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THE EFFECT OF STRESS AND SEASON ON SOME HAEMATOLOGICAL AND BIOCHEMICAL PARAMETERS OF THREE ANTELOPE SPECIES IN THE KALAHARI GEMSBOK NATIONAL PARK

F. LE R. FOURIE

Eugène Marais Chair of Wildlife Management
Department of Zoology
University of Pretoria
Pretoria
0001

M. T. VAN OUWERKERK

Department of Zoology
University of Pretoria
Pretoria
0001

Abstract – A seasonal study of some haematological and biochemical parameters of three antelope species (springbok *Antidorcas marsupialis*, oryx *Oryx gazella* and blue wildebeest *Connochaetes taurinus*) was conducted during January, April and June in the Kalahari Gemsbok National Park. A communal increase in two blood parameters during April was observed in all three species studied. These increases correlated with optimal range conditions. Active pursuit of individuals as well as drug induced immobilisation proved to place animals under stress influencing haematograms extensively.

Introduction

The Kalahari Gemsbok National Park (KGNP) is situated in the low rainfall (< 250 mm p.a.) semi-arid northwestern corner (24°15′ – 26°30′S: 20°00′ – 20°45′E) of the Republic of South Africa. Although referred to as a desert, this part of the Kalahari is best described as a southwest arid region according to the 11 point biotic index system of Africa (Odum 1969).

Typical rainfall and ambient temperature patterns exist with a maximum and minimum occurrence during January to April and May to December respectively. Vegetation varies according to season being abundant (February to June) or minimal (July to January), and animals must adapt to existing conditions. A study was undertaken to assess the impact of seasonal environmental variation on various haematological and biochemical parameters of three antelope species. Because of the rather wasteful nature of obtaining blood samples in the KGNP (culling excess animals by rifle) we also used blood samples obtained after active and organised hunting sessions in the Mier area to the south of and adjacent to the KGNP in order to increase the size of the test sample. The trauma of active hunting proved to be detrimental in obtaining 'normal' blood values and we consequently investigated immobilisation as yet another alternative means of augmenting the blood sample size. This paper thus reflects the influences of various factors on the haematograms of animals studied in this specific area.

Materials and methods

The three species of animals selected for this study are common to the KGNP and comprised springbok (*Antidorcas marsupialis*), oryx (*Oryx gazella*) and blue wildebeest (*Connochaetes taurinus*). Male animals were randomly selected at various sites during specific periods of the year *i.e.* January (mid summer), April (late summer) and June (mid winter). Animals were totally at ease when culled by a single rifle shot, usually from a distance within 30 metres. No other stress factors were thus involved.

Only one animal in a herd was culled whereafter blood was immediately aspirated directly from the left ventrical into 10 ml heparinized syringes. Blood analyses were completed within three hours in a field laboratory before setting out for further material. The number of animals utilized per specific month ranged from four to eleven for each species.

A limited number of blood samples were collected from animals shot in the adjacent Mier area (two springbok and two oryx) where seasonal hunting is permitted. Animals culled during this exercise proved to be extremely alert and invariably were run in by vehicle to be shot. Blood samples were collected and analysed as previously described.

The effect of a drug combination was thereafter evaluated and two springbok (0,5 mg Etorphine hydrochloride (M-99) and 22,3 mg Xylazine hydrochloride) as well as two oryx (2 mg M-99 and 70 mg Xylazine) were immobilized. These drugs were kindly provided by Reckitt and Colman (S.A.) (Pty) Ltd. and by Bayer Pharmaceuticals (S.A.) (Pty) Ltd.. Blood samples were immediately aspirated from the jugular vein of the drugged animals and thereafter at specific timed intervals. The antidote Diprinorphine hydrochloride (M-50/50 Reckitt and Colman) was thereafter administered and the animals set free.

