

UDC 636.2:591.11:577.16

## IMPACT OF “OLIHOVIT” PREPARATION ON THE ANTIOXIDANT STATUS AND BIOCHEMICAL BLOOD PROFILE OF PREGNANT COWS AND CALVES, DELIVERED BY THEM

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Received on November 15, 2016

**Aim.** To investigate the mechanisms and ways of increasing the immunobiological status of the organism of productive animals at different stages of development (prepartum and early postpartum periods) using the complex vitamin-mineral preparation “Olihovit”. **Methods.** Immunological and biochemical methods of blood analysis, indices of milk and colostrum and the methods of variational statistics. **Results.** It was established that the preparation has stimulating effect on hemopoiesis and the activity of the antioxidant protection system. Positive dynamics of the increase in the body weight while using the investigated preparation was determined. **Conclusions.** Reliable increase in the average daily gain of calves was proven.

**Keywords:** vitamin-mineral preparation, antioxidant status, hematological indices.

**DOI:** 10.15407/agrisp3.03.060

### INTRODUCTION

One of the methods of improving the quality, reducing costs and preserving the competitiveness of the animal production may be found in the application of biologically active substances, decreasing the negative environmental impact [1]. It is known that the most intense growth of fetus in cows takes place during the interlactation period, when the diet is deficient in the content of vitamins, micro- and macroelements and nutrients. The course of calving, postpartum period, the viability and future productivity of newborn calves depends on the conditions of keeping within this period [2]. Additional introduction of vitamin and mineral complexes into the diet, especially during the periods of calving, is a vital prerequisite of successful calving and the development of future generations.

The elaboration of vitamin-mineral preparations requires thorough studies. Both excess and shortage of vitamins may have acute negative effect. As for micro-elements, it is important to consider their interaction

among themselves and their capability of acting as agonists or antagonists (Fig. 1).

The correction of the diet of down calvers using such deficient microelements as copper, cobalt, zinc, and manganese in different ratios facilitates the prevention of obstetric and gynecologic diseases, the increase in mating and the reduction of the service period. The main purpose of introducing the vitamin-mineral additives is the stimulation of the course of the first and second stages of the calving, the prevention of after-calving pathologies, and the increase in immune reserves, etc.

The aim of this study was to investigate the effect of the complex of vitamins and mineral substances on the immunobiological reaction of the organism of down calvers and the calves, delivered by them [3, 4].

### MATERIALS AND METHODS

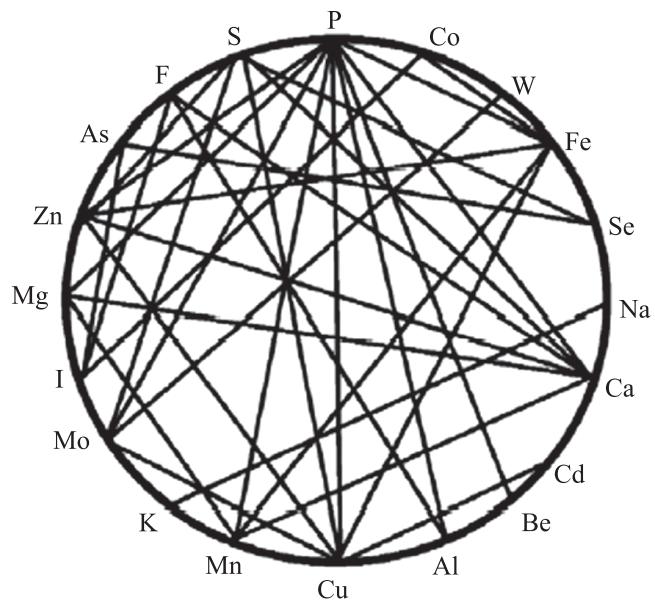
The experiment was conducted in the state enterprise, experimental farm “Obroshyne” in Pustomytyv district, Lviv region, using full-grown cows, divided into the control and experimental groups of

