EFFECTS OF HAEMONCHUS CONTORTUS IN PEN-FED VERSUS PADDOCK SHEEP AS DETERMINED BY PLASMA CORTISOL AND INSULIN-LIKE GROWTH FACTOR 1

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SUMMARY

The effects of a "trickle" infection of *Haemonchus contortus* on paddock sheep and on sheep transferred to a feedlot are reported. Faecal egg counts showed that all these sheep were extremely resistant to an infection with *H. contortus. No* stress effects as determined by sustained increases in plasma cortisol *were* found during a seven week experimental period, during which occasions.

A reduction in live weight occurred in the pen-fed sheep until they became accustomed to the lucerne pellets. In the pen-fed sheep, but not in the paddock sheep, the effects of a sub-clinical infection of *H. contortus were* observed to cause a significant but transient decrease in plasma insulin-like growth factor 1 (IGF-1). The reduced plasma IGF-1 concentrations suggest that a further decline in nutritional status occurs in infected sheep, already suffering some nutritional deprivation due to diet alteration.

INTRODUCTION

Doses of infective *Haemonchus contortus* larvae given to sheep twice weekly as a "trickle" infection allow differences in the onset and expression of protective immunity against the parasite to be observed (Barger et al. 1985). It is also known that stressful environmental conditions can alter host resistance to disease by effects on the immune system (Kelley 1980). Further, plasma cortisol has been found to be elevated in certain disease states or by acute stressors in the sheep (Shutt *et al.* 1988). In contrast, plasma insulin-like growth factor 1 (IGF-1) concentrations were reduced in calves following restricted feed-intake and exposure to internal parasites (Elsasser et *al.* 1988), and plasma IGF-1 is thought to reflect nutritional status (Isley *et al,* 1983).

As part of a program studying the interaction of environmental stressors with nutrition and internal parasites in the sheep, the present paper reports on the effects of these interactions, as assessed by changes in the endocrine system. Observations were made of changes in plasma cortisol and IGF-1 concentrations following a "trickle" infection of *H. contortus* in the paddock sheep, and in sheep exposed to the stress of being transferred to a feedlot.

MATERIALS AND METHODS

Experimental animals were three-year old, non-pregnant Merino ewes selected on the basis of uniform live weight and allocated either to feedlots, or paddock grazing on established phalaris and white clover pastures, The pen-fed sheep *were* allowed sufficient lucerne-based pellets to maintain live weight (approximately 600 g pellets/head/day). All sheep had been treated for the

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control of helminthosis with closantel and broadspectrum anthelmintics (Dash 1986). Half of the pen-fed sheep, and half of the paddock sheep were then given oral doses of 750 infective larvae of *H. contortus* twice weekly (a "trickle" infection) for seven weeks. Faecal egg counts were carried out weekly after infection. Jugular blood samples were also collected, and live weights were determined on four occasions, corresponding to pretreatment, one week, four weeks, and seven weeks after the experiment commenced with the transfer of the sheep to the feedlots. The blood samples were collected into 10 ml heparinized vacutainers and after centrifugation were stored frozen until assayed for cortisol and IGF-1. Results are reported on seven sheep from each of the four groups-of sheep.

The radioimmunoassay of cortisol in plasma samples was performed as previously described by Fell *et al.* (1985). For the measurement of IGF-1 in plasma a specific radioimmunoassay was used which utilised a rabbit antiserum Tr10 (Baxter *et al.* 1987). Prior to radioimmunoassay plasma samples *were* acidified with glycine-HCl for 24 h essentially by the method of Underwood *et al.* (1982), and ultrafiltration was then carried out on the acidified plasma in microconcentrators (Centricon 30, Amicon Scientific, Australia), to remove possible interference from binding proteins. Aliquots of the ultrafiltrates were dried under nitrogen, and then taken up in assay buffer for radioimmunoassay.

RESULTS

As shown in Table 1 mean plasma cortisol concentrations declined in both pen-fed and paddock sheep over an experimental period of seven weeks, in which half of the sheep were exposed to *H. contortus*. After seven weeks plasma cortisol concentrations were significantly lower ($P<0.01$) than pre-treatment levels in the pen-fed sheep, with or without infection with H. contortus. A similar though less significant reduction in plasma cortisol occurred in the paddock sheep.

