

## COMPARISON OF BIOCHEMICAL AND HEMATOLOGICAL PARAMETERS IN TWO DIFFERENT BREEDS OF HORSES (ARABIAN AND BARBE) IN ALGERIA

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### ABSTRACT

Specific reference values (in range) are needed for each animal species for the appropriate interpretation of the results obtained from serum biochemical and haematological tests. The aim of this study was to determine ranges of Haematological and Biochemical parameters of pure-bred Arabian and Barbe mares to evaluate the need for defining reference values for different breed groups.

20 clinically healthy mares from one farm (Chaouchaoua HARAS-Tiaret, Algeria) were divided into two groups. Groups 1 and 2 comprised 10 Arabian Mares and 10 Barbe Mares (age from 04 to 09 years). Glucose, urea, albumin, aspartate aminotransferase (AST), Gamma Transferase (GT), Creatinine phospho kinase (CPK), Sodium (Na), Potassium (K) concentrations were measured in the blood serum samples.

Haematological profile were determined, red blood cell parameters (MCV, MCH, MCHC), number of leucocytes and the percentage of their respective platelets kinds, There were significant Breed related differences for most of the biochemical and Haematological parameters, (Potassium) $P= 0.006^*$ ,(urea) $P= 0.000^{***}$ , (GGT), (Albumin)  $P= 0.043^*$ ,  $P= 0.041^*$ , except for: AST, CPK, RBC, Hb, Fibrinogen.

The results of the present study indicate that there is variation in the values of most of the biochemical and haematological indices of Arabian and Barbe mares in Algeria, ((Haematocrit %)  $P= 0.031^*$  (VCM  $\mu\text{m}^3$ )  $P= 0.022^*$  (MCHC g/dl)  $P= 0.027^*$  Leucocytes  $P= 0.01^*$  (poly éosi  $\text{mm}^3$ )  $P= 0.000^{***}$  (poly baso  $\text{mm}^3$ )  $P= 0.037^*$  (Lympho  $\text{mm}^3$ )  $P= 0.001^{**}$  (Platelets  $\text{M}/\text{mm}^3$ )  $P= 0.012^*$ .

The breed influences some of the resting haematological and biochemical indices in adult Mares. Barbe Mares used for reproduction have higher performances capacity and show a more beneficial adaptation for the Algerian environment compared to Pure Arabian Mares used for reproduction.

Keywords: Mares; Barbe; Arabian; haematological; biochemical.

### INTRODUCTION

Hematological parameters, such as red blood cells (RBC), mean corpuscular volume (MCV), hemoglobin concentration (Hb), packed cell volume (PCV) and total and

differential white blood cell (WBC) count, as well as biochemical parameters such as different enzyme activities, mineral concentrations and other serum substrates help clinicians to evaluate the health status of horses and diagnose infectious and some

parasitic diseases, as well as to observe a recovery period. They also provide important information about the response to therapy, strength and systemic effect of disease and the metabolic state of the individual animal or herd [1]. Some of the hematological and biochemical values are of great interest in horses subjected to standardized field exercise tests [2-3]. The hematological and serum biochemical reference values of various horse breeds may differ due to genetic factors and/or various environmental factors (e.g. nutritional quality, availability of water, parasites and climate). For instance, warm blood horse breeds have a lower number of erythrocytes, lower hemoglobin, hematocrit and blood volume than warm blood horses.

Hematological data are available for several horse breeds, such as American miniature horse [4], Andalusian horse [5], Arab horse [6], Thoroughbred horse [7], Lipizzan horse [8], Quarter horse [9], Yugoslav Trotter [10], as well as for horses in general [11]. For Croatian working horse breeds, some hematological parameters are available for Murinsulaner horses [12]. However, no data on hematological and biochemical parameters have been published for the Barbe horse breeds.

The aim of this study was to determine and compare hematological and biochemical reference values for Barbe and Arabian Mares breeds remove combined in Algerian environment, as blood samples were collected prior to formal closure of their respective studbooks and all the individuals were kept together under the same conditions. Our aim was also to compare those reference values with already known reference values for other breeds of cold blood horses, warm blood horses and horses in general. We further aimed to determine any differences associated with breeds of the studied Mares.

