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STUDY OF THE EFFICIENCY OF USING BIOGENIC METALS FOR FEEDING CALVES

Oksana Shkromada, Tetiana Fotina, Roman Petrov

Breeding young cattle is important, especially during the transition period from dairy feeding to concentrated dry feed. The paper presents the results of the use of biogenic metal nicotinates for weaning calves to improve metabolism in animals.

The aim of research. *To investigate the effect of biogenic metal nicotinates: Zn, Cu, Fe, Co, Mn on the biochemical parameters of blood in calves after weaning.*

Materials and methods. *The study was carried out during 2021 in the conditions of LLC "Agrofirma Lan", Sumy region, Sumy district, Kindrativka, Ukraine for breeding young cattle. The calves of the experimental group were given compound feed and a premix of nicotinates of biogenic metals: Zn, Cu, Fe, Co, Mn, manufactured by PPronos Agro" (1 g per 1 kg of feed). In the control group, combined feed and a premix with metal sulfates were used for 30 days.*

Results. *An increase in the level of total protein in the body of calves of the experimental groups was established by 16.12 % in comparison with the control ($p \leq 0.05$). Also, in experimental animals, the activity of the enzymes alanine aminotransferase and aspartate aminotransferase was higher than the physiological norm, which shows an insignificant effect of nicotinates of biogenic metals on internal organs and systems. In the experimental group of calves, the level of magnesium was probably higher by 52.38 % and potassium – by 14.94 % compared to the control group ($p \leq 0.05$). It was found that the animals of the experimental groups probably had more zinc by 34.96 %; copper – by 35.72 %; iron – by 92.29 %; manganese – by 41.13 %; selenium – by 3.22 % and cobalt – by 98.33 % compared to the control ($p \leq 0.05$).*

Conclusions. *The positive effect of the use of biogenic metal nicotinates on the metabolism of calves at weaning has been proven. It was found that the level of total protein in the body of calves of the experimental groups was probably higher by 16.12 %, magnesium - by 52.38 %; potassium - by 14.94 %. When determining the content of inorganic substances, it was found that the animals of the experimental groups probably had more zinc by 34.96 %; copper – by 35.72 %; iron – by 92.29 %; manganese – by 41.13 %; selenium – by 3.22 % and cobalt – by 98.33 % compared to the control ($p \leq 0.05$)*

Keywords: *calves, biogenic metal nicotinates, inorganic substances, blood biochemical parameters*

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1. Introduction

Dairy producers raise dairy calves to replace dairy cows that are culled from the herd for various reasons such as mastitis, infertility, lameness, and low milk production [1].

An additional reason for removing cows from the herd is the sale of cows to other milk producers. Dairy producers who plan to expand their herd often increase the number of cows by keeping more heifers in the herd. This is called internal herd growth [2]. Balanced feeding of calves has many benefits for the producer, lower feed costs and less waste. The increase is focused on 0.8 kg/day, while meeting all the needs of heifers after weaning. The mineral content of the feed is also important in the formulation of the diet [3].

Iron is a component of blood and is involved in enzymes in the electron transport chain, such as cytochrome oxidase and cytochrome enzymes. It is very difficult to quantify the demand because most of the Fe is recycled. Calves require about 150 mg/kg iron, while an adult cow requires only 24 mg/kg [4].

Manganese is essential for the production of superoxide dismutase and can be efficiently absorbed when converted to an amino acid. The Mn requirement is 40 mg/kg diet dry matter. An increase in Mn results in a low fertilization rate. Deficiency can also have the same effect on fertility as toxicity, but it can also cause bone abnormalities. Most of the absorbed Mn is transported to the liver and excreted in the bile. Manganese is found in bones, liver and hair [5].

Zinc is found in several metalloenzymes such as RNA polymerase, carbonic anhydrase, and several others that affect nutrient metabolism. Zinc absorption mainly occurs in the small intestine [6]. The Zn requirement for

a growing heifer is 200–300 mg and 22.8 mg/kg of ration. About 50 % of the Zn in milk is absorbed by the calves. Zn deficiency leads to hoof weakness, impaired ovarian growth and parakeratosis (scaly skin) [7].

Copper is part of the cytochrome oxidase system required for the electron transport chain. Cu is an integral part of the enzyme lysyl oxidase, which catalyzes the cross-linking of desmosine in collagen and elastin, which are essential for bone strength. It is also involved in the synthesis of ceruloplasmin, which is necessary for the transport of Fe for the synthesis of hemoglobin and superoxide dismutase, which functions as an antioxidant. Copper requirements are low, with a lactating cow requiring 0.15 mg/kg milk. In adult cattle, it is assimilated by only 1–5 %, in newborns – by 70 %. In diets high in S and Mo, Cu absorption is reduced. Copper is usually fed to animals via sulfate (inorganic form), but chelated copper (organic form) increases the absorption of Cu [3].

