## PRELIMINARY BIOCHEMICAL AND HEMATOLOGICAL INVESTIGATIONS ON THE EUROPEAN BISON (*BISON BONASUS* L., 1758) FROM THE VÂNĂTORI NEAMȚ RESERVATION

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**Keywords:** European bison, hematological and biochemical indices, different ages **Abstract:** The paper discusses the preliminary results of some biochemical and hematological analyzes performed on blood samples taken over from a group of European bison coming from Sweden and brought to the Vânători Neamţ Natural Park in the year 2009. Nine usual hematological and fifteen biochemical indices have been determined, a comparison being then made between the values recorded in two and three year-old individuals, as well as between the mean values recorded in European bison and the literature data provided on free cervides (deer) and domestic bovines

(cow).

#### **INTRODUCTION**

The European bison, the biggest herbivorous mammal of Europe, belonging to the *Bovidae* family, the *Bison* genus, is a wild ruminant that once lived in the hardwood forest and silvo-steppe ecosystems of the temperate zones. The European bison is an emblematic species for the Carpathians, yet the anthropic activities and, especially, its wild hunting reduced drastically - almost up to extinction - the free European bison populations of Romania, as early as the first decades of the XIX<sup>th</sup> century.

That is why, beginning with the 50'ies of the last century, a National Program for completing the European bison populations has been launched, which involved bringing up of several animals from Poland, Ukraine, Slovakia and other European countries, installed either in captivity, in zoological parks and reservations (Hateg Silvut, Vânători Neamţ etc.) or in controlled freedom, in fenced reservations, such as those of Neagra Bucşani-Târgovişte (160 ha) and Vânători Neamţ (180 ha). In Poland and Ukraine, the free European bison populations have already attained hundreds of individuals while, in Romania, having more than 50% of the Carpathian surface, efforts are now in progress for increasing the semi-free effectives and for the reintroduction of completely free European bison representatives.

According to the Habitat Directive, the European bison is an especially important species at communitary interest, so that the strategies for European bison conservation became mandatory for all countries of the European Union.

#### MATERIALS AND METHOD

The present study analyzes the preliminary results of the investigations performed on blood samples taken over from four female European bison, brought from Sweden and introduced in the Natural Park of Vânători Neamț in July 2009. Two females were two year-old, the other two - three year-old.

Taking over of the blood samples required tranquilization of the animals, through puncturing of the jugular vein, 2 mL Vacutainer-type tubes being employed.

Laboratory investigations were performed on an ABX Micros VET ABC Hematological Analyzer and an Accent 200-type Biochemical Analyzer, the following 9 hematological indices being considered: hemoglobin - Hb (g/dL blood), packed cell volume - PCV (%), number of red blood cells - RBC (x  $10^6/\mu$ L), mean corpuscular volume - MCV ( $\mu$ m<sup>3</sup>/red blood cell), mean corpuscular hemoglobin - MCH (pg Hb/red blood cell), mean corpuscular hemoglobin concentration - MCHC (g Hb/dL red blood cell), number of leukocytes (x  $10^3/\mu$ L), lymphocytes (%) and neutrophils (%). The biochemical indices under investigation were: glucose (mg/dL), cholesterol (mg/dL), total proteins (g/dL), albumins (g/dL), calcium (mg/dL), magnesium (mg/dL), iron ( $\mu$ g/dL), phosphorous (mg/dL), urea (mg/dL), uric acid (mg/dL), are plasmatic amylase (u/L).

#### **RESULTS AND DISCUSSION**

The Romanian literature of the field makes no mention of any studies devoted to the physiology of the bison; equally, the literature of other countries provided only few data on the biochemical composition and values of the main hematological indices of wild ruminant mammals, the bison included. As a reference study, mention should be made of the monograph dedicated to the physiology of the European bison (Gill, 1999), synthesizing the investigations developed, between 1954 and 1999, by the author, on this species. Also, older studies, on the breathing properties and some blood biochemical parameters in the European bison should be also cited (Haines *et al.*, 1977; Kita *et al.*, 1990), along with those discussing the macro- and micro-elements present in the hair, blood and natural food of the European bison, and the digestibility of natural food in various seasons (Kowalczyk *et al.*, 1976; Kosla, 1993).

The results of the hematological analyses discussed in the present paper are synthesized in Table 1, while data on the biochemical content of blood plasma are listed in Table 3.

Analysis of the hematological indices here considered (Table 1) evidences an increasing tendency of these parameters with the age of the females under study. Thus, the mean value of hemoglobin in three year-old European bison females is 31.8% higher than that of the two year-old ones. In a similar manner, the PCV and the RBC mean number are 7.5 and 17.4%, respectively, higher in three year-old females, comparatively with the younger ones. Stress should be laid on the compensating decrease of the MCV mean values in three year-old females, which is 7.5% lower than the values recorded in two year-old females.

