

Haematology and blood chemistry of *Cebus apella* in relation to sex and age

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Abstract: An effective health care program entails the prevention, diagnosis and treatment of medical problems. A knowledge of baseline values in clinically normal individuals is essential for determining the limits between good health and disease and for understanding the changes produced by pathogenic agents. However, very little information is currently available concerning the blood chemistry and haematological values of different species of monkeys, particularly new-world primates. The values of some haematological and chemical parameters in *Cebus apella* were determined. The aim of the present work was to verify the effect of age and sex on normal blood values. Blood samples were collected once a year for two successive years from 36 monkeys living in large captive social groups. Significant differences between males and females were found for AST, GGT, urea nitrogen and creatinine, erythrocytes, haemoglobin and haematocrit. Significant differences between juveniles and adults were found for calcium, AST, alkaline phosphatase, inorganic phosphorus, glucose, neutrophils, lymphocytes and serum protein parameters.

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Introduction

A knowledge of haematological and biochemical values of normal blood is essential for determining the limits between health and disease and for understanding the changes produced by pathogenic agents. Clinical haematology and clinical chemistry provide a useful aid for differential diagnosis in humans, and in animals used as laboratory models, as well as in comparative physiology and ethological studies.

Normal ranges of blood chemistry values have been reported for some species of non-human primates, most of which belong to the genus *Macaca* [3, 4, 6, 9, 10]. In contrast, little information is available on new-world monkeys, even for those most commonly used in the laboratory. For example, there is little information concerning the normal haematology and blood chemistry of the capuchin monkey, *Cebus apella* [12].

The capuchin monkey is a new-world monkey that is widely distributed in Central and South America. In recent years, capuchin monkeys have been used increasingly in biomedical and ethological research [1, 13].

Age and sex effects are shown to be the rule and to interact in many haematological and biochemi-

cal parameters, both in humans and in other non-human primates [3, 7, 8, 11, 15].

In a previous publication, preliminary data on 31 blood parameters of capuchin monkeys were reported [14]. The present work is a continuation of that study in which further samples have been considered, also to verify the effect of age and sex on normal blood values.

Materials and methods

Study subjects

Thirty-six healthy captive capuchin monkeys, 16 males and 20 females, classified into the following two age classes: juvenile 1–4 years (9 individuals) and adult > 5 years (27 individuals), were studied during their annual check-up.

The monkeys were housed in social groups, each group in an indoor–outdoor cage (indoor: 1.70 × 1.90 × 2.60 m; outdoor: the same size). Cages were furnished with perches and slides; a variety of plastic toys and wooden blocks were provided on a daily basis.

Monkeys were fed *ad libitum* with monkey chow. In addition, fruit and vegetables were supplied each afternoon. Twice a week the animals received

an extra meal of cheese mixed with eggs, bran, oats, sugar, salt, vitamins and minerals. Water was provided *ad libitum*.

According to a basic veterinary examination, all animals were considered healthy at the time of blood sampling. This was confirmed by the animals' behaviour in the colony as judged by the observers.

Veterinary check-up

Blood samples were collected once a year in the same month (October) for two successive years. This was to avoid possible seasonal effects on blood values, which are likely to influence some parameters such as growth pattern [5].

During the annual check-up, animals were placed in squeeze cages and sedated by an intramuscular injection of ketamine hydrochloride (7–10 mg/kg of body mass). While immobilised and under veterinary supervision, rectal temperature was taken using a paediatric thermometer and 5-ml blood samples were collected from the jugular vein between 09:00 and 11:00 h. Two millilitres of blood were placed in a lithium heparin anticoagulant tube for haematological analysis. The remainder was centrifuged for 10 minutes at 2,000 rpm within 3 hours from the time of blood collection. Serum was collected to determine serum biochemical values.

Haematological determinations were performed within 8 hours following the collection of the samples using standard haematological techniques. Total erythrocyte counts, leukocyte counts, haematocrit and haemoglobin values were performed electronically using a Seac H12. In order to obtain the white blood cell count, one thin blood smear was made of each sample, which was air dried, fixed with methanol, stained with Wright's Giemsa and read by optical microscopy.

The serum samples were analysed using an auto-analyzer (RA 1,000 Tachicon-Bayer). Table 1 summarises the methods used. All haemolysed serum samples were excluded.

Moreover, during the annual check-up the animals were tested for toxoplasmosis and leptospirosis, weighed and inspected for overall health. Faecal samples were also collected for bacteriological and parasitological tests.

Statistics

The haematological parameter values collected over the two years were averaged for each animal.

Standard statistical methods were used to process the data. The Mann–Whitney test (*U*-test)

was performed to determine significant differences between compared values.

