EFFECT OF EXPERIMENTAL *Haemonchus contortus* INFECTION ON HEMOGLOBIN CONCENTRATION AND PACKED CELL VOLUME OF DOES

M. M. R. Howlader¹, S. S. Capitan, S. L. Eduardo², N. P. Roxas and C. C. Sevilla

Institute of Animal Science, University of the Philippines at Los Baños, College, Laguna 4031, Philippines

Summary

Twelve Philippine does of 3.5-5.0 years old were used in this study. They were divided into three groups, T₁, T₂ and T₃. Four animals were randomly allocated to each group. Before infection, they were dewormed and housed in individual pens with concrete floors. They were provided with a uniform management. They were infected orally with a single dose of three levels (0, 15,000 and 30,000) of infective *Haemonchus contortus* larvae. Blood samples were collected from the jugular vein of each animal at fortnightly intervals for 17 fortnights. Hemoglobin (Hb) concentration was determined using hemoglobinometer and packed cell volume (PCV) by microhematocrit methods. Animals in infected groups showed significantly ($p < 0.05$) lower hemoglobin values than the control except during fortnight 1 for group 2 and fortnights 1, 2, 5, 6 and 7 for group 3. Hemoglobin concentration did not significantly ($p > 0.05$) differ between the animals in infected groups throughout the sampling period except fortnight 2. The PCV values of animals in infected groups were significantly ($p < 0.05$) lower than the control for most of the sampling periods. The PCV values of animals in group 2 did not significantly ($p > 0.05$) differ from group 3.

(**Key Words** : Effect, *Haemonchus contortus*, Philippine Doe, Hemoglobin Concentration, Packed Cell Volume)

Introduction

Haemonchus contortus, a nematode parasite occurs in the abomasum of goats. Traditionally, this parasite of goats is regarded as a major problem in the hot and humid areas of the world. The principal feature of infection with this parasite is anemia. The fourth larval and adult stages of this worm suck blood and in addition, move and leave wounds that hemorrhage from the abomasal wall of the host. A blood sucking *H. contortus* can suck about 0.05 ml blood per day in sheep (Clark et al., 1962). The understanding of the effect of this parasite on hemoglobin concentration and packed cell volume is essential to reduce the losses caused by this infection in does. Reports of a number of studies have been published on the hematological parameters of pasture grazing sheep

predominantly infected with this parasite (Cobon and O' Sullivan, 1992; Albers et al., 1989). However, no attempt was made to evaluate the effect of *H. contortus* infection on hemoglobin concentration and packed cell volume of Philippine does. The present study evaluated the effect of infection on Hb concentration and PCV values of adult female Philippine goats (does) challenged by three levels of single dose of *H. contortus* larvae while the animals were offered a uniform feed in a confined house for a long period of time.

Materials and Methods

This experiment was conducted at the Institute of Animal Science Farm, University of the Philippines at Los Baños, College, Laguna, Philippines.

Twelve adult female Philippine goats (does) of 3.5-5.0 years old were randomly allocated into three treatment groups (T₁, T₂ and T₃). The animals were housed in individual pens with concrete floors to ensure that adventitious infections with nematode parasites do not occur. They were fed a concentrate mixture at equivalent to 1% of their liveweight. The concentrate feed was compounded using copra meal (50.5%), tricalcium

¹ Address reprint requests to Dr. Matiar Rahman Howlader, Senior Scientific Officer, Bangladesh Livestock Research Institute, Savar, P.O. Savar Dairy Farm, Dhaka 1341, Bangladesh.

² College of Veterinary Medicine, University of the Philippines at Los Baños, College, Laguna 4031, Philippines.

Received November 8, 1995

Accepted June 15, 1996

phosphate (1.7%), rice bran (29.5%), molasses (15.2%), urea (1.0%), salt (1.0%), limestone (1.0%) and vitamin-mineral (0.1%). Napier grass (*Pennisetum perpureum*) and Guinea grass (*Panicum maximum*) were offered *ad libitum* to satisfy the dry matter requirement of the animals.

Haemonchus contortus infective larvae were obtained by culturing the feces of kids harboring monospecific infections of the parasite. Each animal in the treated groups T₁, T₂ and T₃ was fed orally with a single dose of 0, 15,000 and 30,000 *H. contortus* infective larvae in 10 ml physiological saline solution, respectively.

