Final Report

<u>Title of Project :</u>

Some Biochemical and Hematological Indices in Different Breeds of Camels in Saudi Arabia .

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Table of Contents

Item :

- 1. Introduction
- 2. Literature review
- 3. Materials and Methods
 - a. Animals
 - b. Collection of blood samples
 - c. Determination of hematological parameters
 - d. Determination of biochemical parameters
 - e. Statistical methods
- 4. Results
- 5. Discussion
- 6. References

<u>1. Introduction</u>

Information on the normal hematological and biochemical values in indigenous camels is largely inadequate . Variation in the blood constituents due to breed (Grundel 1988) and sex (Majeed et al 1980, Hussein et al 1982, 1983) were reported. Such profile are useful in diagnosis of diseases. The objective of this study is to investigate the following :

- 1. Comparative hematological and biochemical studies in different breeds of camels in Saudi Arabia .
- Determination of hematological and biochemical indices in male and female healthy camels.

2. Literature Reviw

Camels belong to the Sub Order Tylopoda of the Artiodactyls, which first appeared in the fossil record of the Tertiary period (about 2 million years ago). camels are adapted in evolutionary terms in such a way that they use ingested water very economically. Water is continuously recirculated in the camel's gut, from duodenum and colon via the blood stream into the fore stomach. The alimentary tract functions as a water reservoir and the rate of water turnover is low (Yagil, 1985). The water turnover is cut down by reducing metabolism, renal loss and changes in erythrocyte shape, the glomerular filtration rate and renal plasma flow are 2-4 times higher in sheep than in camels. Furthermore, the nephron in the camel is twice as long as in cows or goats (for review see Al-Dughaym et al. 1998). These features may account for major differences in physiological and biochemical characteristics between camels and other species (Homeida et al. 1981). These features may account for major differences in physiological and biochemical characteristics between camels and other species (Homeida et al. 1981, Oukessou et al. 1992, Al-Busadah, 1998). The total number of camels in Saudi Arabia is over one million. These animals are producing meat, milk, hides and participating in racing (Sebag Al-Higin) and endurance events .

In recent years a considerable amount of research has been carried out on the blood chemistry of the camel . Much of this has taken place in India, Egypt and Sudan, and to a lesser extent in Israel . Unfortunately, many of the results appear to be contradictory, the anomalies perhaps arising from different methods of analysis and the difficulties of reproducing the same conditions in exactly the same way . Some of the differences can be explained by seasonal and nutritional factors and by the effects of sex and the rut but many anomalies are unexplained .

Dehydration is the principal factor affecting blood chemistry. Although it is known that PCV remains almost constant, what happens to total serum proteins in not established Reports from Macfarlane in Australia and other workers in India (Ghosal, Appanna and Dwaraknath, 1975) indicate a rise in total serum proteins but a Sudanese worker has recorded a lowering of total serum proteins , due to a fall in globulins while albumin rose from 40.7 to 75.5 per cent (Hassan , 1971). In these experiments the erythrocyhte (red cell) count, which is normally about 7.24 x 10° per 100 ml of serum (Banerjee , Bhattacharjee and Singh, 1962, Al-Busadah and Osman, 2001), rose slightly . This rise may be related to the longer half-life (12 days) and survival time (150 days) of red blood cells during dehydration when compared with 8 and 120 days respectively in hydrated camels (Yagil, Sod-Moriah and Meyerstein, 1974). The lengthening of survival time may contribute to the water

conservation ability. In the same series of experiments the acid-base parameters were also studied . The pH rose from 7.25 to 7.32 and pCO₂ and from 29.4 to 48.0 mm of mercury. Sodium and magnesium rose while blood pO_2 and calcium levels fell. These changes have also been interpreted to an additional mechanism to conserve body water (Yagil, Etzion and Berlyne, 1975).

