

Influence of temperature and age on hemato-biochemical results in East Balkan pigs raised under extensive breeding system

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Abstract

The experiment was carried out in the Scientific Centre of Agriculture – Sredets with indigenous East Balkan pigs divided in three age groups – suckling piglets (n = 10), growers (n = 10) and sows (n = 10). Seven hematological and nine blood biochemical parameters were determined. The pigs raised under extensive breeding system – in semi-open buildings without providing the optimum temperature for the different pig categories depends on the outside temperature. The changes in hematological and blood biochemical parameters at four temperature ranges (TR) – I (up to 8 °C), II (9–15 °C), III (16–22 °C) and IV (over 23 °C) were controlled. Blood samples were taken during the months of February, May, July and October. The results were analyzed through two-way ANOVA to assess the influence of the age and temperature on the hematological profile of the animals.

The lowest monthly average values were measured in the period from December to March – 1,8 °C–6,0 °C. The next temperature interval – from 9.6 °C to 14.6 °C was observed in the months of April, October and November and optimal temperature values – from 16 °C to 20 °C, in May, June and September. The highest average monthly temperature was established during the period July–August – above 23.0 °C.

The leukocyte's number is influenced by both the temperature and the age of the pigs. For all controlled categories values were lower in II and III compared to I and IV TR, which could be explained by differences in temperature. The differences obtained for the number of lymphocytes and granulocytes are not significant and does not show any age and temperature dependence. Significant influence was found at the red blood cells number (P < 0.001), hemoglobin (P < 0.001) and hematocrit (P < 0.01), as well as in the most biochemical parameters including glucose, creatinine (P < 0.001), total protein (P < 0.001), albumin (P < 0.001), alanine aminotransferase (P < 0.001), aspartate aminotransferase (P < 0.01), triglycerides (P < 0.05) and cholesterol (P < 0.01).

Key words: pigs, East Balkan breed, hematological and blood biochemical results, age, temperature, effect

Introduction

The ability of the organism to maintain physiological balance with environmental conditions

is a key factor in maintaining the productivity of animals. The growth and development of the animals are associated with complex of biological interactions that occur as a result of oxida-

tive processes in the blood and affect its composition. In this aspect, the study of hematological and biochemical profiles is an important method for monitoring the health status of farm animals (Chemshirova, 2000).

Breeding system is one of the determining factors for reaching a certain level of metabolic processes, and an adaptation-protective mechanism. East Balkan pigs reared in extensive systems in semi-open sheds without the possibility of providing an optimal temperature regime for different age categories, and depending on the ambient temperature is the only native breed of pigs in our country, with high adaptability to extreme climatic conditions.

Although blood is characterized by relative constancy, it is still remains a dynamic system. The main factors affecting its variability include animal age (Czech et al., 2017), the way of feeding and growing (Kovalenko et al., 2013; Perevoiko and Kosilov, 2014), and environmental conditions (Mayengbam and Tolengkomba, 2015). On the other hand, by knowing the specificity of the biochemical parameters of the blood, an objective opportunity is created for assessing of the health status of the animals (Thorn et al., 2000). From a practical point of view, the study of hematological parameters makes it possible, on the one hand, to reveal the physiological and biochemical determination of the traits and on the other as an interior indicator to predict productivity (Cemshirova, 2000).

The aim of study was to follow the changes of the haematological and biochemical profile of East Balkan pigs raised under extensive breeding system depending on the age and ambient temperature.

Material and methods

The study was carried out in the Scientific Center of Agriculture – Sredets for a one-year period with three age groups ($n = 10$) of East Balkan pigs – suckling (group A), growers (group B) and sows (group C). The animals were reared extensively, in semi-open sheds with single-row boxes, serving aisle and a walk-in yard.

The sows were bred daily for grazing on natural grassland in the Strandzha Forest Fund, as the duration of grazing depending on the climatic parameters and the condition of the pastures used. Feeding was done with ground organic feeds. Growing pigs were fed with organically manufactured certified feed (barley and wheat) depending on age and live weight in the amount of 0.3 kg for pig up to 0.8 kg. The sows were fed in the evening after pasture with the same feed in the amount of 2 kg per day. The pigs had *ad libitum* access to water.