Results and discussion

In literature, haematological data regarding *Cebus apella* are very rare. The only available data on this species are those published by Rosner et al. [12] in which only the values relative to males and females are compared. For this reason, it is very difficult to compare the results presented in this study with other data.

For the normal range data, comparisons were made between male and female, and between juvenile and adult values, for each test performed.

Table 2 compares blood baseline parameters of both males and females. Significant differences in biochemistry parameters were found for AST, GGT, urea nitrogen and creatinine ($P < 0.01$ and $P < 0.05$). These parameters were lower in males than in females, except for creatinine, which was higher in males. Interestingly, the inorganic phosphorus and glucose values were on average higher in females than in males, although the difference was not statistically significant. The other blood biochemical values were not statistically

Table 1. Methods of analysis used for serum chemistry values

Analysis	Methods	Units
Calcium	CPC	mg/dl
Aspartate aminotransferase (AST)	IFCC, 37°C	U/L
Alanine aminotransferase (ALT)	IFCC, 37°C	U/L
γ -Glutamyl transpeptidase (GGT)	Szasz, SCE, 37°C	U/L
Alkaline phosphatase	IFCC, AMP, 37°C	U/L
Phosphorus	Phosphomolybdate, 340 nm	mg/dl
Glucose	Hexokinase, 340 nm	mg/dl
Total protein	Biuret	g/dl
Urea nitrogen	Urease GLDH	mg/dl
Creatinine	Jaffe, kinetic	mg/dl
Total bilirubin	Blanked diazo	mg/dl
Cholesterol	Enzymatic	mg/dl
Triglycerides	GPO, Trinder	mg/dl
Lactate dehydrogenase (LDH)	L → P, 340 nm, Tris 37°C	U/L
Magnesium	Xylidyl blue	mmol/L
Uric acid	Uricase, trinder	mg/dl
Creatine phosphokinase (CPK)	IFCC-NAC activated, 37°C	U/L
Albumin	Electrophoretic	%
α_1 globulin	Electrophoretic	%
α_2 globulin	Electrophoretic	%
β globulin	Electrophoretic	%
γ globulin	Electrophoretic	%

Table 2. Mean, standard deviation (SD) and range for blood biochemistry, haematological and serum protein parameters for males and females¹

	Females			Males			
	n	Mean $\pm$ SD	Range	n	Mean $\pm$ SD	Range	<i>U</i> -test
Blood parameter							
Calcium	20	9.18 $\pm$ 0.61	7.95–10.7	16	9.02 $\pm$ 0.37	8.55–10	N.S.
AST	20	48.18 $\pm$ 10.77	23–66.5	16	42.22 $\pm$ 8.78	29.5–58	<i>P</i> <0.05
ALT	20	36.73 $\pm$ 18.78	17–108	16	34.38 $\pm$ 7.97	24–48	N.S.
GGT	20	95.53 $\pm$ 26.05	63–160	16	71.06 $\pm$ 14.32	34–94.5	<i>P</i> <0.01
Alkaline phosphatase	20	135.65 $\pm$ 108.46	43.5–410	16	145.22 $\pm$ 61.12	39–244	N.S.
Phosphorus	20	3.43 $\pm$ 0.77	2.3–5.0	16	2.95 $\pm$ 0.83	1.9–4.9	<i>P</i> = 0.05
Glucose	20	98.5 $\pm$ 41.97	35–192	16	76.22 $\pm$ 29.09	39–164	<i>P</i> = 0.05
Total protein	20	7.30 $\pm$ 0.55	6.3–8.35	16	7.25 $\pm$ 0.5	6.6–8.6	N.S.
Urea nitrogen	20	16.38 $\pm$ 5.23	8.5–31	16	13.44 $\pm$ 4.82	7.0–26	<i>P</i> <0.05
Creatinine	20	0.61 $\pm$ 0.12	0.45–0.94	16	0.71 $\pm$ 0.13	0.41–0.91	<i>P</i> <0.01
CPK	20	377.5 $\pm$ 224.2	131–964	16	364.88 $\pm$ 276.69	115–1009	N.S.
Total bilirubin	20	0.52 $\pm$ 0.05	0.5–0.7	16	0.51 $\pm$ 0.05	0.5–0.7	N.S.
Cholesterol	20	162.13 $\pm$ 53.55	82–272	16	136 $\pm$ 26.44	98–184	N.S.
Tryglycerides	20	92.7 $\pm$ 41.18	48–221	16	89.53 $\pm$ 26.41	63.5–176	N.S.
Magnesium	20	2.2 $\pm$ 0.27	1.63–2.63	16	2.15 $\pm$ 0.34	1.33–2.49	N.S.
LDH	20	284.85 $\pm$ 126.17	91–677	16	263.19 $\pm$ 63.94	183.5–414	N.S.
Uric acid	20	3.39 $\pm$ 1.24	0.84–5.88	16	3.46 $\pm$ 0.88	1.79–4.73	N.S.
Haematological parameter							
Erythrocytes	20	5.43 $\pm$ 0.62	3.76–6.31	16	6.27 $\pm$ 0.44	5.63–7.31	<i>P</i> <0.01
Leukocytes	20	7.21 $\pm$ 2.09	3.3–12	16	7.72 $\pm$ 1.84	4.3–12.15	N.S.
Haemoglobin	20	13.39 $\pm$ 1.28	9.6–15.25	16	15.01 $\pm$ 0.78	13.8–16.2	<i>P</i> <0.01
Haematocrit	20	40.94 $\pm$ 5.72	27.6–51.75	16	45.27 $\pm$ 3.60	37.65–52.7	<i>P</i> <0.01
Neutrophils	20	59.1 $\pm$ 9.65	41.5–79	16	63.16 $\pm$ 5.98	51–72	N.S.
Basophils	20	0	0	16	0	0	N.S.
Eosinophils	20	0.03 $\pm$ 0.11	0–0.5	16	0	0	N.S.
Lymphocytes	20	41.63 $\pm$ 10.64	21–63	16	36.78 $\pm$ 5.96	28–49	N.S.
Monocytes	20	0	0	16	0	0	N.S.
Serum protein parameter							
Albumin	20	63.92 $\pm$ 3.49	58.1–71.3	16	63.85 $\pm$ 4.91	54.1–69.7	N.S.
α_1 globulin	20	2.15 $\pm$ 0.45	1.5–2.9	16	2.0 $\pm$ 0.53	1.35–3.3	N.S.
α_2 globulin	20	5.7 $\pm$ 2.65	2.6–10.7	16	5.65 $\pm$ 2.05	2.9–9.7	N.S.
β globulin	20	14.58 $\pm$ 3.45	8.2–22.1	16	15.27 $\pm$ 3.16	9.45–22	N.S.
γ globulin	20	13.66 $\pm$ 3.47	7.6–20.4	16	13.23 $\pm$ 2.83	9.95–18.6	N.S.