Hematological indices		Eu	ropean bi	son					
	Two year-old		Three year-old		Mean	Free deer <sup>(1)</sup>	Cow <sup>(2)</sup>	Bison / Deer (%)	Bison / Cow (%)
	M.V.	%	M.V.	%				(70)	(70)
Hemoglobin (g/dL)	11	100	14.5	131.8	12.7	15.9	11.5	79.9	110.4
PCV (%)	33.5	100	36	107.5	35	51.44	35	68	100
RBC (x 10 <sup>6</sup> /µL)	5.75	100	6.75	117.4	6.25	20.7	7.5	30.2	83.3
MCV (µm <sup>3</sup> )	58.7	100	54.3	92.5	56.5	24.78	50	228	113
MCH (pg)	19.2	100	21.8	113.5	20.5	7.67	14	26.7	146.4
MCHC (g/dL)	32.8	100	40.3	122.9	36.6	30.91	31.4	118.4	116.6
Leucocytes (x $10^3/ \mu L$ )	8	100	9.4	117.5	8.7	9.1	8	95.6	108.7
Lymphocytes (%)	23.65	100	25.6	108.2	24.65	36.63	60	67.3	41.1
Neutrophils (%)	31.3	100	65.6	209.6	48.5	57.25	30	84.7	161.7

Table1. Comparative hematological data in some ruminants

M.V. – mean values; <sup>(1)</sup> – after Gupta et al., 2007; <sup>(2)</sup> – after Duncan and Prasse, 1986.

Such modifications might be explained by the adaptative response of the breathing function in three year-old European bison females, which has to assure a higher oxygen outfit - corresponding to a heavier bodily weight - both through hemoglobin synthesis stimulation (a 31.8% increase in hemoglobin) and through an increased breathing exchange surface, by orientation of erythropoiesis towards the production of a higher number (+17.4%) of red blood cells, each with a 7.5% lower volume, which amounts to a total exchange surface larger than in the case of two year-old females.

The tendency towards an additional hematological prosperity in three year-old females, comparatively with the younger ones, is also evidenced by the other indices under analysis, regarding both the hemoglobin charge of each red blood cell and the number of total leukocytes/ $\mu$ L blood and of the ratio of lymphocytes and, especially, neutrophils. The same

tendency has been also evidenced (Feldman *et al.*, 2000) in the young deer of Virginia (*Odocoileus hemionus columbianus*), in the first 150 days after birth (Table 2).

Specificity	Age (days)								
specificity	1-9	10 - 29	30 - 89	90 - 139	> 140				
RBC (x 10 <sup>6</sup> /µL)	7.51	7.79	9.67	10.6	10.46				
Hemoglobin (g/dL)	9.33	11.32	13.45	14.04	15.43				
PCV (%)	29.7	32.2	37.4	38.7	42.1				
Leucocytes (x 10 <sup>3</sup> /µL)	5.3	5	4.5	2.8	3.6				

Table 2. Hematological changes in the young deer of Virginia (Feldman et al., 2000)

On the other hand, a comparison made among all mean values recorded in the European bison representatives and literature data on the free cervides and cows shows that, as to hemoglobin and PCV, 20-30% lower values appear in the semi-free bison, *versus* the free deer, and up to 10% lower ones, respectively, than in cows. Also, as a compensation, the MCV values are 13-128% higher in European bison than in cows and free cervides, which may be explained by the effect of the captivity condition, known as inducing chronic stress, capable of stimulating both erytropoiesis and hemoglobinsynthesis, thus assuring the breathing homeostasis. Such a phenomenon was noticed, too, in the Vancouver deer, values of 10.1 g Hb/dL in the wild groups, and of 14.6 g Hb/dL, respectively, in the captive animals being recorded by Gupta *et al.*, 2007. Also, the PCV registered a mean value of 34.5% in the free groups and of 38.7%, respectively, in the captive ones, while the number of red blood cells increased from 5.5 mil./ $\mu$ L in the free deer to 12.7 mil./ $\mu$ L in the captive ones.

As to the biochemical indices analyzed in the sanguine plasma of the European bison, the same increasing tendency of the mean values with the age of the animals may be evidenced (Table 3).

