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HEMATOLOGIC AND BLOOD CHEMISTRY VALUES OF THE MASAI OSTRICH (*STRUTHIO CAMELUS*)

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ABSTRACT: Normal mean values for hematocrit, hemoglobin concentration, erythrocyte and leukocyte counts, hematimetric indices, erythrocyte dimensions, glucose, urea, uric acid, cholesterol, creatinine, total bilirubin, serum aspartate aminotransferase, serum alanine aminotransferase, alkaline phosphatase, creatinine phosphokinase, lactic dehydrogenase, inorganic phosphorus, chloride, total plasma protein, sodium, potassium, calcium, and magnesium were obtained from the blood or plasma of four Masai ostriches (*Struthio camelus*) when juveniles at 5 mo of age and as adults 1 yr later in the Barcelona Zoo (Spain). Young ostriches had significantly lower concentrations of hematocrit, hemoglobin concentration, calcium, and magnesium, and higher levels of total protein and potassium, than the adult individuals. The rest of the parameters were not significantly different between the two age groups. The data obtained provide reference values for Masai ostriches.

Key words: Blood chemistry values, hematology, Masai ostrich, *Struthio camelus*, normal reference values, age variation, captive study.

INTRODUCTION

There is a paucity of information on the hematology and blood chemistries of many wild avian species, especially for ratite birds. Ostriches are peculiar flightless bird with vestigial wings and well-developed legs allowing them to run for several minutes at a velocity of 50 to 70 km/hr and for long distances (Cramp, 1985). Hematological values of the Masai ostrich (*Struthio camelus massaicus*) were obtained by Gulliver (1875). Later, De Villiers (1938) described physical properties of the blood of the ostrich and more recently, Leonard (1982) listed hematological data including red and white blood cell counts and hemoglobin. Stoskopft et al. (1982) in a pool of eight blue necked ostriches (*Struthio camelus austrealis*) described a few hematologic parameters and the plasma protein concentration.

Clinical hematology and chemistry is a useful aid for the diagnosis of disease and illness in caged birds (Woerpel and Rosskopf, 1984). Therefore, it is important to establish hematological reference values in the ostrich and other ratites in order to have baseline information. This paper reports on baseline hematological and biochemical values in healthy juveniles and adult Masai ostriches.

MATERIALS AND METHODS

The ostriches were born in the Tel Aviv Zoo (Tel Aviv, Israel), subsequently purchased by the Barcelona Zoo (Barcelona, Spain), and maintained in 500 m² fenced pens. The diet of the animals consisted of a mixture of fresh vegetables plus a vitamin supplement (Pecutrin® Bayer, AG Leverkusen, Federal Republic of Germany). Water was available ad libitum. All animals were apparently healthy with no clinical signs of disease.

Blood samples were obtained on two occasions from the same individuals: once when the ostriches were 5-mo-old (juveniles), and 1 yr later (adults). To reduce the variation associated with diurnal changes in blood (Dolnik, 1973), the blood samples were taken from the brachial vein at the same hour (10:00 to 11:00 A.M.). Blood samples were drawn with a heparinized syringe. The cellular portion of the blood was removed by centrifugation to stabilize different enzyme systems. A small blood sample was separated to immediately conduct the hematological analyses, which were always completed within 2 hr after blood withdrawal. The plasma tubes were immediately frozen and the biochemical analyses were conducted subsequently.

The hematocrit was determined using a heparinized capillary tube and centrifuging the blood in a micro-hematocrit centrifuge (Gri-Cel, Barcelona, Spain) for 6 min at 11,500 rpm.

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TABLE 1. Hematologic values in the captive ostrich.

Measurement	Unit	Juveniles	Adults
Hematocrit	%	37.0 (2.1)*	48.0 (2.4)
Erythrocytes	$\times 10^6/mm^3$	1.91 (0.28)	2.42 (0.37)
Hemoglobin	g/100 ml	13.3 (0.39)	15.6 (0.89)
Mean corpuscular volume	μm^3	196.9 (31.2)	201.1 (25.1)
Mean corpuscular hemoglobin	pg	70.5 (9.0)	65.3 (7.0)
Mean corpuscular hemoglobin concentration	%	35.9 (3.1)	32.5 (3.0)
Leukocytes	$\times 10^3/mm^3$	19.5 (13.7)	21.0 (8.0)

* Mean (standard deviation).

