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Hematological and serum biochemical indices in healthy bonnet macaques (*Macaca radiata*)

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Abstract

Background—Blood reference values for bonnet macaques (*Macaca radiata*) are limited. The goal of this study was to determine reference ranges for hematological and serum biochemical indices in healthy, socially housed bonnet macaques for males and females over a range of ages.

Methods—Blood hematological and serum biochemical values were obtained from 50 healthy bonnet macaques of both sexes and aged 10–234 months.

Results—Age and sex differences were present in a number of measures. Globulins, total protein, and creatinine (CREAT) values were highest among older subjects, while alkaline phosphatase, albumin, and phosphorus values were higher in juveniles. Sex differences were present in concentrations of red blood cells and CREAT, with higher values in males.

Conclusion—The blood parameter data reported here as age-specific reference values for laboratory-housed, healthy bonnet macaques may be used to inform clinical care and laboratory primate research.

Keywords

blood; CBC; chemistry; clinical pathology; monkey; nonhuman primate; parameter; reference range; reference range; wellness

Introduction

Hematological and serum biochemistry measures provide information that is essential for assessment of health, diagnosis of pathology, and understanding of disease processes. Furthermore, because many blood parameters change during normal development and maturation, there is potential for their use as biological markers of age-related decline [6]. The extent to which hematological and serum biochemistry measures are useful for either clinical or research use depends, however, on the availability of reference ranges against which a clinical or research sample can be compared and evaluated. Understanding sources of individual variation, including differences between species, sexes, and age groups is important because it can provide a meaningful context for interpreting data and a range of expected values for healthy animals. Identifying other factors or conditions that may significantly influence blood parameters is also essential to informing comparisons of blood parameters between populations or reference ranges.

Reference hematological and serum biochemistry data are available for many non-human primate species with long histories in laboratory research [18] including, for example, chimpanzees [6], cynomolgus macaques, [2] and rhesus macaques [3, 4, 9]. Previous research has contributed not only basic reference range data for the species, but also detailed information for specific age and sex classes within those species. Reports of both age and sex differences in blood hematologic and biochemical measures underscore the importance of obtaining both age and sex specific data. For example, males and females differ in red blood cell counts (RBC) and creatinine (CREAT) concentrations, while alkaline phosphatase (ALP) and phosphorus (PHOS) concentrations [17, 22] vary with age.

Among the macaques, hematological and serum bio-chemical parameters have been most extensively investigated predominantly in rhesus macaques [1, 9, 10, see 1 for published data on these parameters]. Consideration of subject characteristics such as age and sex are important to meaningful evaluation of an individual animal's data within the context of reference data for its species. Thus, identifying factors that influence individual variation in a healthy animal's hematological and serum biochemical values is important to using these data for health assessments and clinical care. Parallel to other primates, the results of previous studies of macaques demonstrate age-related differences in a number of blood parameters, including increased mean corpuscular volume (MCV) and mean corpuscular hemoglobin concentration (MCHC) values and decreased concentrations of ALP in older animals. Sex differences are also present, with higher CREAT values in males [1].

Given significant age and sex-related differences in rhesus macaques, both factors may play a role in individual variation in hematological and biochemical parameters in other species of macaques, including the bonnet macaque. A few investigations of bonnet macaques have demonstrated similarities to rhesus macaques in patterns of age and sex differences [see 15] in hematologic and biochemical parameters [7,14]. For example, with regard to age differences, ALP values were significantly lower in adult bonnet females when compared to adult males and juveniles [14] and with regard to sex differences, CREAT values were marginally higher in adult bonnet males compared to adult females, while RBC concentrations were significantly higher in bonnet males than females [14].

Although there are published reports on hematological and serum biochemical indices in bonnet macaques [12-14], they are from studies of animals in which the dietary and experimental conditions vary from the bonnet macaques in our study and from most macaques maintained in US laboratories. For example, two of the studies presenting reference values for bonnet macaques use subgroups of animals that were wild-caught, individually housed, and fed a laboratory diet that, while nutritionally balanced, is not commercially available to a wide number of primate research facilities in the US [14, 18]. Thus, one aim of our study was to provide hematological and serum reference values for bonnet macaques maintained under experimental and husbandry conditions more similar to those of other macaques in US facilities that provided data for other published reference ranges.

The study reported here provides hematological and serum biochemical data in a laboratory colony of 50 healthy bonnet macaques. The data provide reference values for this species under these laboratory conditions. We report average values and their variation within this population for each measure, as well as statistical evaluation of sex and age differences in those parameters for which previous studies in macaques have demonstrated age-related change or significant differences between males and females.