The data on average, minimum and maximum air temperatures were obtained on the basis of our own measurements and compared with the data from the meteorological station in Burgas. The results obtained were divided into four temperature ranges (TR) – I (up to 8 °C), II (9–15 °C), III (16–22 °C) and IV (above 23 °C) depending on their influence on the animal organism of different age groups.

Blood sampling was done by puncturing anterior *vena cava*, the blood was then poured into test tube containing anticoagulant. Blood samples were taken in February, May, July and October. Haematological analysis included the following parameters: WBC, number of Lymphocytes and Granulocytes, RBC, HGB, HCT and PLT, determined by using automatic hematology analyzer EXIGO. Serum biochemical analysis was performed by automatic biochemical analyzer SYNCHRON CX9 PRO and included determination of glucose, urea, TP, ALB, ASAT, ALAT, TG and cholesterol.

Data were statistically evaluated using the StatSoft 6 statistical analysis program (STATISTICA for Windows). Other Significance Tests – Difference Between Two Means were used to determine statistically significant differences.

Results and discussion

The climate of the Municipality of Sredets is typical of the Trans-Mediterranean. The average annual temperature is 11–12 °C (Figure 1). The lowest monthly average values were measured in the period from December to

March – 1.8 °C–6.0 °C. The next temperature range – from 9.6 °C to 14.6 °C was observed in the months of April, October and November, optimal temperature values – from 16 °C to 20.4 °C were measured in May, June and September. The lowest monthly average temperature was recorded in January – 1.8 °C with deviation from 11.2 °C to 9.3 °C and the highest in July – 23.1 °C, with deviation from 11.8 °C to 37.3 °C.

The results of the haematological analysis are presented in Table 1. The leukocyte count is more strongly influenced by age and less by ambient temperature. In suckling and grower pigs the values of this indicator significantly ($P \leq 0.01$) exceed those of the sows by 34.45% and 36.26%. From a physiological point of view, the number of leukocytes in young animals is increased due to the intensity of metabolic processes associated with their growth and development. The temperature was more influential in young animals. The lowest leukocyte count was measured over the III TR, followed by II and the highest in I and IV. Obviously, both the very low and the very high temperature affect almost equally the number of leukocytes. With increasing age, their numbers decrease and in older animals the values are closer.

Lymphocyte levels across different temperature ranges ranged from $7.78 \times 10^9/l$ to $10.33 \times 10^9/l$ in suckling and from $8.47 \times 10^9/l$ to $11.42 \times 10^9/l$ in growing pigs. Slightly lower are the val-

ues from $5.64 \times 10^9/l$ to $8.36 \times 10^9/l$ in sows. Increased lymphocyte values in young pigs could be explained by their function in the organism related to the immune defense and immunoglobulin synthesis. The immune memory is provided by the lymphocytes, and its role is to produce antibody synthesis upon repeated encounter with a similar antigen. The main function of lymphocytes is to promote hemostasis. They are also involved in inflammation by secreting cytokines and chemokines, molecules that the immune system uses as chemical messages to promote the interaction, communication, and behavior of cells in the immune system.

A similar trend for higher values in the young animals is found in the platelets count. In addition to their blood clotting function, platelets contain histamine and serotonin, directly related to the normal function of the digestive system.

The results obtained could be explained by the fact that animals that have completed their growth on the one hand have a lower degree of metabolic processes and on the other, with an adaptive mechanism to changes in temperature. This ascertainment is especially true for East Balkan pigs bred environmentally for thousands of generations.

Not only significant differences are observed with respect to granulocyte values, which do not show a significant effect of age and temperature on their amount.