Trace elements are usually used in feed in the form of inorganic salts – sulfates and chlorides. Such salts are readily soluble and easy to mix. However, their biological action is complicated by the phenomenon of chemical antagonism of metals. For example, iron is a chemical antagonist for zinc and zinc is a chemical antagonist for copper. Being in a dissociated, readily soluble state, metal ions are able to react with other chemicals that enter the intestines along with food and create a thick mixture called chyme. At the same time, there is also the likelihood of chemical reactions, as a result of which trace elements form insoluble compounds and precipitate, as a result of which they cannot be used in metabolic processes. These problems are solved by using chelates – combinations of microelements with organic substances, for example, with amino acids [8]. Scientists have determined that the replacement of inorganic zinc, copper and manganese in the diet of sows with their chelated (organic) complexes provided a comparative advantage over the traditional diet from inorganic mineral sources [9].

The aim of research is to investigate the effect of nicotinates of biogenic metals: Iron, Zinc, Manganese, Copper, Cobalt on the biochemical parameters of blood in calves after weaning.

2. Materials and methods of research

The study was carried out during 2021 in the conditions of LLC "Agrofirma Lan", Sumy region, Sumy district, Kindrativka, Ukraine for raising cattle. Experimental and control groups of 10 calves were formed after weaning each.

The calves of the experimental group were given compound feed and a premix of nicotinates of biogenic metals: Iron, Zinc, Manganese, Copper, Cobalt, manufactured by "Kronos Agro" (1 g per 1 kg of feed) (Table 1). In the control group, combined feed and a premix with metal sulfates were used for 30 days (Table 1).

Table 1

The composition of premixes for addition to feed for calves of different groups

Experiment group	Control group
– Iron nicotinate 53.77 g (iron 10 g)	– Iron sulfate 53.77 g (iron 10 g)
– Zinc nicotinate 57.09 g (zinc 12 g)	– Zinc sulfate 57.09 g (zinc 12 g)
– Manganese nicotinate 43.59 g (manganese 8 g)	– Manganese sulfate 43.59 g (manganese 8 g)
– Copper nicotinate 9.69 g (midi 2 g)	– Copper sulfate 9.69 g (midi 2 g)
– Cobalt nicotinate 0.31 g (cobalt 0.06 g)	– Cobalt sulfate 0.31 g (cobalt 0.06 g)
– Filler zeolite up to 1 kg	– Filler zeolite up to 1 kg

All animal studies were carried out in accordance with Directive 2010/63/EU as amended by Regulation (EU) 2019/1010 and approved by the conclusion of the Ethics and Bioethics Commission of the Faculty of Veterinary Medicine of Sumy National Agrarian University Protocol No. 6 dated 09/18/2021.

The calves received 20 samples of blood serum of cattle (calves) and conducted biochemical tests: total protein – SOP-BP-02-2017; albumin – SOP-BP-25-2018; urea – SOP-BP-03-2017; inorganic phosphorus – SOP-BP-04-2017; total Calcium – SOP-BP-05-2017; total cholesterol – SOP-BP-07-2017; ALT – SOP-BP-08-2017; AST – SOP-BP-09-2017; globulins, protein coefficient (A/G), de Ritis index (AST/ALT), Ca/P, urea nitrogen – by calculation; Magnesium – SOP-BP-06-2017; Potassium – SOP-BP-11-2017; combined samples – experimental group and control group – vitamin E – SOP-BP-12-2018; vitamin A – SOP-BP-14-2018, inorganic microelements (Zn, Cu, Fe, Mn, Se, Pb, Ni, Sr, Co, Br) – SOP-BP-15-2018 [10].

The statistical analysis of the experiments was carried out using the Microsoft Excel for Windows 2010 program. The results obtained in the work were statistically calculated using the Fisher-Student method, taking into account the statistical errors and the likelihood of the compared indicators. Indicators with a level of more than 95 % ($p \leq 0.05$) were considered possible.

3. Research results

Weaning calves from milk is a difficult animal process for farmers. Nowadays, cattle calves are always weaned in the traditional way, when calves and cows are usually kept in the same unit for 90–180 days, mainly on compound feed. The work investigated the effectiveness of two types of premixes for calves at weaning, which will help facilitate adaptation to solid feed and improve the metabolism of animals (Table 2).

According to the results of the study (Table 1), it can be said that the use of the premix with metal sulfates in the food of calves in the control group and the nicotinates of biogenic metals in the experimental group did not have a negative effect on protein metabolism in the animal body. All characteristics were within the physiological norm. However, the level of total protein in the body of calves in the experimental groups was 16.12 % higher than in the control ($p \leq 0.05$). Therefore, it

is possible to state the fact that in animals that received a premix based on nicotinates of biogenic metals, the protein was better absorbed.

In the blood of calves, lipid metabolism (cholesterol) was within the normal range in the experimental and control groups (Table 3).

