THE EFFECTS OF TICK (*BOOPHILUS MICROPLUS*) INFESTATIONS ON THE BLOOD COMPOSITION OF SHORTHORN × HEREFORD CATTLE ON HIGH AND LOW PLANES OF NUTRITION

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Summary

The effects of heavy infestations of tick (*B. microplus*) on the blood composition of Shorthorn × Hereford steers on low and high planes of nutrition were studied.

Steers fed wheat straw *ad libitum* for 50 days and infested with tick larvae on five occasions to a total dose of 9 g carried an average of 5324 mature female ticks and lost 18·1% empty body weight, as compared with 10·5% loss in tick-free animals fed similarly. The tick-infested animals showed significantly greater reductions from the initial values in haematocrit, haemoglobin, serum albumin, total protein, total cholesterol, triglyceride, total lipid, and amylase activity than the tick-free animals.

Steers fed high-quality lucerne hay *ad libitum* for 58 days and infested with six doses of tick larvae to a total dose of 28 g carried an average of 5534 ticks. They maintained their initial empty body weight but tick-free control animals gained 18·1%. There were significant increases in haematocrit, serum albumin, and total lipid in the tick-free group. Haematocrit, serum total cholesterol, and phospholipid were significantly decreased and the free : total cholesterol ratio significantly increased in the tick-infested animals.

Part of the effects of tick infestation may have been associated with depressed feed intake but the results are compatible with the view that the tick exerts a direct influence on its host's metabolism by secretion of a toxin.

I. Introduction

Depression of animal performance by cattle tick, *Boophilus microplus*, is well known (Francis 1960; Little 1963; Johnston 1969). However, much of the literature reports work on grazing cattle and little attention has been paid to the relationships between growth rates, nutritional status of the hosts, and the magnitude of tick burdens.

Blood composition changes were investigated in tick-infested Shorthorn × Hereford steers on a diet deficient in protein and energy and on a diet which was adequate (O'Kelly and Seifert 1969). The results measured the combined influence of poor nutrition and tick infestation but did not evaluate the influence each exerted independently. The first experiment described in this paper attempts to separate the effects of tick from the effects of malnutrition on the blood picture of tick-infested Shorthorn × Hereford steers fed a diet designed to reduce body weight.

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O'Kelly and Seifert (1969) also reported that Shorthorn × Hereford steers subjected to moderate tick infestations carried light tick burdens on high planes of nutrition, and made substantial weight gains in a 62-day experimental period. It is important to know to what extent high-quality feed counterbalances the influence of increasing tick numbers. Heavy tick infestations of cattle on adequate diets depress growth rate and produce alterations in blood composition; these give information which may help to show how the parasite affects its hosts' metabolic processes.

The second experiment reported in this paper describes the body weight changes and alterations in blood composition of Shorthorn × Hereford steers fed high-quality lucerne hay *ad libitum* and infested with large doses of tick larvae.

II. Materials and Methods

(a) *Design of Experiments*

(i) *Experiment 1: Steers on Low-quality Feed*

Twelve Shorthorn × Hereford steers, approximately $1\frac{1}{2}$ years old, were used in the experiment. The initial susceptibility to ticks was assessed while they were at pasture and each animal was infested with 0·5 g tick larvae (equivalent to about 10,000 larvae) followed by a similar dose 7 days later. The number of adult female ticks between 0·45 cm and 0·8 cm counted on the left side of the animal on days 19 to 23 after dosing with larvae was used as the index of susceptibility (see O'Kelly and Seifert 1969). The means of the two counts for each animal are referred to as the selection counts. The steers were then ranked on their susceptibility to ticks and allocated by restricted randomization to two groups of six animals. Each group was yarded separately for the experimental period of 50 days (from March 24 to May 13) and all animals were fed *ad libitum* and had access to shade and water. The diet for both groups was wheat straw sprayed with a 1 : 20 dilution of molasses (4 oz per animal per day). One group was kept tick-free. Each animal in the other group was given an initial dose of 1 g of tick larvae followed by four weekly doses of 2 g each. Female ticks maturing were counted as described above, and the total of counts following all five infestations, multiplied by two, was used as the measure of the tick load on each animal.

Animals were weighed before and after a 16-hr overnight fast. The mean initial empty body weights (lb ± standard error) for the tick-free and tick-infested animals were 448 ± 15 and 472 ± 17 respectively.