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5–7 animals by the principle of analogues. The diet of the animals was balanced in the main nutrients and consisted of hay, made of different herbs, silage, fodder roots and concentrated feeds. The cows in the experimental group were given intramuscular injections of the vitamin-mineral complex "Olihovit" (KELA, Belgium) in the dose of 0.5 ml per 10 kg of the body weight of the animal 30 days and 1–2 days prior to the anticipated calving, and the cows in the control group were injected the normal saline solution in the dose of 10 ml per animal in the abovementioned periods. "Olihovit" contains vitamins A, D<sub>3</sub>, PP, E, B<sub>1</sub>, B<sub>2</sub>, B<sub>4</sub>, B<sub>5</sub>, B<sub>6</sub>, B<sub>8</sub>, B<sub>12</sub>, microelements Co, Mg, Cu, Zn, Mn, and an irreplaceable aminoacid methionine.

The blood for immunological and biochemical studies was sampled from the jugular vein one month and 1–2 days prior to the anticipated calving, and also on the 8<sup>th</sup> day after calving, the blood of the calves, delivered by them, 3 h after feeding colostrum, – at the age of 8 and 30 days.

The number of erythrocytes and leukocytes was determined in heparin-stabilized blood in Goryaev chamber, the content of hemoglobin – using Drabkin's method; the total protein in blood serum – by the biuret reaction; the content of lipid hydroperoxides in blood plasma – according to Myronchyk, TBA-active products – according to Korobeinikova, glutathione peroxidase activity – by the rate of glutathione oxidation in the presence of hydroperoxide of tertiary butyl (Moin) and the content of reduced glutathione in blood erythrocytes – according to Butler.



**Fig. 1.** Interaction of microelements in the organism

During the investigations the course of calving and postpartum period of cows was observed (the control of productivity and preservation of calves).

The obtained digital data were statistically processed using Microsoft Excel software for personal computers by common methods of variation statistics with the determination of mean values ( $M$ ), their standard error ( $m$ ) and the reliability of differences, determined using Student's  $t$ -criterion.

## RESULTS AND DISCUSSION

The analysis of morphological blood indices is relevant for the estimation of the functional state of the im-

**Table 1.** Hematological indices and the content of total protein in blood serum of pregnant cows ( $M \pm m$ ;  $n = 4$ )

Index	Group	Survey period		
		30 days prior to calving	1–2 days prior to calving	8 days after calving
Erythrocytes, t/l	C	$3.50 \pm 0.17$	$4.92 \pm 0.50$	$3.39 \pm 0.15$
	E	$3.88 \pm 1.18$	$5.18 \pm 0.57$	$4.08 \pm 0.14^*$
Leukocytes, g/l	C	$6.17 \pm 0.22$	$9.38 \pm 0.39$	$6.32 \pm 0.35$
	E	$6.42 \pm 0.37$	$10.73 \pm 1.36$	$7.35 \pm 0.29^*$
Hemoglobin, g/l	C	$112.52 \pm 1.77$	$109.13 \pm 0.94$	$107.04 \pm 1.22$
	E	$110.03 \pm 2.51$	$113.30 \pm 1.27^*$	$110.52 \pm 2.27$
Total protein, g/l	C	$76.13 \pm 1.70$	$74.67 \pm 0.63$	$71.22 \pm 1.26$
	E	$76.75 \pm 1.41$	$75.61 \pm 1.07$	$74.74 \pm 1.15$

Note. In this table and the following ones the statistically reliable differences between the investigated indices for the animals of the experimental group compared to the indices for the animals of the control group are as follows: \* $p < 0.05$ ; \*\* $p < 0.01$ ; C – control; E – experimental group

mune system and the general physiological condition of the organism of animals. As seen from the presented results (Table 1), the number of erythrocytes and leukocytes in the blood of cows of the experimental group increased reliably 8 days after calving ( $p < 0.05$ ) compared to the animals of the control group. Leukocytes, also called white blood cells, reflect the total responsiveness of the organism. It should be noted that leukocytes produce lymphokines, interleukins, leukotrienes, enzymes with bactericide effect.

