HAEMATOLOGIC AND BIOCHEMICAL PROFILE OF GRAZING BOER-GOATS SUPPLEMENTED WITH COWPEA SEED HULLS AND COMMERCIAL CONCENTRATE.

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ABSTRACT: The objective of the study was to investigate the haematological and biochemical indices of grazing/browsing Boer-goats before and after supplemented with either cowpea seed hulls or commercial concentrate. Weanling Boer-goats (n=36) were assigned 3 treatments comprising of 4 animals each replicated three times in a completely randomized design (CRD) matrix. There were no variations ($p>0.05$) in mean values of haematological parameters of the supplemented and non-supplemented goats except for mean corpuscular haemoglobin (MCH) and mean corpuscular volume (MCV). Both values increased significantly ($p<0.05$). For clinical chemistry analysis, the goats fed different diet treatments had elevated *creatinine* and total plasma proteins, but still within the normal range. There was also noticeable subclinical *phosphotaemia* as goats fed different diets or diet combinations had low phosphorus level at the beginning of the study but slightly increased at the end of the study.

Keywords: haematology, biochemical indices, cowpea seed hulls, commercial concentrate, browse/graze, Boer-goats

Introduction

Clinical haematology and blood biochemical studies are scientific procedures used in veterinary medicine to evaluate health status of animals (Sowande *et al*., 2008). The protocols have opened up in-depth understanding of population processes, physiological conditions and ecological relationships in animal populations (Franzmann, 1972; Seal *et al*. 1975). Animal nutritionists undertake haematological and serum biochemistry studies to investigate feed production and utilization to ascertain cyclical pattern of weight gain and losses between seasons (Sowande *et al*., 2008). Haematological parameters together with plasma metabolites can provide information that could show effects of ingested diets (Jenni-Elermann, 1998; Spinu *et al*., 1999).

Clinical haematology and blood chemistry are usually influenced by diseases, nutritional stress, body condition, sex, age, diet, circadian rhythms, captivity, etc.(Woerpel *et al*., 1984; Palomeque *et al*.,1991; Spinu *et al*., 1999; Quintavalla *et al*., 2001). Some workers reported to have observed varied blood value indices in feral pigs that showed habitat characteristics and adaptation (McIntosh and Pointon, 1981). In a similar study, Singer and Ackerman (1981) observed some bloody value correlates of animals' conditions during good seasons and/or when fed poor feed. This study was carried out to determine the effects of supplementing cowpea seed hulls or commercial concentrate on grazing weanling Boer goats.

Materials and Methods

Experimental Area

The study was conducted at Mantshwabisi Government ranch, located about 95 km from Gaborone (Capital City) in Kweneng District, Botswana. The local climate of the area is classified as hot and semi-arid with mean daily temperature of 30^{o}C; the area has mean rainfall of 153.4 mm per annum, which is normally received in summer months (December– April)(Meteorology, 2013). The geographic position of the ranch is between the coordinates:
S 24^{0} 10$^{'}$ 47.4" and E 25^{0} 16' 36.1" at an altitude of 1174 m above sea level. The vegetation of the area can be described as shrub savannah or wood land savannah.

Chemical analyses

The dry matter (DM) of feed of gazing/browsing samples were determined by drying the feed samples at 65^{o}C for 24h and ash obtained by igniting the samples in a muffle furnace at 550^{o}C for 6hours. Nitrogen content was measured by the Kjeldahl method (AOAC, 1990) and crude protein calculated as N x 6.25. Acid detergent fibre (ADF), neutral detergent fibre (NDF), acid detergent lignin (ADL) and ether extract (EE) content of the forage/feeds were determined using the method by Van Soest *et al*. (1991).

Experimental animals, feeding and management

A total of 36 Boer goats aged 15±2.3 months old with body weights (BW) 26.32 ± 6.36 kg and of mixed sex were used in this study. Natural pasture was mainly used as basal diet for the goats. However, experimental goats were supplemented with either commercial feed or cowpea seed hulls at the rate 300 g per goat per day before they were released to graze/browse. The control group grazed/browsed the natural pasture without supplementation. Furthermore, all the goats were given dicalcium phosphate x salt licks and fresh clean water on free choice basis. All the experimental goats were de-wormed before the start of the experiment with Nalsacur anthelmintic at 2.5 ml per 10 kg BW. The animals were housed in sufficiently ventilated pens with concrete floors at night after grazing during the day. For the three different treatments, different ear tag colour codes were used for ease of identification of the goats. The colour code for the control was red, commercial supplement, green and for cowpea seed hulls, yellow.