Blood sugar has a fairly wide range of levels ranging from 74 to 140 mg glucose per 100 ml (Chavanne and Boue, 1950; Kumar and Banerjee, 1962; Maloiy, 1972; Yagil and Berlyne, 1977). In general it seems that blood glucose levels are higher in females than in males and slightly lower in the dry season than when feed is green. The levels are higher and more variable than those found in cattle under similar conditions. When large infusions of glucose are given to fully hydrated camels, the glucose is readily removed in the urine with only a slight increase in blood levels. Following dehydration, blood glucose levels increase considerably but excretion in the urine is reduced. Conversely insulin levels fall with dehydration but injection of glucose led to the balance being restored. These data have led to the conclusion that camels are not only acclimatized to conserving body water under desert conditions but are able to adapt very quickly to acute non-physiological stresses to prevent loss of body water.

The specific gravity of whole blood is about 1.05 in both summer and winter in India, that of serum 1.02 and of plasma 1.03. Haemoglobin values are about 11.7 g per 100 ml, red cels $5.54 - 7.2 \times 10^6$ per milliliter and white cells 11.8-18.1 x 10^3 per milliliter . None of these is affected by age or sex of the animal .

Chloride levels are generally higher in camels than in other species. They rise with maturity, but are not affected by pregnancy, lactation or health . Inorganic phosphorus, copper and iron are also higher than in other species (Ayoub, Awad and Bayyazaeed, 1960; Benerjee, Bhattacharjee and Sinjgh, 1962; Bhattacharjee and Banerjee, 1962; Ghosal, Appanna and Dwaraknath, 1973; Lakhotia , Bhargava and Mehrotra, 1964).

Haematological and biochemical analysis of blood can often provide valuable information regarding health and sickness of animals. Only limited information on serum biochemistry and haematology of one humped camel is available (Lakhotia et al 1964, Barakat & Abdel-Fattah 1971, ghosal et al 1975, Ghodslan et al 1978, Al-Ani et al 1992, rezakhani et al 1997, Osman and Al-Busadah, 2000), but in most of these studies, the number of animals used were very low and the animals were from different climatic conditions. Thus the values obtained in one country could not be taken as standard in other countries having different climate. Since the camel is an adaptable species , the standard haematological and serum biochemical values need to be determined in a number of animals in variable environmental conditions.

3. Materials and Methods :

a. Animals :

This study was conducted in the Camel Research center of King Faisal University in the city of Al-Ahsa, Eastern Region where the climate is subtropical with mild winter and hot summer (Laben 1980). Twenty healthy camels of each of Majaheem , Maghateer and Awarik breeds were used in this study. The selection of these breeds was based on the

finding that they are the most numerous and widely distributed camels in Saudi Arabia (Al-Eknah et al 1997). Each breed of Animals were housed together . Animals were fed on natural pasture . In addition each camel was offered craked barley, Berseem, hay and free supply of mineral salt licks. Water was provided *ad libitum*.

b. Collection of blood samples :

Blood samples 10ml were collected from jugular vein in two sets. One containing EDTA and the other without EDTA for serum seperation .

c. Determination of haematological parameters :

Erythorcyate sedimentation rate (ESR) was determined by westeregen sedimentation tubes. Packed cell volume (PCV) was determined by microhaematocrit method . Haemaglobin (Hb), number of red blood cells (RBC) and Leucocytes (TLC) were determined by electronic counter (Model ZB1, Coulter Electronics, Hialeah, USA) . Thin blood smears for differential TLC were air dried, fixed in double distilled methanol and stained with Giemsa. At least 200 cells were counted under light microscope. Mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC) were calculated according to formulae of Coles (1986).

d. Determination of biochemical parameters

The VETTEST 8008 biochemical analyzer (Sanofi Animal Health Ltd., England) was used to determine the serum concentration of total protein, blood urea nitrogen, creatinine and cholesterol. Alanine aminotransferase (ALT), aspartate aminotransferase (AST), lactic dehydrogenase (LDH) and Creatinine kinase (CK) were determined using specific kits.