¹Values recorded during two annual check-ups. n, number of samples; N.S., not significant.

significant, and it is reasonable to conclude that the ranges presented are applicable to both sexes.

The haematological data (Table 2) show significantly lower values of erythrocytes, haemoglobin and haematocrit in females. This could be attributed to menstruation, as is recognised in the human species. *C. apella*, in fact, is a menstruating monkey and cycle length is of about 20 days [2]. The white blood cell count was not statistically significant in either males or females. None of the serum protein values show statistically significant differences.

The values obtained in this study are generally in agreement with those reported by Rosner et al. [12] for this species, in particular the significant differences found between males and females in haemoglobin, erythrocytes and haematocrit. Different husbandry could account for the small

number of differences between Rosner's data and ours.

Table 3 compares blood baseline values between juveniles and adults. Significant differences in biochemical parameters were found for calcium, AST, alkaline phosphatase, inorganic phosphorus and glucose ($P < 0.01$ and $P < 0.05$). All parameters show higher values in juveniles. The higher values in alkaline phosphatase and inorganic phosphorus may be due to differences in osteoblast activity and bone development associated with skeletal growth. This is true in human beings where alkaline phosphatase and inorganic phosphorus values in serum vary considerably according to age and sex.

Significantly, the value of creatine phosphokinase (CPK) is on average higher in juveniles than in adults, although the difference is not statistically significant. The higher value of CPK, a muscle

Table 3. Mean, standard deviation (SD) and range for blood biochemistry, haematological and serum protein parameters for juveniles and adults

