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Blood profile in normal one humped dromedary (Camelus dromedarius) camels in Libya. Part 2: Effect of breed variation on biochemical and haematological blood profile

Anwar M Abdalmula[1], Fahima A Alnagar[1], Amal O Buker[2], Fathia M Benashour[1], Ismail M Abograra[3], Mansur E Shmela[4]

[1]Department of Physiology, Biochemistry and Nutrition, Faculty of Veterinary Medicine, University of Tripoli, Tripoli, Libya.

[2]Department of Anatomy, Histology and Embryology, Faculty of Veterinary Medicine, University of Tripoli, Tripoli, Libya.

[3]National Centre of Animal Health, Tripoli, Libya.

[4]Department of Preventive Medicine, Genetics & Animal Breeding, Faculty of Veterinary Medicine,University of Tripoli, Tripoli, Libya.

1. Abstract

As little is known about the blood profile of camels in libya, this article is the second of a 4-part series describing the biochemical and haematological blood profile in Libyan camels. In Part 1 of these manuscripts, the overall blood biochemical and haematological mean values of camels in Libya were determined, parts 2-4 evaluates the effects of breed, gender and age respectively on these values. Blood samples were collected from three camel breeds, namely, Fakhreya, Sirtaweya and Mahari, and the levels of enzymes, metabolites, electrolytes and haematological indices were measured. The blood of the Sirtaweya breed showed (*i*) higher levels of aspartate aminotransferase (AST), albumin and Phosphorus (Ph), than the other two breeds, (*ii*) higher levels of lactate dehydrogenase (LDH), amylase (AMS) and total proteins than the Fakhreya breed and (*iii*) higher levels of glucose, triglycerides, total cholesterol, Very Low Density Lipoprotein (VLDL), Low Density Lipoprotein (LDL), Calcium (Ca), Packed Cell volume (PCV), Mean Corpuscular Volume (MCV) and Albumin/Globulin (A/G) ratio than the Mahari breed. The Fakhreya breed had (*i*) higher levels of urea, Iron (Fe), Haemoglobin (Hb), Mean Corpuscular Haemoglobin (MCH) and neutrophils number than the other two breeds, (*ii*) higher levels of glucose, A/G, LDL, Ca, PCV, MCV and monocytes number than the Mahari breed and (*iii*) higher levels of erythrocyte osmotic fragility, MCH and Mean Corpuscular Hemoglobin Concentration (MCHC) than the Sirtaweya breed. The Mahari breed had (*i*) higher levels of globulin than the other two breeds, (*ii*) higher levels of AMS than the Fakhreya breed and (*iii*) higher levels of erythrocyte osmotic fragility, Erythrocyte Sedimentation Rate (ESR), MCHC than the Sirtaweya breed. The tested blood parameters in the three Libyan breeds in this study were affected by breed variations.

Key words: Camel breeds, blood profile, biochemistry, haematology, Libya

2. Introduction

Camel is a ruminant animal being raised for food (milk and meat) production, burden, transport, agricultural work, beauty contest and pleasure as sport animals in pastoral and agro-pastoral areas (Wardea, 1998). Camels belong to the Camelidae family which include three genera; *Camelus*, *Lama* and *Vicugna* (Ramadan and Inoue-Murayama, 2017). The *Lama* and *Vicuna* genera originate from the Andes Mountains of South America and include two domestic species (lama and alpaca) and two wild species (guanaco in genus *Lama*, and vicuna in genus *Vicugna*) (Ramadan and Inoue-Murayama, 2017). The camel genus (*Camelus*) include three species; the dromedary or one- humbed camel (*camelus dromedarius*), the bactrian or two-humped camel (*camelus bactrianus*) and wild bactrian camels (*Camelus ferus*) (Mukasa-Mugerwa, 1981; Ramadan and Inoue-Murayama, 2017). The dromedary camels are inhabitant of the dry arid zones of North Africa, Ethiopia and eastern part of Asia (Wardeh, 2004). The domesticated bactrian camels live in the cold steppes and deserts in Central Asia and China while the wild bactrian camels populate the desert of Gobi (Wardeh, 2004; Ramadan and Inoue-Murayama, 2017).

