig. 3, animal 1, head), the proportions of all the stages are about equal on some areas, suggesting no movement. On other animals (fig. 3, animal 3, head — no adult females), the same area may show completely different proportions. Animal 5 (Fig. 3) showed a consistently high number of all stages on the head ; possibly this was related to the fact that this was the only portion of this resistant (but unrestrained) animal that could not be self-groomed. No consistency of pattern was noted except on a broad scale, the major portion of the larval population occurred anterior to the flank region. Nymphs were more equally distributed over the animal than the larvae, and the majority of the females occurred on areas posterior to the flank (Fig. 4). This data can be interpreted in two ways :

1) The population moves in an anterior to posterior direction as it matures. While it is generally considered that once established, a tick does not generally move about on its host (except at ecdysis), recent work (Bennett, 1974) has shown that *Boophilus microplus* has the capability to attach and detach at mid-instar and that, on occasion, as much as 10 % of the female population can do this nightly. Ammah-Attoh (1966) described similar movement for *Boophilus decoloratus*. In view of this, a shift of the maturing population in a posterior direction is quite feasible. The different proportions of the various tick stages on various parts of the host could thus be explained by a selection of ' preferred sites ' by the ticks, such preferred sites for the adult females being primarily towards the posterior, especially the peri-anal-scrotum-escutcheon-inner thighs region.

2) There is differential survival of the various tick stages on different parts of the host. If this were the case, certain areas of some animals must be antagonistic to one or more stages of the tick, and in some animals (Fig. 3, animal 3, head) particularly antagonistic to one stage. Reasons for such variable skin reactions are difficult to formulate.

The pattern of the distribution of the tick population on these hosts was broadly similar in that areas such as the flank, dewlap, forelegs, etc., consistently supported lower populations of ticks than other areas. There was also considerable variation in detail between the individual hosts, probably related, in part, to the imposed conditions of the study. However, differences noted between animals 1, 2 and 5 and animals 3 and 4, the members of each group kept under similar conditions, must be due to variability between individual hosts. The resistant hosts differed from the susceptible in that the mortality of the larvae of 24-48 hours of age was higher (ROBERTS, 1968 ; BENNETT, 1974). Those larvae which successfully established on the resistant hosts continued to develop and the subsequent distribution of the ticks on the host was similar to that noted for susceptible animals (Figs. 2 and 3). Similar conclusions were drawn by ROBERTS (1968).

The great variability of the distribution of a population of ticks has been commented on by LEGG (1930) and WILKINSON (1955). This study indicates the inherent variability of the surface of the host as it relates to the attachment and maturation of a tick population and explains in part the irregularities of distribution noted by Legg and Wilkinson although offering no reasons for the variability of the host surface. This study also stresses the complexity and variability of results encountered while studying the host-parasite relationships of *Boophilus microplus* and the folly of generalizing too extensively about this parasite.

ACKNOWLEDGEMENTS

This work was carried out while the author was with the Division of Entomology, CSIRO, Long Pocket Laboratories, Indooroopilly, Queensland, Australia. It is a pleasure to acknowledge the advice and assistance of Dr. R. H. WHARTON throughout this study. Messers Utech, Newman, Carnell and Wooderson lent invaluable assistance in the tedious process of removing ticks from the animals. Misses G. MULLINGAN and D. MCBRYDE, in addition to removing ticks from the calves, analyzed much of the data. Miss Judy FITZPATRICK prepared the illustrations and Mr. P. R. WILKINSON provided much valuable criticism of the manuscript.

LITERATURE CITED

AMMAH-ATTOH (V.), 1966. — Development of *Boophilus decoloratus* (Koch, 1844) on the ox. — Nature, London, **209** : 629-630.

BENNETT (G. F.), 1969. — *Boophilus microplus* (Acarina : Ixodidae) : experimental infestations on cattle restrained from grooming. — Exp. Parasitol., **26** : 323-328.

BENNETT (G. F.), 1974. — *Boophilus microplus* (Canestrini) (Acarida : Ixodidae) on the bovine host. I. Mortality during developmental cycle. — Acarologia. (in press).

HITCHCOCK (L. F.), 1955. — Studies on the parasitic stages of the cattle tick, *Boophilus microplus* (Canestrini) (Acarida : Ixodidae). — Aust. J. Zool., **3** : 145-155.

LEGG (J.), 1930. — Some observations on the life history of the cattle tick (*Boophilus australis*). — Proc. Roy. Soc. Queensland, XLI : 121-132.

ROBERTS (J. A.), 1968 *a*. — Acquisition by the host of resistance to the cattle tick, *Boophilus microplus* (Canestrini). — J. Parasitol., **54** : 657-662.

ROBERTS (J. A.), 1968 *b*. — Resistance of cattle to the tick, *Boophilus microplus* (Canestrini). I. Stages of the life-cycle of the parasite against which resistance is manifest. — J. Parasitol., **54** : 667-673.

SNOWBALL (G. J.), 1956. — The effect of self-licking by cattle on infestations of cattle tick, *Boophilus microplus* (Canestrini). — Aust J. Agric. Res., **7** : 227-232.

WILKINSON (P. R.), 1955. — Observations on infestations of undipped cattle of British breeds with cattle tick, *Boophilus microplus* (Canestrini). — Aust. J. Agric. Res., **6** : 655-665.