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Biochemical Disturbances Associated with Haemonchosis in Sheep

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الاضطرابات الحيوية الكيمائية المرتبطة بالإصابة بدودة الهيمونكس في الضأن

الملخص : ارتفعت معدلات تركيز AST و ALT في مصل الدم ، والفسفور واليوريا والبوتاسيوم في الحيوانات المصابة. إن ارتفاع زيادة مستوى AST و ALT يدل على تأثيرات مرضية في العضلات الهيكلية والنسيج الحشوي للكبد وذلك نتيجة التهاب المعدة النزيفي وتحلل شحوم الكبد . ويمكن إرجاع سبب ارتفاع معدل الفسفور في دم الحيوانات المصابة إلى علاقة تبادلية مع معدل الكالسيوم في الدم واستخدام الكربوهيدرويدات . كما يمكن إرجاع سبب ارتفاع معدل تركيز اليوريا والبوتاسيوم إلى الجفاف وإلى ازدياد معدل تكسير البروتين وإلى حالة الحماض (acidosis) نتيجة لارتفاع معدل فقد السوائل وارتفاع معدل تحطيم الشحوم والبروتين والكربوهيدريدات. وقد أدى ذلك إلى انخفاض معدلات تركيز الكوليسترول والنحاس والبروتين الكلي والزنك والكالسيوم والجلوكوز والكلور يد والحديد والمغنيسيوم في دم الحيوانات المصابة . ويعزى سبب انخفاض هذه المكونات الكيماوية إلى فقدان كبير في الدم وإلى الاضطرابات المعدية والمعوية الناتجة عن الإصابة بدودة الهيمونكس والتي تؤدي إلى ضعف في الامتصاص و/ أو عمليات بناء وهدم هذه المكونات الغذائية . لذلك يوصي بوجوب إعطاء الحيوانات المصابة العناصر الغذائية الدقيقة والكبيرة ومحاليل الجفاف كعلاج مساند بالإضافة إلى العلاج الكيميائي .

ABSTRACT: The serum concentrations of aspartate amino transferase (AST), alanine amino transferase (ALT), phosphorus, urea and potassium increased in infected animals. The increased AST and ALT levels suggested pathology of the skeletal muscle and hepatic parenchyma due to haemorrhagic gastritis and fatty degeneration of the liver. An increase in the serum phosphorus levels in infected animals may be attributed to a reciprocal relationship with serum calcium and carbohydrate utilization. An increase in urea and potassium concentrations might be due to dehydration and increased breakdown of proteins, and acidosis as a result of excessive fluid loss and increased catabolism of fat, protein and carbohydrate. The serum concentrations of cholesterol, copper, total protein, zinc, calcium, glucose, chloride, iron and magnesium decreased in infected animals. The decrease in the serum concentrations of these substances was attributed to the heavy blood loss and gastro-intestinal disturbances caused by haemonchosis leading to poor absorption and/or metabolism of these nutrients. It is recommended that in addition to an anthelmintic treatment, infected animals should be provided with essential micro/macro-elements and rehydration salts as supportive therapy.

Haemonchosis is one of the most important parasitic diseases of sheep in Pakistan causing significant economic losses amounting to US$448,000 per annum (Iqbal *et al.*, 1993). The disease is caused by *Haemonchus (H.) contortus*, a voracious haemophagus parasite that brings about an insidious drain on production with an estimated overall loss of 24% in meat and 40% in wool in young lambs (Hussain and Akram, 1967). Studies on the biochemical changes associated with haemonchosis have been carried out (Herlich, 1962; Albees and Lajambre, 1983). Reports with a narrow spectrum of parameters are also available (Hamid *et al.*, 1981; Ahmed *et al.*, 1990). This paper describes in more detail the biochemical disturbances associated with haemonchosis in the Lohi breed of sheep.

Materials and Methods

EXPERIMENTAL ANIMALS: Twelve, one-year old Lohi sheep of either sex and of similar age and body weight were used. The animals were treated with coumaphos (Bayer, Liverkusen, Germany) and tetramisole (Star Laboratories Ltd., Pakistan) for ecto and endo-parasites, respectively. The animals were ensured to be free from parasites based on gross and faecal examination (Soulsby, 1982). The animals were randomly divided into two equal groups i.e. infected

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and uninfected (control), housed in separate pens and offered similar ration.