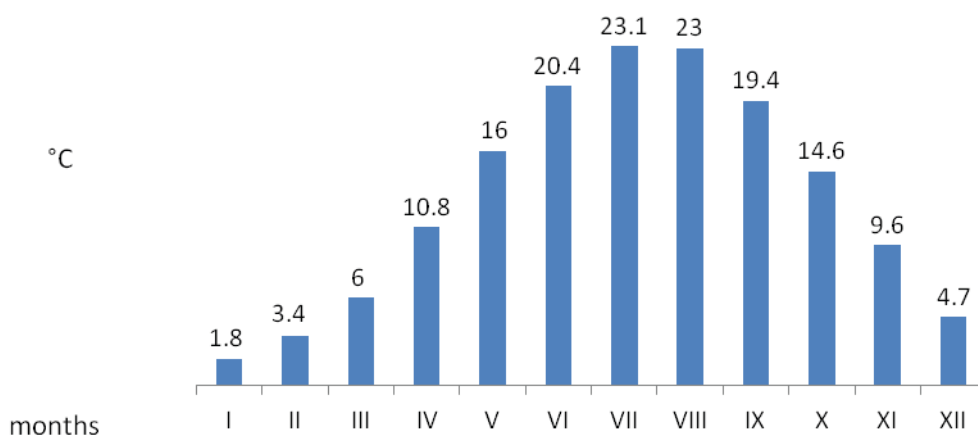


Fig. 1. Monthly average temperature over the study period, °C

Table 1. Effect of age and temperature on the hematological parameters in East Balkan pigs

Parameters	Suckling (group A) N = 10				Growing (group B) N = 10				Sows (group C) N = 10						
	Temperature ranges (TR)														
	I	II	III	IV	Signifi- cances	I	II	III	IV	Signifi- cances	I	II	III	IV	Signifi- cances
WBS, x 10 ⁹ /l	26.95	21.56	20.9	25.35		27.18	22.26	20.99	25.61		17.55	18.46	16.49	17.97	A-C* B-C*
Lymphocytes, x 10 ⁹ /l	7.78	8.8	10.33	8.47		9.0	11.42	8.53	6.79		6.84	8.36	7.02	5.64	B-C*
Granulocytes, x 10 ⁹ /l	8.12	10.37	7.23	13.42		6.77	12.2	9.96	14.52		8.04	8.25	6.8	10.23	
PLT, x 10 ⁹ /l	331.4	415.4	549.0	409.2	I-II*** I-III*** I-IV** II-III* III-IV*	301.8	402.2	484.2	316.1		254.8	187.6	222.0	339.3	I-II*** I-III*** II-IV** II-III** III-IV***
RBS, x 10 ⁹ /l	7.69	6.04	7.41	8.76	I-II** II-III** III-IV**	7.57	6.96	6.90	7.42		6.37	5.47	5.99	5.97	
HGB, g/l	133.6	113.7	128.5	144.1		120.0	136.9	123.8	127.1		136.0	127.	133.6	122.3	I-IV*
HTC, %	49.99	38.11	43.03	46.51	I-II**	43.93	33.56	41.74	41.13		53.17	35.11	42.12	39.16	I-II** I-III* I-IV** I-III*

* P < 0.05; ** P < 0.01; *** P < 0.001

Erythrocyte values are higher in young animals compared to sows. In pre-weaned pigs the temperature is strongly and significantly ($P \leq 0.01$) influenced by this indicator. In the low and high temperature ranges the values increase to $7.69 \times 10^9/l$ at I TR and $8.76 \times 10^9/l$ at IV TR, while the other two are lower. In growing and adult pigs, erythrocyte values are within physiological norms and are close to those of Nikolova et al. (2015).

The hemoglobin levels were significantly different at different TR in the three pig categories. Significant fluctuations in values are observed in suckling pigs in which they are higher in extreme I TR (133.6 g/l) and IV TR (144.1 g/l). For the other two age groups the differences were smaller. A reliable ($P \leq 0.01$) effect on hematocrit values is exerted by temperature. In all controlled categories pigs with lower values were found at II TR and III TR at which the temperature was within or close to favorable compared to I TR and IV TR, especially during the lowest temperature period (49.99% in mammals, 43.93% in young pigs and 53.17% in sows).

The results of the biochemical analysis (Table 2) show that glucose levels are lower in II TR and III TR compared to I TR and IV TR. The age of the pigs also had an effect on this indicator. The lowest values were found in sows compared to the other two categories.

Urea has higher values in suckling pigs and in I TR and IV TR. The level of urea in the blood is an indicator of protein absorption from ration. Considering the fact that for suckling pigs' main food is milk, with a high absorption rate (up to 98%), it is logical that the level of urea at this age category should be higher than the other two ones.

Creatinine values are lower in suckling and growing pigs and less affected by temperature. Considering that creatinine is a by-product of the decomposition of creatine phosphatase in muscle, the results are logical, since young animals have the lower muscle mass compared to the mature sows.