Materials and methods

Animals

Fifty bonnet macaques ranging in age from 10 to 234 months were included in this study (see Table 1). For the purposes of age group comparisons, subjects within the age range of 10–23 months were grouped as juvenile, 24–59 months as adolescent (i.e. the period from puberty through full adult maturation), and 128–234 months as fully mature adult. All animals were captive bred and group-housed in indoor-outdoor pens at the Wake Forest University Primate Center. Animals were housed in same-sex and mixed-sex groups. Diet consisted of commercially produced monkey chow, (Purina lab diet no. 5038) fed twice daily, and supplemented with fresh fruits and vegetables once daily. All adult females were confirmed to be nonpregnant via ultrasonography done prior to blood sampling. Physical examinations were performed during the sampling procedure and all animals contributing data for hematological and biochemical reference ranges had no signs of illness prior to blood sampling. All environmental conditions, procedures, and handling of animals were in strict compliance with the Institutional Animal Care and Use Committee and all experimental procedures were conducted in accordance with the Guide for the Care and Use of Laboratory Animals [11].

Blood sampling

Animals were fasted for 18 hours before sample collection. They were anesthetized using ketamine hydro-chloride (10 mg/kg, IM, KetasefIM; Fort Dodge Inc) prior to sampling blood via the femoral vein. Two 2-ml blood samples were collected. The first sample was drawn into a tube containing no anticoagulant and was allowed to clot for 1 hour before centrifugation and submission for serum biochemical analyses. The second was drawn into a

tube containing EDTA anticoagulant, hand mixed several times, and placed on wet ice for further hematological analyses.

Hematological and serum biochemical analyses

All samples collected for this study were sent to a commercial diagnostic laboratory (IDEXX Laboratories Inc, Westbrook, ME, USA). Whole blood and serum samples were submitted to analysis using IDEXX wellness panels (Total Health Plus™, Test code 1601). The analysis returned the values presented in Tables 2 and 3.

Statistical analyses of hematological and serum biochemical indices

Means and standard deviations were calculated for each measure as a function of age group. Analysis of variance (ANOVA) was used for comparisons of hematological and serum biochemical measures among the three age groups (juvenile, adolescent, adult) and t-tests were used for comparison of males and females. There was a larger age range in our adult sample (128–234 months), however, our analyses showed no significant differences between the oldest animals and the other animals in the adult sample and therefore the values were combined. Evaluation of age and sex effects was performed for those measures previously shown to be influenced by age or sex in other nonhuman primates [Age: hemoglobin (HOB), hematocrit (HCT), MCV, MCHC, lymphocytes, ALP, platelet count (PLAT CT), total protein (TP), albumin (ALB), globulin (GLOB), CREAT, glucose (GLU), and PHOS; Sex: RBC, HOB, HCT, MCV, CREAT, and GLU]. Significant main effects detected in ANOVA were followed with Fisher's post-hoc comparisons. *P*-values <0.05 are reported as statistically significant.

Results

Hematology

Hematological values (mean ± SD) for each age group and for the entire population are shown in Table 2, while age and sex differences are summarized in Table 4. There were significant age effects for PLAT CT, $F^{2,45} = 4.29$, $P = 0.02$. PLAT CT values were higher in adults (377.37 ± 131.24) when compared to adolescents (284.19 ± 82.97), $P = 0.006$. Juveniles' PLAT CT did not differ significantly from either adults or adolescents. There was a significant main effect of sex on RBC, $t = -2.29$, $P = 0.03$, such that RBC were higher in males (6.01 ± 0.62) than in females (5.66 ± 0.42). No other age or sex-related differences were found in analyzed hematological parameters.

Serum biochemistry

Serum biochemical values (mean ± SD) for each age group, as well as the entire subject sample, are summarized in Table 3. Table 4 provides a summary of significant age and sex differences. GLOB values were significantly different as a function of age, $F^{2,47} = 39.56$, $P < 0.001$. Adult GLOB values (3.62 ± 0.49) were higher than those of adolescents (2.87 ± 0.19) and juveniles (2.53 ± 0.21), $P < 0.0001$ and $P < 0.0001$, respectively. Similarly, adolescents had higher GLOB values than those of juveniles, $P = 0.02$. TP concentrations showed age effects as well, $F^{2,47} = 11.69$, $P < 0.0001$. TP concentrations were significantly higher in adults (7.40 ± 0.47) compared to adolescents (7.07 ± 0.32) and juveniles ($6.69 \pm$

0.20), $P = 0.007$ and $P < 0.0001$. Adolescents had higher TP values than juveniles, $P = 0.01$. ALB concentrations also varied significantly between age groups, $F^{2,47} = 14.09$, $P < 0.0001$ and were lower in adults (3.77 ± 0.35) compared to adolescents (4.20 ± 0.22) and juveniles (4.16 ± 0.15), $P < 0.0001$ and $P = 0.001$.