PREPARATION OF INOCULUM FOR EXPERIMENTAL INFECTION: Female *H. contortus* worms collected from abomasum of naturally infected sheep, washed in lukewarm water, placed in 0.9% normal saline and incubated at 37°C for 24 h. The released ova of *H. contortus* were collected and cultured as described by Thienpont *et al.* (1979). After seven days of incubation, third stage infective larvae (L_3) were purified with Baerman's apparatus (Urquhart *et al.*, 1988). Six inocula, each equivalent to 15,000 larvae, were absorbed separately on Whatman's filter paper No. 40. The larvae were exposed to direct sunlight (18-22°C) for 15 minutes to enhance larval activity and then transferred into gelatin capsules. One gelatin capsule was given orally to each sheep in the infected group. Faecal examination of all the experimental animals were conducted on alternate days to monitor the infection and/or parasite free status of the control animals. The intensity of infection was calculated on the basis of eggs per gram of faeces (Soulsby, 1982).

COLLECTION OF SAMPLES: Three grams faecal and 10 ml blood samples were collected from infected and uninfected control animals at day 0,7,14,21,35 and 56 post experiment (PE). Faecal samples were examined for haemonchus eggs per gram of faeces as described by Thienpont *et al.* (1979). The blood was centrifuged at 3,000 rpm for 10 minutes and the serum was separated for biochemical analysis. The concentrations of serum aspartate amino transferase (AST), serum alanine amino transferase (ALT), glucose, cholesterol, total protein and urea were detected using an automatic chemistry analyzer (Model FP 90, M/S Laboratory Systems, Finland) with specific respective reagent kits (E. Merck. Frankfurter Straße 250, D-6100 Darmstadt 1). The samples were processed as described by AOAC (1984) and trace minerals/electrolytes were detected by an atomic absorption spectrophotometer and a flame photometer (Khan and Tayyab, 1988). The concentration of phosphorus was determined with a spectronic-20 spectrophotometer and that of chloride with a chloride meter (Anonymous, 1954).

STATISTICAL ANALYSIS: The data were analyzed by using a "t" test (Steel and Torrie, 1981). Comparisons between observations on infected and uninfected animals were made at day 0,7,14,21,35 and 56 PE.

Results and Discussion

The serum concentrations of AST, ALT, phosphorus, urea and potassium continuously increased with the advancement of infection as was evident from the eggs per gram of faeces (Table 1). Significant differences ($P<0.01$) were noted between infected and uninfected animals from day 7 to 56 PE (Figure 1 A-E). The increased concentrations of AST and ALT suggested pathology of the skeletal muscle and/or the hepatic parenchyma due to haemorrhagic gastritis and fatty degeneration of the liver (Frankel *et al.*, 1970; Soulsby, 1982). An increase in serum phosphorus levels in infected animals may be attributed to a reciprocal relationship with serum calcium and carbohydrate utilization (Simesen, 1971). Similar results have also been reported by Zajicek *et al.* (1976) and Hamid *et al.* (1981). An increase in urea concentration during *H. contortus* infection had also been reported previously by Correa *et al.* (1978) and might have been due to dehydration and increased breakdown of proteins (Frankel *et al.*, 1970), a characteristic feature of haemonchosis. An increase in serum potassium concentration was also reported by Evans *et al.* (1963). This may have been attributed to acidosis as a result of excessive fluid loss and increased catabolism of fat, protein and carbohydrate (Tasker, 1971).

