

## EFFECT OF THE SELECTED IMMUNOSTIMULATORS ON PHYSIOLOGICAL AND PRODUCTION PARAMETERS OF SOWS

### WPLYW WYBRANYCH IMMUNOSTYMULATORÓW NA WSKAŹNIKI FIZJOLOGICZNO-PRODUKCYJNE U LOCH

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#### ABSTRACT

The aim of the study was to determine the effect of immunostimulation of sows during the perinatal period (Biostymine, Lydium-KLP) on physiological-production parameters of the sows: haematological and biochemical blood indices, colostrum and milk composition and fatty acid profile, physical (pH) and cytological (somatic cells count - SCC) parameters as well as results of reproduction and rearing of piglets. None effect of the examined immunostimulators on the most of the studied traits and indices was found. There were the changes in pH, energy level and composition of milk. Additionally, the changes in fatty acid profile in milk fat were recorded; they consisted in significantly lower or higher participation of certain fatty acids in the samples, collected from the sows which received Biostymine, as compared to the group, receiving Lydium-KLP and/or groups which did not receive any additive.

**KEY WORDS:** sows, immunostimulation, blood, colostrum, milk, fertility, rearing piglets

#### STRESZCZENIE

Celem pracy było określenie wpływu immunostymulacji loch w okresie okołoporodowym (Biostymina, Lydium-KLP) na wskaźniki fizjologiczno-produkcyjne u loch: hematologiczne i biochemiczne krwi, skład siary i mleka oraz profil wybranych kwasów tłuszczowych, parametry fizyczne (pH) i cytologiczne (liczba komórek somatycznych - LKS), a także wyniki rozrodu i odchowu prosiąt. Nie stwierdzono wpływu badanych immunostymulatorów na większość badanych cech i wskaźników. Wystąpiły zmiany w pH, energetyczności i składzie mleka. Stwierdzono ponadto zmiany profilu kwasów tłuszczowych w tłuszczu mleka polegające na istotnie niższym lub wyższym udziale niektórych kwasów tłuszczowych w próbkach pobranych od loch, którym podano Biostyminę w porównaniu do grupy otrzymującej Lydium-KLP i/lub grupy bez dodatku.

**SŁOWA KLUCZOWE:** lochy, immunostymulacja, krew, siara, mleko, płodność, odchów prosiąt

## STRESZCZENIE SZCZEGÓŁOWE

Celem pracy było określenie wpływu immunostymulacji loch (mieszanie F1 polska biała zwistoucha x wielka biała polska, wieloródki:pierwiastki ~2:1, n=28) w okresie okołoporodowym (grupy doświadczalne: Biostymina (E1) i Lydium-KLP (E2) oraz grupa kontrolna bez dodatku (C)) na wskaźniki hematologiczne (RBC, WBC, HCT, MCV, WBC, leukogram) i biochemiczne krwi (ALB, GLU, BUN, TP, ALP, TRIG, CHOL, HDLC, VLDL, LDL), skład siary i mleka (sucha masa, białko, tłuszcz, laktoza), profil wybranych kwasów tłuszczowych (C14:0, C16:0, C16:1, C18:1, C18:2, CLA, C18:3, C20:4) oraz parametry fizyczne (pH) i cytologiczne (liczba komórek somatycznych - LKS), a także wyniki rozrodu i odchowu prosiąt. Nie stwierdzono wpływu badanych immunostymulatorów na większość badanych cech i wskaźników, a jedynie korzystne zwiększenie energetyczności mleka oraz zawartości suchej masy, białka i tłuszczu w mleku loch E1 w porównaniu z E2 i C ( $P \leq 0,01$  lub  $P \leq 0,05$ ) przy jednocześnie wysoko istotnie lub istotnie podwyższonym pH mleka w grupie E1 w stosunku do grup E2 i C. Stwierdzono różnice w profilu kwasów tłuszczowych ( $P \leq 0,01$  i  $P \leq 0,05$ ), polegające na niższym udziale kwasów C14:0, C16:0, C16:1 i wyższym kwasów C18:0, C18:1 i CLA w próbkach mleka loch, którym podano Biostyminę (E1) w porównaniu z grupą E2 i/lub C. Uzyskane rezultaty powinny być zweryfikowane na większym materiale badawczym.

## INTRODUCTION

The supreme purpose of immunostimulation is to induce the antigen-independent cellular or humoral mechanisms of non-specific defence against pathogenic factors. In practice, immunostimulating agents of the 1<sup>st</sup> and 2<sup>nd</sup> generation are applied. Their health-promoting and/or production-stimulating effect was revealed [1, 2, 14]. The preparations, containing antibodies of egg yolk (IgY) [9,15] or methanol extract of aloe [13] are also successfully employed.