Among the estimated 28 million camels worldwide, around 82 and 14 different breeds are documented for dromedary and bactrian camels respectively (FAO, 2018). The names of the camel breeds and types are usually reflect the locality or country where the animals are raised, the people who breed them or the animals colour rather than the purpose of rearing for work, riding, meat or milking breeds (Mukasa-Mugerwa, 1981).

The camel population in Libya is 62 thousands camels and camels are all dromedary (FAO, 2018). According to Wardeh (2004), the Libyan camels can be classified into four breeds; Sirtaweya, Fakhreya, Maghrebeya and Mahari breeds (Wardeh, 2004). The Sirtaweya breed is found mainly in the middle coastal zone in Libya. They are medium in size, hump is not well developed and selected females, under good feeding conditions, produce 3000-4000 kg of milk per 305 days. The Fakhreya breed is found in the southern and western areas from Benghazi and is well known for their milk production (3500 kg/year) under natural grazing conditions. The Maghrebeya breed is found in most coastal zones of the North African territories that extend from Egypt to Morocco. They are medium in size with small but pointed hump and reared for meat and milk production as

well as used for all kinds of agricultural, industrial and draft purposes. The Maghrebeya camels generally respond to feeding and might gain about 700-1000 grams per day during the first year under intensive management conditions. The Mahari breed is found in the Sahara from Mauritania in the west to the eastern boarders of Sudan in the east, and in Libya is located in the western south of Libya. They are medium in size, light in color and mainly utilized for riding, pack purpose and milk production. Bakory described two more breeds in south of Libya known as Alarabya and Altebesti (Bakory, 2012). Alarabya camels are medium in size, large head with white, light brown and gray body colour while Altebesti camels are characterized by small size, small hump with yellow and sand body colour. In addition, Bakory documented genetic as well as morphological variations among the four Mahari, Sirtaweya, Alarabya, Altebesti breeds (Bakory, 2012).

The blood profile's data that was generated in Libyan camels in the first part of this series (Abdalmula et al., 2018) was subdivided in this second part and the effect of breeds variation on the studied blood parameters was investigated and compared with similar studies performed elsewhere.

3. Materials and Methods

3.1 Animals

Camels were chosen randomly and based on their availability from three different breeds, Fakhreya, Sirtaweya and Mahari breeds, with different ages and of both sexes with a total of sixty six apparently healthy camels. Forty two camels were chosen from the Fakhreya breed, from a farm located in Tarhuna city in the north west of Libya. Thirteen camels were chosen from The Mahari breed located in Obari city in the south west of Libya. Eleven camels were chosen from the Sirtaweya breed, from a farm located in Wadi Alrabee area in the suburban area of Tripoli, in the north west of Libya. The Fakhreya and Mahari breeds were grazing naturally while the Sirtaweya breed was housed and fed on barley grain and concentrates for the purpose of meat production.

3.2 Blood collection

Blood samples were collected in the summer time of the year. Thirteen millilitre of blood were collected from the jugular vein of each camel by disposable plastic syringe and a 19G

needle. Three millilitre of blood were distributed into EDTA anti coagulant containing tubes for haematological analysis while the remained ten millilitre of blood were distributed into clean dry plain tubes for serum analysis. All blood samples were transferred on ice to laboratory at the Faculty of Veterinary Medicine, University of Tripoli, Tripoli, Libya. The blood allowed to clot and after centrifugation at 5000rpm for 15 min, the serum samples were aliquoted in dry clean Eppendorf capped tubes and stored at -80º C for later analysis.