About 5 ml blood was collected in glass vials from the jugular vein of each animal at fortnightly intervals. Blood samples were treated with oxalates anticoagulant to prevent blood clotting. Hemoglobin concentration was determined using hemoglobinometer method and packed cell volume (PCV) by microhematocrit method (Coles, 1980). Fecal samples were collected directly from the rectum of each animal. The number of eggs per gram (EPG) of feces was determined using a modified method (Gordon and Whitlock, 1939) at fortnightly intervals. Four (4) grams of feces were mixed with 60 ml saturated sodium chloride (NaCl) solution. A portion of the fecal suspension was examined using a McMaster's slide. The EPG of feces was calculated by adding the egg counts of two chambers which was multiplied by 50 to represent the EPG.

A split-plot-in-time analysis of variance was used to test for differences between different levels of larval infection, time periods and larval infection by period interaction. Comparison based on the least significant difference at $p < 0.05$ was done between the means of T₁, T₂ and T₃ at each time period.

Results and Discussion

Fecal egg count

Average fortnightly fecal egg counts of does are presented in figure 1. The worms that were fed did not start laying their eggs during fortnight 1. Egg production by the induced worms started during the second fortnight and followed an irregular pattern in 2 groups of infected animals. The EPG of feces of animals in infected groups indicated the patency of infection from 3rd fortnight and irregular fluctuations in egg production were found till 14th fortnight. From the 15th fortnight the EPG of feces of animals in groups 2 and 3 showed higher values which persisted till the end of the experiment. Some fecal samples of animal in control and infected groups showed insignificant number of nematode parasite eggs other than *Haemonchus* during the sampling periods.

Figure 1. Average fortnightly fecal egg per gram (EPG) of feces of does as affected by different levels of stomach worm (*Haemonchus contortus*).

Hemoglobin concentration

There was significant ($p < 0.01$) interaction effect of *H. contortus* and duration of infection on hemoglobin concentration (table 1). Hemoglobin levels of the animals in infected groups were significantly ($p < 0.05$) lower than the control except during fortnight 1 for group 2 and fortnights 1, 2, 5, 6 and 7 for group 3. No significant ($p > 0.05$) difference in hemoglobin concentration was found between the animals in two infected groups throughout the experiment except during the second sampling period. The animals in control group registered fluctuating values of hemoglobin. The animals in treatment group 2 had significantly ($p < 0.05$) lower values from the second fortnight to the end of the experiment. Similarly, animals in treatment group 3 registered significantly ($p < 0.01$) lower values from the third fortnight to the end of the study. The postinfection fall in hemoglobin concentrations in groups 2 and 3, suggests the development of anemia. Evans et al. (1963) reported that the anemia caused by the blood sucking of *H. contortus* was severe in sheep where plasma protein concentrations and liveweights were reduced. In the present study, liveweights of infected animals were severely reduced. At the end of this study,

TABLE 1. AVERAGE FORTNIGHTLY HEMOGLOBIN CONCENTRATIONS OF DOES AS AFFECTED BY DIFFERENT LEVELS OF STOMACH WORM (*HAEMONCHUS CONTORTUS*)

Hemoglobin concentration (g /100 ml blood)				
Time (F)	Treatment (T)			F-Mean
	T ₁ (0 larva) ¹	T ₂ (15,000 larvae) ¹	T ₃ (30,000 larvae) ¹	
F1	9.57abcA	9.83aA	9.94aA	9.78
F2	9.75aA	8.66bB	9.30aA	9.24
F3	9.60abA	8.65bB	8.25bB	8.83
F4	9.13a-dA	7.90bcdB	8.26bB	8.43
F5	8.65cdA	7.60cdB	8.13bcAB	8.13
F6	8.36dA	7.40cdB	8.09bcAB	7.95
F7	8.45dA	7.64cdB	8.21bAB	8.10
F8	9.05a-dA	8.16bcB	7.58bcdB	8.26
F9	8.80bcdA	7.75cdB	7.35bcdB	7.97
F10	8.86a-dA	7.48cdB	7.25cdB	7.86
F11	9.00a-dA	7.49cdB	7.48bcdB	7.99
F12	8.68bcdA	7.26cdeB	7.65bcB	7.86
F13	8.98a-dA	7.18deB	7.67bcB	7.94
F14	9.01a-dA	7.71cdB	7.41bcdB	8.05
F15	8.50dA	7.04defB	6.75deB	7.43
F16	9.14a-dA	7.54efB	6.30eB	7.33
F17	9.49abcA	6.31fB	6.00eB	7.27
T-Mean	9.00	7.68	7.74	8.14

¹ Average of four replications. Means in the same column with a common small letter and in a row with similar capital letter are not significantly ($p > 0.05$) different.

on an average, animals in treatment groups 2 and 3 lost 4.65 and 4.88 kg, respectively, from 18.30 and 18.30 kg initial liveweights. On the other hand, animals in control group gained 0.75 kg over their 20.05 kg initial liveweight by the same period. However, plasma protein concentration was not studied.