(ii) *Experiment 2: Steers on High-quality Feed*

The same twelve Shorthorn × Hereford steers were used; the experimental design was the same as in experiment 1; the two new groups were formed so that half the animals that had previously been infested were in each of the new groups; the treatment period was 58 days (from June 26 to August 23); and the feed presented to the animals, in the same way as in experiment 1, was good-quality lucerne hay. All animals were drenched with Neguvon to control internal parasites. One group was kept tick-free. Starting on day 3 of the treatment period, each animal in the other group was infested with six successive doses of tick larvae at 7-day intervals. Doses 1 and 2 were 3 g each, 3 and 4 were 5 g each, and 5 and 6 were 6 g each. Tick counting was performed as in experiment 1.

The mean initial empty body weights (lb ± standard error) for the tick-free and tick-infested groups were 426 ± 21 and 445 ± 22 respectively.

(b) *Analytical Procedures*

Analytical procedures for the blood lipid analyses have been described previously (O'Kelly 1968*a*). Haematological techniques and analyses of serum proteins, electrolytes, and plasma protein-bound iodine (P.B.I.) have been described in a previous paper (O'Kelly and Seifert 1969).

Serum amylase (E.C. 3.2.1.1) was assayed by the saccharogenic method of Henry and Chiamori (1960). The results for each parameter were subjected to one-way analysis of covariance (Snedecor 1956), in which account was taken of the initial differences between animals in that parameter.

III. Results

(a) Experiment 1: Steers on Low-quality Feed

The total number of ticks counted on one side of each animal in the infested group during treatment averaged 2662 ± 1190. In this period, the mean change in empty body weight was $-18 \cdot 1\%$ compared to $-10 \cdot 5\%$ in the tick-free animals.

Table 1

SERUM LIPID LEVELS OF STEERS ON LOW-QUALITY FEED

Values given are means ± standard errors

Determination	Day	Treatment: Tick-free	Treatment: Tick-infested
Total cholesterol (mg/100 ml)	0	63·3 ± 2·4	66·8 ± 3·8
	21	51·0 ± 3·7	37·8 ± 3·1
	50	48·9 ± 2·3	30·7 ± 2·4
Free cholesterol (mg/100 ml)	0	12·3 ± 0·9	13·1 ± 0·6
	21	14·8 ± 1·5	11·5 ± 1·3
	50	12·7 ± 0·9	8·1 ± 0·9
Ratio of free to total cholesterol (%)	0	19·3 ± 0·9	19·6 ± 0·5
	21	29·9 ± 1·3	29·9 ± 1·6
	50	26·0 ± 1·4	26·5 ± 3·1
Phospholipid (mg/100 ml)	0	88·6 ± 3·3	91·3 ± 2·8
	21	67·9 ± 5·0	57·4 ± 3·9
	50	61·7 ± 3·7	49·2 ± 4·8
Triglyceride (mg/100 ml)	0	32·3 ± 4·7	25·0 ± 3·1
	21	18·7 ± 0·7	15·7 ± 1·5
	50	22·2 ± 2·6	14·5 ± 1·4
Non-esterified fatty acids (μ-equiv/l)	0	459 ± 87	342 ± 27
	21	402 ± 43	381 ± 48
	50	621 ± 51	633 ± 58
Total lipids (mg/100 ml)	0	243·6 ± 13·9	245·6 ± 10·9
	21	173·9 ± 10·9	139·7 ± 8·5
	50	174·8 ± 9·0	126·9 ± 9·0

At the end of treatment the mean body weights of the tick-free animals were significantly higher ($P < 0 \cdot 01$) than those in the tick-infested group. Weight loss was correlated ($r = 0 \cdot 882$, $P < 0 \cdot 05$, 4 d.f.) with the total number of ticks carried in the treatment period, the animals carrying the most ticks losing the most weight. The correlations between the selection counts and the tick counts from each infestation were not significant, except in the case of the count from the initial 1-g infestation ($r = 0 \cdot 893$, $P < 0 \cdot 05$, 4 d.f.).

The results of the blood composition analyses are given in Tables 1–3.

Day 21.—The tick-infested animals showed significantly lower concentrations of serum albumin ($P < 0 \cdot 001$), globulin ($P < 0 \cdot 05$), total protein ($P < 0 \cdot 01$), total cholesterol ($P < 0 \cdot 05$), total lipid ($P < 0 \cdot 05$), and amylase activity ($P < 0 \cdot 01$), and had a lower albumin : globulin ratio ($P < 0 \cdot 05$) than the tick-free group.

