

## Parasitic Infection in Association with Serum Copper, Phosphorus, and Haematological values in Sheep and Goats of Central Trinidad

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### Abstract

A study was done to investigate the possible association between parasitic infections, serum copper phosphorus and haematological values in sheep and goats of Central Trinidad. Single faecal, whole and clotted blood samples were taken from one month to four year old sheep (n=66) and goats (n=66) of local farms. Complete blood counts and the degree of parasitic infection were determined on whole blood and faecal samples, respectively. Total protein, fibrinogen, copper (Cu) and phosphorus (P) concentrations were determined in plasma and serum, respectively. Fifty five percent of sheep and 85% of goats displayed a mild to moderate degree of mixed parasitic infections. Haemoglobin, PCV, and Cu values varied significantly ( $P < 0.05$ ) with the degree of parasitic infection in goats and slightly in sheep ( $p < 0.07, 0.05$ ). Lower Hb and PCV values were found in goats without an apparent parasitic infection and also with an occasional and a 2+ degree of parasitic infection. Lower mean serum Cu concentrations were found in sheep without evidence of parasitic infections and in goats with both 1+ and 4+ degree of mixed infections. 25% to 37% of parasitized sheep and goats had serum Cu ( $< 0.5$  mg/L) and P ( $< 40$  mg/L) i.e below critical levels (BCL). The degree of mixed parasitic infections was somewhat related to the lowered Hb and PCV mean values and serum Cu concentrations more so in goats than in sheep. Both regular treatment for parasitism and mineral supplementation would impact positively on the growth and productivity of sheep and goats of Central Trinidad.

**Key words:** Copper (Cu), Phosphorus (P), below critical level (BCL), Haemoglobin, PCV, degree of parasitic infection

### Introduction

Coccidian and gastrointestinal nematode infections as mixed or single infections are major parasitic conditions influencing the sheep and goat industry in both tropical and temperate climates<sup>1</sup>. In Trinidad, Adogwa et al<sup>2</sup> demonstrated in a controlled study, that sheep administered copper heptonate IM and infected with gastrointestinal nematode eggs, had lowered serum copper (Cu) and haemoglobin (Hb) levels for several weeks compared to untreated controls. Coop, Sykes and Angus<sup>3</sup> have also shown via larval dosing that the presence of adult worms in the intestinal mucosa of growing lambs caused an inhibition of intestinal P absorption, defective bone matrix formation with accompanying depressed plasma P levels<sup>3</sup>.

Although the economic importance of these conditions is speculated upon, no field study has been done in Trinidad to demonstrate whether parasitic infections and Cu and P deficiencies occur concurrently at sheep and goat farms of Trinidad. As such, the study

described here provides preliminary information on the predominance of gastrointestinal intestinal nematode (GIN) infections, concurrent mixed coccidian and *Moniezia* spp. infections, its related haematology, and Cu and P deficiencies of sheep and goats of several farms of Central Trinidad. It is hoped that this information will be used to advise local sheep and goat farmers on methods of improving their economic livelihood via adequate mineral supplementation in conjunction with an appropriate parasitic control programme.

### Materials and Methods:

#### *Farms/ Locations*

Samples for this study were collected from sixty-six goats and sixty six sheep between the ages of one month and four years from farms of Central Trinidad (Table 1). Most samples were collected in the late dry season (Feb-May, 1987, 89), and in a four month period of the wet season (June – September 1987, 89).

**Table 1.** Blood samples collected from sheep and goats of certain areas of Central Trinidad

Sheep		Goats	
Location	Samples	Location	Samples
Petrotrin Sheep Farm	16	Couva	28
Chaguanas	17	Chaguanas	27
Mon Jaloux Livestock Farm	15	Sugarcane Feeds Centre	11
Sugarcane Feeds Centre	18		
<b>Total Sheep (n)</b>	<b>66</b>		<b>66</b>

The samples collected represented a sub-sample of a wider study to evaluate the mineral status and haematological values of sheep and goats of Central Trinidad.

Farms were located on a variety of soil types consisting of clay, sandy clay loams and fine sandy loams of Central Trinidad. Total rainfall in the late dry and wet seasons ranged from 14 to 47mm and 1248 to 1542mm, respectively, depending on farm or location.

Whole blood samples were collected from lambs and kids and adult sheep and goats of several small farms and three state owned farms or at other locations in Central Trinidad. Pertaining to state owned farms, sheep were managed intensively at the Mon Jaloux Livestock Farm and at the Sugar Cane Feeds Centre; and semi-intensively at the Petrotrin Sheep farm and at the Chaguanas location. Goats were reared intensively at the Sugar Cane Feeds Centre and semi-intensively at the Couva and Chaguanas locations. Sampling represented approximately 15% of the animals at the Mon Jaloux Livestock farm and 15 to 45% at other farms or locations.