The purpose of the study was to determine the effect of immunostimulating the sows during the perinatal period (Biostymine, Lydium-KLP) on haematological and biochemical blood parameters, colostrum and milk composition and profile of the selected fatty acids and physical (pH) and cytological (SCC) parameters as well as on the results of reproduction and rearing of piglets.

## MATERIAL AND METHODS

The research material consisted of 28 sows of crossbreds F1 (multiparous: primiparous sows ~ 2:1) of Polish Large

White x Polish Landrace breeds and their progeny. The animals were classified into three groups. The sows from the experimental group 1 (E1; n=10 heads) received Biostymine (1 ml/head) for three successive days after parturition and those ones from experimental group 2 (E2; n=10 heads) obtained Lydium-KLP in the quantity of 0.02 mg/kg of body weight on the first day after parturition. The control sows (C group, n=8 animals) did not receive any immunostimulators.

During the duration of the experiment, the sows received the full-ration concentrate of "nursing sow" type (NS); during pregnancy, the feed was administrated twice and during lactation – three times a day, in conformity with the standards. During the first week of life, the piglets received milk substitute and from the 5<sup>th</sup> – 6<sup>th</sup> day, they were additionally ad libitum fed the mixture Prestarter I. The mixture NS contained 2270 kcal, 17% of crude protein and 1.0% of lysine in 1 kg. Prestarter I had 2416 kcal, 20% of crude protein and 1.5% of lysine.

The sows and piglets were covered with the standard prophylactic programme.

The selected reproduction traits of sows and the production traits of their progeny were controlled. The litters were weighed at birth and the piglets were individually weighed at weaning at the age of 3 weeks ( $\pm 0.1$ kg). The metabolic changes in sows were evaluated from the parameters of complete blood and serum and the functional ones of mammary gland were assessed on the ground of testing serum and milk.

Blood samples for haematological tests were collected from the sows in the presence of anticoagulant (EDTA) on the 104<sup>th</sup> day of pregnancy and on the day of piglets' weaning; the blood samples were collected from neck vein before the morning feeding. The following determinations were carried out in full blood: haemoglobin concentration (HB), number of erythrocytes (RBC) and leukocytes (WBC), haematocrite (HCT), mean volume of red cells (MCV) and number of blood plates (PLT). The determinations were carried out in haematological analyser ABACUS of Diatron company. White blood cell composition, number of neutrophilic, acidophilic and basophilic granulocytes as well as lymphocytes and monocytes was determined.

Blood samples for biochemical determinations were collected to clean test tubes and then, after formation of coagulum, they were centrifuged (10 min., 3500 r.p.m). In blood serum, the following determination were carried out: albumin (ALB), glucose (GLU), urea nitrogen (BUN), total protein (TP), alkaline phosphatase (ALP), cholesterol (CHOL), triglycerides (TRIG) and fractions of lipoproteins with great, small and a very small density (HDL, LDL, VLDL). The determinations

Table 1. Morphological blood indices of sows

Traits	Group			SE	P		
	E1	E2	C				
in the 104 day of pregnancy							
RBC, $10^{12} \cdot l^{-1}$	5.02	5.96	5.11	0.223	0.198		
HCT, $l \cdot l^{-1}$	0.30	0.34	0.31	0.009	0.184		
MCV, fl	61.20	58.00	61.00	1.183	0.480		
HGB, $mmol \cdot l^{-1}$	6.38	7.03	6.53	0.162	0.265		
WBC, $10^9 \cdot l^{-1}$	12.06	14.93	12.10	0.599	0.110		
Leukocitic, %	Granulocytes	Neutrophiles stick	0.80	1.00	0.71	0.196	0.822
		Neutrophiles segmentation	66.20 <sup>A</sup>	41.00 <sup>Aa</sup>	57.14 <sup>a</sup>	3.047	0.016
		Eosinophiles	1.60	2.83	3.43	0.549	0.417
		Basophiles	0.40	0.00	0.00	0.110	0.288
		Lymphocytes	31.40 <sup>A</sup>	55.00 <sup>Aa</sup>	38.71 <sup>a</sup>	2.911	0.016
PLT, $10^9 \cdot l^{-1}$	123.80	215.67	159.43	19.922	0.215		
at piglets weaning							
RBC, $10^{12} \cdot l^{-1}$	5.03	5.53	5.15	0.187	0.557		
HCT, $l \cdot l^{-1}$	0.29	0.31	0.30	0.009	0.774		
MCV, fl	59.80	56.80	59.25	1.060	0.518		
HGB, $mmol \cdot l^{-1}$	6.57	6.79	6.61	0.169	0.863		
WBC, $10^9 \cdot l^{-1}$	13.12	13.96	14.89	0.660	0.533		
Leukocitic, %	Granulocytes	Neutrophiles Stiuk	0.80	1.80	0.88	0.309	0.387
		Neutrophiles segmentation	52.80	57.00	56.00	2.655	0.817
		Eosinophiles	3.60	1.20	3.25	0.590	0.260
		Basophiles	0.00	0.00	0.13	0.058	0.564
		Lymphocytes	42.80	40.00	38.50	2.910	0.824
PLT, $10^9 \cdot l^{-1}$	244.60	251.00	258.88	24.244	0.968		