Haematological and biochemical analyses were performed according to the following procedures (apparatus or method indicated in parentheses):

Blood cell counts (Haemacytometer and dilutent);
Micro-haematocrit (Micro-fuge);
Haemoglobin concentration (according to Kleihauer & Betke 1957);
Total plasma protein (Hitachi refractometer);
Blood glucose (Mannheim Boehringer Biochemica test combination);
Blood urea nitrogen (Merck test combination);
Lactic acid and cholesterol concentrations (Mannheim Boehringer Biochemica test combination); and
Malate dehydrogenase and Lactate dehydrogenase enzyme activities (Mannheim Boehringer Biochemica test combination).

The red cell indices mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were obtained by calculation.

Statistical differences were computed by means of students' t-test for paired values and results are given as mean ± Standard Deviation where applicable.

Results

Basic seasonal descriptions of 13 parameters for each species are presented in Tables 1, 2 and 3. Communal seasonal variations in the different species were evident in blood glucose and blood urea nitrogen concentrations. Each species, however, reflected various significant seasonal fluctuations in several other parameters. Small sample sizes invariably affected statistical computations and results obtained during this study should be considered as of indicative rather than absolute tendencies.

Table 1

Seasonal haematological and biochemical data for springbok (Antidorcas marsupialis)

Parameter	JUNE	JANUARY	APRIL
n	8	5	11
RBC ($\times 10^{-6}$ mm^3)	13,7 ± 2,6	14,7 ± 2,8	13,2 ± 1,8
WBC ($\times 10^{-3}$ mm^3)	3,5 ± 1,9	3,0 ± 2,9	4,8 ± 4,9*
Hct (%)	45,8 ± 1,1	47,1 ± 1,2	46,2 ± 2,0
Hb (g/ℓ)	167,0 ± 12,0	158,0 ± 9,0	163,0 ± 12,0
Glucose (mmol/ℓ)	2,6 ± 0,5	2,6 ± 0,2	4,1 ± 0,1**
Urea (mmol/ℓ)	24,3 ± 2,7	23,3 ± 1,7	27,0 ± 2,9
Lactic acid (mmol/ℓ)	0,99 ± 0,7	1,2 ± 0,9	1,12 ± 1,0
Cholesterol (mmol/ℓ)	1,9 ± 0,3	1,7 ± 0,3	1,9 ± 0,3
Total protein (g/ℓ)	75,0 ± 11,0	66,0 ± 9,0	68,0 ± 8,0
MDH (U/ℓ)	67,9 ± 20,0	49,0 ± 17,0	66,0 ± 22,0
LDH (U/ℓ)	180,0 ±104,0	106,0 ± 90,0	130,0 ±114,0
MCV (μ^3m)	33,3 ± 1,81	32,0 ± 1,0	35,0 ± 1,6*
MCH (ng)	12,2 ± 1,09	10,7 ± 0,1	12,4 ± 0,9
MCHC (%)	36,5 ± 4,1	33,6 ± 2,1	35,3 ± 3,8

* $P<0,10$
** $P<0,5$

Figure 1 reflects the percentage deviation observed in the haematograms of animals culled in the Mier area opposed to values obtained from animals shot in the KGNP (KGNP values were regarded as being 100%). Culling of animals in Mier was executed during the same week and under similar veld conditions as in the Kalahari Gemsbok National Park. Small sample sizes rendered statistical analyses obsolete and a general comparison is reflected in Fig. 1.

Table 2

Seasonal haematological and biochemical data for oryx (Oryx gazella)