Sheep were of Barbados Black Belly and West African Hair type origin. Goats were of local Saanen and Anglo-Nubian breeds and of the Creole goat dwarf type origin. Except for the Mon Jaloux Livestock farm, sheep and goats were either zero grazed or allowed grazer mainly on native bamboo grass (*Paspalum fasciculatum*), para grass (*Bracharia mutica*) and tapia grass (*Sporobolus indicus*).

Sheep at the Mon Jaloux Livestock farm were kept on a urea-molasses bagasse-based feed, to which no other mineral supplements were added except sodium chloride. Sheep and goats were occasionally dewormed at various farms using Oxfendazole at the recommended dosage of 5mg/kg (Synantic, Syntex Agribusiness).

*Sample collection*

Approximately two and one half millilitres of blood drawn by venipuncture, was collected in 5mls bottles

containing the potassium salt of EDTA (ethylene diamine tetra acetic acid) as the anticoagulant. Five millilitres of blood were also collected in acid-washed demineralised tubes.

The clotted blood was centrifuged within four hours of collection to remove serum that was stored at -20°C.

*Haematology*

Packed cell volume (L/L) was determined by the microhaematocrit methods. A coulter haemoglobin meter was used to determine haemoglobin concentration (g/l). The total WBC was calculated with the aid of a haemocytometer. The differential leucocyte count (x 10<sup>9</sup>/L) was performed on a stained blood smear (i.e. the number of each leucocyte type in total of 100 cells). Total plasma protein (g/L) was determined by Millar’s precipitation methods. Reticulocyte counts were done on blood smears stained with new methylene blue, from animals exhibiting low Hb (< 80 g/L) and PCV (< 21 L/L) values.

*Chemical Analysis*

Serum Cu levels (mg/L) were determined according to Fick et al.<sup>6</sup> using a Pye Unicam 2900 Atomic Absorption Spectrophotometer equipped with a PU9090 data graphics system. Phosphorus was determined colorimetrically according to Cavell<sup>7</sup>, using a Pye Unicam PU8600 UV/Vis spectrophotometer.

*Parasitology*

Single faecal samples were collected from rectum or immediately after defaecation of sheep and goats. Faecal samples were examined for helminths and coccidian by faecal defaecation. Five-gram samples of faeces were mixed with 25 gm. saturated NaCl in a wide-mouthed bottle and mixed to a homogeneous suspension, followed by straining and centrifugation at 1500 r.p.m. for 3-5 minutes. Saturated NaCl was added again and a clean slide placed at the top to remove a drop of fluid to examine for eggs and oocysts. At least ten microscopic fields were then examined for eggs or larvae at low power field (LPF) and reported as follows (Table 2):

**Table 2.** Degree of Faecal Infection per Microscopic Field

Parasitic Mixed Infections	Microscopic Observation	Coded Result
I	No eggs seen	None seen
II	Less than 1 egg/ LPF	Occasional
III	1 - 2 eggs/ LPF	+
IV	2 - 4 eggs/ LPF	++
V	4 - 6 eggs/ LPF	+++
VI	More than 6 eggs/ LPF	++++

### Statistical Analysis

Degree of parasitic infection, haematological parameter means and serum Cu and P means were tested in sheep and goats separately by a one way analysis of variance using the general linear model procedure (Genstat Data analysis software release 8.21, University of Reading, 1989). Means were tested among the degree of parasitic infection, haematological values, including Cu and P concentrations.

### Results

#### Parasitic infection and serum Cu and P level

The prevalence of gastrointestinal nematodes GIN (I to VI) mixed with other infections (coccidia and *Moniezia* spp.) as observed and concurrent Cu and P deficiencies in sheep and goats are shown in Tables 3 and 4. The parasitic eggs identified were those of species of trichostrongyles, and strongyloide, Coccidia, and the cestode *Moniezia* spp. The trichostrongyle eggs identified were of the species *Haemonchus contortus* and the coccidia identified was that *Eimeria* spp.

55% of sheep and 85% of goats showed a mild to moderate degree (II — VI) of parasitic infections from various faecal collections. Of the parasitized sheep 29% had serum Cu (< 0.5 mg/L) and 32% had P (< 40 mg/L) below critical levels (BCL). 37% of parasitized goats had serum Cu (< 0.5 mg/L) and 25% P (< 40 mg/L) below critical levels (BCL). Note however, that most sheep and goats had low serum Cu (< 0.65mg/L) and P (< 45 mg/L)<sup>9</sup>.

