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Physiological And Biochemical Status Of Lactating Cows That Receive Antioxidants And Sorbents.

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ABSTRACT

Hydrolytic and oxidative processes are observed in feed mill during storage. They lead to accumulation of lipid oxidation products (peroxides, aldehydes, ketones, etc.) that reduce farm animals productivity by inhibiting ruminal digestion due to its toxicity to animals. Hence, antioxidants and sorbents should be introduced into farm animals diet. For this purpose, the studies on Black and white lactating cows were conducted. 4 groups (one control and three test ones) with 10 cows in each were formed. Cows in the control group were fed by the basic feeding plan, that included on-site produced feed mill, and cows from the three test groups had antioxidant epofen, sorbent toxisorb and epofen and toxisorb in combination added to the basic plan feeding. The authors established that epofen and toxisorb in combination significantly increased metabolic nitrogen by 11.2%, protein nitrogen by 22.7%, infusoria count in forestomachs by 30.1%, cellulolytic activity of rumen content by 22.5% and increase of Flavobacterium vitarumen count by 22.9% in rumen chyme content in the 3rd test group in comparison with the control. Due to this increase, cows from the 3rd test group significantly better digested dry feed by 3.2%, organic matter by 3.3%, crude protein by 3.6%, crude fiber by 4.5% and nitrogen-free extractive substances by 4.2% in comparison with the control cows. Biochemical analysis showed that cows from the 3rd test group had higher levels of serum total protein by 7.6%, albumin by 10.5% and glucose by 10.2% than in the control. Bactericidal activity parameters in 3rd test group exceeded the ones in the control by 5.16% and nonspecific resistance parameters – by 28.1%. Hence, introduction of epofen and toxisorb in combination into a lactating cow diet optimizes intermediate metabolism and enhances nonspecific immunity.

Keywords: productive stage of life, feeding diet, elements of nutrition, red biological fluid, antioxidants, sorbents, morphobiochemical parameters, rumen metabolism, nitrogen and minerals.

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INTRODUCTION

Modern technological solutions in animal breeding focus on production of qualitative, competitive and safe feed products, based on all biogenetic factors that produce living objects. In these conditions, the most efficient potential realization of a farm animal can be achieved only by implementation of beneficial feeding plans and utilization of high-quality feed [1, 2].

It is known, that these days farm animal breeders focus on production cost cuts, which makes an increasing share of farms switch to utilization of on-site produced feed mill [3, 4].

However, milk production has higher standards of requirements to ecological safety and quality of feed mill composition. It was determined that long-term feed storage results in degradation of some vitamins and bioactivity inhibition of a number of enzymes due to lipid metabolic processes in maize, wheat and barley, that are characterized by synthesis of intermediate peroxides. In addition, the stored feed mill is prone to contamination with mold fungi, like *Aspergillus Lavus* and *Aspergillus parasiticus*, which leads to the growth and accumulation of aflatoxin B₁, T-2 toxin, ochratoxin A, etc. It is highly likely that they have negative impact on farm animals general metabolism and production growth [5]. On-site production practice shows that it is impossible to prevent these processes or to avoid using contaminated crude grain in the production of feed mill for milking animals.

A number of scientists highlight that it is obligatory to use chemical substances, like antioxidants and sorbents, that neutralize negative influence of microscopic toxic fungi and harmful intermediate fatty metabolites [6, 7].

Thus, the only way for breeders to provide high productivity and ecological safety of milk in animals, that are fed with on-site produced feed mill, is to introduce antioxidants and sorbents into animals diet.

The purpose of the present study was to evaluate antioxidant epofen and mycotoxin sorbent toxisorb utilization in milking cows diet along with on-site produced feed mill in order to increase cows milk productivity, improve milk characteristics and enhance animals metabolism.

MATERIALS AND METHODS

The study design included 4 groups (one control group and three test groups) containing 10 cows in each one, that were selected by analogue pairs based on physiological status, age and productivity [8, 9].

Cows in the control group were fed according to the basic feeding plan. Cows in 1st test groups had antioxidant epofen 3 g/head added to the basic feeding plan, 2nd group – sorbent toxisorb 1.5 kg/ton of concentrated feed, 3rd group – epofen 3g/head in combination with sorbent toxisorb 1.5 kg/ton of concentrated feed.

RESULTS AND DISCUSSION

It is known that the main part of digestion and absorption of feed mill in ruminants is performed in rumen, which is considered to be a unique peculiarity of these animal species.

Although rumen cells do not secrete endogenous enzymes, the elements of feed are finely digested due to microbiological processes that are going in rumen. At the same time, high level of microflora activity in rumen primarily depends on the conditions, created by the range and quality of feed. Intermediate complex biochemical hydrolysis products include peptides, amino acids and ammonia, that are used in microorganisms protein synthesis.