Creatinine values differed significantly between the three age groups, $F^{2,47} = 20.24$, $P < 0.0001$. Adult (1.02 ± 0.21) CREAT values were significantly higher when compared to adolescents (0.78 ± 0.11) and to juveniles (0.69 ± 0.06). As expected, there was a significant main effect of age on ALP values ($F^{2,47} = 47.52$, $P < 0.0001$), with adults' ALP values (133.58 ± 67.79) lower than those of either adolescents (750.77 ± 256.49) or juveniles (936.22 ± 398.08), $P < 0.0001$ and $P < 0.0001$, respectively. Juveniles' ALP values were marginally greater than adolescent ALP values, $P = 0.06$.

Phosphorous levels showed significant age effects, $F^{2,47} = 6.24$, $P = 0.004$, with adult values (3.39 ± 0.76) that were significantly lower compared to those of adolescents (4.08 ± 1.16) and juveniles (4.73 ± 0.85), $P = 0.03$ and $P = 0.001$. There was also a significant age effect for GLU, $F^{2,47} = 15.15$, $P < 0.0001$. Juvenile and adult GLU values were not different; however, adolescent GLU values (50.82 ± 5.79) were lower to both juvenile (66.11 ± 8.98) and adult (67.95 ± 14.74) values, $P = 0.0006$ and $P < 0.001$.

Males and females differed significantly in CREAT levels, $t = -2.03$, $P = 0.05$, such that males (0.93 ± 0.23) had higher levels compared to females (0.17). No other age or sex differences were found in analyzed serum biochemical parameters.

Discussion

The results of our study provide detailed information about hematological and serum biochemical parameters over a range of ages for bonnet macaques that are socially housed in a laboratory setting. Direct comparisons across species and laboratories are challenged by significant variation in housing, care, and methodological parameters. Although blood parameters are reported, obtaining reference values from healthy animals is often not the principal focus of the studies reporting such measures and thus, their interpretation is difficult. The goal of this study was to provide reference ranges for a group of healthy captive bonnet macaques, as well as identification of age and sex differences that may contribute to individual variation. Together our findings increase the available data against which values obtained for clinical or experimental purposes may be evaluated. Many of our findings converge with the pattern of age and sex differences observed in other primate studies. For example, our results are consistent with previous findings for RBC, GLOB, CREAT, ALP, and PHOS values in several non-human primate species including chimpanzees [6], rhesus macaques [4], tonkean macaques [17], as well as previous reports on bonnet macaques [14].

Hematological analyses revealed age-related differences in PLAT CT and sex-related differences in RBC concentrations. Platelet number values were highest in adult bonnets, a finding also reported in rhesus macaques [4]. Our finding of increased RBC concentrations in males is consistent with previous data in chimpanzees, capuchin monkeys, and bonnet

macaques [6, 14, 18]. No other significant differences in other hematological parameters were observed between age groups or sexes.

Age group differences were apparent in several serum biochemical measures. For example, the increase in TP concentrations with age was due to significant increases in GLOB with respect to age. The TP component of serum is composed of ALB and GLOB [5]. ALB proteins serve mainly as carrier proteins and osmotic regulators in the blood, while GLOB function ranges from its importance in secondary hemostasis to adaptive immune responses via the formation of gamma GLOB proteins such as immunoglobulins [5]. Our finding of increased GLOB may represent an increase in the production of immunoglobulin proteins with age and is consistent with previous reports in chimpanzees [6] and rhesus macaques [3, 4]. The decrease in ALB in older monkeys may reflect an increased demand of carrier proteins and protein metabolism during early development and maturation [5]. Age-related differences in TP, ALB, and GLOB were not observed in other studies of macaques [e.g., rhesus (7), and bonnet (14)]; however, a number of factors could contribute to the apparent discrepancy in results. In the study of bonnet macaques [14], the juvenile animals were reared in captivity, while the adult animals were of 'wild origin,' which may have long-term consequences on carrier proteins and immune related measures. Furthermore, the adult animals were fairly young (6-10 years old). In the previous study of rhesus macaques [7] comparisons were between adult and aged monkeys; this truncated range did not allow for evaluation of age effects in a manner similar to that reported here. Taken together, these findings suggest further study is warranted to determine the consistency of age-related differences in TP, ALB, and GLOB in macaques.