#### Parasitic infection, serum Cu and P levels and Red blood cells values

Haemoglobin and PCV values did not vary with the degree of parasitic infection, however, these values were slightly lower in sheep with a 1+ (III) and 4+ (VI) degree of parasitic infection. Lowest mean serum Cu concentration (P< 0.05) were found in sheep with a

2+ (IV) degree of parasitic (Table 5). However mean Cu concentrations found in sheep without an apparent infection and those with an occasional infection were also below critical level (< 0.5 mg/L)<sup>9</sup>. Haemoglobin and PCV vales varied significantly (P< 0.05) with the degree of parasitic infection in goats (Table 6). Lowest values were found in goats without an apparent parasitic infection (I) and a 2+ degree (IV) of parasitic infection. However goats without an apparent infection (I) and a 2+ degree (IV) of parasitic infection (II) also had mean Hb and PCV values (80 g/L) below expected values for goats. The lowest serum Cu concentration was found in sheep without an apparent parasitic infection. However goats with a 1+ and 4+ degree (III & IV) of infection also had mean Cu concentrations below critical level<sup>9</sup> (BCL) (< 0.5 mg/L). Animals with low Hb and PCV values exhibited a low reticulocyte count (<1%) indicative of a non-regenerative anaemia.

#### Parasitic infection and white blood cell values

Absolute and percent lymphocyte counts were significantly higher in sheep mildly parasitized (occasional, 1+ & 2+) than in other sheep. Percent eosinophil counts were significantly higher in mildly parasitized goats than in other goats. However, fibrinogen concentrations were highest (P<0.01) in heavily parasitized (4+) goats (Table 5). Note however, that most goats had high absolute white blood cell (13.0 x 10<sup>9</sup>/L) and neutrophil counts (> 5.0 x 10<sup>9</sup>/L).

### Discussion

Haemoglobin and PCV values of sheep were generally similar to values reported for sheep of temperate origin<sup>10</sup>. However; values were higher than those found in West African Dwarf<sup>11</sup>, Nigerian Crossbred sheep<sup>12</sup> and indigenous Ethiopian sheep<sup>13</sup>. Haemoglobin and PCV values in goats were similar to those reported for West African Dwarf breeds<sup>11</sup> and Indian Chegu goats<sup>14</sup>. However, values were lower than those reported for indigenous goats of Swaziland<sup>15</sup>, East Africa<sup>16</sup> and

India<sup>17</sup>. The low Hb and PCV values with a concurrently low reticulocyte count, suggestive of a non-regenerative anaemia found in mildly parasitized goats was similar to the findings in Nigerian dwarf breeds<sup>18</sup>.

Comparison of the total white blood cell count and differential leucocyte counts in both apparently normal sheep and goats with those of normal sheep and goats of temperate origin revealed higher values for the total white blood counts, neutrophil, eosinophil and basophilia counts in this study. The WBC values found in sheep were generally similar to those reported for West African Dwarf breeds.<sup>11</sup> The high WBC counts found more often in normal kids than in adult goats have also been observed in kids and goats of West African origin<sup>11</sup>, Swaziland<sup>15</sup> and Cashmere origin<sup>14</sup>. The high white blood cell count found in this and other tropical studies suggest a phenomenon of kids and goats reared in tropical environments.

The rise in percent eosinophil (eosinophilia) found in mildly parasitized goats has been found in other studies<sup>11</sup>. This eosinophilia was also associated with a general rise in neutrophil counts in these animals. The low Hb, PCV values with accompanying leukocytosis, neutrophilia and eosinophilia were probably due to the presence of mild to moderate numbers of adult worms of *Haemonchus contortus* and *Strongyloides* spp. causing blood and protein losses in the GI tract of these animals. The concurrent *Eimeria* spp and *Moniezia* spp. infections found in some cases indicate an added parasitic burden in these animals. Consequently, declines of both Hb and PCV levels especially in goats were probably associated with blood loss from parasitic infection<sup>19</sup> and reduced erythropoietic activity from Cu deficiency<sup>20</sup>. The level of parasitic infection found in more than half of sheep and goats would impact negatively on growth and production in these animals. The level of parasitic infection, including depressed serum Cu and P levels found in more than half of sheep and goats could be indicators of depressed growth and production in these animals<sup>21</sup>. Both regular treatment for parasitism and mineral supplementation would impact positively on the productivity of sheep and goats of Central Trinidad.