In the end, microbial protein, synthesized by microorganisms, becomes a nutritional element in the animal digestive tract.

It is well-known that microbiological protein has higher biological value than plant protein. Besides, rumen microflora promotes synthesis of a great number of polysaccharides that are absorbed in small intestine after a complex of digestive transformations.

Infusoria actively accumulate polysaccharides. It is known that an important adaptive microorganism capability is accumulation of polysaccharides in their cells, which is relevant for both protozoans and their hosts, because in the process of digestion they are metabolized into monosaccharides and absorbed into bloodstream.

The rate of rumen processes influences on variability of chyme metabolites level. For this reason, the rate of rumen metabolism is characterized by the concentration of different metabolite groups.

The authors evaluated the parameters of rumen metabolism in animals that received epofen and toxisorb in addition to the basic feeding plan (Table 1).

Table 1: Parameters of rumen metabolism in cows, (n=3)

Parameter	Group			
	Control	1 st test	2 nd test	3 rd test
Nitrogen, mg%:				
- total	122.4±5.9	132.2±4.53	129.6±3.97	136.2±6.14
- nonprotein	46.3±3.22	43.4±4.22	44.6±2.32	42.8±2.68
- protein	76.1±4.38	88.8±5.78	85.0±2.88	93.4±4.68
- ammonia	16.2±4.12	14.4±2.32	14.9±2.38	13.8±1.89
- of pH media	6.84±0.12	7.02±0.10	6.98±0.09	7.08±0.08
VFA, mmol/100 ml	11.12±0.17	11.89±0.14	11.70±0.17	12.00±0.16
including, %: acetic acid	62.16±0.29	64.98±0.32	64.46±0.30	65.39±0.37
propionic acid	19.89±0.20	20.02±0.22	19.96±0.24	20.16±0.14
oleic acid	12.56±0.18	9.69±0.16	10.48±0.20	9.14±0.19
<i>Flavobacteriumvitarumen</i> , ths/ml	122±1.8	144±1.9	138±2.1	150±2.2
Infusoria, ths/ml	568±3.2	712±2.9	690±3.0	739±2.5
Cellulose activity, %	14.35±0.47	16.98±0.51	16.44±0.45	17.59±0.62
Protease activity, %	43.12±0.38	45.98±0.41	45.64±0.34	46.42±0.35

Content of total protein and nitrogen types indicates on the level and rate of nitrogen containing transformations and synthesis of microbial protein in rumen. The data in Table 1 shows that inclusion of epofen and toxisorb into cows basic feeding plan had positive impact on microbial synthesis in rumen. In comparison with the control group, cows in the 3rd test group had metabolic nitrogen level in chyme increased by 11.3%, protein nitrogen level increased by 22.8% and ammonia nitrogen level decreased by 14.8%. Normal pH in rumen media is 6.5 – 7.4. In the 3rd test group this parameter increased by 0.24 unites in comparison with the control group, which was associated with improvement of microflora species quality, their general activity, rate of synthesis and absorption of organic acids and ammonia, improvement of rumen and reticulum motor function.

In the 3rd test group, where cows received epofen and toxisorb, infusoria count in forestomachs significantly increased by 30.0% and cellulolytic activity in rumen increased by 22.5% (P>0.95) in comparison with the control group.

The highest bacteria F count growth was registered in rumen of cows from the 3rd group and was equal to 22.9% (P>0.95) in comparison with the control group. This bacteria species synthesizes vitamins. Thus, indication of epofen and toxisorb into the basic feeding plan stimulates activation of biosynthetic processes in rumen.

Volatile fatty acids are the final products of carbohydrate metabolism in rumen. That is why, the rate of protein biosynthesis in rumen depends on the rate of fermentative activity processes that provide energy for rumen microorganisms growth. It is possible that the content of bacterial protein can be identified by the

size and volume of hexoses, fermented in rumen, and the rate of VFA synthesis. Based on these facts, the authors identified VFA concentration in rumen chyme, their composition and volume by their type.

It was determined that introduction of epofen and toxisorb in combination into the basic feeding plan of milking cows increased VFA content in rumen chyme. In the 3rd group total VFA content increased by 7.9% in comparison with the control group, which indicated on enhancement of fermentation rate in rumen under the influence of the introduced feed additives. It should be noted that fermentation processes were propionate oriented.

Acetic acid content, which is the main source of milk fat, in rumen fluid of cows from the 3rd group was higher by 5.1% (P>0.95) in comparison with the control group.

The results of physiological studies showed that combined indication of epofen and toxisorb provided highest level of digestion activity due to improvement of microflora species quality in upper gastrointestinal tracts, which enhanced the synthesis of proteinases, cellulases and amylases. This resulted in enhancement of hydrolysis of crude protein, crude fiber and nitrogen-free extractive substances (NFE) consumed by cows (Table 2).