For the measures of ALP and PHOS we saw age-related decreases in serum levels. Alkaline phosphatase is important in the diagnoses of hepatic and bonerelated diseases [2]. Although several different isoenzymes of ALP exist in the liver, bile duct, kidney, bone, and placenta, high levels of ALP in young animals is likely caused by increases in the specific bone isoenzyme during growth and high rates of metabolism by bone-forming cells known as osteoblasts [5]. Age effects found in our study are consistent with previous reports in bonnet macaques as well [14]. PHOS, like ALP, is found to be increased in times of bone metabolism, and rapid growth as would be expected in the younger animals. Our finding of a decrease in PHOS with age is likely consistent with increases in levels of this mineral during development in juveniles and is similar to age-related differences in ALP reported previously in capuchin monkeys and chimpanzees [6, 22, respectively] and with no differences between adults and aged macaques [7].

Glucose was significantly lower in adolescents when compared to both juveniles and adults. Lower blood GLU concentrations in adolescents may be attributed to increased insulin levels and increased GLU metabolism in adolescents [5, 19]. The higher GLU values in adults are also consistent with the likelihood that heavier animals would have increased GLU concentrations [19]. A previous study in bonnet macaques reported that there were no age effects for GLU [14]; however, the age classifications in the previous study were based on body weight rather than known chronological age. In our sample, GLU is significantly and positively correlated with body weight, $r(48)=0.48$, $P=0.0003$. Further study is needed to address the consistency of age-related differences in GLU in bonnet macaques and to

directly determine whether environmental factors or method of age classification, or both, may contribute to the discrepancy between the previous report and our findings.

Finally, we found age and sex-related differences in CREAT levels. CREAT is a nitrogenous waste product found in muscles and is produced at a fairly constant rate that is dependent on muscle mass. CREAT levels are often increased in blood due to muscle damage [2]; however, our finding of increased CREAT in adults and in males can likely be attributed to higher average muscle masses in these subject groups [2, 17]. This finding is also consistent with previous reports of sex effects for CREAT in bonnet macaques [trend, see 14] and previous reports of both age and sex differences in Tonkean macaques [17].

Beyond species differences and potential differences in a range of husbandry conditions, there are also experimental conditions that are known to influence hematological and biochemical measures. For example, there is a group of studies that report that ketamine, and other anesthetics, affect several blood hematological and serum biochemical values in non-human primates. Measures affected by ketamine include, but are not limited to: HGB, white blood cell, GLU, and TP concentrations and blood lipid parameters [12, 18]. The studies that report significant effects of ketamine on these measures also vary widely by collection methodology, including order effects in drug presentation [12, 18], handling conditions, and potential for acute stress effects in no-drug conditions where blood sampling is performed on awake, restrained animals [8, 12, 18, 20]. One study compared the effects of a number of different anesthesia formulations by using a randomized design and found no effects on hematological measures due to anesthesia type [21]. In light of these mixed findings and gaps in the literature, it is difficult to clearly determine the effects of anesthesia and sample collection procedures on hematological measures. These findings suggest that further experimental study is needed to identify whether – as well as the extent to which – anesthesia and collection procedures influence these parameters. The findings also point to the need for a larger number of reference datasets to provide a more accurate context for comparison and emphasize the importance of specifying sample collection conditions along with reference range data. At the same time, these considerations underscore that sample values collected for clinical or experimental procedures should ideally be evaluated against reference data collected under similar conditions.

Together these considerations highlight not only challenges to direct comparisons across laboratories and species, but also some of the difficulties in construction of useful reference ranges from which to evaluate the degree to which an individual's data may vary from norms. They also call attention to the importance of obtaining data from healthy animals in specific study populations, using a standard method, in order to provide the most meaningful comparison data against which to monitor their health via measurement of hematological and serum biochemical measurements. In the case of many laboratory studies, blood sample collection under ketamine anesthesia would constitute the 'normative' conditions for a blood draw taken during clinical treatment and some experimental procedures. Accordingly, the data presented here serve as appropriate baseline hematological and serum biochemical reference ranges following the use of ketamine anesthesia in bonnet macaques.