## **MATERIALS AND METHODS**

### **Animals**

This study was conducted in center of Chaouchaoua (HARAS) Tiaret-Algeria. A total of 20 mares clinically healthy, pure-bred Arabian and Barbe kept by the HARAS were used to determine the effect of the Breed of the animal on the normal ranges of serum biochemical and Hematological parameters.

The farms selected were representative samples of the farms located in the Tiaret region, Algeria. The region is one of the coldest regions in Algeria with an average daily high temperature of only 23 degrees centigrade. Several months of the year it is warm to hot at temperatures continuously above 25 degrees centigrade, sometimes up to 37 degrees. Animals used for the study were served appropriate nutritional regimen and good health status achieved before the sampling.

All horses were healthy and showed no sign of abnormality during the study period. Before the start of the trial, All mares that were reproductively active and pregnant in their late gestational stage were excluded from the study.

### **Blood Collection**

Blood samples were collected from the animals between February to March/May 2016, 15 Days post Foaling and these were handled with care to minimize stress-induced effects. To reduce circadian variations, all samples were collected between 8:00 AM and 10:00 AM, before the animals were fed. The animals were fed twice a day with hay and oats. They did not receive any mineral supplements in their diet. Drinking water was given ad libitum. None of the animals performed physical

exercise before blood samples were collected. All samples were collected through the jugular vein using 18/19 gauge needle of each animal into venipuncture (Improvacuter®, evacuated blood collection tube for in vitro diagnostic use) anticoagulant used should contain one not lithium, citrate and EDTA. Blood sample bottle should be used to collect whole blood for haematological . The serum was separated by centrifugation at 3.500 rpm for 15 minutes and the aliquots were kept in 4°C and transported to laboratory for analysis.

### **Biochemical and Haematological Analyses**

#### **Biochemical parameters**

Concentrations of serum Glucose, blood urea nitrogen (BUN), albumin, aspartate aminotransferase (AST), Gamma Transferase (GT), Creatinine phospho kinase (CPK), Sodium (Na), Potassium (K) were measured by commercial kits (Biolabo SA, France) using a biochemical autoanalyser (AIRONE 200, Medisis Medical Systems Ltd., Italy), known to be appropriate for veterinary use.

#### **Haematological parameters**

Haematological indices were established by the use of haematology analyser (Automatic) with the use of the haematology analyser ADVIA 2120 (Siemens AG, Munich, Germany): red blood cell count (RBC), haematocrit (HCT), haemoglobin concentration (HGB), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), number of platelets (PLT), white blood cell count (WBC) and the percentage of particular kinds of WBC in the total number of leucocytes.

### **Statistical Analysis**

Statistical analysis of the data were performed using the SYSTAT 12 © (SYSTAT 12 © Copyright 2007) software. The results obtained were verified using ANOVA and Student's t-test for independent samples and the level of significance was set at  $P < 0.05$ . The presented results are expressed as mean and standard error of the mean (SEM, calculated from the standard deviation, using the formula  $SD = SE \cdot \sqrt{n}$ ,  $n$  being the size of the workforce per lot).

### **RESULTS**

Tables 1, 2 and present the mean values of some biochemical and Haematological parameters levels in pure-bred Arabian and Barbe mares according to Breeds. Significant variations were observed in the values of, Albumin, GGT, Leucocytes, Ht, MGV (mean corpuscular volume), MCHC, Platelets, Basophils ( $P < 0.05$ ), K, BUN, Neutrophils, Eosinophils, Lymphocytes and cortisol ( $P < .001$ ) among the two different breeds. Mean serum concentrations, and mean serum activities of AST, Na, glucose, CPK, RBC, HB, Monocytes and Fibrinogen did not differ significantly among the different breeds groups ( $P > 0.05$ ).

### **DISCUSSION**

#### **Haematological Parameters**

The haematological parameters constitute the first blood indices applied for the assessment of horse health, performance and a training degree [13], Majority of haematological indices determined in the present study were within the normal range of reference values specified for horses cited by (laboratory Vet

Agro Sup ( Lyon ), [14,15,16], which proves good health and proper condition of the tested animals. The values of RBC count, HCT value and Hb concentration determined in the current experiment for pure Arabian breed and Barbe were close to those obtained in earlier studies with no significant differences between breeds.