Parameter	JUNE	JANUARY	APRIL
n	5	6	5
RBC ($\times 10^{-6}$ mm^3)	9,8 ± 1,1	9,9 ± 2,2	10,2 ± 1,0*
WBC ($\times 10^{-3}$ mm^3)	5,9 ± 0,9	3,6 ± 0,4	6,9 ± 1,1
Hct (%)	36,3 ± 1,1	38,9 ± 0,8	37,9 ± 1,5
Hb (g/ℓ)	114,0 ± 16,0	118,0 ± 9,0	121,0 ± 13,0*
Glucose (mmol/ℓ)	4,4 ± 0,05	3,8 ± 0,04	4,6 ± 0,02*
Urea (mmol/ℓ)	18,8 ± 2,0	18,3 ± 2,3	21,0 ± 1,4*
Lactic acid (mmol/ℓ)	2,19 ± 0,06	1,81 ± 0,07	1,76 ± 0,11*
Cholesterol (mmol/ℓ)	1,8 ± 0,4	1,8 ± 0,3	22 ± 0,2**
Total protein (g/ℓ)	72,0 ± 4,0	69,0 ± 10,0	70,0 ± 12,0
MDH (U/ℓ)	83,0 ± 19,0	66,0 ± 29,0	77,0 ± 30,0
LDH (U/ℓ)	209,0 ± 81,0	189,0 ± 93,0	170,0 ± 66,0*
MCV (μ^3m)	36,9 ± 0,3	39,3 ± 4,1	37,2 ± 1,2
MCH (ng)	11,5 ± 0,2	11,9 ± 1,8	11,9 ± 0,9
MCHC (%)	31,3 ± 0,4	30,3 ± 2,1	31,9 ± 2,0

* $P<0,10$

** $P<0,5$

Table 3

Seasonal haematological and biochemical data for blue wildebeest (Connochaetes taurinus)

Parameter	JUNE	JANUARY	APRIL
n	7	6	8
RBC ($\times 10^{-6}$ mm^3)	8,2 ± 1,1	7,9 ± 2,1	8,1 ± 1,9
WBC ($\times 10^{-3}$ mm^3)	5,3 ± 1,1	6,0 ± 4,3	4,3 ± 2,1**
Hct (%)	44,1 ± 3,8	44,2 ± 2,6	43,9 ± 1,8
Hb (g/ℓ)	137,0 ± 16,0	130,0 ± 10,0	141,0 ± 9,0*
Glucose (mmol/ℓ)	4,0 ± 1,0	3,8 ± 0,7	4,9 ± 1,1**
Urea (mmol/ℓ)	26,0 ± 2,7	20,2 ± 2,3	28,3 ± 3,1**
Lactic acid (mmol/ℓ)	1,1 ± 0,1	1,6 ± 1,0	1,4 ± 0,5
Cholesterol (mmol/ℓ)	1,6 ± 0,3	1,6 ± 0,2	2,1 ± 0,4*
Total protein (g/ℓ)	78,0 ± 9,0	76,0 ± 6,0	77,0 ± 12,0
MDH (U/ℓ)	55,0 ± 24,0	71,0 ± 28,0	78,0 ± 17,0*
LDH (U/ℓ)	260,0 ± 71,0	180,0 ± 66,0	220,0 ± 53,0
MCV (μ^3m)	53,7 ± 3,2	55,9 ± 2,1	54,2 ± 4,9
MCH (ng)	16,7 ± 1,9	16,5 ± 1,1	17,4 ± 2,1*
MCHC (%)	31,1 ± 2,6	29,4 ± 3,1	32,1 ± 2,1