	Juveniles			Adults			
	n	Mean $\pm$ SD	Range	n	Mean $\pm$ SD	Range	U-test
Blood parameter							
Calcium	9	9.52 $\pm$ 0.6	8.9–10.7	27	8.65 $\pm$ 0.54	7.95–10.1	$P < 0.05$
AST	9	51.83 $\pm$ 5.01	47–63	27	43.43 $\pm$ 10.74	23–66.5	$P < 0.05$
ALT	9	34.89 $\pm$ 7.44	21.5–48	27	35.94 $\pm$ 16.7	17–108	N.S.
GGT	9	77.56 $\pm$ 9.74	63–91	27	87.02 $\pm$ 27.68	34–160	N.S.
Alkaline phosphatase	9	242.61 $\pm$ 99.93	79–410	27	105.67 $\pm$ 53.31	39–244	$P < 0.01$
Phosphorus	9	4.11 $\pm$ 0.85	2.95–5.0	27	2.92 $\pm$ 0.56	1.9–4.3	$P < 0.01$
Glucose	9	125 $\pm$ 43.52	73–192	27	76.46 $\pm$ 27.40	35–174	$P < 0.01$
Total protein	9	7.16 $\pm$ 0.47	6.3–7.65	27	7.31 $\pm$ 0.55	6.45–8.6	N.S.
Urea nitrogen	9	15.94 $\pm$ 6.71	8.0–31	27	14.78 $\pm$ 4.71	7.0–26	N.S.
Creatinine	9	0.64 $\pm$ 0.13	0.49–0.94	27	0.66 $\pm$ 0.14	0.41–0.91	N.S.
CPK	9	445.72 $\pm$ 231.49	185–898	27	347.28 $\pm$ 248.9	115–1009	N.S.
Total bilirubin	9	0.51 $\pm$ 0.04	0.5–0.62	27	0.51 $\pm$ 0.05	0.5–0.7	N.S.
Cholesterol	9	166.89 $\pm$ 50.56	107.5–253.5	27	145.06 $\pm$ 42.68	82.5–272	N.S.
Tryglycerides	9	98.06 $\pm$ 31.65	67–172.5	27	89.04 $\pm$ 36.27	48–221	N.S.
Magnesium	9	2.16 $\pm$ 0.37	1.63–2.53	27	2.18 $\pm$ 0.29	1.33–2.62	N.S.
LDH	9	294.22 $\pm$ 57.68	211.5–379	27	268.89 $\pm$ 113.85	91–677	N.S.
Uric acid	9	4.05 $\pm$ 1.09	2.89–5.88	27	3.21 $\pm$ 1.01	0.84–4.7	N.S.
Haematological parameter							
Erythrocytes	9	5.96 $\pm$ 0.38	5.41–6.57	27	5.76 $\pm$ 0.76	3.76–7.31	N.S.
Leukocytes	9	6.58 $\pm$ 1.51	4.3–8.75	27	7.72 $\pm$ 2.05	3.3–12.15	N.S.
Haemoglobin	9	14.31 $\pm$ 0.69	13.3–15.2	27	14.04 $\pm$ 1.51	9.6–16.2	N.S.
Haematocrit	9	44.37 $\pm$ 4.98	37.65–51.75	27	41.02 $\pm$ 9.06	4.55–52.7	N.S.
Neutrophils	9	56.39 $\pm$ 5.46	46.5–63.5	27	62.41 $\pm$ 8.7	41.5–79	$P < 0.05$
Basophils	9	0	0	27	0	0	N.S.
Eosinophils	9	0	0	27	0	0	N.S.
Lymphocytes	9	44.1 $\pm$ 5.71	36.5–53	27	37.93 $\pm$ 9.55	21–63	$P < 0.05$
Monocytes	9	0	0	27	0	0	N.S.
Serum protein parameter							
Albumin	9	66.91 $\pm$ 3.56	59.45–71.3	27	62.88 $\pm$ 3.84	54.1–69.25	$P < 0.01$
α_1 globulin	9	2.32 $\pm$ 0.39	1.8–2.8	27	2.0 $\pm$ 0.5	1.35–3.3	$P < 0.05$
α_2 globulin	9	4.26 $\pm$ 2.04	2.9–9.6	27	6.15 $\pm$ 2.31	2.6–10.7	$P < 0.05$
β globulin	9	15.95 $\pm$ 1.92	12.2–19.05	27	14.53 $\pm$ 3.6	8.2–22.1	N.S.
γ globulin	9	10.56 $\pm$ 2.41	7.6–15.65	27	14.44 $\pm$ 2.79	9.95–20.4	$P < 0.01$

¹Values recorded during two annual check-ups. n, number of samples; N.S., not significant.

enzyme, may be due to a more intense activity in juveniles than in adults.

Significant differences in haematological parameters between juveniles and adults were found only for neutrophils (higher in adults, $P < 0.05$) and lymphocytes (higher in juveniles, $P < 0.05$), as observed also in human species.

Significant differences in serum protein values between juveniles and adults were found for albumin, α_1 , α_2 and γ globulin ($P < 0.01$ and $P < 0.05$). Albumin and α_1 globulin parameters displayed lower values in adults, while α_2 and γ globulin in juveniles. We have so far been unable to account for this difference, especially because we are aware of no other published data on haematological parameters and serum protein values for this species.

Interestingly, our results refer to animals housed in social groups in highly enhanced and stimulating cages and on a very rich diet. This justifies hopes that our data are as close to the natural situation as possible.

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