Experimental study design

Thirty-six Boer goats were partitioned into live-weight groups of four animals (two males and two females), with three replicates for each treatment, and within a live-weight group, goats were assigned dietary treatments in a completely randomized design (CRD). The first group was grazed/browsed with no supplemental diet (control); the second group of goats was offered 1.2 kg of commercial feed at 08h30 and then released to graze/browse the natural pasture; the third group of goats was also offered 1.2 kg of cowpea seed hulls at 08h30 and also released to graze/browse the natural pasture. The study comprised of a 14-day preliminary period of feed adjustment which was started on 15 June to 29 June; and the study finished on 26 August, 2013.

Both at the beginning and end of the feeding trial (42 d), two sets of blood samples were collected from each animal by jugular vein puncture using a 10 ml 20 gauge syringe and 10 gauge needles. One set of the blood samples (5ml) was collected into plastic tubes containing the anti-coagulant ethylene diamine-tetra-acetic acid (EDTA) for the determination of haematological parameters. The other set of blood samples (10 ml) was collected into anti-coagulant free tubes, and the blood coagulated at room temperature thereafter centrifuged for 5 minutes at 3000 rpm. The supernatant sera were stored in a freezer for biochemical analysis.

Blood analysis

The blood was analysed using the QBC® Vet Test analyser (IDEXX Laboratories inc. Westbrook, Maine, USA) for blood parameters including haematocrit, differential cell count and haemoglobin concentration. Briefly, fresh blood was filled into venous tubes and the samples were centrifuged for about 10 minutes before reading with the QBC® VetAutoread haematology™ analyser. In addition, the analyser reported the following parameters; mean corpuscular volume [MCV], mean corpuscular haemoglobin [MCH] and mean corpuscular haemoglobin concentration [MCHC] calculated from values of red blood cells [RBC], and haemoglobin [Hb] concentration. Serum total protein, serum urea, creatinine, albumen, serum calcium and phosphorus were determined by the use of Pentra C200, Horiba Medical equipment, clinical chemistry analyzer (Co. Ltd., France).

Statistical analysis

The variables analysed were haematological profile (RBC, WBC, PCV, Hb, MCV, MCH, and MCHC), clinical chemistry (total protein, albumin, creatinine, urea, calcium and phosphorus). Data were analyzed using the MIXED procedures of the Statistical Analysis Systems Institute (2008). Haematological parameters and chemical constituents of blood were analysed and pre-experimental haematological and clinical chemistry of the sera values were also used as covariates in analyzing post-experimental haematological and biochemical values. Means were compared by Dunnette t-test in the general linear model (GLM) procedure. Differences of means with $p<0.05$ were accepted as representing statistically significant differences. Repeated measures were taken on individual goats from the start of the trial and at the end of the study.

Results

Hematologic profile of the goats

Table 1 shows the mean values of white blood cells (WBC), haemoglobin (Hb), red blood cells (RBC), packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), and mean corpuscular haemoglobin concentration (MCHC) in Boer-goats grazed/browsed natural pasture and Boer-goats grazed/browsed natural pasture supplemented with either cowpea seed hulls or commercial concentrate. There was no significant variation ($p>0.05$) in mean values of haematological parameters of goats grazed/browsed and supplemented and those that were grazed/browsed without supplementation, except for the values of MCV and MCH, which were above the normal range. Both values increased significantly ($p<0.05$).

Table 1: Effect of natural browsing/grazing and natural browsing/grazing supplemented with cowpea seed hulls and commercial feed/concentrate on clinical haematology of Boer goats at Mantshwabisi Government ranch.