3.3 Biochemical analysis

The serum activity of aspartate aminotransferase (AST, L-aspartate/2-oxoglutarate as a substrate), alanine aminotransferase (ALT, L-alanine/2-oxoglutarate as a substrate), lactate dehydrogenase (LDH, Pyruvate/NADH+H^+ as a substrate), alkaline phosphatase (ALP, p-nitrophenylphosphate as a substrate), gamma glutamyl transferase (GGT, Gulpa Carboxy/glycyglycine as a substrate), amylase (AMS, 2-chloro-4-nitrophenyl α-D-maltotriose as a substrate) and the concentration of glucose (glucose oxidase method, GOD-PAP), cholesterol (cholesterol oxidase method, CHOD-PAP), cholesterol-High Density Lipoprotein (HDL, cholesterol oxidase method after precipitation by phosphotungstic acid/magnesium chloride, CHOD-PAP), triglyceride (glycerol-3-phosphate oxidase method, GPO-PAP), urea (Berthelot modified method), creatinine (kinetic test without deproteinization), total protein (biuret method), albumin (bromocresol green method), calcium (Ca, O-cresolphtaleine method), inorganic phosphorus (Ph, ammonium molybdate method), magnesium (Mg, calmagite method) and iron (Fe, ferrozine method) were measured by commercial kits (Biomaghreb, Ariana, Tunisia) and the values were calculated according to the manufacturer instructions using Jenway spectrophotometer, Model 6500 (Bibby Scientific Ltd, Stone, Staffordshire, United Kingdom). Sodium (Na) and potassium (K) were measured using EasyLyte analyser that uses ion selective electrode technology. Globulin levels were calculated by subtraction of albumin content from the total protein value, cholesterol-Very Low Density Lipoprotein (VLDL) level was calculated by dividing triglyceride level on 5 while cholesterol- Low Density Lipoprotein (LDL) level was calculated by subtraction of the cholesterol-VLDL and cholesterol-HDL from the total cholesterol value.

3.4 Haematological analysis

The EDTA- anti coagulated blood was used to determine the haemoglobin concentration (Hb, g/dl), packed-cell volume (%), Fragility (% of haemolysis), Erythrocyte sedimentation rate (ESR, mm/hr), counts of red blood cells (RBC, $x10^6/mm^3$) and white blood cells (WBC, $x10^3/mm^3$). Haemoglobin concentration was determined following Sahli's method (Van Kampen and Zijlstra, 1961). Packed–cell volume was estimated by haematocrit capillary tube and centrifuged at 600 *g* for 20 minutes. Haematocrit value was read and recorded according to Schalm *et al.* (Schalm et al., 1975). Red blood cells and white blood cells were counted using haemocytometer and counted at x40 objective of phase contrast microscope according to Schalm *et al* (Schalm et al., 1975). The haematological indices mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH) and mean corpuscular haemoglobin concentration (MCHC), were calculated from the erythrocytic series values. The differential cell count was enumerated on slides with Giemsa stain and performed counting a minimum of 100 cells under a light microscope according to Schalm *et al.* (Schalm et al., 1975). Erythrocyte sedimentation rate (ESR) was determined by Westergren Method according to Bull *et al.* (Bull et al., 1993). Erythrocyte osmotic fragility was determined according to Benson and Swallen (Benson and Swallen, 1964).

3.4 Statistical analysis

Results are expressed as mean ± SEM. Data were analyzed using GraphPad Prism statistical software (version 6.0b; GraphPad Software Inc, La Jolla, CA, USA). Analysis of data between groups was performed using one-way ANOVA with Dunn's multiple comparison tests and statistical significance between groups was accepted at $p < 0.05$.

4. Results

The serum enzyme activities of ALT, AST, ALP, LDH, GGT and AMS measured in the serum of the camels involved in this study are shown in table 1. The AST activity was higher in the Sirtaweya breed than the other two breeds. LDH activity was higher in the serum of the Sirtaweya breed than the Fakhreya one. The AMS activity was higher in the serum of the Sirtaweya and Mahari breeds than the Fakhreya breed. ALT, ALP and GGT

enzymes did not show significantly different activities in the serum of the three studied camel breeds.

The mean ± SEM concentrations of glucose, total proteins, albumin, globulin, urea, creatinine, triglycerides, cholesterol and lipoproteins measured in the serum of the camels involved in this study are shown in table 2. Glucose, LDL and A/G values were significantly higher in the serum of the Sirtaweya and Fakhreya breeds than the Mahari breed. Triglycerides, total cholesterol and VLDL values were significantly higher in the serum of the Sirtaweya breed than the Mahari breed while total proteins and albumin values were significantly higher in the serum of the Sirtaweya breed than the Fakhreya breed for total proteins and higher than the Fakhreya and Mahari breeds for albumin. Globulin values were significantly higher in the serum of the Mahari breed than the Fakhreya and Sirtaweya breeds while urea values were significantly higher in the serum of the Fakhreya breed than the Mahari and Sirtaweya breeds. Creatinin and HDL did not show significant differences between the three studied breeds.