* $P<0,10$

** $P<0,5$

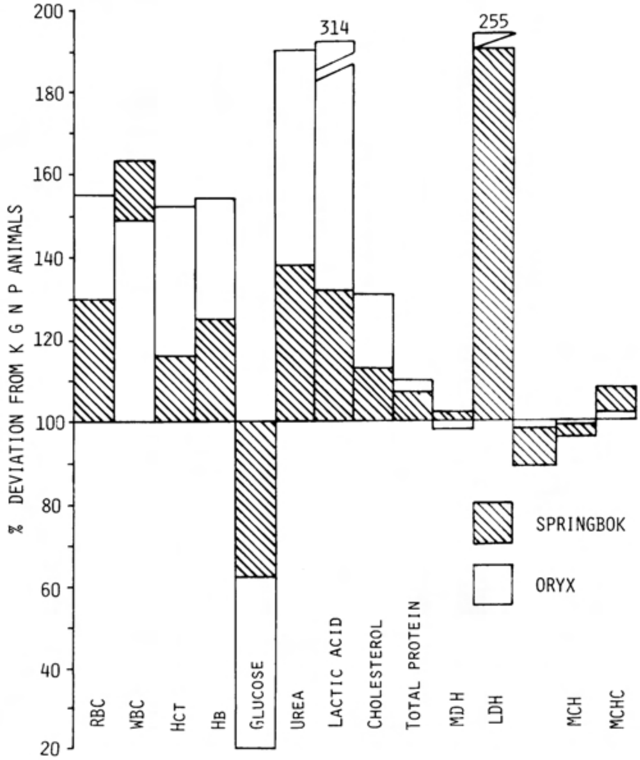

Fig. 1. Percentage deviation observed in haematograms of animals culled in the Mier area opposed to animals obtained from the Kalahari Gemsbok National Park (KGNP). (KGNP animal values were considered 100%.)

Immobilization of animals was achieved by utilizing a drug combination of Etorphine hydrochloride (M-99) and Xylazine hydrochloride (Rompun). This drug combination clearly affected the blood profile over a period of time and induced respiratory irregularities in springbok whilst oryx required subsidiary drug treatment to obtain satisfactory immobilization. Results obtained are given in Tables 4 and 5.

Discussion

Various researchers have embarked on the possibility of utilizing certain parameters as predicative measures for body and range habitat conditions (Le Resche, Seal, Karns & Franzman 1974; Blakenship & Varner 1975; Barrett & Chalmers 1976). According to these authors certain parameters could be regarded as of promise such as cholesterol, free fatty acids, blood urea nitrogen, glucose and probably total plasma proteins. In the present study blood glucose and blood urea nitrogen concentrations were significantly elevated towards late summer correlating with optimal range and visual animal condition. These results are in agreement with the work of Le Resche *et al.* (1974) and correspond to the period of optimum range conditions during which animals should be in a peak anabolic state. This

Table 4
Haematological and biochemical changes observed in time after drug administration (Oryx gazella)

Parameter	Time: 49 min	52 min	70 min
Dose: 2 mg M-99 + 70 mg Xylazine			
Antidote: 4 mg M50–50			
n = 1			
RBC ($\times 10^{-6}$ mm^3)	9,82	9,52	7,62
WBC ($\times 10^{-3}$ mm^3)	5,9	8,1	8,9
Hct (%)	36,3	35,3	35,7
Hb (g/ℓ)	114,0	110,0	87,0
Glucose (mmol/ℓ)	4,4	5,1	5,6
Urea (mmol/ℓ)	18,8	20,0	20,0
Lactic acid (mmol/ℓ)	2,2	2,6	3,3
Cholesterol (mmol/ℓ)	–	–	–
Total protein (g/ℓ)	–	–	–
MDH (U/ℓ)	68,8	84,3	84,3
LDH (U/ℓ)	167,0	200,0	417,0
MCV (μ^3m)	37,0	37,1	46,9
MCH (ng)	11,6	11,6	11,5
MCHC (%)	31,3	31,2	24,5
T–ambient (°C)	15,0		
T–body (°C)	40,0	39,9	40,1
Heart rate	–	–	–
Respiratory rate	–	–	–

consistent increase in blood urea nitrogen is probably the result of protein intake exceeding protein requirements and the surplus amino acids being deaminated so that the resulting keto acids can be utilized in energy metabolism. However, Franzman & Bailey (1977) are of the opinion that increased blood urea nitrogen values could also result from catabolism of body tissue during periods of low protein intake.

Glucose concentration concomitantly increased with improved range conditions as reflected in the values for April, which indicate that the intake of glycogenic precursors increased during optimal conditions. None of the animals used revealed hypoglycaemic plasma during the study period but age differences, health and excitability could well have influenced results obtained (Franzman 1972).