Table 2

SERUM PROTEINS, AMYLASE ACTIVITY, PLASMA ELECTROLYTES, AND PROTEIN-BOUND IODINE OF STEERS ON LOW-QUALITY FEED

Determination	Day	Treatment	
		Tick-free	Tick-infested
Serum albumin (g/100 ml)	0	2·55± 0·05	2·77±0·07
	21	2·65± 0·08	2·25±0·10
	50	2·14± 0·12	1·80±0·08
Serum globulin (g/100 ml)	0	4·29± 0·27	4·29±0·16
	21	4·00± 0·09	3·75±0·12
	50	3·90± 0·02	3·79±0·07
Albumin/globulin ratio(%)	0	60·4 ± 3·3	65·2 ±3·5
	21	66·5 ± 2·5	60·2 ±2·9
	50	54·8 ± 3·1	47·4 ±2·1
Serum total proteins (g/100 ml)	0	6·73± 0·23	7·02±0·16
	21	7·31± 0·16	6·68±0·19
	50	6·54± 0·11	5·86±0·17
Serum amylase (units/100 ml)	0	130·4 ±20·6	135·9 ±5·8
	21	47·0 ±10·4	29·8 ±5·1
	50	43·6 ± 9·6	21·9 ±2·6
Plasma sodium (m-equiv/l)	0	141·6 ± 0·7	140·6 ±0·6
	50	141·4 ± 0·7	139·4 ±1·0
Plasma potassium (m-equiv/l)	0	4·63± 0·10	4·51±0·17
	50	5·01± 0·16	4·38±0·18
Plasma protein-bound iodine (μg/100 ml)	0	3·74± 0·18	4·80±0·16
	21	2·36± 0·08	2·55±0·22
	50	2·29± 0·16	2·42±0·28

Both tick-infested and tick-free groups showed significant decreases in phospholipid ($P < 0 \cdot 01$), triglyceride ($P < 0 \cdot 05$), and P.B.I. ($P < 0 \cdot 01$), and a significant increase ($P < 0 \cdot 001$) in the free : total cholesterol ratio.

Day 50.—The tick-infested animals showed significantly greater reductions from the initial values in haematocrit ($P < 0 \cdot 001$), haemoglobin ($P < 0 \cdot 001$), albumin ($P < 0 \cdot 05$), total protein ($P < 0 \cdot 001$), total cholesterol ($P < 0 \cdot 001$), triglyceride

($P < 0\cdot05$), total lipid ($P < 0\cdot01$), and amylase activity ($P < 0\cdot001$) than the tick-free animals.

There was a significant increase ($P < 0\cdot01$) in plasma non-esterified fatty acid and a significant decrease ($P < 0\cdot001$) in phospholipid and P.B.I. in both groups. The tick-infested animals showed significantly lower amounts of plasma sodium ($P < 0\cdot05$), potassium ($P < 0\cdot05$), and free cholesterol ($P < 0\cdot01$) than the tick-free group. Mean corpuscular haemoglobin concentrations were not significantly different from the initial values.

Table 3

HAEMATOLOGICAL VALUES OF STEERS ON LOW-QUALITY FEED

Determination	Day	Treatment	
		Tick-free	Tick-infested
Haematocrit (%)	0	$33\cdot4\pm0\cdot5$	$33\cdot3\pm0\cdot5$
	21	$32\cdot2\pm1\cdot1$	$28\cdot8\pm1\cdot7$
	50	$29\cdot4\pm0\cdot5$	$20\cdot7\pm1\cdot9$
Haemoglobin (g/100 ml blood)	0	$14\cdot3\pm0\cdot3$	$14\cdot3\pm0\cdot3$
	21	$13\cdot6\pm0\cdot4$	$12\cdot4\pm0\cdot9$
	50	$13\cdot1\pm0\cdot3$	$9\cdot5\pm0\cdot9$
Mean corpuscular haemoglobin concn. (%)	0	$42\cdot7\pm0\cdot3$	$43\cdot1\pm0\cdot4$
	21	$42\cdot2\pm0\cdot5$	$42\cdot8\pm1\cdot1$
	50	$44\cdot6\pm0\cdot5$	$45\cdot5\pm0\cdot6$

(b) *Experiment 2: Steers on High-quality Feed*

The body weight changes for the two groups and the mean weekly tick counts for the tick-infested group are presented in Figure 1. The total number of ticks counted on one side of each animal in the infested group during treatment averaged 2769. At the end of the treatment period the mean empty body weight of the tick-infested group was the same as at the beginning, whereas the tick-free group had gained $18\cdot1\%$.