Table 1 Comparison of mean (± s.e.) plasma cortisol concentrations (nmol/l) between four groups of seven sheep, either pen-fed or grazing in a paddock, with or without infection with *Haemonchus contortus* larvae

	Pen-fed + *H. contortus*	Pen-fed	Paddock + *H. contortus*	Paddock
Pre-treatment	84 ± 17	68 ± 12	82 ± 20	42 ± 6
Week 1	47 ± 11	47 ± 9	99 ± 11	42 ± 9
Week 4	69 ± 17	36 ± 8*	64 ± 15	56 ± 7
Week 7	28 ± 17**	19 ± 5**	35 ± 10	30 ± 8

Means significantly different (* $P<0.05$, ** $P<0.01$) relative to pre-treatment values. Student's t-test.

Changes in live weight are shown in Table 2 along with changes in plasma IGF-1 concentrations in the pen-fed sheep with *or* without *H. contortus*. A significant reduction in live weight ($P<0.05$) occurred in both pen-fed groups one week after transfer from the paddock, with recovery occurring by week four. In contrast, a significant reduction in mean plasma IGF-1 concentrations ($P<0.01$) only occurred in the first week after the pen-fed sheep were exposed to the "trickle" infection *of H. contortus* (Table 2).

Table 2 Comparison of mean (±s.e.) insulin-like growth factor 1 concentrations (ng/ml) and live weights (kg) in two groups of seven pen-fed sheep, with or without infection with *Haemonchus contortus* larvae

	Pen-fed + *H. contortus*		Pen-fed	
	IGF-1	Live weight	IGF-1	Live weight
Pre-treatment	228 ± 20	46.1 ± 1.6	212 ± 29	45.4 ± 1.2
Week 1	125 ± 18**	40.1 ± 1.6**	202 ± 26	40.8 ± 1.5*
Week 4	272 ± 22	43.6 ± 1.8	233 ± 26	43.6 ± 1.9
Week 7	274 ± 24	43.1 ± 1.9	185 ± 21	43.3 ± 2.2

Means significantly different (* P<0.05, ** P<0.01) relative *to* pretreatment values. Student's t-test.

In the paddock sheep, as shown in Table 3, no significant change occurred in either live weight or mean plasma IGF-1 concentrations over a seven week experimental period in which half the sheep were exposed to the "trickle" infection of *H. contortus*.

Faecal egg counts remained at a low control level throughout the experimental period in all four groups of sheep. At the end of the experimental period, during week eight, faecal egg counts ranged from less than 100 eggs/gram to only 3,800 eggs/gram (900 ± 180; mean ±s.e. for all infected sheep).

Table 3 Comparison of mean (± s.e.) plasma insulin-like growth factor 1 concentrations (ng/ml) and live weights (kg) in two groups of seven sheep grazing in a paddock, with or without infection with *Haemonchus contortus* larvae

	Paddock + *H. contortus*		Paddock	
	IGF-1	Live weight	IGF-1	Live weight
Pre-treatment	188 ± 14	46.8 ± 1.7	224 ± 27	47.7 ± 2.0
Week 1	185 ± 11	48.4 ± 2.0	226 ± 31	51.0 ± 2.3
Week 4	188 ± 19	48.4 ± 1.8	232 ± 35	49.6 ± 2.0
Week 7	161 ± 13	49.1 ± 1.9	232 ± 32	49.7 ± 1.8

DISCUSSION

Faecal egg counts showed that the sheep used in the present experiments were extremely resistant to infection with *H. contortus,* and the intensity of resistance of some mature Merino sheep to *H. contortus* has been well documented (cf. Adams 1989). Thus an anticipated stress effect, as determined by an elevated plasma cortisol, did not occur in response to the "trickle" infection of *H. contortus*. The reduction in plasma cortisol by the end of the experiment can probably be attributed to habituation of the sheep to the handling procedures.

A significant decline in live weight (P<0.05) did occur when sheep were transferred to a feedlot, until the sheep became accustomed to eating only lucerne pelleted feed. In the pen-fed sheep experimentally infected with *H. contortus,* this was associated with a significant reduction (P<0.01) in plasma IGF-1 concentrations after one week, although live weight and IGF-1 concentrations had recovered by week four. Reduced feed intake accompanied by a parasitic infection in calves has been shown to have an additive effect on lowering plasma IGF-1 levels (Elsasser et *al.* 1988), and this would be consistent with our observations.

A drain in protein and energy for host defence is a characteristic response to infection (Campbell 1983), and is reflected in a decline in nutritional status. Maintenance of plasma IGF-1 has been shown to be sensitive to changes in protein and energy (Isley 1983). However, in the present experiments, measurement of plasma IGF-1 appears to be useful as an indicator of reduced nutritional status only in parasite infected sheep already suffering some nutritional deprivation.

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