TABLE 1

Analysis of faeces in haemonchus infected animals

Days PE*	Eggs per gram of faeces					
	Animals					
	1	2	3	4	5	6
0	0	0	0	0	0	0
7	238	0	356	258	178	0
14	8955	7845	8674	8753	7934	8962
21	14226	16789	15642	16324	14567	16539
35	22340	25477	23459	22380	24567	26740
56	26590	28769	34087	29845	20769	32568

Note: none of the uninfected animals passed parasite eggs in the faeces.
*PE: post experiment

The serum concentrations of cholesterol, copper, total protein, zinc, calcium, glucose, chloride, iron and magnesium decreased ($P<0.01$) with the advancement of the disease in infected animals compared with the uninfected ones, from day 7 to 56 PE (Figure 1 F-N). A decrease has been reported previously in the serum concentrations of glucose (Herlich, 1962; Hamid *et al.*, 1981), total protein (Herlich, 1962; Siddiqua *et al.*, 1989; Ahmad *et al.*, 1990), copper and iron (Hamid *et al.*, 1981; Albees and Lajambre, 1983) and calcium (Mossalam *et al.*, 1975; Zajicek *et al.*, 1976; Hamid *et*

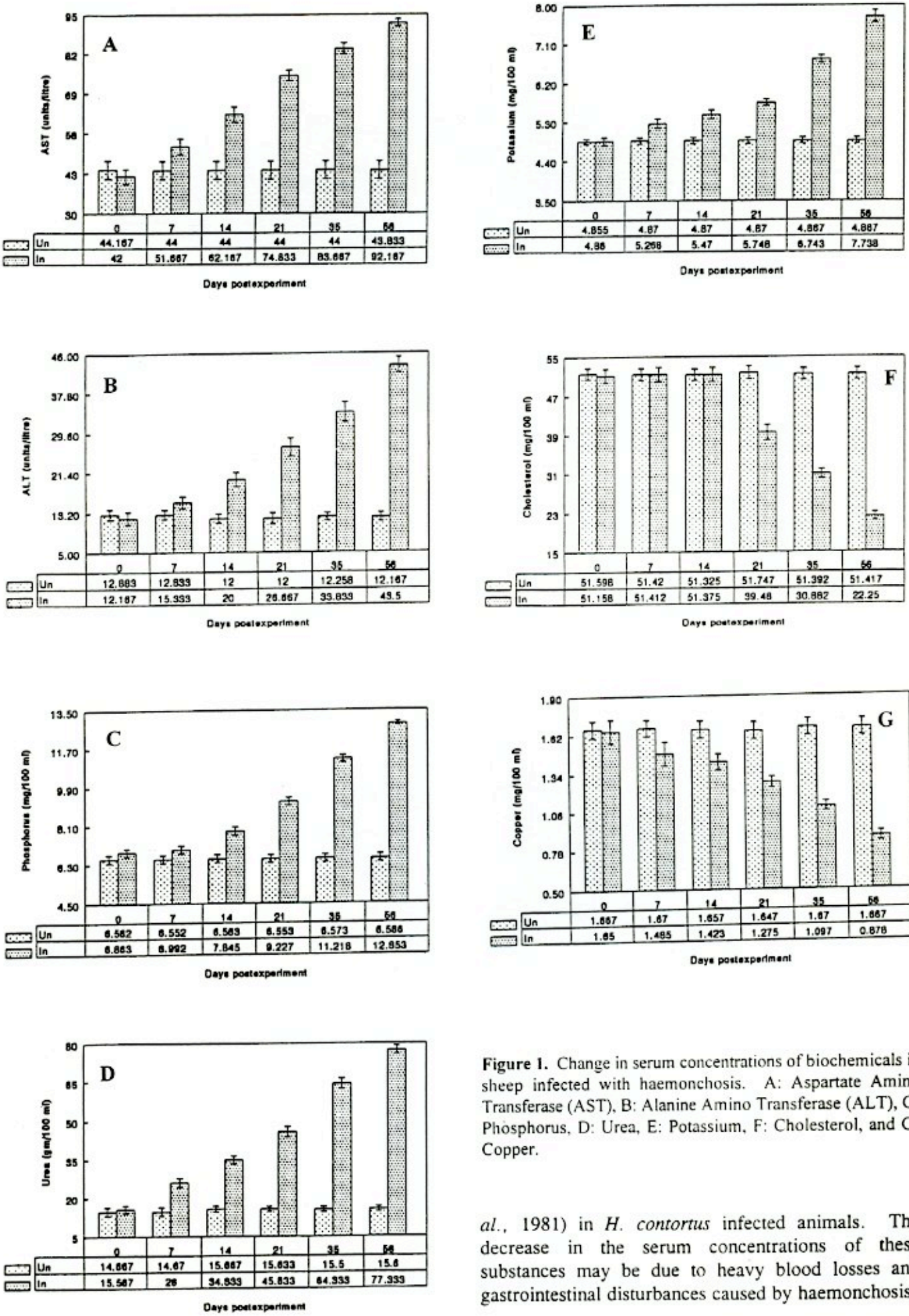
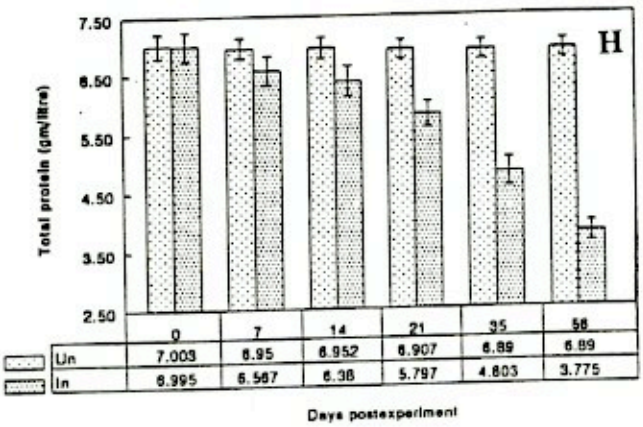
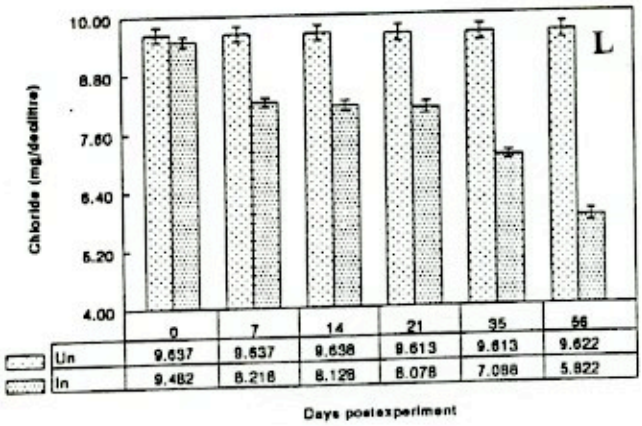
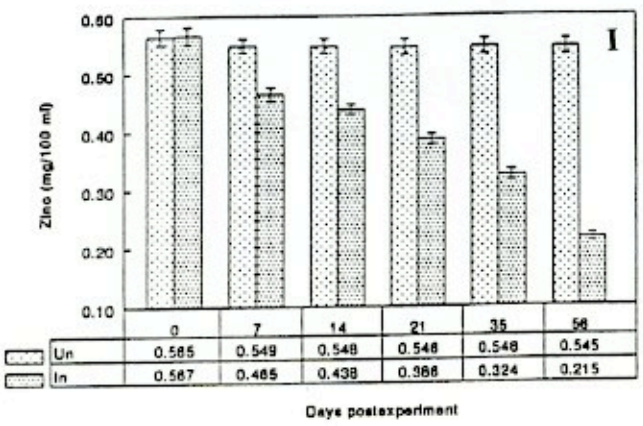