PARAMETER	BEFORE TREATMENT WITH CSH	AFTER TREATMENT WITH CSH	BEFORE TREATMENT WITH CF	AFTER TREATMENT WITH CF	INITIAL TREATMENT WITH NP	END OF TREATMENT WITH NP	REFERENCE RANGE
WBC x106/µl	10.26 ± 1.64	10.64 ± 1.76	12.81±2.45	8.34 ± 0.80	9.26± 0.60	8.46 ± 0.52	(4 - 13)
Hb (g/l)	8.29 ± 1.02	8.25 ± 0.90	7.34 ± 0.53	7.36 ± 0.44	7.90± 0.15	8.09 ± 0.53	(8 - 12)
RBCx1012/l	9.87 ± 1.23	9.12 ± 0.01	9.18 ± 2.23	8.27 ± 0.25	9.46± 0.24	7.98 ± 0.08	(8 - 18)
PCV (%)	28.11 ± 3.51	26.10± 2.83	26.6 ± 1.14	23.22 ± 0.74	26.73±0.71	22.27 ± 0.24	(22 - 38)
MCV (f/l)	28.4 ± 3.36	28.70±3.04	29.0±0.60	28.11± 0.11	28.27±0.20	28.00± 0.00	(16 - 25)
MCH (pg)	8.41 ± 1.00	9.08 ± 1.00	8.06± 0.59	8.96 ± 0.57	8.37 ± 0.13	10.13± 0.68	(5 - 8)

WBC- white blood cells; Hb – hemoglobin; RBC – red blood cells; PCV – packed cell volume; MCV – mean corpuscles volume; MCH – mean corpuscle hemoglobin; CSH – cowpea seed hulls; CF – commercial feed/concentrate; NP – natural pasture

Clinical chemistry

The results for biochemical indices of Boer-goats grazed natural pasture alone and Boer-goats grazed natural pasture supplemented with either cowpea seed hulls or commercial concentrate are presented in Table 2. There was no statistically significant variation ($p>0.05$) in the mean biochemical parameters of Boer-goats grazed natural pasture alone and those grazed natural pasture supplemented with either cowpea seed hulls or commercial concentrate, except in the mean values of calcium ($p<0.05$). There was a slight increase in the calcium level of goats that were not supplemented; this was evident at the beginning of the trial and at the end of the study. On the other hand, phosphorus levels also slightly increased in all the goats either supplemented or not. However, plasma albumin showed a slight decline after supplementation with cowpea seed hulls. Although creatinine level was within normal ranges in all the goats fed different diets or diet combinations, it was observed to be on the high side.

Table 2: Effect of natural browsing/grazing and natural browsing/grazing supplemented with cowpea seed hulls and commercial feed/concentrate on clinical biochemistry of Boer goats at Mantshwabisi Government ranch.

PARAMETER	BEFORE TREATMENT	AFTER TREATMENT	BEFORE TREATMENT	AFTER TREATMENT	INITIAL TREATMENT	END OF TREATMENT	REFERENCE
	WITH CSH	WITH CSH	WITH CF	WITH CF	WITH NP	WITH NP	RANGE
Protein	61.65 ± 1.72	59.72 ± 1.27	61.15 ± 0.74	62.03 ± 0.72	61.40 ± 1.27	61.02 ± 0.61	(59 - 74)
Calcium	2.53 ± 0.03	2.43 ± 0.01	2.53 ± 0.04	2.44 ± 0.03	2.66 ± 0.49	2.58 ± 0.05	(2.13- 2.55)
Urea	5.61 ± 0.58	4.55 ± 0.25	6.51 ± 0.39	4.57 ± 0.41	5.86 ± 0.49	2.58 ± 0.05	(5.4 - 11.8)
Creatinine	73.91 ± 2.75	97.87 ± 2.43	77.56 ± 3.03	90.96 ± 2.59	78.07 ± 2.48	102.06 ± 3.26	59.7 - 134.8
Phosphorus	1.69 ± 0.12	2.11 ± 0.12	1.85 ± 0.17	2.98 ± 0.10	1.40 ± 0.14	2.12 ± 0.13	(2.42- 3.97)

Discussions

The packed cell volume (PCV) and mean corpuscular haemoglobin concentration (MCHC) values observed in this study are in agreement with what other workers have established (Mason *et al*., 1989; Mbassa and Poulsen, 1993). There was no significant difference between values of blood components from goats grazed/browsed natural pasture alone and those grazed natural pasture and supplemented with either cowpea seed hulls or commercial concentrate, except for mean corpuscular haemoglobin (MCH) and mean corpuscular volume (MCV). Both values increased significantly. Mean corpuscular haemoglobin (MCH) evaluates the concentration of haemoglobin present in the blood whereas the mean corpuscular volume (MCV) determines the cell size. Haemoglobin plays an important role in supplying oxygen in the animal body. The over production of MCH is often caused by either megablastic anaemia due to dietary deficiencies in foliate vitamin or heat stress (Borges *et al*., 2013), the latter could possibly have been the cause as goats were grazed/browsed in scorching heat from the sun.