**Table 1. Serum biochemical metabolites concentration (mean  $\pm$  SEM) between Arabian and barbe breed mares in postpartum**

Serum biochemical metabolites	Mares bred		P value
	Barbe (n=10)	Pure Arabian breed (n=10)	
Sodium mmol/l	138.200 $\pm$ 0.742	139.800 $\pm$ 0.593	0.109 NS
Potassium mmol/l	3.688 $\pm$ 0.115	3.827 $\pm$ 0.074	0.006*
Glucose g/l	0.810 $\pm$ 0.015	0.799 $\pm$ 0.016	0.622 NS
Urea g/l	0.233 $\pm$ 0.013	0.270 $\pm$ 0.009	0.000***
AST U/l	180.300 $\pm$ 11.031	180.300 $\pm$ 10.527	0.251 NS
CPK U/l	176.900 $\pm$ 14.797	175.900 $\pm$ 10.918	0.957 NS
GGT U/l	16.200 $\pm$ 1.511	12.300 $\pm$ 0.967	0.043*
Albumine g/l	28.300 $\pm$ 0.473	29.500 $\pm$ 0.477	0.041*
Cortisol nmol/l	148.742 $\pm$ 8.717	192.230 $\pm$ 25.720	0.004 **

NS: not statistically significant, AST: aspartate aminotransferase, CPK: -creatin Phosphokinase, GGT : Gamma Glutamyl Transferase, \* P= 0.05, \*\*\* P< 0.001

**Table 2. Serum haematological metabolites concentration (mean  $\pm$  SEM) between Arabian and barbe breed mares in postpartum**

Serum hematological metabolites	Mares bred		P value
	Barbe (n=10)	Pure Arabian breed (n=10)	
Red Blood Cells M/mm <sup>3</sup>	7.453 $\pm$ 0.162	7.511 $\pm$ 0.174	0.07 NS
Haemoglobin g/dl	13.520 $\pm$ 0.263	13.550 $\pm$ 0.286	0.072 NS
Haematocrit %	34.940 $\pm$ 0.657	36.240 $\pm$ 1.172	0.031 *
VCM $\mu$ m <sup>3</sup>	46.800 $\pm$ 1.031	48.400 $\pm$ 0.897	0.022 *
MCH pg	18.170 $\pm$ 0.146	17.990 $\pm$ 0.196	0.376 NS
MCHC g/dl	38.690 $\pm$ 0.738	36.240 $\pm$ 0.196	0.027 *
Leucocytes	8.100 $\pm$ 0.407	7.400 $\pm$ 0.400	0.01 *
poly éosi mm <sup>3</sup>	123.35 $\pm$ 1.470	164.820 $\pm$ 2.457	0.000***
poly baso mm <sup>3</sup>	246.38 $\pm$ 0.873	190.560 $\pm$ 0.677	0.037 *
Lympho mm <sup>3</sup>	4 290.700 $\pm$ 2.622	3 003.78 $\pm$ 1.992	0.001 **
Mono mm <sup>3</sup>	1 084.640 $\pm$ 3.273	1 324.30 $\pm$ 4.377	0.073 NS
Platelets M/mm <sup>3</sup>	187.100 $\pm$ 7.418	156 700.00 $\pm$ 6.321	0.012 *
Fibrinogen g/l	1.849 $\pm$ 0.068	1.717 $\pm$ 0.106	0.069 NS

NS: not statistically significant, MCV-mean corpuscular volume, MCH-mean corpuscular haemoglobin, MCHC-mean corpuscular haemoglobin concentration, \* P= 0.05, \*\* P< 0.01, \*\*\* P< 0.001

Red blood cell parameters (MCV, MCH, MCHC) indicate efficiency of hemoglobin synthesis and its oxygen transport capacity. Grondin and Dewitt [17] (MCV, MCHC) revealed a significant difference between the two breeds with a p Value = 0.02, the highest value of which was recorded among Pure Arabian mares. The statistical analysis revealed a non significant difference between these with a p. Value = 0.37 of which the greatest value is recorded in Barbes mares.

The present study show significant differences in the resting WBC count and their kinds between pure Arabian mares and Barbe. All our results are included in the ranges of the usual values quoted by the laboratory Vet Agro Sup (Lyon) and definitely higher than those quoted by [18].