The serum total protein in the controlled pig categories is relatively high, but no differences are established, especially in sows, which does

not show a significant effect of temperature on the amount of total protein in the organism.

Albumins are a major source of protein synthesis in organs. For particular categories pigs, their levels varied from 31.1 g/l to 44.9 g/l in pre-weaned, from 33.52 g/l to 41.64 g/l in growing pigs and from 40.57 g/l to 43.31 g/l in sows and is within the limits of the physiological norm. An increased level of values was observed in the lower TR in the three controlled animal categories.

The aminotransferases ASAT and ALAT, as a part of cellular enzymes, are especially important as they make the connection between protein and carbohydrate metabolism. The enzymatic activity data of these two indicators show significantly higher values when breeding at lower temperatures. In our opinion, the differences are the result of a higher level of metabolic processes at low TR in late autumn and winter to maintaining body temperature.

Lipids are a group of organic compounds of different structure but with similar physicochemical properties including triglycerides, cholesterol, fatty acids and others. Data on the amount of triglycerides in the blood serum of pigs show that pre-weaned and growing pigs have higher levels in all TR compared to sows. In our view, these results are logical, since young animals use a higher level of nutrition, whereby part of the calories consumed with the ration are transformed into triglycerides to provide energy to the body. Higher triglyceride values in younger animals and their decrease with age are reported by Markova et al. (2018).

Significant effects of temperature and age have been found on mammalian cholesterol levels.

Conclusion

Environmental temperature and age are key factors with varying degrees of impact on the hematological and biochemical parameters of Eastern Balkan pigs raised under extensive breeding system. The WBC count is more strongly influenced by pig age and less by temperature but the ambient temperature had a stronger and more reliable effect on the RBC, HGB, HCT, which are

Table 2. Effect of age and temperature on the biochemical parameters in East Balkan pigs

Parameters	Suckling (group A) N = 10				Growing (group B) N = 10				Sows (group C) N = 10						
	I	II	III	IV	Signifi- cances	I	II	III	IV	Signifi- cances	I	II	III	IV	Signifi- cances
Glucose, mmol/l	8.646	5.390	6.28	7.419		9.574	7.428	5.63	8.01		5.650	4.694	4.30	5.9	A-C* B-C*
Urea, mmol/l	7.870	5.230	5.73	8.216		6.090	4.600	3.76	6.57		5.270	4.310	4.86	6.50	A-C*
Creatinine, µmol/l	97.100	83.400	78.50	92.350		135.200	93.400	98.4	115.1	A-B* I-II** I-III**	130.3	139.70	115.2	160.9	A-C*
TP, g/l	74.90	63.98	60.58	64.9	I-II** I-III**	78.21	71.63 b	63.34	73.87	I-III*	78.64	83.28	80.12	89.0	
ALB, g/l	44.900	31.100	36.33	39.62	I-II** I-III**	41.64	33.52	33.93	37.42	I-II** I-III**	43.31	40.57	41.33	43.17	
ASAT, u/l	99.80	35.70	64.25	94.19	I-II** II-IV**	93.50	59.60	81.89	76.7	I-II** II-III**	92.80	86.20	74.60	88.35	
ALAT, u/l	80.10	51.90	69.00	76.6	I-II** II-IV**	54.40	45.30	70.67	46.23	A-B*	61.40	67.20	65.00	76.71	A-C*
TG, mmol/l	1.272	1.271	1.178	1.192		0.933	1.088	1.007	1.117		0.775	0.934	0.99	0.832	A-C*
Cholesterol, mmol/l	4.02	3.13	3.53	3.32		3.38	2.83	4.37	3.08		2.30	2.44	2.72	2.14	A-C* B-C*

* $P < 0.05$; ** $P < 0.01$; *** $P < 0.001$

higher in the extreme temperature ranges. Different influence is also found in biochemical parameters. Age had a greater effect on triglyceride levels, whereas glucose, urea, aminotransferases ASAT and ALAT and albumin had higher values at lower temperature.

In all the indicators studied, the tendency for a less pronounced influence of the studied factors in the sows is maintained, which could be explained by the fact that the animals that completed their growth have a lower degree of metabolic processes and on the other with an adaptive mechanism for changes in ambient temperature.

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