Feeding the cows of the experimental group with "Olihovit" preparation during the last month before calving promoted the increase in the hemoglobin level in their blood during all the periods of investigations, but the results turned out to be unreliable.

It was identified that the indices of hemoglobin and protein for calves, delivered by the cows of the experimental group, were higher than those for calves, delivered by cows of the control group (Table 2).

The introduction of protein-vitamin-mineral probiotic preparation BioDarin in the doses of 0.5, 1.0, and 1.5 kg per 100 kg of the grain mixture increased the content of erythrocytes, hemoglobin and leukocytes in the blood of animals in spring. There was an increase in total protein in blood serum of heifers in spring compared to autumn. The analysis of intergroup differences by the content of total protein in blood serum demonstrated the preference of this index for heifers, which received BioDarin. The results of the investigation testify to a higher productivity level for heifers of the experimental groups, which were fed with the additive in their diet [5].

The study of changes in the system of metabolic homeostasis of cows in the non-grazing time is relevant for the estimation of physiological state of the organ-

ism [6, 7]. An important pathogenic link in the development of the immunodeficient states of pregnant cows is the dysfunction of peroxide oxidation of lipids (POL) and, as a result, a reduction in antioxidant protection (RAP) and the inactivation of the enzymes of energy exchange. The formation of TBA-active products as a result of the breakage of polyunsaturated fatty acids, conditioned by free radicals, is also one of negative consequences of POL. Their concentration in blood serum reflects the activity of POL processes in the organism and serves as a marker of endogenous intoxication degree [8].

It was determined that the level of the final POL product in blood serum of the cows of the experimental group 1–2 days before calving and on the 8<sup>th</sup> day after calving was reliably lower than that for cows, which were not introduced "Olihovit" preparation ( $p < 0.05$ ) (Fig. 2).