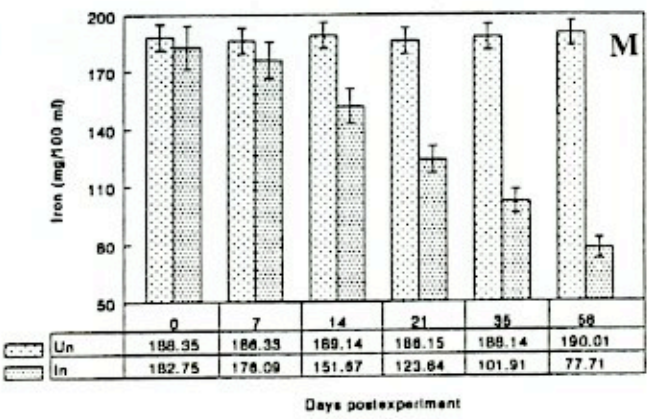
Figure 1. Change in serum concentrations of biochemicals in sheep infected with haemonchosis. A: Aspartate Amino Transferase (AST), B: Alanine Amino Transferase (ALT), C: Phosphorus, D: Urea, E: Potassium, F: Cholesterol, and G: Copper.

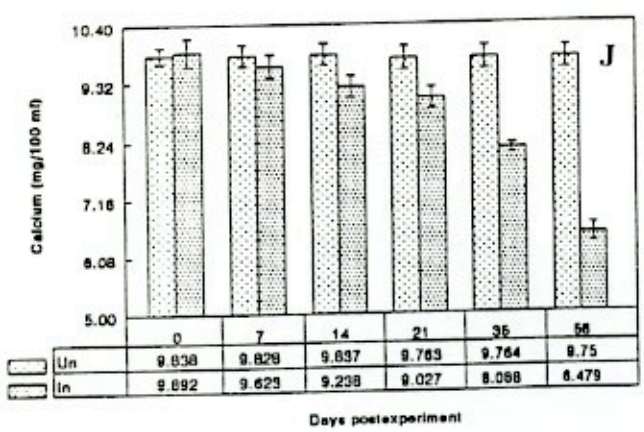
al., 1981) in *H. contortus* infected animals. The decrease in the serum concentrations of these substances may be due to heavy blood losses and gastrointestinal disturbances caused by haemonchosis

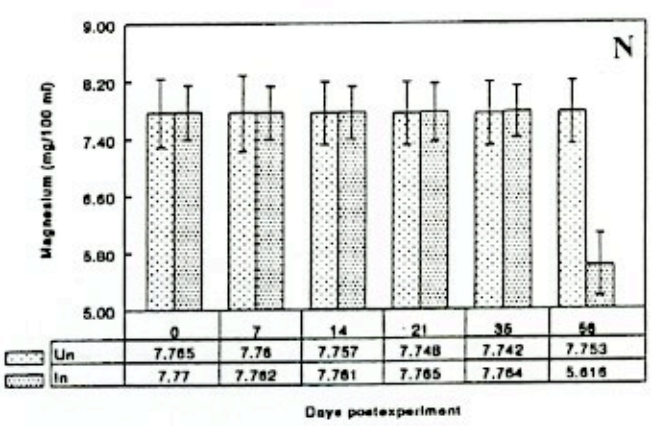












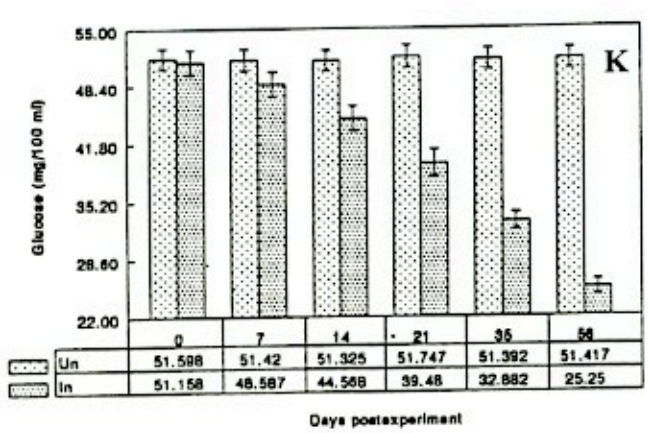


Figure 1. Change in serum concentrations of biochemicals in sheep infected with haemonchosis. H: Total Protein, I: Zinc, J: Calcium, K: Glucose, L: Chloride, M: Iron, and N: Magnesium.

leading to poor absorption and/or metabolism.

The findings of some previous studies are incongruous with our results. For example, phosphorus (Herlich, 1962; Mossalam *et al.*, 1975), potassium (Herlich, 1962) and cholesterol (Correa *et al.*, 1979).

This contradiction could be a reflection of breed differences (Todd *et al.*, 1978; Preston and Allonby, 1979) and geoclimatic factors that affect the severity of infection.

Conclusion

In view of the biochemical disturbances in haemonchus infected sheep, it is recommended that infected animals be given feed supplements of essential micro/macro-elements and rehydration salts in addition to undergoing routine deworming. This would expedite recovery and may enhance an animal's ability to resist haemonchosis infection.

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