In summary, it is important for clinical and experimental purposes to identify not only the reference range for a population, but also whether – and to what degree – a specific subject group may vary from those ranges. The establishment of reference ranges for healthy animals serves clinical diagnosis and treatment needs by providing comparison data against which an individual's measures can be evaluated should specific health concerns arise. The reference values generated by this study add to the existing knowledge of hemodynamic measures in non-human primates, assist the clinical care of laboratory bonnet macaques, and serve as a comparative research resource.

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Table 1

Bonnet Macaque demographics

Maturational category	Males	Females	Total	Mean age at test (in months)
Juvenile (10–23 months)	3	6	9	16.48 ± 0.53
Adolescent (24–59 months)	11	11	22	39.58 ± 0.47
Adult (128–234 months)	5	14	19	172.70 ± 1.99
Total	19	31	50	

Table 2Hematological measures in bonnet macaques (*Macaca radiata*) with age group specific reference values

Maturational category parameter	Units	Juvenile N = 9 Mean $\pm$ SD	Adolescent N = 22 Mean $\pm$ SD	Adult N = 19 Mean $\pm$ SD	All Ages N = 50 Mean $\pm$ SD
White blood cell	Thous/ul	12.2 $\pm$ 3.5	11.2 $\pm$ 4.0	8.7 $\pm$ 1.8	10.36 $\pm$ 3.5
Red blood cell	Million/ul	5.9 $\pm$ 0.2	5.9 $\pm$ 0.6	5.7 $\pm$ 0.5	5.78 $\pm$ 0.5
Hemoglobin	g/dl	13.0 $\pm$ 0.2	12.9 $\pm$ 1.4	12.7 $\pm$ 1.3	12.82 $\pm$ 1.2
Hematocrit	%	41.0 $\pm$ 0.8	40.3 $\pm$ 4.2	40.1 $\pm$ 4.3	40.34 $\pm$ 3.9
Mean corpuscular volume	fL	70.3 $\pm$ 2.8	68.4 $\pm$ 4.0	70.8 $\pm$ 3.9	69.68 $\pm$ 3.9
Mean corpuscular hemoglobin	pg	22.1 $\pm$ 0.7	21.9 $\pm$ 1.2	22.3 $\pm$ 1.4	22.07 $\pm$ 1.2
Mean corpuscular hemoglobin concentration	g/dl	31.6 $\pm$ 0.4	32.0 $\pm$ 0.7	31.5 $\pm$ 1.1	31.71 $\pm$ 0.9
Segmented neutrophils	%	50.1 $\pm$ 17.5	48.1 $\pm$ 12.3	49.7 $\pm$ 11.5	49.06 $\pm$ 12.7
Lymphocytes	%	46.1 $\pm$ 17.0	46.6 $\pm$ 12.0	43.0 $\pm$ 10.6	45.1 $\pm$ 12.3
Monocytes	%	2.8 $\pm$ 0.9	4.9 $\pm$ 2.1	5.4 $\pm$ 2.0	4.73 $\pm$ 2.1
Eosinophils	%	1.0 $\pm$ 0.8	0.4 $\pm$ 0.5	1.9 $\pm$ 1.0	1.1 $\pm$ 1.0
Basophils	%	0.0 $\pm$ 0.0	0.0 $\pm$ 0.0	0.0 $\pm$ 0.0	0 $\pm$ 0.0
Platelet count	Thous/ul	312.1 $\pm$ 48.2	284.2 $\pm$ 83	377.4 $\pm$ 131	326 $\pm$ 108

Table 3Serum biochemical measures in bonnet macaques (*Macaca radiata*) with age group specific reference values