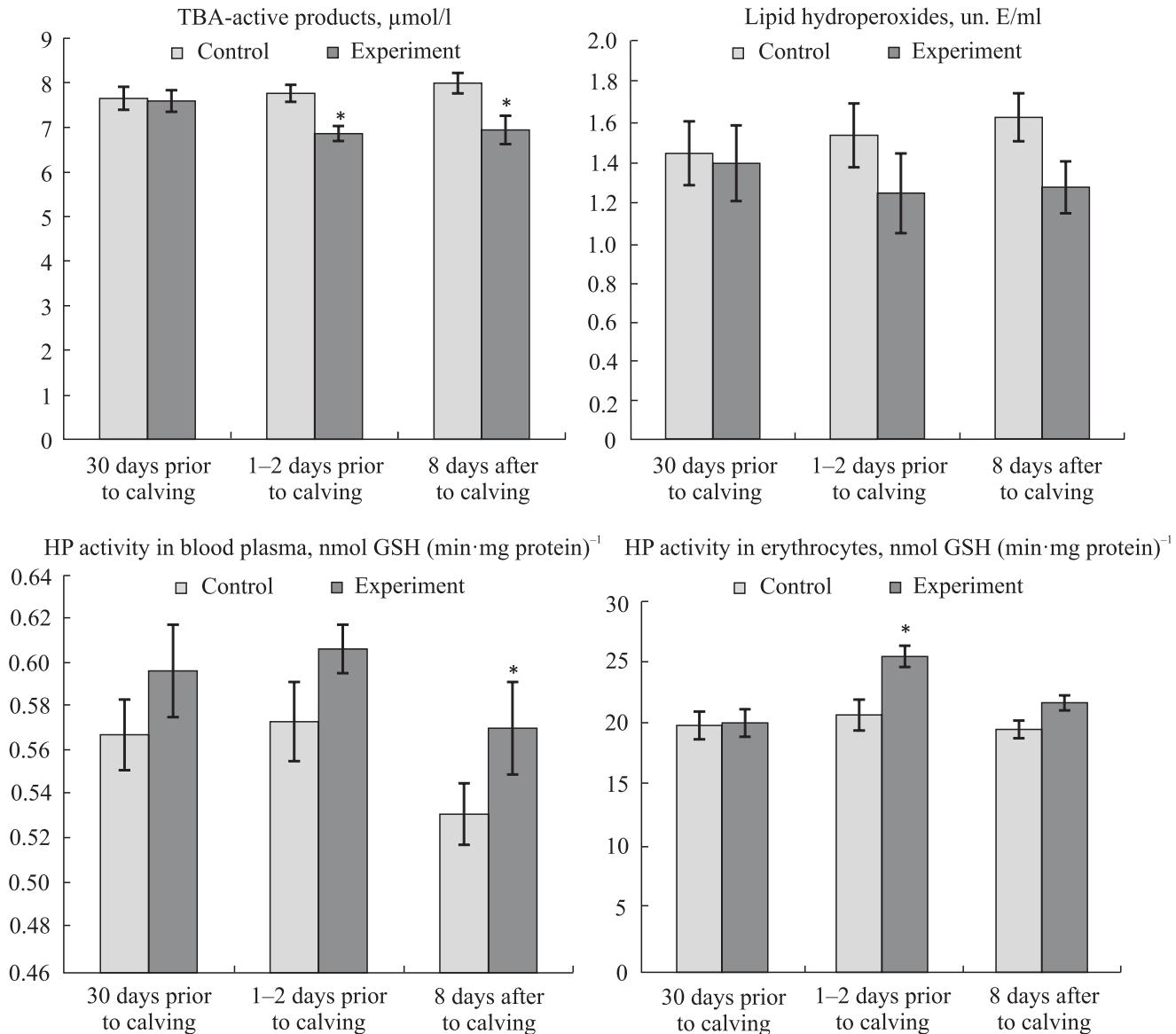
During the mentioned periods there was a tendency to the reduction in the level of the intermediate POL product – lipid hydroperoxides in the animals of the experimental group. The level of POL products is regulated by RAP enzymes. The activity of glutathione peroxidase (HP) in erythrocytes was reliably higher for cows of the experimental group 1–2 days prior to calving compared to the animals in the control group ( $p < 0.05$ ). On the 8<sup>th</sup> day after calving, this index for animals, fed with vitamin-mineral complex, was 10 % higher. The activity of HP in blood plasma and the content of reduced glutathione (one of the components of non-enzymatic RAP link) did not have any significant changes.

The active protection of young animals, calves in particular, involving natural resistance mechanisms, often fails to achieve the optimum effect due to com-

Table 2. Hematological indices and the content of total protein in blood serum of investigated calves ( $M \pm m$ ;  $n = 4$ )

Index	Group	Age of calves			
		3 h	3 days	8 days	30 days
Erythrocytes, t/l	C	4.87 ± 0.46	3.83 ± 0.14	4.08 ± 0.40	3.72 ± 0.25
	E	5.02 ± 0.52	4.63 ± 0.68	4.40 ± 0.48	4.43 ± 0.29
Leukocytes, g/l	C	7.18 ± 0.57	6.50 ± 0.71	6.07 ± 0.22	6.93 ± 0.23
	E	7.82 ± 0.58	7.32 ± 0.39	7.57 ± 0.54	7.60 ± 0.35
Hemoglobin, g/l	C	70.56 ± 0.81	73.21 ± 0.73	75.21 ± 1.05	84.42 ± 0.90
	E	72.99 ± 1.19	73.51 ± 1.59	77.21 ± 0.45	86.35 ± 1.19
Total protein, g/l	C	56.62 ± 1.09	57.34 ± 0.63	59.94 ± 0.81	64.76 ± 0.93
	E	58.39 ± 1.94	59.73 ± 0.65	62.03 ± 1.47	66.48 ± 0.98

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**Fig. 2.** The content of products of the peroxide oxidation of lipids and the indices of antioxidant protection in the blood of experimental cows ( $M \pm m; n = 4$ )

mon metabolic disorders, including the reduction in the activity of the antioxidant system [9]. Microelements are known for some pro-oxidant effect. The consumption of the mixture of  $\text{CuSO}_4$ ,  $\text{CoCl}_2$ ,  $\text{KI}$  by calves promotes the increase in the activity of the enzymatic link of RAP, in particular, there is an increase in catalase and peroxidase activity [10, 11].