Mean values marked with the same capital letters AA are statistically significantly different at ( $P \leq 0.01$ ), small letters aa at ( $P \leq 0.05$ )

were performed by the so-called dry chemical method, with the application of VITROS DT 60 II System, using diagnostic kits of ICN Instruments Polska Ltd. The 50-ml samples of colostrum and bulk milk were collected in the presence of preservative (Mlekostat CC) during parturition and on the day of weaning the litter. Active acidity (pH) of colostrum and milk was determined by pH-meter pH 211 of Hanna Instrument company; the somatic cell count (SCC) was determined by Somacount 150 of Bentley company. In the samples of colostrum and milk, the basic composition, i.e. dry solids, fat, protein and lactose, was determined by IR interferometry method, using Milkoscan FT 120 of Foss Electric company.

The energy level in colostrum and milk was calculated. Fat of colostrum and milk was extracted by Röse-Gotlieb method according to PN-A-86122. Esters of fatty acids were separated by a gas chromatography; the analyses

were conducted in two repetitions and the mean result was considered. The results were statistically developed, using one-factor variance analysis, with the application of the least square method and computer programme SPSS. The numerical values of somatic cells were subject to logarithmic transformation.

## RESULTS AND DISCUSSION

Value of blood parameters is determined by species, gender, age of animals, physiological state, stress and diseases as well as diet and management conditions [6, 7, 18]. In the own studies, none differences between groups E1, E2 and C in respect of haematological and biochemical blood parameters of pregnant and nursing sows were indicated. The exception concerned quantitative differentiation of segmented granulocytes and

Table 2. Biochemical indices in sows serum

Traits	Group			SE	P
	E1	E2	C		
in the 104 day of pregnancy					
ALB, g·l <sup>-1</sup>	37.60	43.17	41.67	1.123	0.182
GLU, mmol·l <sup>-1</sup>	4.09	3.17	3.53	0.159	0.114
BUN, mmol·l <sup>-1</sup>	4.07	5.06	4.79	0.185	0.142
TP, mmol·l <sup>-1</sup>	63.80	70.00	68.11	2.225	0.568
ALP, U·l <sup>-1</sup>	53.40	53.83	54.25	4.652	0.997
TRIG, mmol·l <sup>-1</sup>	0.71	0.73	0.58	0.067	0.548
CHOL, mmol·l <sup>-1</sup>	1.75	2.13	1.93	0.084	0.258
HDLC, mmol·l <sup>-1</sup>	0.69	0.87	0.88	0.043	0.184
VLDL, mmol·l <sup>-1</sup>	0.28	0.29	0.23	0.028	0.571
LDL, mmol·l <sup>-1</sup>	0.74	0.96	0.81	0.066	0.428
CHOL/HDL	2.20	2.47	2.19	0.083	0.292
at piglets weaning					
ALB, g·l <sup>-1</sup>	40.60	40.67	40.89	0.761	0.985
GLU, mmol·l <sup>-1</sup>	2.39	3.09	2.87	0.181	0.336
BUN, mmol·l <sup>-1</sup>	3.64	3.93	4.21	0.170	0.402
TP, mmol·l <sup>-1</sup>	70.60	65.50	66.78	1.485	0.418
ALP, U·l <sup>-1</sup>	38.00	49.17	52.33	4.255	0.391
TRIG, mmol·l <sup>-1</sup>	0.45	0.32	0.33	0.023	0.084
CHOL, mmol·l <sup>-1</sup>	1.59	1.87	1.85	0.074	0.307
HDLC, mmol·l <sup>-1</sup>	0.74	0.85	0.87	0.031	0.230
VLDL, mmol·l <sup>-1</sup>	0.18	0.13	0.14	0.010	0.194
LDL, mmol·l <sup>-1</sup>	0.68	0.89	0.81	0.060	0.402
CHOL/HDL	2.18	2.23	2.07	0.079	0.678