Hemoglobin concentration was measured by adding 10 μl of well-mixed whole blood to 5 ml of Drabkin's reagent (Drabkin and Austin, 1935). After 10 min this was centrifuged at 2,500 rpm for 5 min in order to avoid interference of the lysed nuclei during the determination of the optical density which was measured in a spectrophotometer set a 540 μm. The erythrocyte number (RBC) was counted in a hemocytometer Thomas chamber after the sample was diluted in saline (Ferrer, 1929). The leukocyte count (WBC) was determined using a hemocytometer (Ferrer, 1929). The hematimetric indices, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were calculated rom the RBC count, hematocrit, and hemoglobin concentration. The morphometric determination of average RBC length and width were measured from selected smears. These were fixed with methanol and stained with May-Grünwald and Giemsa solutions, as described by Lucas and Jamroz (1961). The cells were examined at 1,000× (oil immersion objective) and their dimension estimated by means of a calibrated eyepiece (Nikon; Tokyo, Japan). Measurements of 50 erythrocytes were taken from different smears selected for excellence of staining and internal cytology. Ratios of maximum length to width were calculated as an index of the deviation of RBC's from a spherical shape. All parameters were determined following the methods described for avian blood (Lucas and Jamroz, 1961).

TABLE 2. Measurements of erythrocytes in the captive ostrich.

Measurement	Unit	Juveniles	Adults
Cell length	μm	13.18 (1.17)*	13.30 (1.09)
Cell width	μm	7.49 (0.84)	7.53 (0.79)
Cell length/cell width		1.76 (0.15)	1.77 (0.19)
Nucleus length	μm	5.47 (0.65)	5.58 (0.72)
Nucleus width	μm	2.89 (0.40)	2.96 (0.48)
Nucleus length/nuclear width		1.94 (0.37)	1.89 (0.41)

* Mean (standard deviation).

Glucose (glucofix menagent, Menarini, Via Sette Santi 3, 50131 Firenza, Italy), urea (urea HF menagent, Menarini), uric acid (uric acid HF menagent, Menarini), cholesterol (cholesterol HF menagent, Menarini), creatinine (3385 merckotest, Merck, Frankfurter Strasse 250, D-6100 Darmstadt 1, Federal Republic of Germany), total bilirubin (total bilirubin menagent, Menarini), serum aspartate aminotransferase (AST) (3397 merckotest, Merck), serum alanine aminotransferase (ALT) (3398 merckotest, Merck), alkaline phosphatase (AP) (alkaline phosphatase menagent, Menarini), creatinine phosphokinase (CPK) (14328 merck-1-test, Merck), lactic dehydrogenase (LDH) (3349 merck-1-test, Merck), and chloride (chlorofix menagent, Menarini), were determined using commercial kits. These techniques are commonly used in avian veterinary diagnosis. Total plasma proteins (TP) were measured using the Lowry et al. (1951) technique. Sodium and potassium were analyzed by flame emission spectrophotometry (Varian-875, Varian International AG, Viaduckstrasse 65, CH-4011 Basel, Switzerland) and calcium, magnesium and inorganic phosphorus by inductively coupled plasma (ICP spectrophotometer Jobin Yvon-JY-38 VHR, ISA Instruments, Division Jobin Yvon, 16-18 rue du canal Longiumeau, Cèdex, France).

RESULTS AND DISCUSSION

Tables 1, 2 and 3 summarize the hematologic and biochemical values for the juvenile and adult ostriches as well as the mean and standard deviation of the four individuals from each group. The mean values between adults and young birds were compared with the Student's t-test. Statistical significance was considered at $P < 0.05$.

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TABLE 3. Blood chemistry values in the captive ostrich.