The consumption of "Olihovit" preparation by down calvers facilitates further lower level of POL and higher degree of RAP in the blood of calves, delivered by them. As seen from the data in Table 3, as early as 3 h after receiving the first portion of colostrum and on the 3<sup>rd</sup> and 8<sup>th</sup> days of life, the level of TBA-active products in the calves of the experimen-

tal group was reliably smaller compared to the animals of the control group ( $p < 0.05–0.01$ ). As for lipid hydroperoxides, the reliable difference compared to the control group animals was noted on the 3<sup>rd</sup> and 8<sup>th</sup> days of life ( $p < 0.01$ ). The decrease in POL level was facilitated by the enzymatic link of RAP – the activity of HP enzyme increased reliably in erythrocytes and blood plasma of experimental group animals on the 1<sup>st</sup> and 3<sup>rd</sup> days after birth compared to the control group ( $p < 0.05–0.01$ ). HP breaks both hydrogen peroxide and peroxide compounds, formed due to the oxidation of unsaturated fatty acids, which are the fastest to be involved in the peroxidation processes by free radical derivatives of oxygen [12]. As

**Table 3.** The content of products of the peroxide oxidation of lipids and the indices of antioxidant protection in the blood of experimental cows ( $M \pm m$ ;  $n = 4$ )

Index	Group	Age of calves			
		3 h	3 days	8 days	30 days
TBA-active products, $\mu\text{mol/l}$	C	4.74 $\pm$ 0.28	4.31 $\pm$ 0.21	3.95 $\pm$ 0.19	3.65 $\pm$ 0.20
	E	3.48 $\pm$ 0.19**	3.09 $\pm$ 0.19**	3.13 $\pm$ 0.15*	3.17 $\pm$ 0.16
Lipid hydroperoxides, un. E/ml	C	0.400 $\pm$ 0.005	0.387 $\pm$ 0.006	0.359 $\pm$ 0.007	0.311 $\pm$ 0.017
	E	0.316 $\pm$ 0.017	0.298 $\pm$ 0.018**	0.291 $\pm$ 0.016**	0.268 $\pm$ 0.021
LH activity in erythrocytes, nmol GSH ( $\text{min} \cdot \text{mg protein}$ ) <sup>-1</sup>	C	35.97 $\pm$ 1.29	36.05 $\pm$ 0.96	39.27 $\pm$ 1.66	54.56 $\pm$ 2.80
	E	41.72 $\pm$ 0.94*	41.96 $\pm$ 1.24**	42.63 $\pm$ 1.30	55.63 $\pm$ 0.83
LH activity in blood plasma, nmol GSH ( $\text{min} \cdot \text{mg protein}$ ) <sup>-1</sup>	C	0.434 $\pm$ 0.012	0.453 $\pm$ 0.012	0.481 $\pm$ 0.013	0.646 $\pm$ 0.014
	E	0.487 $\pm$ 0.010*	0.493 $\pm$ 0.011*	0.492 $\pm$ 0.012	0.675 $\pm$ 0.010
Reduced glutathione, $\mu\text{mol/ml}$	C	0.58 $\pm$ 0.05	0.51 $\pm$ 0.06	1.22 $\pm$ 0.03	1.51 $\pm$ 0.06
	E	0.66 $\pm$ 0.04	0.69 $\pm$ 0.04*	1.20 $\pm$ 0.07	1.69 $\pm$ 0.06