Serum calcium, copper, potassium, sodium and zinc levels were determined by Shimadzu AA6800 Atomic Absorption Spectrophotometer.

Serum protein electrophoresis was performed on cellulose acetate plate using the EA-4 electrophoresis apparatus (Shanghai Medical Apparatus and Struments Factory, Shanghai, China) according to the method of Henry et al (1974).

Statistical analysis :

Data were analysed by one way ANOVA, using GLM procedure of SAS (Goodnight et al 1986) and Duncani's multiple range test was used to detect significant differences among means .

Results

Results of erythrocytic indices and leucocytic series are shown in Table1. Statistical analysis showed non-significant breed or sex effect (P values varied between 0.1 to 0.8). Therefore results for breed and sex for each parameter were poled and one mean for all camel is given in Table1. The lymphocytes were the predominant cells of total leucocyte count ($51.45 \pm 1.7\%$) followed by the neutrophils ($37.45 \pm 0.71\%$), few monocytes ($6.99 \pm 0.65\%$) and eosinophils ($4.86 \pm 1.22\%$) and rarely basophils ($0.82 \pm 0.19\%$) were the main feature of leucocytic series. The PCV was $25.85 \pm 1.2\%$, RBC $7.72 \pm 0.51 \times 106/\mu$ L, Hb $12.7 \pm 0.519/d$ L and ESR 8.1 ± 0.42 mm/8 hours.

Serum protein values are shown in Table2. There was no statistical differences between either breeds or sexes (P values varied between 0.1 to 0.6). Albumin was the predominant serum protein and -globulin was the predominant globulin . The A/G ratio was more than one .

Non-significant difference in blood urea nitrogen, creatinine, cholesterol, enzymes and trace elements were shown among breeds and sexes (Table 3).

Item	Majaheem Camels (20)		Maghateer Camels (20)		Awarik Camels (20)		Mean of All	Range
	Males (9)	Females (11)	Males (10)	Female (10)	Males (10)	Female (10)	camels (60)	(60)
WBC $(10^{3}/\mu L)$	20.1 ± 0.44	19.5 ± 0.32	18.9 ± 0.31	19.1 ± 0.41	20.2 ± 0.38	19.6 ± 0.33	19.6 ± 0.51	12.1 – 25.3
Lymphocytes(%)	50.3 ± 1.6	50.2 ± 1.7	49.6 ± 1.5	49.4 ± 1.3	50.1 ± 1.5	51.6 ± 1.4	50.13 ± 1.7	41.2 - 56.4
Monocytes (%)	6.87 ± 0.51	7.2 ± 0.50	6.7 ± 0.49	7.1 ± 0.43	7.2 ± 0.48	6.9 ± 0.44	6.99 ± 0.62	3 - 8
Neutrophils (%)	37.21 ± 1.8	38.1 ± 2.0	36.9 ± 1.3	37.1 ± 1.5	38.0 ± 1.4	37.5 ± 1.3	37.45 ± 0.71	20.3 - 45.1
Eosinophils (%)	5.1 ± 0.50	4.1 ± 0.51	6.19 ± 0.41	5.89 ± 0.43	4.21 ± 0.51	3.88 ± 0.33	4.86 ± 1.22	0.00 - 8.1
Basophils (%)	0.52 ± 0.15	0.49 ± 0.20	0.61 ± 0.11	0.51 ± 0.12	0.49 ± 0.13	0.52 ± 0.15	0.52 ± 0.19	0.00 - 2.1
RBC (10 ⁶ /µL)	8.12 ± 0.15	7.13 ± 0.14	8.2 ± 0.22	7.3 ± 0.21	7.9 ± 0.17	7.7 ± 0.16	7.72 ± 0.51	5.1 - 9.1
PCV (%)	25.0 ± 0.23	26.2 ± 0.21	24.9 ± 0.27	25.2 ± 0.25	27.3 ± 0.27	27.4 ± 0.26	25.85 ± 1.2	23.6 - 30.1
Hbg/dL	13.2 ± 0.15	12.6 ± 0.14	11.9 ± 0.18	12.4 ± 0.19	13.1 ± 0.20	13.2 ± 0.15	12.7 ± 0.51	9.2 - 14.1
ESR (mm/8hr)	8.90 ± 0.18	7.92 ± 0.20	8.1 ± 0.21	7.7 ± 0.20	8.8 ± 0.18	8.3 ± 0.18	8.1 ± 0.42	6.1 – 11.1
MCV(fL)	32.31 ± 0.73	30.11 ± 0.68	31.1 ±0.62	32.3 ± 0.66	33.1 ± 0.63	30.1 ± 0.71	31.46 ± 0.64	28.4 - 35.1
MCH (pg)	17.51 ± 0.38	16.90 ± 0.32	16.8 ± 0.31	15.9 ± 0.41	16.1 ± 0.41	17.1 ± 0.39	16.71 ± 0.73	11.2 ± 20.3
MCHC (g/dL)	52.8 ± 1.60	48.9 ± 1.51	47.7 ± 1.5	49.2 ± 1.4	47.9 ± 1.4	48.2 ± 1.4	49.1 ± 2.1	40.0 - 60.1