The observations of leucocyte counts in this study were slightly lower than those reported by Waziri *et al* (2010), except for goats supplemented with cowpea seed hulls. This showed that most of the goats were on healthy condition and the cowpea seed hulls had either some deleterious effects or increased animal bone marrow activities (Waziri *et al*., 2010). An increase of leucocytes can be an indication of either stressful condition such as disease development or increment of bone marrow activities (Waziri *et al*., 2010). Stress stimulates the release of leucocytosis inducing factor (LIF) and colony stimulating factors (CFS) which are known to increase haemopoeitic activities and blood cells mobilization into the circulatory system (Dellmann and Brown, 1987). The elevation of haemopoeitic activity is suggested to have been due to lack of feed intake by the goats as seen at the beginning of the study.

The mean values of plasma proteins were also observed to be on high side than those reported in Sahel goats (Kamalu *et al*., 1988), but within normal range. There was no significant variation in calcium level between the goats grazed/browsed basal diet at the beginning of the trial and at the

end of the study. The elevation of plasma calcium in goats grazed/browsed alone and/or grazed/browsed and offered supplements was possibly due to skeletal calcium release, increased intestinal calcium absorption, or decreased renal calcium excretion. Calcium regulates phosphorylation of endogenous proteins in the nervous system through the activation of calmodulin-dependent protein kinases and it also controls excitability of nerves and muscles. Overproduction of calcium has the effect of reduced excitability of pre-and post ganglionic nerve fibres (Borges *et al*., 2013). Moreover, a high level of calcium in the blood tends to interfere with phosphorus absorption, a condition that may lead to deficiency of phosphorus in animals.

Although phosphorus level appeared low in all goats fed three treatments before supplementation, it later upsurged as shown at the end of the study. This showed that there was subclinical *hypophosphotaemia* in the goats before treatments. *Hypophosphotaemia* is an electrolyte disturbance condition in which an animal abnormally has low level of phosphorus in the blood. This often occurs in malnourished animals which may have consumed large quantities of carbohydrates thereby removing phosphates from the blood creating high phosphorus demands by cells (Barcia *et al*. (1997). Therefore, the elevation of phosphorus level in goats at the end of the study was due to replenishment of phosphorus through controlled release of parathyroid and calcitonin hormones.

The creatinine, albumin and urea levels from goats differed from the report by Kamalu *et al*. (1988). The steadily increase of *creatinine* levels was attributed to the recycled urea as a response to limited dietary protein intake from the natural pasture (Gwaze *et al*., 2010). Incremental *creatinine* in the blood is caused by reduced filtration effects in the kidneys and increased production due to muscle catabolism (Wisloff *et al*., 2003). However, although the *creatinine* level seemed high in the goats fed three different diet treatments or treatment combinations, the levels were within normal limits. The difference in *creatinine* levels was influenced by crude protein content of the diets or diet combinations fed the goats (Gwaze *et al*., 2010). The plasma albumin was slightly lower in goats grazed/browsed and supplemented with cowpea seed hulls and those which were solely fed control diet only. Low levels of albumin are interpreted as under nutrition in animals (Wisloff *et al*., 2003). It was observed that goats which were grazed/browsed without being supplemented had inadequate nutrients supply. But goats which were grazed/browsed and supplemented with cowpea seed hulls had low levels of albumin due to lack of feed intake which was evident at the time when the supplement was first introduced.

Conclusion

The comparison of conventional grazing/browsing and grazing/browsing with supplementation of goats showed differences in both *haematologic and biochemical* profile changes. Goats grazed/browsed and supplemented had higher mean corpuscle haemoglobin (MCH) and mean corpuscle volume (MCV) than goats grazed/browsed without supplementation. Moreover, biochemical analysis showed higher *creatinine* and plasma proteins levels in supplemented goats.

All sets of goats used in the trial showed *hypophosphataemia* at the start of the experiment, but the condition improved quite substantially following di-calcium phosphate licks offered to goats.

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Literature cited

Association of Official Analytical Chemistry. 1990. Official Method of Analysis (12th edition) Washington D.C. USA.

Aukerman, R. C. 1981. The Basal Reader Approach to Reading. New York:John Wiley & Sons.