Tick doses 2, 4, and 6 were 3-week-old larvae in contrast to 2-week-old larvae for doses 1, 3, and 5. The mean weekly tick counts from doses 2, 4, and 6 (Fig. 1: days 30, 44, and 58) were respectively $51\cdot0$, $51\cdot9$, and $15\cdot5\%$ of the tick counts from doses 1, 3, and 5 (Fig. 1: days 23, 38, and 58). It thus appears that the number of ticks maturing from the second infestation of each dose level is related more to the age, and hence viability, of the larvae than to change in the resistance of the host.

In the tick-infested group, weight change was correlated with the total number of ticks carried during treatment whether expressed as actual weight gain ($r = -0\cdot894$, $P < 0\cdot05$, 4 d.f.) or as a percentage weight change ($r = -0\cdot945$, $P < 0\cdot01$, 4 d.f.). Both groups made comparable weight gains for the first 15 days of treatment (Fig. 1). However there was a loss in weight in the tick-infested animals on day 23, coinciding with the maturing of the first crop of adult female ticks. The greatest loss of weight in the tick-infested animals was recorded on day 51 and this also coincided with the maturation of the greatest number of adult female ticks. Shedding this heavy crop of ticks resulted in the animals making a mean weight gain of 13 lb between days 51 and 58 (Fig. 1).

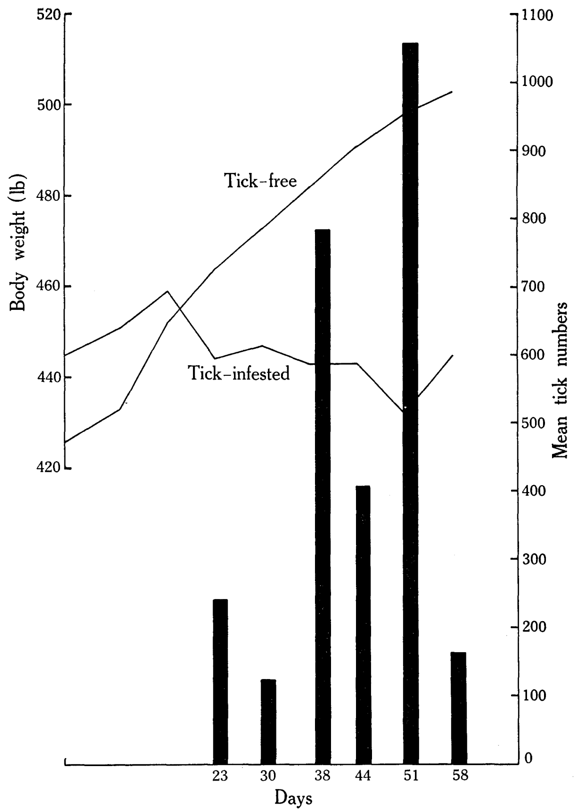

Fig. 1.—Change in body weights in the tick-infested and tick-free groups of steers on high-quality lucerne hay, and the mean tick numbers per animal in the infested group. Tick counts shown on days 23 and 30 were from 3-g doses, on days 38 and 44 from 5-g doses, and on days 51 and 58 from 6-g doses.

The results of the blood composition analyses are given in Tables 4 and 5.

There were no significant differences between the groups in any of the parameters determined on the initial day. Significant increases ($P < 0\cdot05$) from the initial values were found in haematocrit, haemoglobin, serum albumin, the albumin : globulin ratio, and plasma total lipid in the tick-free group.

There were significant decreases from the initial values in haematocrit ($P < 0\cdot001$), haemoglobin ($P < 0\cdot001$), plasma total cholesterol ($P < 0\cdot05$), phospholipid ($P < 0\cdot05$), and total lipid ($P < 0\cdot05$), and a significant increase ($P < 0\cdot001$) in the free : total cholesterol ratio in the tick-infested animals.