The hemoglobin concentrations found in the present study were higher than that of Gallagher (1963) in sheep trichostrongylosis who found 5g/100 ml blood mean hemoglobin value at the peak of the worm egg concentration in the feces. Several additional information were also found. The degree of anemia was increasing at the end of the experiment. Highest fecal egg counts coincided with the lowest hemoglobin concentration values. The decreasing values of Hb were found since 2nd and 3rd fortnight postinfection, respectively, for groups 2 and 3 indicated the poor condition of the animals. Evans

et al. (1963) reported that 51,500 *H. contortus* larvae produced significant changes in the hemoglobin concentration. In their study, hemoglobin concentration went down to about 7 g/100 ml blood by 18 weeks postinfection. In the present study, it took more time to bring down the values of hemoglobin concentration to lower levels. This could be attributed to the lower doses of infection and different host species. Further, Evans and Blunt (1961) argued that *H. contortus* causes substantial losses in Romney Marsh in Australia compared with their counterpart in Great Britain. They explained that it was due to heterozygosity of the breed of the sheep in two countries.

Moreover, the decline in hemoglobin concentration, especially during the late pregnancy could be partly due to the transfer of maternal hemoglobin into the developing fetal circulation or hydroemia effect since blood volume is usually increased at that time. Prakash and Tandon (1978) found Hb mobilization into the developing fetus from the maternal circulation.

Packed cell volume

Significant ($p < 0.01$) interaction of *H. contortus* and the duration of infection influenced PCV as shown in table 2. The hematocrit values of infected does were significantly ($p < 0.05$) lower than the controls for most of the sampling periods except fortnights 1, 3, 4 and 8 for group 2 and 1, 2, 7 and 8 for group 3. There was no significant ($p > 0.05$) difference in hematocrit values between treatment groups 2 and 3. Likewise, there was no significant ($p > 0.05$) differences in PCV values of animals within the control group throughout sampling periods. On the other hand, animals in treatment group 2 registered significantly ($p < 0.05$) lower PCV values. There was a decreasing trend from fortnight 8 until the last sampling period. Almost the same trend was found for animals in treatment group 3. As infections and pregnancy progressed, the difference in PCV values also increased and after kidding, animals in groups 2 and 3 registered the lowest hematocrit values.

Blackburn et al. (1992) also found decreased PCV values as the number of worm increased in *H. contortus* infected goats. The PCV values of animals in treatment groups 2 and 3 during the last 3 and 4 sampling periods, respectively, fell below 20% threshold of normality (Siegmund, 1979). The same results were reported earlier by Blackburn et al. (1991) which could be attributed to chronic effect of infections aggravated during the postparturient period.

Changes in packed cell volume of up to 25% may occur in sheep due to variations in the splenic reservoir

TABLE 2. AVERAGE FORTNIGHTLY PACKED CELL VOLUME OF DOES AS AFFECTED BY DIFFERENT LEVELS OF STOMACH WORM (*HAEMONCHUS CONTORTUS*)

Time (F)	Packed cell volume (%)			F-Mean
	Treatment (T)			
	T ₁ (0 larva) ¹	T ₂ (15,000 larvae) ¹	T ₃ (30,000 larvae) ¹	
F1	28.63aA	28.40aA	28.40aA	28.48
F2	28.10aA	25.08bcB	27.08abAB	26.75
F3	27.93aA	26.43abAB	25.20bcdB	26.52
F4	27.55aA	25.45bcAB	23.63cdeB	25.54
F5	28.79aA	24.20bcdB	23.73cdeB	25.57
F6	27.40aA	24.25bcdB	24.38b-cB	25.34
F7	27.88aA	24.98bcB	25.43bcdAB	26.09
F8	27.99aA	25.38bcA	25.90abcA	26.42
F9	28.33aA	24.23bcdB	23.65cdeB	25.40
F10	27.80aA	24.00bcdB	22.05efgB	24.62
F11	27.75aA	23.00cdeB	20.50fghB	23.75
F12	27.85aA	20.90efB	22.04efgB	23.60
F13	27.55aA	20.88efB	22.91defB	23.78
F14	26.58aA	21.85deB	19.64ghB	22.69
F15	26.08aA	18.64fgB	18.34hiB	21.02
F16	27.79aA	16.74ghB	16.70ijB	20.41
F17	28.84aA	15.59hB	14.64jB	19.69
T-Mean	27.81	22.94	22.60	24.45

¹ Average of four replications. Means in the same column with a common letter, and in a row with similar capital letter are not significantly ($p > 0.05$) different.