Measurement	Unit	Juveniles	Adults
Total protein	g/100 ml	6.22 (0.92)*	3.87 (0.58)
Glucose	mg/100 ml	263.5 (109.9)	207.4 (33.6)
Blood urea nitrogen	mg/100 ml	3.12 (0.43)	2.34 (0.27)
Uric acid	mg/100 ml	9.81 (1.26)	11.17 (2.11)
Cholesterol	mg/100 ml	148.2 (65.1)	116.2 (27.2)
Total bilirubin	mg/100 ml	0.131 (0.072)	0.144 (0.04)
Creatinine	mg/100 ml	0.621 (0.138)	0.641 (0.13)
Lactic dehydrogenase	IU/L	463.3 (168.0)	514.9 (286.1)
Alkaline phosphatase	IU/L	339.1 (135.8)	171.5 (45.9)
Aspartate aminotransferase	IU/L	152.7 (35.2)	190.5 (39.4)
Alanine aminotransferase	IU/L	16.99 (5.71)	20.62 (4.36)
Creatinine phosphokinase	IU/L	570.8 (172.0)	933.0 (269.0)
Sodium	g/L	3.95 (0.09)	3.69 (0.26)
Potassium	mg/100 ml	18.42 (3.18)	6.64 (0.98)
Chloride	mg/100 ml	37.56 (2.64)	31.29 (4.36)
Calcium	mg/100 ml	10.61 (0.63)	18.07 (1.62)
Phosphorus	mg/100 ml	11.95 (2.40)	13.71 (2.92)
Magnesium	mg/100 ml	1.74 (0.15)	2.24 (0.19)

* Mean (standard deviation).

Significant differences were observed in the hematocrit, hemoglobin concentration, total protein, potassium, calcium, and magnesium. All of these, except total protein and potassium, were higher in the older animals.

There was a considerable range in the values of certain parameters including the white blood cell count, glucose, AP, LDH, and CPK. However, no significant differences were found between ages of the birds. birds.

The levels of hemoglobin were similar in adult birds (De Villiers, 1938) or slightly higher (Stoskopf et al., 1982) than those previously described. The values for mature red blood cells (RBC) were slightly higher than those already reported for *Struthio camelus* (De Villiers, 1938; Leonard, 1982; Stoskopft et al., 1982). The hematocrit and RBC values were higher from the ostriches in the present study than those of the closely related rhea, *Rhea americana* (Timoshevskaya et al., 1983), although in general the hematological values, RBC, hemoglobin, hematocrit and hematimetric indices are within the range of most birds (Balasch et al., 1973, 1974, 1976; Cooper, 1975; Elliot et al., 1974; Hawkey et al., 1983; Palomeque et al., 1980).

Young Masai ostriches had significantly lower packed cell volumes and hemoglobin concentrations than the adults. The same differences have been described in many species. Kocan and Pitts (1976) noted a lower hematocrit in canvasback *Aythya valilsineria* ducklings versus adults. Hawkey et al. (1984a, 1984b) found lower hemoglobin values and red cell counts in captive rosy (*Phoenicopterus ruber ruber*) and chilean (*Phoenicopterus chiliensis*) juvenile flamingos than in adults. Fallaw et al. (1976) reported that female guineas (*Numida meleagris*) 8 wk of age had significantly lower hematocrit than other female age groups. According to Nirmalan and Robinson (1971) the increase in hemoglobin concentration could be due to the changes in blood volume per unit of body weight. The increased hemoglobin content per unit of volume of blood may have been a reflection of the decreased volume of blood per unit body weight. This age-related hematological difference does not seem to be characteristic of all avian species. There are many exceptions (Gessaman et al., 1986).

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The white blood cells (WBC) for the ostrich were in the same range as that described by De Villiers (1938). They are in agreement with those listed for the bird species compiled by Leonard (1982), although the variation is large.

The total protein levels of the adults were statistically different from the young ostriches. The former values were very similar to those reported previously in other species (Elliot et al., 1974; Gee et al., 1981; Perry et al., 1986), although lower than those reported previously for *Struthio camelus* (Stoskopf et al., 1982). The total protein of the juveniles was slightly higher than that of the adults; this is associated with high metabolic rates in relation to rapid tissue and feather growth, and as a consequence of high protein demands. In a previous study, Viscor et al. (1984) described an inverse relationship between hematocrit and plasma proteins in pigeons (*Columba livia*), gulls (*Larus ridibundus*), and chickens. The higher the value for the hematocrit, the lower was the value for plasma protein concentration. This was interpreted as a mechanism by which birds with higher hematocrit, such as the pigeon, can use a low plasma protein concentration to maintain the apparent viscosity of the blood at a similar level as other species having a lower hematocrit.

The blood urea nitrogen of the ostrich was similar to some of the concentrations reported for different birds (Gee et al., 1981; Leonard, 1982), although there is a great variability across different species of birds. Nevertheless, our results fall within the lower range of that reported previously. According to Sturkie (1965) high blood urea determinations may result from ingesting large quantities of animal protein; protein, especially that derived from nucleoprotein catabolism, results in increased urea production in birds. Since the ostrich is a vegetarian, this could explain the relatively lower values in this bird compared to other species. However, Lewandowski et al. (1986) concluded that since birds are uricotelic and produce uric acid as the major nitrogenous end product of metabolism, urea nitrogen is not a useful test of renal function in birds.