Barcia, J.P., Strife,C.F. and Langman, C.B. 1997. "Infantile Hypophosphotaemia: Treatment Options to Control Hypercalcium, Hypercalciuria, and Chronic Bone Demineralization." *Journal of Paediatrics 130: 825-828.*

Borges, S.A., Ariki, J. and Cummings, K.R. 2013. Dietary electrolyte balance for broiler chickens exposed to thermo-neutral and heat stress environments. *Poultry Science, 82, 428-435.*

Dellmann, H. and Brown, E.M. 1987. Histology of the digestive systems, In: Textbooks of Veterinary Histology, 3ed. Lea and Febiger 229.

Franzmann, 1972. Environmental sources of variation of bighorn sheep physiologic values. Journal of Wildlife Management 36:924-932.

Gwaze, F.R., Chimoyo, M. and Dzama, K. 2010. Nutritionally-related blood metabolites and faecal egg counts in indigenous Nguni goats of South Africa. South African Journal of Animal Science, 40 (Issue 5, Supplement 1).

Jenni-Elermann, S. and Jenni, L. 1998. What can plasma metabolites tell us about metabolism, physiological state and condition of individual birds? An over view of Biological *Conservation of Fauna., 102:312-319.*

Kamalu, N., Wen, F., Troutt, T.R., Crowe, C.T. and Chung, J.N. 1988. 'Particle dispersion by ordered motion in turbulent mixing layers', ASME Cavitation and Multiphase Flow Forum, FED., 64:150-154.

Mason, V.C., Cook, J.E., Cooper,E.M., Hoadley,C.J. and Cockbin,J.E. 1989. Oven and stack ammoniation of grass hays: voluntary intake, digestibility and faecal nitrogen excretion in sheep in relation to hay composition, *Animal Feed Science Technology, 24:313.*

Mbassa, G.K. and Poulsen, J.S.D. 1993. Reference ranges for haematological values in landrace goats. *Small Ruminant, Res. 9,367-376.*

McIntosh, G.H. and Pointon, A. 1981. The Kangaroo Island strain of pig in biomedical Research. *Aust. Vet.* 57: 182 – 185.

Metereology Department. 2013. Ministry of Science and Infrastructure Gaborone, Botswana.

Palomeque, J., Pinto, D. and Viscor, G. 1991. Haematlogic and blood chemistry values of the Masai Ostrich (*Struthio camelus). J. Wildl. Dis.*27 (1): 34-40.

Quintavalla, F., Bigliardi, E. and Betroni, P. 2001. Blood biochemical baseline values in the ostrich (*Struthio camelus),* 61-71, Universta degli Studi di Parma Annali della. Facolta di Medicina Veterinaria Vol. XXI.

SAS Institute. 2008. SAS User's Guide Statistic, Version 9.2, SAS Institute Inc. Cary NC. USA.

Seal, C. J., A. Sarker, and D. S. Parker. 1989. Rumen propionate production rate and absorption of fermentation end-products into the portal vein of forage and forage-concentrate fed cattle. Proc. Nutr. Soc. 48:143A..

Sowande, O.S., Aina, A.B.J., Oguntona,E.B., Fanimo, A.O., Unaka, V.U., Hassan, T.A. and Oseni, M.O. 2008. Performance, blood constituents and mineral balance of West African dwarf sheep fed preserved elephant grass, layers droppings and cassava peel diets during dry season. Nig. J. of Animal Prod. 35:90- 102.

Spinu, M., spinu, O. and Degen, A. 1999. Haematological and immunological variables in a domesticated and wild sub-species of Ostric (*Struthio camelus*). Br. Poult. Sci. 40:613-618.

Van Soest, P.J. and Mason, V.C. 1991. The influence of Maillard Reaction upon the nutritive value of fibrous feed. Anim. Feed Sci. Technol. 32:45-53.

Waziri, M.A., Ribadu, A.Y. and Sivachelvan, N. 2010. Changes in the serum proteins, haematological and some serum biochemical profiles in the gestation period in the Sahel goats. Vet arhiv 80(2),215-224.

Wisloff, H., Flaoyen, A.,Ottensen, N and Hove, T. 2003. *Narthecium ossifragum*(L.)Huds.Causes kidney damage in goats: Morphologic and functional effects. Vet. Path. 40,317-327.

Woerpel, R. and Rosskopf, W.J. 1984. Clinical experience with avian laboratory diagnostics. Vet. Clin. North. Am. Small. Anim. Pract. 14:249-272