Table 2: Digestibility coefficient of nutrients in the feeding plan of cows in the study groups, (n = 3)

Parameter	Group			
	Control	1 st test group	2 nd test group	3 rd test group
Dry matter,%	66.2±0.42	69.0±0.48	68.6±0.48	69.4±0.52
Organic matter, %	67.9±0.49	70.4±0.50	70.0±0.46	71.2±0.44
Crude protein, %	64.9±0.29	67.6±0.37	67.2±0.34	68.5±0.34
Crude fiber ,%	62.4±0.42	66.3±0.44	66.1±0.48	66.9±0.52
Crude fat ,%	56.8±0.37	59.4±0.35	58.6±0.42	60.0±0.32
NFE,%	78.6±0.57	82.0±0.51	81.6±0.60	82.8±0.57

Data in the Table 2 shows that cows in the 3rd test group digested and absorbed dry matter more actively by 3.3%, crude protein – by 3.6%, crude fiber – by 4.5% and NFE – by 4.2% in comparison with the control group (P>0.95).

1st and 2nd test group parameters were lower than in the 3rd test group, but higher than in the control group.

To evaluate utilization capacity of protein, the main plastic material obtained by milking cows from feed, the authors investigated nitrogen balance during physiological study (Table 3).

Table 3: Nitrogen utilization balance in the studied cows, g/head, (n = 3)

Parameter	Group			
	Control	1st test group	2nd test group	3rd test group
Taken with food	281,14±1,34	280,62±1,08	281,43±1,85	281,06±1,02
Selected from:				
- fece	98,76±2,34	90,93±2,48	92,38±1,96	88,51±2,83
- urine	92,12±1,22	86,69±1,69	88,91±3,08	86,89±1,95

- milk	78,72±1,74	87,06±2,04	85,45±1,44	89,12±1,81
Digested	182,38±2,33	189,69±1,54	189,05±0,97	192,55±1,28
Nitrogen balance, g	11,54±0,47	15,94±0,44	14,69±0,38	16,54±0,50
Used nitrogen for milk, %:				
- from adopted	28,00±1,27	31,02±0,97	30,36±1,28	31,71±1,02
- from the digested	43,16±1,92	45,89±1,96	45,02±1,80	46,28±1,67

The study of nitrogen balance showed that nitrogen consumption was similar in all the test groups and in the control group. However, cows from the test groups had higher digestibility coefficient of nitrogen in comparison with the control group due to the indicated drugs. In the 3rd test group it was higher by 10.17 g or 5.6% in comparison with the control group (P>0.95).

Nitrogen excretion with milk protein complied with the level of productivity of the studied cows.

Introduction of biologically active additives epofen and toxisorb into the basic feeding plan of cows contributed to better digestion and absorption of nitrogen fraction of feed and its transformation into lactoproteins. Thus, in the 3rd test group nitrogen excretion with milk was higher by 10.4% in comparison with the control group (P>0.95). In the 3rd test group the cows utilization of nitrogen from the digested amount for milk synthesis was higher by 3.71% (in absolute units) in comparison with the control group (P>0.95).

At the same time, the lowest nitrogen accumulation was observed in cows in the control group. Still, in all the groups it was positive. In the 3rd test group nitrogen accumulation level was higher by 43.3% per day in comparison with the control group (P>0.95).

Nitrogen accumulation rate in the body indirectly indicates on favorable pregnancy development and normal organism preparation for lactation. Enhanced transformation of feed nutrients and energy into milk determines the rate physiological and biochemical processes in cows.

Blood is a body medium that reflects all the milk production alterations in a cow organism during lactation.

The rate of rumen metabolism is directly influenced by biologically active substances included into the feeding plan, what, it its turn, has impact on cow intermediate metabolism. For this reason, the authors evaluated morphobiochemical blood parameters of the studied cows (Table 4).

Table 4: Cow morphological blood parameters (n=3)

Parameter	Group			
	Control	1st test group	2nd test group	3rd test group
Erythrocytes, 10 ¹² / l	6,08±0,20	7,14±0,22	6,94±0,21	7,56±0,17
Leukocytes, 10 ⁹ / l	10,18±0,27	10,26±0,35	10,26±0,31	10,35±0,32
Hemoglobin, g/ l	100,02±1,02	112,81±1,35	110,59±1,42	109,54±1,21
Methemoglobin, %	3,44±0,29	2,55±0,30	2,43±0,27	1,55±0,32
Total protein, g/ l	72,14±0,42	76,70±0,47	75,68±0,35	77,68±0,33
Albumin, g/ l	31,12±0,26	33,87±0,18	33,21±0,22	34,41±0,28
Globulins, g/ l	41,02±0,25	42,83±0,32	42,47±0,18	43,27±0,40
including α-globulins	9,31±0,12	9,44±0,08	9,44±0,14	9,52±0,08
β-globulins	7,98±0,07	8,29±0,06	8,31±0,09	8,21±0,10
γ-globulins	23,73±0,17	25,10±0,16	24,72±0,10	25,54±0,08
A / G ratio	0,758±0,03	0,790±0,06	0,782±0,05	0,795±0,04