Maturational category parameter	Units	Juvenile N = 9 Mean $\pm$ SD	Adolescent N = 22 Mean $\pm$ SD	Adult N = 19 Mean $\pm$ SD	All Ages N = 50 Mean $\pm$ SD
Alkaline phosphatase	U/l	936.2 $\pm$ 398	750.8 $\pm$ 257	133.6 $\pm$ 67.8	549.6 $\pm$ 411
Alanine aminotransferase	U/l	21.1 $\pm$ 4.0	21.9 $\pm$ 4.2	24.9 $\pm$ 13.2	22.9 $\pm$ 8.8
Aspartate aminotransferase	U/l	30.6 $\pm$ 14.6	30.3 $\pm$ 14.0	21.3 $\pm$ 11.4	26.9 $\pm$ 13.6
Creatine kinase	U/l	855.6 $\pm$ 594	712.7 $\pm$ 398	669.4 $\pm$ 490	722.1 $\pm$ 468
Gamma glutamyl transferase	U/l	75.9 $\pm$ 35.8	71.1 $\pm$ 30.9	42.5 $\pm$ 12.6	61.1 $\pm$ 29.9
Amylase	U/l	271.8 $\pm$ 121	343.7 $\pm$ 83.3	248.6 $\pm$ 50.3	294.1 $\pm$ 90.1
Lipase	U/l	29.6 $\pm$ 19.6	23.4 $\pm$ 13.4	17.1 $\pm$ 8.0	22.0 $\pm$ 13.3
Albumin	g/dl	4.2 $\pm$ 0.2	4.2 $\pm$ 0.2	3.8 $\pm$ 0.3	4.0 $\pm$ 0.3
Total protein	g/dl	6.7 $\pm$ 0.2	7.1 $\pm$ 0.3	7.4 $\pm$ 0.5	7.1 $\pm$ 0.4
Globulin	g/dl	2.5 $\pm$ 0.2	2.9 $\pm$ 0.2	3.6 $\pm$ 0.5	3.1 $\pm$ 0.5
Total bilirubin	mg/dl	0.2 $\pm$ 0.1	0.2 $\pm$ 0.1	0.1 $\pm$ 0.1	0.2 $\pm$ 0.1
Direct bilirubin	mg/dl	0.1 $\pm$ 0.0	0.1 $\pm$ 0.0	0.1 $\pm$ 0.0	0.1 $\pm$ 0.1
Indirect bilirubin	mg/dl	0.1 $\pm$ 0.0	0.1 $\pm$ 0.1	0.1 $\pm$ 0.1	0.1 $\pm$ 0.1
Blood urea nitrogen	mg/dl	19.7 $\pm$ 2.0	17.0 $\pm$ 4.6	14.5 $\pm$ 3.2	16.5 $\pm$ 4.1
Creatinine	mg/dl	0.7 $\pm$ 0.1	0.8 $\pm$ 0.1	1.0 $\pm$ 0.2	0.9 $\pm$ 0.2
Glucose	mg/dl	66.1 $\pm$ 9.0	50.8 $\pm$ 5.8	67.9 $\pm$ 14.7	60.1 $\pm$ 13.3
Calcium	mg/dl	9.4 $\pm$ 0.3	9.4 $\pm$ 0.4	9.1 $\pm$ 0.4	9.3 $\pm$ 0.4
Phosphorus	mg/dl	4.7 $\pm$ 0.8	4.1 $\pm$ 1.2	3.4 $\pm$ 0.8	3.9 $\pm$ 1.1
Total CO ₂	mEq/l	21.3 $\pm$ 3.8	24.2 $\pm$ 1.2	24.0 $\pm$ 1.9	23.6 $\pm$ 2.3
Chloride	mEq/l	108.8 $\pm$ 2.0	111.4 $\pm$ 1.7	110.5 $\pm$ 1.6	110.6 $\pm$ 1.9
Potassium	mEq/l	4.3 $\pm$ 0.5	4.2 $\pm$ 0.3	4.5 $\pm$ 0.5	4.3 $\pm$ 0.5
Sodium	mEq/l	146.2 $\pm$ 2.2	149.5 $\pm$ 1.9	146.9 $\pm$ 1.9	147.9 $\pm$ 2.4
Anion gap	mEq/l	20.3 $\pm$ 3.6	18.1 $\pm$ 1.8	17.0 $\pm$ 2.0	18.1 $\pm$ 2.5

Table 4

Summary of findings on age and sex differences

Parameter	Finding
Hematology	
Platelet count	(Adults > Adolescent) = Juvenile
Red blood cell	Males > Females
Serum biochemistry	
Globulin	Adults > Adolescents > Juvenile
Total protein	Adults > Adolescents > Juvenile
Albumin	Adults < (Adolescent = Juvenile)
CREAT	Adults > (Adolescent = Juvenile)
Alkaline phosphatase	Adults > Adolescents >> Juvenile
Phosphorus	Adults < (Adolescent = Juvenile)
Glucose	Adults > Adolescents < Juvenile
CREAT	Males > Females

>, significantly greater; <, significantly less; =, no difference; >>, marginally greater.

CREAT, creatinine.