PLT count in both groups of animals was similar to the reference values given by Vet Agro Sup (Lyon) and definitely higher than those quoted by [18]. The fibrin that occurs during the second phase of haemostasis, comes from the cleavage of fibrinogen by thrombin. In addition, fibrinogen also plays a role in the inflammation. It is part of the proteins of the acute phase of inflammation, because by generating fibrin, it can "sequester" lesions and initiate healing processes [19].

The statistical analysis revealed a non significant difference between these with a P = 0.069, the highest value of which is recorded in Barbes mares. The fibrinogenemia recorded in our mares is similar to usual values quoted by the laboratory Vet Agro Sup (Lyon) [14,15,16].

### **Biochemical Parameters**

Serum urea and creatinine concentrations are common indices reflecting protein metabolism and efficiency

of renal functions. Moreover, creatinine is a breakdown product of muscular creatine phosphate and its level depends directly on muscle mass and its activity.

Sodium is the main cation of the extracellular compartment. It is primarily responsible for the majority of fluid exchanges across epithelial barriers. Other physiological roles of sodium are: co-factor for many metabolic reactions, co-transporter of organic molecules and diffusion of action potentials in the nervous system.

The recorded serum sodium levels in the study subjects are among the usual values and are similar to the usual reference values as reported by Vet Agro Sup (Lyon) [20,14,15] and slightly higher than those cited by [18].

Values of potassium as observed in this present study indicated  $3.68 \pm 0.115$  in Barbes mares and  $3.82 \pm 0.7$  in pure Arabian mares.

The statistical analysis revealed a very significant difference between the latter with a P = 0.006, the highest values of which are recorded in Ps-Arab mares.

The serum potassium recorded in the subjects in our study is one of the usual values and is similar to those cited by Vet Agro Sup Laboratory (Lyon) [20,21,14,18] and [19].

Glucose and lipid metabolism indices reflect energy economy and its fluctuations in a living organism. The resting level of blood glucose is fairly stable and it generally ranges from 3.1 to 6.2 mmol\*l [22].

In our experiment, the blood glucose levels recorded are among the usual values

and are similar to those cited by Vet Agro Sup (Lyon) [20,14-18].

Albumins are the major (48-76% of TP) and the most osmotically active protein fraction of a horse serum [22], Albumin is a negative protein of the acute phase of inflammation. That is, in case of inflammation, pro-inflammatory cytokines induce a decrease in albumin synthesis, so hypoalbuminemia may be present. It is uncommon in horses (about twenty percent of cases) [1].

Our values recorded in the two breeds studied, barbes and pure Arabian mares are of the order of  $28.30 \pm 0.473$  g / L and  $29.50 \pm 0.477$  g / L respectively. The statistical analysis revealed a significant difference between the latter with a  $P = 0.041$ , the highest value of which is recorded among Pure breed Arabian mares.

Albuminemia recorded in our mares is one of the usual values and is similar to those cited by Vet Agro Sup (Lyon) [20,14-18].

AST is an enzyme induced, found in high concentration in the liver and in the muscles (heart and skeletal) of all species, it is a non-specific enzyme that needs to be analyzed in combination with other variables [1]. A significant increase in activity may be due to liver or muscle pain, or hemolysis because erythrocytes contain AST.

In our experiment, the activities of these enzymes did not differ between barb and Arabian mares.

GGT: The statistical analysis revealed a significant difference between the latter with a  $P = 0.043$  whose greatest value is recorded among Barbes mares.

The serum GGT concentrations recorded in our mares are among the usual values and are similar to those cited by the Vet Agro Sup (Lyon) laboratory [14-18], and well below the values cited by Cornell University [20].

CPK: The statistical analysis revealed a non significant difference between the latter with a  $P = 0,957$  of which the greatest value is recorded in Barbes mares. The serum CPK levels recorded in our mares are among the usual values and are similar to those cited by Vet Agro Sup (Lyon) [20,14-18].

## CONCLUSION

In conclusion Analyzes of some biochemical and hematological parameters and their comparisons between the two experimental breed showed significant differences.

In terms of breed, parameters close to the usual values in the Barbes breed compared to the Arabian, which explained that Barbes mares have an adaptive power to local environmental factors that is very important compared to the Arabian mares.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

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