Table 3. Basic composition and selected physical and cytological parameters of sows colostrum and milk

Traits	Group			SE	P
	E1	E2	C		
Colostrum					
Energy, kcal	1528,58	1663,76	1539,38	52,693	0,484
Total solids, %	25.52	27.21	25.57	0.693	0.502
Protein, %	15.80	16.63	16.11	0.548	0.809
Lactose, %	3.69	3.55	3.64	0.071	0.684
Fat, %	4.98	5.97	4.93	0.421	0.501
pH	6.16	6.11	6.08	0.026	0.507
SCC x 1000	1122.57	2486.20	1504.67	466.052	0.429
LN SCC	6.63	7.17	6.85	0.249	0.640
Milk					
Energy, kcal	1103,97 <sup>Aa</sup>	965,19 <sup>A</sup>	965,99 <sup>a</sup>	19,537	0,011
Total solids, %	19.06 <sup>ab</sup>	17.88 <sup>a</sup>	17.91 <sup>b</sup>	0.197	0.036
Protein, %	.3.88	4.08 <sup>a</sup>	3.59 <sup>a</sup>	0.077	0.050
Lactose, %	5.83	6.23	6.08	0.068	0.063
Fat, %	7.58 <sup>Aa</sup>	5.91 <sup>A</sup>	6.43 <sup>a</sup>	0.198	0.006
pH	6.94 <sup>A</sup>	6.81 <sup>Aa</sup>	6.92 <sup>a</sup>	0.019	0.013
SCC x 1000	4541.00	2891.60	3028.71	819.992	0.661
LN SCC	7.81	7.27	7.55	0.250	0.649

Mean values marked with the same capital letters AA are statistically significantly different at (P≤0.01), small letters aa at (P≤0.05)

Table 4. The profile of fatty acids (% of a sum) in colostrum fat and milk of sows.

Acid	Group			SE	P
	E1	E2	C		
	Colostrum				
C <sub>14:0</sub>	1.92	1.99	2.06	0.105	0.852
C <sub>16:0</sub>	25.24	25.35	25.43	0.557	0.988
C <sub>16:1</sub>	3.16	2.75	3.19	0.101	0.213
C <sub>18:0</sub>	5.57	6.11	5.27	0.188	0.250
C <sub>18:1</sub>	33.30	31.98	30.75	0.426	0.062
C <sub>18:2</sub>	14.79	16.75	16.06	0.346	0.101
CLA	0.41	0.31	0.35	0.015	0.061
C <sub>18:3</sub>	1.29	1.45	1.42	0.040	0.249
C <sub>20:4</sub>	1.06	1.19	1.08	0.022	0.094
	Milk				
C <sub>14:0</sub>	2.98 <sup>A</sup>	3.88 <sup>AB</sup>	3.22 <sup>B</sup>	0.070	0.000
C <sub>16:0</sub>	29.95 <sup>Aa</sup>	35.40 <sup>A</sup>	33.33 <sup>a</sup>	0.579	0.003
C <sub>16:1</sub>	6.97 <sup>Aa</sup>	10.95 <sup>A</sup>	9.60 <sup>a</sup>	0.421	0.003
C <sub>18:0</sub>	5.58 <sup>A</sup>	4.13 <sup>Aa</sup>	4.93 <sup>a</sup>	0.133	0.001
C <sub>18:1</sub>	36.23 <sup>AB</sup>	28.33 <sup>A</sup>	30.96 <sup>B</sup>	0.690	0.000
C <sub>18:2</sub>	8.13	8.53	8.54	0.153	0.501
CLA	0.56 <sup>Aa</sup>	0.31 <sup>A</sup>	0.41 <sup>a</sup>	0.021	0.000
C <sub>18:3</sub>	0.66	0.76	0.70	0.020	0.086
C <sub>20:4</sub>	0.49	0.46	0.46	0.014	0.606

Mean values marked with the same capital letters AA are statistically significantly different at ( $P \leq 0.01$ ), small letters aa at ( $P \leq 0.05$ )

lymphocytes in pregnant sows (Tables 1, 2). The number of red blood cells of sows varies during pregnancy and lactation periods [6]. In pregnancy, RBC, HGB and HCT are decreased what is related to mobilization of the sow's haemoglobin to blood circulatory system of fetus and to the increase of blood plasma volume [18]. The decrease is often maintained till the end of lactation [4]. Other haematological indices in own studies remained on a stable level, irrespectively of the experimental factor and physiological state what is consistent with the literature data in this respect [6, 16].