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**Table 3:** Percentages of parasitism, serum copper and phosphorus below critical level (BCL) in sheep

<b>*Total</b>	<b>Parasitic criteria</b>	<b>% Parasitism</b>	<b>% Cu<sup>a</sup> BCL</b>	<b>% P<sup>b</sup> BCL</b>
I	None apparently present	45	32	18
II	Occasional trichostrongyle ova or Occasional coccidial oocysts	17	9	9
III	1+ trichostrongyle ova or 1+ trichostrongyle ova plus occasional Coccidial oocysts or 1+ trichostrongyle Ova plus occasional coccidial oocysts plus moniezia spp. or 1+ trichostrongyle Ova plus occasional coccidial oocysts plus occasional strongylodides ova or 1+ coccidian only	14	3	6
IV	2+ trichostrongyle ova or 2+ trichostrongyle Ova plus occasional strongyloides ova or 2+ Trichostrongyle ova plus occasional moniezia Plus occasional coccidial oocysts	12	11	5
V	3+ trichostrongyle ova plus occasional Coccidial oocysts or 3+ coccidian only	2	0	2
VI	4+ trichostrongyle ova plus occasional Coccidial oocysts	11	6	5

\*Total (n) = 66

a Critical level 0.5 mg/L for serum Cu (Underwood and Suttle, 1999)

b Critical level 40 mg/L for serum P (Underwood and Suttle, 1999)

**Table 4:** Percentages of parasitism, serum copper and phosphorus below critical level (BCL) in goats

<b>*Total</b>	<b>Parasitic criteria</b>	<b>% Parasitism</b>	<b>% Cu<sup>a</sup> BCL</b>	<b>% P<sup>b</sup> BCL</b>
I	None apparently present	15	11	2
II	Occasional trichostrongyle ova or Occasional coccidial oocysts	23	8	9
III	1+ trichostrongyle ova or 1+ trichostrongyle ova plus occasional Coccidial oocysts or 1+ trichostrongyle Ova plus occasional coccidial oocysts plus moniezia spp. or 1+ trichostrongyle Ova plus occasional coccidial oocysts plus occasional strongylodides ova or 1+ coccidian only	26	15	8
IV	2+ trichostrongyle ova or 2+ trichostrongyle Ova plus occasional strongyloides ova or 2+ Trichostrongyle ova plus occasional moniezia plus occasional coccidial oocysts	11	5	3
V	3+ trichostrongyle ova plus occasional Coccidial oocysts or 3+ coccidian only	12	3	2
VI	4+ trichostrongyle ova plus occasional Coccidial oocysts	14	6	3

\*Total (n) = 66

a Critical level 0.5 mg/L for serum Cu (Underwood and Suttle, 1999)

b Critical level 40 mg/L for serum P (Underwood and Suttle, 1999)

Table 5: Degree of parasitic infection (I - VI), serum copper, phosphorous and haematological values in all sheep

Blood	I		II		III		IV		V		VI		
	(n)	(30)	(11)	(8)	(9)	(14)	(12)	(1)	(7)				
% <sup>a</sup>	45	17	14	12	14	12	2	11	11				
Parameter	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Significance
Hb (g/L)	108	3.36	103	6.2	89	6.53	98	6.92	110	19.6	89	7.4	p<0.07
PCV (L/L)	0.31	0.01	0.29	0.02	0.27	0.02	0.28	0.02	0.32	0.06	0.26	0.02	p<0.10
MCHC (g/L)	373	3.9	349	7.1	334	7.5	347	8	344	22.6	346	8.5	NS
Cu (mg/L)	0.39	0.028	0.4	0.052	0.56	0.054	0.32	0.058	0.54	0.163	0.47	0.061	p<0.05
P (mg/L)	44.94	2.681	39.05	4.428	44.44	4.895	48.07	5.550	31.20	14.685	41.83	5.550	NS
WBC x10 <sup>9</sup> /L	10.7	1.19	17.3	2.14	12.3	2.25	12.3	2.39	18.5	6.76	12.3	2.55	NS
Neutrophil x10 <sup>9</sup> /L	5.56	0.06	5.26	1.06	3.84	1.12	4.1	1.19	5.18	3.36	4.58	1.27	NS
Lymphocyte x10 <sup>9</sup> /L	3.24	0.766	8.91	1.37	6.53	1.445	6.88	1.532	11.1	4.335	5.6	1.638	p<0.01
Eosinophil x10 <sup>9</sup> /L	0.57	0.183	1.24	0.327	1.13	0.344	0.55	0.367	1.48	1.034	0.7	0.391	NS
Monocyte x10 <sup>9</sup> /L	0.17	0.036	0.13	0.065	0.091	0.069	0.12	0.073	0.19	0.206	0.15	0.078	NS
Basophil x10 <sup>9</sup> /L	0.08	0.047	0.15	0.083	0.16	0.088	0.05	0.093	0.19	0.264	0.03	0.1	NS
Protein (g/L)	65	1.7	65	3.1	64	3.3	65	3.5	74	9.9	65	3.7	NS
Fibrinogen (g/L)	3	0.28	2.4	0.52	1.7	0.54	1.9	0.58	1	1.64	2.3	0.62	NS
%	1.6	0.27	1.1	0.47	0.9	0.5	1.1	0.53	1	1.5	1.3	0.57	NS
Basophil	0.08	0.047	0.15	0.083	0.16	0.088	0.05	0.093	0.19	0.264	0.03	0.1	NS
%	0.72	0.39	0.5	0.7	1.22	0.74	0.5	0.78	1	2.21	0.29	0.84	NS
Protein (g/L)	65	1.7	65	3.1	64	3.3	65	3.5	74	9.9	65	3.7	NS
Fibrinogen (g/L)	3	0.28	2.4	0.52	1.7	0.54	1.9	0.58	1	1.64	2.3	0.62	NS