Plasma composition		Eı	iropean b	oison	Free	Cow <sup>(2)</sup>	Bison	Bison		
	Two yea	ar-old	Three y	ear-old	Mean	deer (1)	Cow	/ Deer	/ Cow	
Organic macro-components										
	M.V.	%	M.V.	%				%	%	
Glucose (mg/dL)	71.25	100	78.15	109.7	74.7	97.66	58.5	76.5	127.7	
Cholesterol (mg/dL)	133.6	100	171	128	152.3	106.81	127.5	142.6	119.5	
Proteins (g/dL)	5.44	100	6.58	120.9	6.01	6.83	7	88	85.9	
Albumins (g/dL)	3.03	100	3.39	111.9	3.21	3.87	7.2	82.9	44.6	
Ionic content										
Calcium (mg/dL)	9.43	100	7.96	84.4	8.69	10.27	9.7	84.6	89.6	
Magnesium (mg/dL)	1.65	100	3.14	190.3	2.4	-	2,35	-	102.1	
Iron (µg/dL)	273.4	100	311.8	114	292.6	-	-	-	-	
Phosphorus (mg/dL)	4	100	5.8	145	4.9	8.31	6.1	59	80.3	
Catabolits										
Urea (mg/dL)	22.1	100	27.1	122.6	24.6	53.08	16.4	46.3	150	
Uric acid (mg/dL)	0.9	100	1.47	163.3	1.19	-	-	-	-	

Table 3. Comparative biochemical data in some ruminants

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Diagram composition		Eı	ıropean b	ison	Free	Cow (2)	Bison	Bison		
Plasma composition	Two year-old		Three year-old		Mean	deer <sup>(1)</sup>	Cow	/ Deer	/ Cow	
Creatinine (mg/dL)	1.35	100	1.22	90.4	1.29	1.95	1.2	66.1	107.5	
Total bilirubine (mg/dL)	0.19	100	0.28	147.4	0.24	0.33	0.4	72.7	60	
Plasmatic enzymes										
ALAT (u/L)	46.6	100	47.5	101.9	47.1	30.38	21	155	224.3	
ASPT (u/L)	34.7	100	105.2	30.3	70	42.88	77.5	163.2	90.3	
Amylase (u/L)	12	100	10.7	89.2	11.4	-	69	-	16.5	

M.V. – mean values; <sup>(1)</sup> – after Gupta et al., 2007; <sup>(2)</sup> – after Boyd, 1984.

At the level of organic macro-compounds (glucose, cholesterol, total proteins and albumins), this increase is constant - ranging between 9.7 and 29% - as to the ionic content, with the only exception of calcium - recording values between 14-90.3% - while, in the case of catabolytes - with the exception of creatinine - the mean values in the three year-old European bison females are 22.6-63.3% higher than in younger animals. In the opinion of the authors, such modifications express a more intense physiological vigour in the three year-old females, as a result of a more intense metabolization of nutrients and of the accumulation of increased reserves of organic and mineral compounds following the increased weight and size of the animal.

In the case of European bison, the activity of some plasmatic enzymes evidences, both in the case of ALAT and ASPT, 55-63.2% higher values than in the deer. A comparison between the activities of the two enzymes in the European bison with the reference values recorded for cows shows a different behavior, namely that the ASPT mean activity is quite similar in the two species, while the ALAT activity is 2.3 higher in the bison than in the cow. Generally, a higher activity of such transferases suggests a more intense assimilation of the ketoacids resulted from polyglucide digestion and a corresponding increase of the aminoacids reserve for proteinosynthesis. Nevertheless, the activity of amylase is 8.5% lower in the European bison than in the cow.

### CONCLUSIONS

An extra hematological prosperity was registered in the three year-old female European bison, comparatively with the two year-old ones, higher values being recorded for hemoglobin (+13.5%), PCV (+22.9%), number of red blood cells (+17.5%), as well as for MCV (+13.5%), MCH (+22.9%), MCHC (+22.9%) and number of leukocytes (+17.5%).

In the three year-old European bison females, the mean MCV value is 7.5% lower than in younger females, as due to the adaptation of the breathing function through the increase of the gaseous exchange surface.

The values of hemoglobin, PCV and number of red blood cells in the European bison grown under semi-controlled conditions are 20.1-69.8% lower than in the wild deer, being comparable or about 10% higher than those recorded for cow.

The biochemical indices analyzed in the blood plasma of the bison show the same increasing tendency of the mean values with age, as follows: organic macro-compounds - with 9.7-29%, ionic content (calcium excepted) - 14-90.3% and catabolytes (creatinine excepted) - 22.6-63.3%.

The activity of some plasmatic enzymes (ALAT and ASPT) recorded in European bison evidences 55-63.2% higher values than in the deer while, comparatively with domestic ruminant animals, the ALAT activity is 2.3 higher and that of amylase - 5 times lower.

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Analele Științifice ale Universității "Alexandru Ioan Cuza", Secțiunea Genetică și Biologie Moleculară, TOM XI, 2010