The mean ± SEM concentrations Na, K, Ph, Ca, Mg and Fe measured in the serum of the camels involved in this study are shown in table 3. Ph values were significantly higher in the serum of the Sirtaweya breed than the other two breeds while the Fe levels was significantly higher in the serum of the Fakhreya breed than the Mahari and Sirtaweya breeds. Ca level was significantly higher in the serum of the Sirtaweya and Fakhreya breeds than the Mahari breed. Na, K and Mg did not show significant difference in the serum of the studies breeds.

The mean ± SEM values of the various haematological parameters studied are shown in tables 4 and 5. The concentration of Haemoglobin and number of neutrophils were significantly higher in the blood of the Fakhreya breed than the other two breeds. The concentration of MCH and the number of monocytes were significantly higher in the blood of the Fakhreya breed than Sirtaweya and Mahari breeds respectively. PCV and MCV values were significantly higher in the blood of the Fakhreya and Sirtaweya breeds than the Mahari breed while the fragility of erythrocytes and MCHC values were significantly higher and lower respectively in the blood of the Sirtaweya breed than the other two

breeds. The ESR values were higher in the blood of the Mahari breed than the Sirtaweya breed. The counts of RBC and WBC, and the number of lymphocytes, eosinophils and basophils were not different in the blood of the three studied camel breeds.

Table 1: Mean ± S.E. of activity of ALT, AST, ALP, LDH, GGT and AMS enzymes in the serum of different camel breeds; Sirtaweya (no=11), Fakhreya (no=42) and Mahari (no=13).

Parameter	Unit	Sirtaweya	Fakhreya	Mahari
ALT	UL^{-1}	7.41±1.95a	5.39±0.79a	3.58±0.70a
AST	UL^{-1}	25.93±4.77a	9.42±1.50b	8.31 ±1.61b
ALP	UL^{-1}	3.05±1.25a	4.08±0.52a	5.86±1.40a
LDH	UL^{-1}	52.25±14.65a	22.68±4.51b	54.92±21.80ab
GGT	UL^{-1}	1.81±0.43a	1.76±0.15a	1.83±0.27a
AMS	UL^{-1}	3.52±0.90a	0.95±0.22b	2.98±0.73a

Values were analysed using one-way ANOVA with Dunn's multiple comparison tests and values with different letters in the same row are significantly different with $p \leq 0.05$.

Table 2. The Mean ± SEM concentration of glucose, total proteins, albumin, globulin, urea, creatinine, triglycerides, cholesterol and lipoproteins the serum of different camel breeds; Sirtaweya (no=11), Fakhreya (no=42) and Mahari (no=13).

Parameter	Unit	Sirtaweya	Fakhreya	Mahari
Glucose	mg dl^{-1}	138.1±4.15a	125.5±5.21a	45.28±3.08b
Total proteins	g l^{-1}	55.16±1.45a	48.87±1.09b	54.27±2.24ab
Albumin	g l^{-1}	36.64±0.60a	30.45±0.61b	25.86±1.45b
Globulin	g l^{-1}	18.52±1.30a	18.42±0.74a	28.41±2.30b
A/G	g l^{-1}	2.07±0.14a	1.81±0.12a	0.99±0.10b
Urea	mg dl^{-1}	37.39±2.72a	47.29±1.61b	35.47±2.80a
Creatinine	mg dl^{-1}	1.58±0.05a	1.48±0.03a	1.46±0.06a
Triglycerides	mg dl^{-1}	36.70±2.62a	32.86±2.60ab	23.25±1.55b
Total cholesterol	mg dl^{-1}	43.84±3.64a	37.29±2.15ab	27.16±3.06b
HDL-cholesterol	mg dl^{-1}	15.15±2.69a	14.81±1.49a	20.11±3.19a
LDL-cholesterol	mg dl^{-1}	21.35±3.46a	15.91±2.42a	2.40±4.29b
VLDL-cholesterol	mg dl^{-1}	7.34±0.52a	6.57±0.52ab	4.65±0.31b

Values were analysed using one-way ANOVA with Dunn's multiple comparison tests and values with different letters in the same row are significantly different with $p \leq 0.05$.

Table 3. The Mean ± SEM concentration of Na, K, Ph, Fe, Ca and Mg in the serum of different camel breeds; Sirtaweya (no=11), Fakhreya (no=42) and Mahari (no=13).