Table1. Mean (±SD) and ranges of haematological values in dromedary camels

* Number of animals in parentheses, WBC, white blood cell count; RBC, erythrocyte count; PCV, packed cell volume; Hb haemoglobin concentration; MCV, mean corpuscular volume; MCH, mean corpuscular haemoglobin; MCHC, mean corpuscular haemoglobin concentration.

Item	Majaheem Camels (20)		Maghateer Camels (20)		Awarik Camels (20)		Mean of All	Range
	Males (9)	Females (11)	Males (10)	Female (10)	Males (10)	Female (10)	camels (60)	(60)
Total protein (g/dL)	7.75 ± 0.21	7.81 ± 0.12	7.78 ± 0.20	7.68 ± 0.22	7.72 ± 0.20	7.76 ± 0.19	7.74 ± 0.68	4.9 - 10.2
Albumin (g/dL)	4.20 ± 0.12	4.31 ± 0.11	4.33 ± 0.12	4.28 ± 0.11	4.30 ± 0.10	4.29 ± 0.10	4.29 ± 0.40	3.1 - 6.2
-globulin (g/dL)	0.87 ± 0.02	0.79 ± 0.01	0.73 ± 0.03	0.68 ± 0.02	0.73 ± 0.03	0.76 ± 0.03	0.76 ± 0.08	045 - 0.96
B-gloublin (g/dl)	0.92 ± 0.06	0.93 ± 0.03	0.92 ± 0.05	0.96 ± 0.04	0.94 ± 0.05	0.93 ± 0.03	0.93 ± 0.04	0.51 – 1.1
-globulin (g/dL)	1.76 ± 0.03	1.78 ± 0.02	1.77 ± 0.03	1.76 ± 0.02	1.75 ± 0.04	1.78 ± 0.03	1.76 ± 0.03	0.76 - 2.1
Total globulin (g/dK)	3055 ± 0.16	3.50 ± 0.17	3.45 ± 0.15	3.40 ± 0.16	3.42 ± 0.16	3.47 ± 0.15	3.46 ± 0.18	1.5 - 5.4
A/G	1.18 ± 0.02	1.23 ± 0.03	1.25 ± 0.02	1.26 ± 0.03	1.25 ± 0.03	1.23 ± 0.02	1.23 ± 0.07	0.8 - 1.6