**Table 4.** The body weight and the average daily gain of experimental calves ( $M \pm m$ ;  $n = 4$ )

Index	Group of animals	
	C	E
Body weight at birth, kg	28.08 $\pm$ 1.87	30.08 $\pm$ 1.68
Body weight at 30 days, kg	44.23 $\pm$ 1.79	51.03 $\pm$ 1.70*
Average daily gain, g	538.3 $\pm$ 28.76	698.25 $\pm$ 30.56**
Body weight at 60 days, kg	59.6 $\pm$ 2.58	68.7 $\pm$ 1.65*
Average daily gain, g	513.58 $\pm$ 52.17	589.17 $\pm$ 49.52

for non-enzymatic component of RAP, there is an increase in the level of reduced glutathione in blood erythrocytes of experimental group calves on the 3<sup>rd</sup> day of life ( $p < 0.05$ ).

Feeding the pregnant cows of the experimental group with the vitamin-mineral complex "Olihovit" 30 days and 1–2 days before calving promoted the improvement of productive traits of calves, delivered by them (Table 4). The body weight of the calves of the experimental group at the age of 30 and 60 days was reliably higher than that for the control group animals ( $p < 0.05$ ). The average daily gain of the calves, delivered by the cows, fed with "Olihovit", was higher for the whole period of studies.

Similar results were obtained by the authors of other vitamin-mineral additives [13]. At the end of the experiment the average body weight of the calves of experimental group was 5.2 kg higher than that for the control group, and amounted to  $112.2 \pm 1.18$  kg. The

average daily gain was also higher for them –  $739.5 \pm 17.75$  g against  $644.8 \pm 12.6$  g. The data were obtained with high degree of reliability ( $p \leq 0.001$ ).

## CONCLUSIONS

It was determined that the introduction of the vitamin-mineral preparation "Olihovit" promotes the increase in the number of erythrocytes and the level of hemoglobin in the blood of cows. It was established that feeding the cows with the complex of vitamins and microelements 40 days and 1–2 days before calving conditions the increase in the immunobiological responsiveness of the organism and further obtaining of calves with higher indices of natural resistance and better productivity features. It was established that calves, delivered by the cows of the experimental group, had 1.3-fold higher average daily gain, and at the age of one month their body weight was 1.15 times higher than the weight of the calves in the control group.

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### **Вплив препарату «Оліговіт» на антиоксидантний статус та біохімічний профіль крові тільних корів і отриманих від них телят**

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**Мета.** Дослідити механізми і способи підвищення імунобіологічного статусу організму продуктивних тварин на різних етапах розвитку (передотельний та ранній постнатальний періоди) за використання комплексного вітамінно-мінерального препарату «Оліговіт». **Методи.** Імунологічні і біохімічні методи дослідження крові, показників молока і молозива та методи варіаційної статистики. **Результати.** Установлено стимулювальну дію препарату на процеси кроветворення і активність системи антиоксидантного захисту. З'ясовано позитивну динаміку нарощання маси тіла при використанні досліджуваного препарату. **Висновки.** Доведено вірогідне зростання середньодобових приростів у потомства.

**Ключові слова:** вітамінно-мінеральний препарат, антиоксидантний статус, гематологічні показники.

### **Влияние препарата «Олиговит» на антиоксидантный статус и биохимический профиль крови тельных коров и полученных от них телят**

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**Цель.** Исследовать механизмы и способы повышения иммунобиологической системы организма продуктивных животных на разных этапах развития (передотельный и ранний постнатальный периоды) при использовании комплексного витаминно-минерального препарата «Олиговит». **Методы.** Иммунологические и биохимические методы исследования крови, показателей молока и молозива и методы вариационной статистики. **Результаты.** Установлено стимулирующее действие препарата на процессы кроветворения и активность системы антиоксидантной защиты. Выяснена положительная динамика нарастания массы тела при использовании исследуемого препарата. **Выводы.** Доказан достоверный рост среднесуточных приростов у потомства.

**Ключевые слова:** витаминно-минеральный препарат, антиоксидантный статус, гематологические показатели.

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