Blood levels are also closely related to stress (Blankenship & Varner 1975) and blood studies of moose (*Alces alces gigas*) populations in Alaska indicated that packed cell volume, haemoglobin concentration, calcium, phosphorus and total protein concentrations were the most useful in evaluating population condition (Franzman & Bailey 1977). Wicht (1979) illustrated a correlation between total plasma protein and blood urea nitrogen in springbok obtained in the Transvaal and Monro & Skinner (1979) indicated that both kidney fat index and indices

Table 5

Haematological and biochemical changes observed in time after drug administration (Antidorcas marsupialis)

Parameter	Time: 10 min	15 min	25 min	30 min	45 min	60 min
Dose: 0,5 mg M-99 + 22,4 mg Xylazine						
Antidote: 1,0 mg M50–50						
n = 2						
RBC ($\times 10^{-6}$ mm^3)	14,2 ± 2,0	14,2 ± 1,19	–	10,5 ± 0,9	9,2 ± 1,4	9,6 ± 2,0
WBC ($\times 10^{-3}$ mm^3)	4,0 ± 1,1	4,7 ± 0,8	–	3,3 ± 0,7	3,0 ± 0,4	3,5 ± 1,0
Hct (%)	58,7 ± 2,3	51,5 ± 4,4	–	46,4 ± 2,0	47,0 ± 2,1	48,0 ± 1,9
Hb (g/ℓ)	189,0 ± 19,0	158,0 ± 33,0	–	143,0 ± 30,0	147,0 ± 28,0	150,0 ± 30,0
Glucose (mmol/ℓ)	2,2 ± 0,1	3,6 ± 0,1	–	4,0 ± 0,3	4,2 ± 0,2	4,4 ± 0,3
Urea (mmol/ℓ)	24,3 ± 1,7	21,2 ± 1,3	–	22,2 ± 2,0	17,8 ± 1,3	35,5 ± 1,8
Lactic acid (mmol/ℓ)	80,93 ± 0,1	0,82 ± 0,1	–	0,71 ± 0,1	0,82 ± 0,1	0,77 ± 0,1
Cholesterol (mmol/ℓ)	–	–	–	–	–	–
Total protein (g/ℓ)	–	–	–	–	–	–
MDH (U/m)	85,9 ± 12,0	51,6 ± 11,0	–	77,3 ± 13,0	67,1 ± 10,0	75,7 ± 9,0
LDH (U/m)	200,0 ± 27,0	150,0 ± 13,0	–	83,0 ± 15,0	150,0 ± 14,0	150,0 ± 10,0
MCV (μ^3m)	41,3 ± 1,1	36,2 ± 0,1	–	44,2 ± 0,2	50,9 ± 2,0	50,0 ± 1,9
MCH (ng)	12,7 ± 0,2	11,1 ± 0,1	–	13,6 ± 0,2	15,9 ± 0,9	15,7 ± 1,1
MCHC (%)	30,6 ± 2,1	30,6 ± 2,2	–	30,8 ± 2,0	31,3 ± 1,9	31,3 ± 2,0
T–ambient (°C)	17,6					
T–body (°C)	39,2 ± 0,2	38,9 ± 0,6	38,8 ± 0,4	37,3 ± 0,6	37,4 ± 0,2	36,9 ± 0,2
Heart rate	66,0 ± 4,0	48,0 ± 8,0	48,0 ± 8,0	48,0 ± 6,0	48,0 ± 7,0	42,0 ± 8,0
Respiratory rate	Irregular					

derived from complete buttock dissection proved to be reliable indices of animal condition in the impala (*Aepyceros melampus*). Although similar results for kidney and body fat indices were described by Wicht (1979) for springbok, he only succeeded to illustrate a correlation between fat and blood urea nitrogen from the blood parameters analysed.

During April and July of this study, when range conditions varied between optimal to reasonable, a consistent increase in total plasma protein was evident in all three studied species. These increased protein levels further substantiate the higher blood urea nitrogen levels observed during this period. With the exception of oryx, haemoglobin content also followed a similar pattern of higher values during April and July which inevitably were reflected in MCH and MCHC calculations. Barrett & Chalmers (1976) indicated statistical differences in MCH and MCV calculations in relation to body temperatures. In our study body temperatures were not monitored but a fluctuating tendency in the previously mentioned parameters did manifest itself. Both springbok and oryx had high red cell counts during January which correlates with a general low haemoglobin value, possibly indicative of lowered protein turnover. This tendency of inverse relationship also exists between range condition and haematocrit values for all three species which is in contrast with the findings of Franzman (1972) who indicated a positive relationship.