TABLE 4

SERUM LIPID LEVELS OF STEERS ON HIGH-QUALITY FEED

Values are means ± standard errors

Determination	Day	Treatment	
		Tick-free	Tick-infested
Total cholesterol (mg/100 ml)	0	80·6 ± 1·9	73·5 ± 4·3
	58	87·5 ± 3·1	56·6 ± 3·8
Free cholesterol (mg/100 ml)	0	16·4 ± 1·2	14·7 ± 1·0
	58	17·9 ± 0·9	14·2 ± 0·9
Ratio of free to total cholesterol (%)	0	20·4 ± 0·3	20·0 ± 0·4
	58	20·4 ± 0·5	25·1 ± 0·5
Phospholipid (mg/100 ml)	0	92·7 ± 3·9	87·2 ± 4·7
	58	102·3 ± 3·2	70·5 ± 4·0
Triglyceride (mg/100 ml)	0	21·7 ± 1·6	19·3 ± 1·9
	58	22·5 ± 2·8	17·0 ± 2·1
Non-esterified fatty acids (μ-equiv/l)	0	386 ± 29	384 ± 28
	58	365 ± 25	399 ± 33
Total lipids (mg/100 ml)	0	251·3 ± 5·2	232·3 ± 10·8
	58	272·0 ± 6·1	185·0 ± 10·3

TABLE 5

SERUM PROTEINS, AMYLASE ACTIVITY, AND HAEMATOLOGICAL VALUES OF STEERS ON HIGH-QUALITY FEED

Determination	Day	Treatment	
		Tick-free	Tick-infested
Serum albumin (g/100 ml)	0	2·01 ± 0·08	2·03 ± 0·20
	58	2·65 ± 0·12	1·85 ± 0·08
Serum globulin (g/100 ml)	0	4·68 ± 0·18	4·68 ± 0·16
	58	4·61 ± 0·19	4·66 ± 0·16
Albumin/globulin ratio (%)	0	43·4 ± 2·8	43·5 ± 4·5
	58	56·7 ± 2·2	40·2 ± 2·6
Serum total proteins (g/100 ml)	0	7·35 ± 0·32	7·18 ± 0·16
	58	8·02 ± 0·26	7·49 ± 0·20
Serum amylase (units/100 ml)	0	132·0 ± 14·7	126·8 ± 12·9
	58	156·4 ± 12·9	92·3 ± 11·5
Haematocrit (%)	0	30·3 ± 1·5	31·2 ± 1·1
	58	35·9 ± 1·4	22·3 ± 0·6
Haemoglobin (g/100 ml blood)	0	13·1 ± 0·8	13·3 ± 0·5
	58	15·6 ± 0·7	9·3 ± 0·3
Mean corpuscular haemoglobin concn. (%)	0	43·0 ± 0·4	42·7 ± 0·16
	58	43·4 ± 0·7	41·7 ± 0·7

IV. Discussion

(a) *Experiment 1: Steers on Low-quality Feed*

The level of non-esterified fatty acid in the plasma controls the amount utilized by the body tissues (Armstrong *et al.* 1961) and is a good indicator of the rate at which fat is being mobilized from adipose tissue (Fredrickson and Gordon 1958; Kronfeld 1965). The elevated plasma non-esterified fatty acid concentrations found in both tick-infested and tick-free animals therefore reflect the increased utilization of these acids as energy substrate.

Hypoalbuminaemia usually accompanies insufficient protein intake and is generally accepted as a diagnostic sign of protein deficiency. The reduction in serum albumin concentration was expected, although the magnitude of the effect was different in the two groups. The tick-infested animals showed a significant lowering of albumin levels at the 21-day sampling in contrast to the tick-free animals, which had normal serum albumin levels at this time. The normal levels in the tick-free group clearly show that the level of serum total protein is not a good measure of protein malnutrition. Serum globulin concentration usually does not decrease until protein deficiency is severe, consequently serum albumin level must be considered a better biochemical index of protein status.

Poor nutrition produces a fall in haematocrit and haemoglobin levels in cattle (Meacham *et al.* 1964; Springell 1968). The nutritional depression in the values of these parameters was clearly demonstrated in the tick-free animals and a similar but significantly greater effect was found in the tick-infested group.

The fall in P.B.I. appears to reflect the nutritional status of the animal and agrees with previous findings (O'Kelly and Seifert 1969). Little is known of the effects of parasitism on electrolytes in the body fluids and it is not possible to deduce what mechanisms contributed to the lowering of the plasma sodium and potassium concentrations in the tick-infested animals.