Parameter	Unit	Sirtaweya	Fakhreya	Mahari
Na	mmol/l	149.1±1.03a	149.5±0.67a	144.4±2.55a
K	mmol/l	5.40±0.20a	4.90±0.12a	4.87±0.23a
Ph	mg dl^{-1}	7.97±0.22a	4.42±0.26b	5.36±0.28b
Fe	mg l^{-1}	0.39±0.19a	1.08±0.14b	0.43±0.27a
Ca	mg dl^{-1}	10.08±0.30a	10.05±0.08a	9.12±0.13b
Mg	mg dl^{-1}	2.74±0.16a	2.45±0.06a	2.50±0.12a

Values were analysed using one-way ANOVA with Dunn's multiple comparison tests and values with different letters in the same row are significantly different with $p \leq 0.05$.

Table 4. Mean ± S.E. of red blood cell values in the blood of different camel breeds; Sirtaweya (no=11), Fakhreya (no=42) and Mahari (no=13).

Parameter	Unit	Sirtaweya	Fakhreya	Mahari
PCV	%	36.74±1.38a	35.62±1.15a	23.77±1.46b
Hb	g/dl	11.06±0.59a	13.44±0.27b	10.95±0.59a
Fragility	%	0.70±0.00a	0.08±0.00b	0.76±0.06b
ESR	mm/hr	20.00±0.00a	27.87±2.85ab	52.36±11.26b
RBC count	10^6/ml	12.53±0.43a	11.52±0.25a	12.06±1.64a
MCV	fL	29.62±1.36a	31.41±1.11a	21.92±1.76b
MCH	pg	8.95±0.54a	11.84±0.30b	10.31±1.03ab
MCHC	g/dl	30.72±2.29a	39.16±1.32b	47.41±3.57b

Values were analysed using one-way ANOVA with Dunn's multiple comparison tests and values with different letters in the same row are significantly different with $p \leq 0.05$.

Table 5. Mean ± S.E. of white blood cell values in the blood of different camel breeds; Sirtaweya (no=11), Fakhreya (no=42) and Mahari (no=13).

Parameter	Unit	Sirtaweya	Fakhreya	Mahari
Total WBC count	10^3/ml	9.45±0.61a	11.28±0.61a	11.29±1.50a
Lymphocytes	10^3/ml	6.41±0.39a	6.27±0.40a	8.45±1.35a
Neutrophils	10^3/ml	1.84±0.20a	3.60±0.31b	1.85±0.23a
Monocytes	10^3/ml	1.07±0.15ab	1.25±0.09a	0.85±0.17b
Eosinophils	10^3/ml	0.03±0.01a	0.03±0.00a	0.03±0.01a
Basophils	10^3/ml	0.03±0.01a	0.03±0.00a	0.03±0.01a

Values were analysed using one-way ANOVA with Dunn's multiple comparison tests and values with different letters in the same row are significantly different with $p \leq 0.05$.

5. Discussion

The overall blood profile's mean of sixty six Libyan camels was recorded in the first part of this series (Abdalmula et al., 2018). The data generated in that study was subdivided in this part to explore the effect of breed variation on the measured biochemical and haematological blood parameters in three selected Libyan breeds. The effect of gender and age are to be investigated in the following two parts of this series.

The variations in the haematology and serum biochemistry parameters observed in the various livestock animals were attributed to many genetic and non genetic factors such as the parameters of breed (Pandian et al., 2012), genotype (Chineke et al., 2006), sex (Daramola et al., 2005), age (Egbe-Nwiyi et al., 2000), physiological status (Alodan and Mashaly, 1999), environment (Vecerek et al., 2002), season (Hussain et al., 1999), nutrition (Madubuike and Ekenyem, 2006), medications (Khan et al., 1994) and management systems (Addass et al., 2012). The breed factor was reported to have an effect on the blood profile measured in various livestock animals such as horse (Lacerda et al.,

2006), cattle (Mohammed et al., 2016), sheep (Antunović et al., 2011), goat (Mohammed et al., 2016), rabbit (Chineke et al., 2006), chicken (Mohammed et al., 2016), poultry (Pandian et al., 2012).

According to the existing literature, few studies compared the blood profile among camel breeds (AL-Busadah, 2007; Aichouni et al., 2010) and both studies did not reveal any significant differences between the studied camel breeds. In the current work, the three participants' Libyan camel breeds showed significant differences in many biochemical and haematological parameters. However, with the absence of any genotype studies performed on the camels' biochemical or haematological parameters examined in this research, it was hard to attribute these differences only to the breed variation. The management systems and; the feeding system in particular, cannot be ruled out.