Lactic acid concentrations in springbok and blue wildebeest peaked during January whilst cholesterol levels were consistently higher during April or April and July. This is in agreement with Bjarghov, Fjellheim, Hove, Jacobsen, Skjenneberg & Try (1976) who found higher serum levels of total lipids and cholesterol in animals fed on balanced pelleted reindeer feed compared to deer kept on a diet of lichens.

They are further of the opinion that body weight, haematocrit and magnesium concentrations could be regarded as effective indicators of nutritional status of reindeer (*Rangifer tarandus tarandus*).

Seasonal variation in the haematograms of the three different species in the study differed considerably. April values for oryx's red cell count, total haemoglobin, blood glucose, urea and cholesterol levels as previously mentioned were all elevated whilst lactic acid concentration and LDH activity decreased significantly when computed against January or July values. Some of these values are also applicable to the other two species. The lower LDH and lactic acid profile for oryx during April, could be indicative of an increased capacity to tolerate stress (Harthoorn 1976b). Franzman & Bailey (1977), however, are critical of LDH as an excitability index.

In a comparative study of this nature, care should be excercised to minimize external factors which could influence the results obtained (Morgan & Upton 1975; Kumar & Bahga 1976; Nangia, Singh & Sukhia 1980). Factors to be considered are for instance time delay during analyses (Hinton & Jones 1978), sampling techniques (Morgan & Upton 1975), different experimenters (Lindena, Bütterner & Friedel 1977), variances incurred due to electronic measuring instruments and its calibration (Burrow, Hendrich & Rapp 1977) as well as the effects of vacutainer tubes on various cell indices (Glickman & Boyd 1977) and the possibility of dehydration due to stress and exercise, to name but a few.

The second part of this study clearly indicated a substantial difference in haematograms for animals culled in the Mier area compared to KGNP animals. From this it can be derived that KGNP animals are well conditioned to motor vehicles resulting in minimal stress when approached and shot. This is also the method of preference compared to results obtained from immobilization or physical restraint (Blankenship & Varner 1975; Barrett & Chalmers 1979). Animals occurring in areas frequently exploited by hunters appear to be under considerably more stress, inducing drastic changes in some blood constituents, following the classical physical stress syndrome attributed to elevated levels of corticosteroid and catacholamines (Jacobsen, Kirkpatrick, Burkhart & Davis 1978; Schalm, Jain & Carrol 1975). Although Fig. 1 only comprises two animals of each species, it seems that oryx tended to be more stressed than springbok. Omitted from this figure, however, is the degree of stress inflicted before the kill. Blood samples obtained from the Mier area were thus not incorporated in the final assessment.

Another method of increasing sample sizes without killing animals is by immobilization. This method was also found insufficient for comparative purposes which is in accordance with the observations of Blankenship & Varner (1975). The necessity of additional dosages of the drug compound in oryx retrospectively probably stems from a too low dose of narcotic which rendered one of the animals only partly immobilized. This invariably could have affected the physiological status of animals and a continuous drift in most parameters was noticed during the immobilization period. These changes are mediated by the central acting mode of Etorphine hydrochloride with a pharmacological profile similar to that of morphine (Harthoorn 1976a). This drug combination is also active on endocrinological level *e.g.* inhibiting *inter alia* insulin secretion resulting in hyperglycaemia (Feldberg & Gupta 1974). In the present study this tendency was verified and the influence of the drug combination was illustrated. Due to the possible effect of this drug on the hormonal status, the physiological homeostasis should be considered endangered resulting in abnormal haematological and biochemical values (Kumar & Bahga 1976; Kitchen 1979). These values were also not incorporated in order to increase sample size.

From this study it is concluded that a general increase in blood glucose concentration and blood urea nitrogen could be associated with optimal range and body condition of three species of animals in the Kalahari Gemsbok National Park. It is further stressed that care should be exercised in obtaining and interpreting haematological data. Results obtained from animals subjected to periodic hunting as well as samples collected from drugged animals should be treated with the necessary interpretive care.

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