(Turner and Hodgetts, 1959). These changes are under the control of the adrenal medulla (Evans, 1957). In the present experiment, all the animals were pregnant, and the decline in PCV values during the late pregnancy might be partly due to the mobilization of maternal cells into the developing fetal circulation. Prakash and Tandon (1978) reported similar findings in cows in India. Even in the animals in control group, there was a decline in PCV values during late pregnancy and early lactation, but it was not statistically significant.

The effect of *H. contortus* on packed cell volume has been reported by many workers (Charleston, 1964; Dineen et al., 1965; Christie et al., 1964; Gallagher, 1963 and Albers et al., 1989). In all studies, different levels of hematocrit values were found. It may be concluded that patent *H. contortus* infection could substantially decrease the hemoglobin concentration and packed cell volume in does. Thus, the malady may contribute in productive and

reproductive performances of the affected animals.

Acknowledgements

We thank Dr. F. F. Peñalba for providing the facility for the conduct of the experiment at the Institute of Animal Science Farm, University of Philippines at Los Baños, College, Laguna, Philippines. We also thank Dr. B. A. Oliveros for his advice during analysis of data.

Literature Cited

- Albers, G. A. A., G. D. Gray, L. F. Le Jambre, L. R. Piper, T. A. Barger and J. S. F. Barker. 1989. The effect of *Haemonchus contortus* on liveweight gain and wool growth in young Merino sheep. *Aust. J. Agric. Res.* 40:419-432.
- Blackburn, H. D., J. L. Rocha, E. P. Figueiredo, M. E. Berne, L. S. Vieira, A. R. Cavalcante and J. S. Rosa. 1991. Interaction of parasitism and nutrition and their effects on production and clinical parameters in goats. *Vet. Parasit.* 40:99-112.
- Blackburn, H. D., J. L. Rocha, E. P. Figueiredo, M. E. Berne, L. S. Vieira, A. R. Cavalcante and J. S. Rosa. 1992. Interaction of parasitism and nutrition in goats: Effects on haematological parameters, correlations and other statistical associations. *Vet. Parasit.* 44:183-197.
- Charleston, W. A. G. 1964. Comparison of blood and bone marrow changes in lambs with haemonchosis and in lambs repeatedly bled. *J. Comp. Path.* 74:223-240.
- Christie, M. G., M. R. Brambell and W. A. G. Charleston. 1964. Resistance to haemonchosis in sheep following phenothiazine treatment of a previous infestation. *Res. Vet. Sci.* 5:202-212.
- Clark, C. H., G. K. Kiesel and C. H. Goby. 1962. Measurements of blood loss caused by *Haemonchus contortus* infection in sheep. *Am. J. Vet. Res.* 23:977-980.
- Cobon, D. H. and B. M. O'Sullivan. 1992. Effect of *Haemonchus contortus* on productivity of ewes, lambs and weaners in a semi-arid environment. *J. Agric. Sci., Cambridge*, 118:245-248.
- Coles, E. H. 1980. *Veterinary clinical pathology*. 3rd Ed. W. B. Saunders Company. London.
- Dineen, J. K., A. D. Donald, B. M. Wagland and Jan Offner. 1965. The dynamics of the host-parasite relationship. III. The response of sheep to primary infection with *Haemonchus contortus*. *Parasit.* 55:515-525.
- Evans, J. V. 1957. The stability of the potassium

- concentration in the erythrocytes of individual sheep compared with the variability between different sheep. J. Physiol. 136:41-59.
- Evans, J. V. and M. H. Blunt. 1961. Variation in the gene frequencies of potassium and haemoglobin types in Romney Marsh and Southdown sheep established away from their native environment. Aust. J. Biol. Sci. 14:100-108.
- Evans, J. V., M. H. Blunt and W. H. Southcott. 1963. The effects of infection with *Haemonchus contortus* on the sodium and potassium concentrations in the erythrocytes and plasma in sheep of different haemoglobin types. Aust. J. Agric. Res. 14:549-558.
- Gallagher, C. H. 1963. Studies on trichostrongylosis of sheep: Plasma volume, haemoglobin concentration and blood cell count. Aust. J. Agric. Res. 14:349-363.
- Gordon, H. and H. Whitlock. 1939. A new technique for counting nematode eggs in sheep faeces. J. Coun. Sci. Ind. Res. 12:50-52.
- Prakash, B. S. and R. N. Tandon. 1978. Studies on haemoglobin, packed cell volume and glucose concentration of Holstein X Tharparkar heifers during late pregnancy and early lactation. Indian J. Dairy Sci. 31:287-289.
- Siegmund, O. 1979. Merck Veterinary Manual, Merck & Co. Inc., New Jersey.
- Turner, A. W. and V. E. Hodgetts. 1959. The dynamic red cell storage function of the spleen in sheep. Aust. J. Expt. Biol. Med. Sci. 37:399-420.