None differences in biochemical indices between the groups E1, E2 and C (Table 2) were found. The changes in activity were recorded during physiological periods (pregnancy, lactation) what stays in accordance with the literature data [6]. The optimal activity of biochemical parameters in pigs is specified in standards [17]. Their values indicate the level of metabolic changes. They are studied in growing pigs, in those ones used in reproduction and in the pigs subjected to effect of stressing agents. Physiological rise of ALP activity is recorded during pregnancy; the level of alkaline phosphatase is lowered during lactation [17] what was observed in the own studies (Table 2). The changes in metabolic profile may indicate the deficit of protein in feed and easily digestible carbohydrates and also, necrotic phenomena. As a result,

tissue metabolism may change into catabolism. Such phenomenon was not observed in the experimental animals and biochemical parameters were found within the limits of reference standards for a given species and group [6, 17]. During lactation period, the decrease of GLU, TRIG and CHOL (Table 2) as compared to pregnancy period was recorded. Žvorc et al. [18] reported on a constant level of glucose in pregnant sows. The same authors [18] and Kudlač et al. [11] showed the increase of GLU level in blood of the sows during lactation, as compared to pregnancy period. The occurring changes in glucose concentration result from physiological changes, which take place in metabolic processes at the beginning of lactation [10]. The lipid management was also discussed by other authors [8].

The level of protein in colostrum and milk of the sows (Table 3) is similar as in the data given in literature in this respect [3, 12]. It confirms the correctness of feeding the experimental animals. The energy level in milk and the content of dry solids and fat was higher in group E1 as compared to E2 and C ( $P \leq 0.01$ ;  $P \leq 0.05$ ). The statistically confirmed differences between the groups were also found for pH of milk. Values obtained for the groups corresponded to the literature data [3]. Somatic cell count in colostrum and milk did not differ significantly between the groups but it was considerably higher in

Table 5. Selected indices of sows' reproduction

Traits	Group			SE	P
	E1	E2	C		
Number of piglets born, head	12.00	11.10	10.70	0.464	0.532
Number of piglets born alive, head	11.88	11.00	10.70	0.473	0.601
Number of stillborn piglets, head	0.13	0.10	0.00	0.051	0.568
Number of weaned piglets, head	9.50	10.00	10.00	0.279	0.722
Litter weight at birth, kg	19.56	17.19	17.00	0.877	0.451
Litter weight at weaning, kg	53.15	57.70	58.10	2.323	0.653
Average piglet's weight at birth, kg	1.66	1.56	1.59	0.044	0.639
Average piglet's weight at weaning, kg	5.58	5.75	5.79	0.164	0.864
Period from weaning to oestrus*, days	5.40	5.57	5.25	0.210	0.799

\* period between weaning and efficient mating

milk as compared to colostrum what was confirmed by literature [5].

Fatty acid profile in milk of sows from E1, E2 and C groups was comparable but many differences ( $P \leq 0.01$ ;  $P \leq 0.05$ ) were recorded (Table 4). In group E1, as compared to E2 and C, the participation of acids: C14:0, C16:0 and C16:1 was lower whereas that one of acids C18:0, C18:1 and CLA was higher. The obtained results should be verified on greater material although the obtained fatty acid profile is supported by literature data on the discussed subject [12]. The results of sows' fertility were quite good and those of piglets rearing should be considered as correct (Table 5). Lack of differences between the groups suggest that the immunostimulators, employed in groups E1 and E2 did not have any effect on the controlled production traits. The subject requires further studies.

## CONCLUSIONS

None effect of Biostymine (group E1) and Lydium-KLP (E2) on the most of the studied traits and parameters was found. Only a favourable increase of energy level in milk and content of dry solids, protein and fat in milk of the sows from E1 group was recorded, as compared to E2 and C groups ( $P \leq 0.01$  or  $P \leq 0.05$ ). There was a simultaneous highly significant or significant increase of pH of milk in group E1 in relation to the remaining groups. Differences in fatty acid profile ( $P \leq 0.01$  and  $P \leq 0.05$ ) consisting in lower participation of acids C 14:0, C16:0 and C16:1 and higher participation of acids C18:0, C 18:1 and CLA in the samples of milk from the sows, fed the addition of Biostymine (E1) as compared to E2

and/or C. The obtained results should be verified on more greater research material.

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