Undernutrition reduces the plasma total cholesterol and phospholipids in cattle to low levels (O'Kelly 1968*b*). A similar result was found in this study, but the combined effect of tick and poor nutrition produced a significantly greater depression in the plasma concentration of total cholesterol than the effect of inadequate nutrition alone. There was also a marked difference in the effects on the plasma levels of free cholesterol. The free cholesterol concentration was unaltered in the tick-free animals, so the reduction in total cholesterol resulted from a reduction in the cholesterol ester fraction with a consequent increase in the free : total cholesterol ratio. Such a dietary disturbance in the balance between these two forms of plasma cholesterol has been reported previously (O'Kelly 1968*b*). Tick infestation resulted in a decrease in the plasma concentrations of both free and esterified cholesterol, the increased free : total cholesterol ratio in the tick-infested animals was then due to a proportionally greater fall in the cholesterol ester fraction.

(b) *Experiment 2: Steers on High-quality Feed*

The increased haematocrit, haemoglobin, and serum albumin and the general unaltered state of the plasma lipid fractions found in the tick-free group are the results expected of cattle gaining weight on a good-quality lucerne hay diet. In contrast, growth rate was arrested in the tick-infested animals.

The lack of weight gains caused by tick infestation must in part be associated with some reduction in feed intake; changes in the concentrations of plasma constituents may then only reflect alterations in the animals' metabolism caused by reduced feed intake. The inability of the tick-infested animals, in contrast to the tick-free animals, to elaborate greater amounts of serum albumin could possibly be explained by a lowered feed intake. This is unlikely, however, to account for the reduced amounts of plasma total cholesterol and phospholipid since the concentrations of these two lipids are not influenced by level of intake on high-quality lucerne hay diets (O'Kelly 1968*b*). For the same reason it is not likely that the increased free : total cholesterol ratio in the tick-infested animals would result from small changes in dietary intake. Also the significant drop in haematocrit would not be expected to result from small reductions in intake of high-quality feed.

A previous study (O'Kelly and Seifert 1969) showed weight gains and significant increases in haematocrit in lightly infested Shorthorn × Hereford steers on high-quality diets. This investigation demonstrates that even an adequate diet cannot counterbalance the depression in growth rate and certain blood parameters caused by heavy tick burdens.

(*c*) *Conclusions*

Loss of appetite has been reported in many parasitic conditions in ruminants e.g. see Bremner 1961) but there is little information about depressed feed intakes in tick-infested cattle. The possibility of anorexia developing in the tick-infested animals must then be considered. In this case, the sequence of changes in blood components (Tables 1–3) in the tick-infested group on low-quality feed, compared to the tick-free group, would be consistent with a more drastic nutritional deficiency. The animals in this experiment were on an *ad libitum* feeding regime and individual feed intakes were not recorded. It is impossible, therefore, to eliminate reduced feed intake in the tick-infested animals as a major contributor to the difference between the treated and control groups in the levels of certain blood components.

However, it is considered that a reduction in feed intake is not likely to account for the lowered haematocrit and serum total cholesterol and phospholipid levels in the tick-infested animals on high-quality feed. Liver dysfunction in these animals is indicated by the elevated free : total cholesterol ratio. The results from both experiments clearly show that serum amylase activity reflects the nutritional status of the animals and is strongly influenced by tick infestation. The blood loss to tick, not more than 50 ml per animal per day in either experiment, would not be expected to be a major factor in producing anaemia in the infested animals (O'Kelly and Seifert 1969). While the experimental results do not permit a precise conclusion they are compatible with the view that many of the changes in the host's blood picture are caused by the tick by direct interference with metabolic processes, probably by secretion of a toxin. More critical studies are needed to delineate the effects of altered feed intake and tick on the host's metabolism.

Almost identical tick counts were produced by 9 g and 28 g of tick larvae on the animals fed low- and high-quality diets respectively. Thus, the percentage yield of mature ticks was about three times greater on low-quality than on high-quality feed, suggesting a marked effect of nutrition on resistance and agreeing with previous findings (O'Kelly and Seifert 1969). However, the two experiments were carried out at different times of the year and the possibility of seasonal variation in tick resistance must be considered. Also, the use of tick larvae of different ages in the high-quality feed experiment further rules out any quantitative comparison between the two results.

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