Table2. Mean (±SD) and ranges of serum protein values in dromedary camels

* Number of animals in parenthesed ; A/G , ALbumin / globulin ratio

Item	Majaheem Camels (20)		Maghateer Camels (20)		Awarik Camels (20)		Mean of All	Range
	Males (9)	Females (11)	Males (10)	Female (10)	Males (10)	Female (10)	camels (60)	(60)
BUN (mmoL/L)	5.3 ± 0.22	5.4 ± 0.23	4.9 ± 0.24	5.1 ± 0.22	4.8 ± 0.23	4.9 ± 0.21	5.06 ± 0.61	3.9 - 6.2
Creatinine (mmoL/L)	0.321 ± 0.04	0.41 ± 0.051	0.35 ± 0.012	0.39 ± 0.021	0.31 ± 0.022	0.38 ± 0.031	0.35 ± 0.051	0.160 - 0.533
Cholesterol (mmoL/L)	2.61 ± 0.11	2.45 ± 0.22	2.51 ± 0.11	2.31 ± 0.12	2.65 ± 0.13	2.55 ± 0.14	2.50 ± 0.21	1.91 – 4.2
Na (mmoL/L)	160.3 ± 16	140 ± 20	155.1 ± 19	162 ± 14	167 ± 15	146 ± 14	155.03 ± 19	100 - 190
K (mmoL/L)	4.6 ± 0.23	4.1 ± 0.22	3.9 ± 0.23	4.3 ± 0.33	4.4 ± 0.31	4.5 ± 0.34	4.3 ± 0.61	2.9 - 6.2
Ca (mg/dL)	10.1 ± 0.61	9.6 ± 0.51	11.2 ± 0.42	10.5 ± 0.51	11.2 ± 0.55	10.3 ± 0.51	10.48 ± 2.1	7.6 – 13.1
Cu (µg/L)	6.5 ± 0.22	6.2 ± 0.22	6.6 ± 0.23	5.9 ± 0.21	6.2 ± 0.24	6.4 ± 0.24	6.3 ± 1.1	4.2 - 8.5
Zn (µg/L)	4.1 ± 0.13	4.3 ± 0.16	4.4 ± 0.21	3.8 ± 0.22	3.9 ± 0.24	4.2 ± 0.25	4.11 ± 0.61	2.9 - 6.1
ALT (Iu/L)	10.2 ± 0.54	11.2 ± 0.56	13.4 ± 0.57	10.6 ± 0.58	10.1 ± 0.56	12.1 ± 0.55	11.23 ± 1.6	8.8 - 14.5
AST (Iu/L)	27.2 ± 1.3	31.2 ± 1.6	25.7 ± 2.1	30.1 ± 2.4	31.2 ± 2.1	28.5 ± 2.2	28.98 ± 3.1	24.1 - 35.1
LD (Iu/L)	250 ± 15	240 ± 4	260 ± 15	266 ± 16	255 ± 14	251 ± 15	253 ± 26.1	225 - 280
CK (Iu/L)	25.3 ± 1.6	24.9 ± 1.4	25.5 ± 2.1	25.6 ± 1.7	25.4 ± 1.8	25.8 ± 1.6	25.4 ± 1.1	29.1 - 30.3

Table3. Mean (±SD) and ranges of serum biochemical values in dromedary camels

* Number of animals in parentheses, Bun, blood urea nitrogen; Na, sodium; K, potassium; Ca, calcium; Cu, copper; Zn, zinc; ALT, alanine amino

transferase ; AST , aspartate amino transferase ; LD , Lactic dehydrogenase ; CK , creatine kinase

Discussion

The haematological values presented in this study were within the reference ranges to those reported elsewhere for the dromedary (Abdelgadir et al 1984; Mehrotra and Gupta 1997). Compared to other species like horse and ruminants camels have more RBC but Lower PCV (Schalm et al 1975). Consequently the MCHC was also higher than in any other species as the PCV is the denonominator in the formula which determines MCHC (Jain 1986). The finding that RBC count was higher and PCV value was lower in the camel compared to other species is because of the smaller elliptical cells pack tighter in the camel. The RBC count was inversely proportional to MCV, the indicator of red cell size. This is in line with the belief that the smaller the size of red cells the greater their number per unit volume of blood (Kerr 1989).