The blood samples were collected from the three camel breeds in the same season and climatic condition and analyzed with the same technical procedures to minimize the variation that could affect the accuracy of the obtained data. However, these camel breeds differ in their location of living, type of feeding and purpose of rearing. The Sirtaweya camels were housed and raised under an intensive management system for the production of meat and their feeding was dependant on concentrates and barely grains. The Fakhreya camels were naturally grazing in a restricted area and reared for the purpose of milk production while the Mahari camels were naturally grazing in open field areas and reared for riding and social shows as well as milk production.

The AST, ALT, ALP, GGT, LDH and AMS enzymes are considered as indicators of the health of the animal and reveal the damage to tissues or cells in the muscle, liver, skeleton and heart (Kataria and Bhatia, 1991). The studied camel breeds in this work showed significant differences only in the serum levels of AST, LDH and AMS enzymes. The Sirtaweya camels had higher levels of AST and LDH than the other two breeds while the Fakhreya camels had a lower level of AMS.

The serum of the three camel breeds in the present work did not show significant difference in the levels of Na, K and Mg. However, the Ph, Fe and Ca electrolytes showed

significantly different concentration among the camel breeds. The Sirtaweya and Fakhreya breeds had significantly high levels of Ph and Fe respectively while the Mahari breed had a low level of Ca when compared to the other two breeds. Pasture is rich with Ca and K and deficit in Ph and Fe (Underwood and Suttle, 2000). This fact was in accordance with the serum low level of Ph observed in the Fakhreya and Mahari camels which were mainly dependant on grazing in their feeding in contrast to the Sirtaweya camels which were reared in intensive management and fed on concentrates. However, the Fe level which is deficit in grass was higher in the Fakhreya serum than the Sirtaweya ones which contradict the same fact.

All of the metabolites measured in the serum of the three camel breeds showed significant differences except creatinine and HDL. The Sirtaweya breed that was raised in intensive management and fed on concentrates showed high values of glucose, total proteins, albumin, triglycerides, total cholesterol, LDL and VLDL when compared to the other breeds which were dependant on the grazing in their feeding. The management system under which animals are kept greatly affect a wide range of haematological and serum biochemical parameters (Addass et al., 2012; Onasanya et al., 2015) and the serum vitamin, protein and lipid concentration are affected by diet and nutrition (Al-Zadjali et al., 2004; Swanson et al., 2004). Although, the blood urea nitrogen concentration decrease in low dietary protein level or hepatic chronic disease and increased in the case of renal failure and body dehydration (Mishra et al., 2013), the Fakhreya and Mahari breeds showed higher urea and globulin levels respectively than the Sirtaweya breed.

The high levels of Hb, PCV, MCV, MCH showed by the Fakhreya camels was in accordance with the high level of Fe in their blood. However, the high haematological levels observed in the Fakhreya camels that were reared in semi-extensive management system were not in accordance with the findings of Olayemi et al. where West African Dwarf (WAD) sheep raised in an intensive management system showed higher level of PCV, Hb and MCV than the WAD sheep reared in an extensive management system (Olayemi et al., 2000). The high osmotic fragility observed in the Sirtaweya erythrocytes was not in consistence with another study of Olayemi et al in which the White Fulani Cattle raised in an extensive study showed higher osmotic fragility of red blood cells when

compared to the cattle raised in an intensive management system (Olayemi et al., 2000). Although the WBC counts of the three camel breeds were not different, the Fakhreya camels showed higher numbers of neutrophils and monocytes than the other two breeds. However, the high level of ESR and globulin observed in the Mahari breed might suggest a higher immunity or slight infection in this breed when compared to the other two breeds.

In conclusion, the differences in the haematological and biochemical values obtained from the three Libyan dromedary breeds in this study has proved the effect of breed variation on blood profile of camels and in accordance with findings observed in the other various livestock animals. Having said that, it is hard at this stage to attribute these blood variations between the Libyan camels to the breed factor only. Although the genetic variation between Libyan camels was documented previously (Bakory, 2012), genotypic studies regarding the haematological and biochemical indices and on a larger number of camels can highlight if the breed variations documented in this work are related to genetic and/or other reasons such as the management and nutrition system.

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