<sup>a</sup> of all sheep

NS - not significant

Table 6: Degree of parasitic infection (I - VI), serum copper, phosphorous and haematological values in all goats

Blood	I		II		III		IV		V		VI		
	(11)		(15)		(16)		(7)		(8)		(9)		
% <sup>a</sup>	15		23		26		11		12		14		
Parameter	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Mean	S.E.	Significance
Hb (g/L)	79	5.5	78	4.53	85	4.26	72	6.64	92	6.21	100	5.85	p<0.05
PCV (L/L)	0.23	0.02	0.24	0.01	0.25	0.01	0.23	0.02	0.27	0.02	0.3	0.02	p<0.05
MCHC (g/L)	352	7.2	334	5.9	334	5.5	324	8.6	335	8.1	331	7.6	NS
Cu (mg/L)	0.4	0.044	0.58	0.038	0.48	0.037	0.54	0.055	0.54	0.051	0.48	0.049	p<0.05
P (mg/L)	47.38	3.026	43.65	2.591	46.34	2.509	47.61	3.793	54.59	3.548	48.70	3.345	NS
WBC x10 <sup>9</sup> /L	16.3	2.01	16.5	1.64	16.9	1.59	12.3	2.4	17.9	2.25	16.7	2.12	NS
Neutrophil x10 <sup>9</sup> /L	7.58	1.252	5.93	1.022	6.35	0.99	5.27	1.496	7.55	1.4	7.81	1.32	NS
Lymphocyte x10 <sup>9</sup> /L	0.69	0.116	0.58	0.095	0.77	0.092	0.51	0.138	0.84	0.13	0.71	0.122	NS
Eosinophil x10 <sup>9</sup> /L	0.97	0.432	1.49	0.352	1.56	0.341	0.41	0.516	0.45	0.583	0.27	0.455	p<0.10
Monocyte x10 <sup>9</sup> /L	0.06	0.081	0.020	0.066	0.34	0.063	0.34	0.1	0.2	0.088	0.4	0.083	p<0.01
Basophil x10 <sup>9</sup> /L	0.22	0.082	0.29	0.067	0.27	0.065	0.09	0.098	0.1	0.092	0.08	0.086	NS
Protein (g/L)	77	2.8	73	2.3	74	2.2	68	3.4	70	3.2	71	3	NS
Fibrinogen (g/L)	3.5	0.63	2.3	0.51	3.6	0.5	2.6	0.75	2.1	0.7	4.8	0.66	p<0.01
%	0.4	0.53	1.2	0.44	2.5	0.42	2.6	0.64	1.4	0.6	2.7	0.56	**
Basophil	0.22	0.082	0.29	0.067	0.27	0.065	0.09	0.098	0.1	0.092	0.08	0.086	NS
%	1.2	0.46	1.7	0.38	1.4	0.36	0.9	0.55	0.5	0.51	0.7	0.49	NS
Protein (g/L)	77	2.8	73	2.3	74	2.2	68	3.4	70	3.2	71	3	NS
Fibrinogen (g/L)	3.5	0.63	2.3	0.51	3.6	0.5	2.6	0.75	2.1	0.7	4.8	0.66	**

<sup>a</sup> of all sheep

NS – not significant