The values obtained in this study for WBC count is comparable to values reported in other studies (Lakhotia et al 1964, Soliman and Shaker 1967, AL-Ani 1992) . However the most frequent white cells are not neutophils but lymphocytes . In this study the percentage of lymphocytes was 50.13 ± 1.7 and neutrophils was 37.45 ± 0.71 . Corresponding values of lymphocyte and neutrophil counts were 29 and 58% in Iranian camels (Ghodsian et al 1978) , 45.9 and 48.11% in Turkmen camels (Rezakhari et al 1997) 50 and 36.6% , in Pakistani camels (Majeed et al 1980) and 56 and 38% , respectively in kenyian cameds (Nyang, ao et al 1997) . These differences could be due to the different breeds of camels used . This in part confirms Majeed et al (1989) findings who observed that lymphocytes and eosinophils appear together to reciprocate the neutrophils in different seasons .

Similar values of serum proteins in this study were obtained by other workers (Soliman and shaker 1967, ghodsian et al 1978, Abdo et al 1987, Mehrotra and Gupta 1989, AL-Ani et al 1992, Nyang, ao 1997). However the mean serum albumin concentration and A/G ratio were significantly higher than those in other ruminants (Sarwar 1992) being more than one. This probably makes it possible to maintain the high colloid osmotic pressure needed for storing water in blood or regulating water balance. Furthermore, it has been shown that the A/G ratio deceased by about 25% when camel was taken from semi – desert pasture to artificial feeding (Ghosal 1975).

The blood urea nitrogen (BUN), creatinine, cholesterol and enzymes were similar to the reference values for cattle (Zongping 2003) and the dromedary camels (Abdelgadir et al 1984, Wahbi et al 1984, Eldiridiri et al 1987, Bengoumi et al 1999). The exceptionally high level BUN in camels in comparison to other livestock is of interest in view of camel's ability to utilize urinary nitrogen at times of poor grazing or water deprivation.

Similar value of AST has been established by several workers (Boid et al 1980, Eldirdiri et al 1987). AST lacks organ specificity but is present in skeletal muscle, cardiac muscle and liver of large animals and pathological changes in these organs elevate the activity of AST in the blood (Kaneko 1989) . Like other animals the serum level of ALT in conjunction with other enzymes may be useful indicator for hepatic or muscular damage (Kaeneko 1980), but kerr (1989) considers ALT as non specific index for liver investigations .

The values of CK presented here was lower than values reported else where (AL-Ali et al 1988, Beaunoyer 1992, Nyang'ao et al 1997). Skeletal muscle are the richest source of serum CK. Therefore it is the most widely used serum enzyme determination in muscular disease of large animals (Kaneko 1989). Normal LD values reported here was similar to that reported by other workers (Nyang'ao et al 1997). LD is not organ – specific and may be of value in conjunction with other enzymes (Coodley 1970). The sodium and potassium concentration were similar to those obtained by Rezakhani et al (1997) in Turkmen camel but higher than those obtained by AL-Ani et al (1992) in Iraqi camels. The mean values of serum calcium in this study are in agreement with those reported by soliman and Shaker (1967), AL-Ani et al 1992 and Rezakhani et al (1997). The values of copper reported in this study are similar to those reported by Faye and Bengoumi (1997). These results confirmed the low values observed on camels in comparisons to other ruminants. It is Known that camels graze more forage trees than grasses and leaves from trees are generally richer in copper than other parts of the plant (Rutagwenda et al 1990). Zinc values are similar to those reported for camels (Berngoumi et al 1995, Faye and Bengoumi 1997), but lower than the deficiency threshold admitted to ruminants which is 7.0µg/100ml (Faye and Bengoumi 1997). Therefore it could be considered that zinc deficient threshold for camel is below 4.9µg/ml.

In Conclusion, there are few valations between the present findings and those from previous workers that may be attributed to the breed differences, nutrition, husbandry or assay methodology.

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