BASQUE, %	58,68±1,36	62,49±0,78	62,29±0,98	63,84±1,52
Content of lysozyme, ug / ml	4,62±0,85	5,61±0,64	5,38±0,78	5,92±0,39
Acetone, mmol/l	0,44±0,02	0,24±0,04	0,31±0,03	0,24±0,04
Glucose, mol/l	3,42±0,06	3,63±0,07	3,59±0,08	3,77±0,08
Calcium, mmol/l	2,32±0,03	2,46±0,05	2,42±0,04	2,51±0,06
Phosphorus, mmol/l	1,54±0,04	1,62±0,05	1,62±0,07	1,69±0,06
Vitamin a, μmol / l	0,22±0,004	0,41±0,006	0,38±0,003	0,46±0,002
Vitamin C, mmol / l	1,23±0,004	2,44±0,006	2,36±0,005	2,59±0,005

Data in Table 4 shows that in the 3rd test group due to biologically active additives, hematopoietic function activity was higher than in the control group, which was confirmed by higher erythrocyte count by $1.48 \times 10^{12}/L$ and higher hemoglobin level – by 9.52 g/L ($P > 0.95$).

The rate of metabolism and protein synthesis in an organism is directly related to the content of total protein and its fractions in serum. It was established that the studied biologically active additives had great impact on the level of protein, its fractions and subfractions content in serum of the studied cows.

Biochemical analysis showed that total protein content, primarily albumins content, in serum was higher in the test groups by 4.56, 3.54 and 5.54 g/L or by 6.3, 4.9 and 7.6%, respectively, than in the control group ($P > 0.95$).

Increased levels of albumins were associated with the impact of the studied biological active substances on activation of synthesis of protein fraction plastic materials and their unitization for milk production. Albumin protein fraction level in cows serum from the 3rd test group was higher by 3.29 g/L or 10.5% ($P > 0.95$) in comparison with the control group ($P > 0.95$).

In the 3rd group globulin fraction protein level was significantly higher by 5.5% ($P > 0.95$) in comparison with the control group.

In the 3rd test group the index was higher by 4.88% than in the control group ($P > 0.95$).

The studied additives decreased the level of acetone in serum of cows from the 3rd test group by 1.8 times ($P > 0.95$) and increased the level of glucose by 10.2% ($P > 0.95$) in comparison with the control group. Total calcium and nonorganic phosphor in all the groups were within the normal values range. However, calcium and phosphor levels in cows blood from the 3rd test group were higher by 8.2% and 9.7% ($P > 0.95$), respectively, in comparison with the control group.

Because of exceptionally high levels of vitamins in cows, the authors evaluated the content of vitamin A and C in cows serum. Their levels in the 3rd group were higher by 2.09 and 2.1 times ($P > 0.95$) in comparison with the control group.

The above mentioned data confirms that epofen and toxisorb exert high biological activity in rumen digestion. This, in its turn, prevents degradation and oxidation of carotenoids, in particular, β -carotene.

The study of nonspecific immunity humoral factors showed significant increase of these parameters in cows from the test groups, in particular, from 3rd test group, that received the studied additives in combination.

It is known that bactericidal activity is associated with the protective capacity of the body to react to microbial agents. This protective mechanism is the first one to face infectious agents in an organism.

It was established that bactericidal activity parameters in cows from the test groups were higher by 3.81, 3.61 and 5.16% (in absolute units), respectively, in comparison with the control group ($P > 0.95$).

Lysozyme is expressed into blood by macrophages and is believed by many scientists to exert immunostimulating and immunoregulating action on protective mechanisms.

For this reason, identification of lysozyme levels in serum is necessary for objective evaluation of nonspecific resistance [10,11,12,13,14].

The study results showed that this parameter was higher in cows from test groups by 21.4, 16.4 and 28.1%, ($P>0.95$) respectively, in comparison with the control group. Since macrophages are the main producers of lysozyme, it can be assumed that functional activity of these cells significantly increased in the study cows.

Thus, combined introduction of epofen and toxisorb to the basic feeding plan of lactating cows, that contains on-site produced feed mill, optimizes intermediate metabolism and enhances nonspecific immunity.

CONCLUSION

The results of the present study confirmed the benefits of antioxidant epofen and sorbent toxisorb feed additives utilization in animal feeding for normal activation of protective immune mechanisms and improvement of physiological and biochemical status of lactating cows.

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