The uric acid level of the ostrich was within the normal range for birds (Gee et al., 1981; Leonard, 1982), although the range was very variable. According to Bell and Sturkie (1965) elevated uric acid levels occur in animals on high protein diets, although Gee et al. (1981) did not find high values in raptors.

The glucose values obtained were compatible with values reported by others for birds. In previous studies (Ferrer et al., 1987; Leonard, 1982) this parameter varies widely. This wide variation in glucose concentrations could be a consequence of the absorptive state of the animal. Also, Bairlein (1983) showed a seasonal rhythm in the blood glucose concentration of caged songbirds.

The cholesterol concentration was similar to the average values described (Gee et al., 1981; Brugère-Picoux et al., 1987) for most species of birds, although the values for captive raptors (Ferrer et al., 1987) and cavasbacks (Perry et al., 1986) were higher. Nevertheless, a wide variation is possible due to the circadian rhythms described in some species (Garcia-Rodriguez et al., 1987) or to the effect of diet on plasma and serum cholesterol in birds (Mori and George, 1978). Lowering the protein intake results in a higher level of serum cholesterol. A decrease in the excretion of cholesterol in the form of bile acids (the metabolic end products of cholesterol) causes high levels of serum cholesterol.

Total bilirubin in the Masai ostrich was close to many of the values reported for other avian species (Gee et al., 1981). Creatinine levels were also in the range of the values described for different birds (Brugère-Picoux et al., 1987; Gee et al., 1981). Creatinine, which is an indicator of renal integrity, showed a variety of concentrations in other studies; this might be explained by its relation with diet (Woerpel and Rosskopf, 1984). Birds fed a diet high

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in animal protein had high creatinine values.

The non-significant, but higher values of alkaline phosphatase (AP) found in young ostriches could be explained by the higher metabolism of the younger animals, since AP activity is related to cell and tissue turnover characteristic of the growing periods (Bell, 1971). The creatinine phosphokinase (CPK) and lactic dehydrogenase (LDH) values were slightly higher than those reported by some investigators (Mitruka and Rawnsey, 1977), possibly because they derive from muscular activity (Spano et al., 1987). The apprehension and restraint of the birds for bleeding could have increased the values of these parameters.

The sodium levels in plasma were similar to the values reported in other birds (Balasch et al., 1973, 1974). The potassium values for young ostriches were statistically different than those of the adults, almost three times greater.

Plasma chloride levels in the Masai ostrich and in other birds (Gee et al., 1981; Ghebremeskel et al., 1989; Perry et al., 1986) were similar and with smaller variations than those of other ions.

Inorganic phosphate levels in ostriches were higher than the values of other species of birds (Gee et al., 1981; Ghebremeskel et al., 1989).

The calcium concentration of ostriches was similar in the young birds and a little higher in the adults than the values reported by Leonard (1982) in 13 different species of birds. They are more in agreement with the concentration that has been found in chickens (Simkiss and Taylor, 1971). The variability observed in birds could have several causes, such as age, sex, time of the last feeding as noted in hawks (Rehder et al., 1982), diurnal rhythms as seen in various raptors (Garcia-Rodriguez et al., 1987), and as occurs during the egg-laying period as seen in Canada geese (Mori and George, 1978). The magnesium concentration in ostriches was within the range of previously reported data for birds (Balasch et al., 1973; Mitruka and Rawnsley, 1977).

We can conclude that the flightless ostrich has an oxygen carrying capacity close to that of other flying birds (Balasch et al., 1974) as reflected by the hematocrit, hemoglobin concentration and RBC count. Ostriches able to run at speeds comparable to other fast animals require, as do flying birds, large quantities of energy, which implies high oxygen transport requirements by the blood.

In summary, significant variations comparing the parameters of this study and the values in other avian species for the hematological and biochemical concentrations examined were revealed. Aside from the influence of biological or ecological factors, some of these variations could result from the different biochemical methods, temperatures at which the assay is performed, and the manufacturer of the kit used (Brugère-Picoux et al., 1987). It is important to standardize the analytical, collection, and processing techniques for hematological and biochemical studies in birds in order to make the future data from different laboratories truly comparable.

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