Table 2

Results of biochemical studies of blood serum samples from calves (M±m, n=10)

Group of calves	Name of the defined parameters, unit of measurement							
	Total protein, g/l	Albumin, g/l	Globulin, g/l	Albumin, %	Globulin,%	(A/G), units	Urea, mmol / l	Urea nitrogen, mg/dl
RD designation for test methods	SOP-BP-02-2017	SOP-BP-25-2018	Calculation				SOP-BP-03-2017	Calculation
experiment	70.56±0.68*****	35.17±0.28	35.39±0.52	49.84±0.17	50.16±0.20	0.98±0.02	4.96±0.08	13.90±0.11
control	60.76±0.45	31.05±0.32	29.71±0.19	51.10±0.94	48.89±0.40	1.04±0.04	4.44±0.15	12.44±0.12
reference values for calves up to 6 months age	55–70	26–35	–	40–60	–	–	3.0–6.5	–

Note: – p≤0.05 compared with the control group

Table 3

Results of biochemical studies of blood serum samples from calves (M±m, n=10)

Group of calves	Name of the defined parameters, unit of measurement						
	Total cholesterol, mmol / l	AST, units/l	ALT, units/l	(AST/ALT), units	Total Ca, mmol/l	Inorganic P, mmol/l	Ca/P, units
RD designation for test methods	SOP-BP-07-2017	SOP-BP-2017	SOP-BP-08-2017	Calculation	SOP-BP-05-2017	SOP-BP-04-2017	Calculation
experiment	2.71±0.05	46.44±0.45	25.38±0.63	1.83±0.03	2.78±0.03	2.29±0.07	1.21±0.05
control	2.69±0.08	46.26±0.28	23.05±0.51	2.01±0.05	2.45±0.06	2.19±0.05	1.12±0.03
reference values for calves up to 6 months age	1.3–4.0	10–70	7–20	–	2.0–3.0	1.8–2.4	–

In research animals, the activity of the enzymes alanine aminotransferase (ALT) and aspartate aminotransferase (AST) was higher than the physiological norm, which shows an insignificant effect of nicotinates of biogenic metals on internal organs and systems. There was also a high content of Ca and P in the blood of calves from the experimental groups, which reduces the risk of rickets.

Vitamins as catalysts of metabolic processes in the body of animals are of great importance, especially for an intensively growing organism (Table 4).

Magnesium is essential for the normal functioning of the nervous system in animals. Potassium levels are important for kidney and heart function. The results of the study show a sufficient level of magnesium and potassium in the blood of calves from the experimental and control groups. However, in the study group, magnesium was probably 52.38 % higher and potassium 14.94 % higher than in the control group (p≤0.05). The introduction of various premixes into the diet of calves did not affect the content of vitamins A and E in the blood serum.

More visual results were achieved when determining the content of inorganic elements in the blood of calves (Table 5).

Table 4

Results of biochemical studies of blood serum samples from calves (M±m, n=10)

Group of calves	Name of the defined parameters, unit of measurement			
	Magnesium, mmol/l	Potassium, mmol/l	Vitamin A, μg/100ml	Vitamin E, μg/ml
RD designation for test methods	SOP-BP-06-2017	C SOP-BP-11-2017	SOP-BP-14-2018	SOP-BP-12-2018
experiment	0.96±0.05*****	5.00±0.04*****	16.12±0.04	0.7±0.01
control	0.63±0.03	4.35±0.02	15.83±0.06	0.7±0.04
Reference values for cows	0.50–1.15	4.3–5.3	12.5–25.0	0.3–2.0

*Note: * – p≤0.05 compared with the control group*

Table 5

Content of inorganic elements in collected blood serum samples of calves by groups (M±m, n=10)

Name of the defined parameters, unit of measure	Group of calves		Reference values
	experiment	control	
RD designation for test methods: SOP-BP-15-2018			
Zinc (Zn), μg %	159.74±2.36*	118.36±3.75	104.00–160.00
Cuprum (Cu), μg %	89.28±1.34*	65.78±2.57	65.00–102.00
Ferum (Fe), μg %	196.81±4.25*	102.35±3.24	84.00–140.00
Manganese (Mn), μg %	5.97±0.05*	4.23±0.05	4.00–6.00
Selenium (Se), μg %	8.32±0.05*	8.06±0.05	7.50–16.00
Plumbum (Pb), μg %	Not found	Not found	–
Nickel (Ni), μg %	2.92±0.05	2.84±0.05	2.80–5.40
Strontium (Sr), μg %	Not found	Not found	–
Cobalt (Co), μg %	5.94±0.05*	3.01±0.05	2.30–5.90
Bromine (Br), mg %	0.86±0.05	0.89±0.05	0.70–1.30

*Note: *–p≤0.05 according to the control group*

According to the results of the studies, a significant difference in the biochemical parameters of the blood of the animals of the experimental groups was established. Thus, when determining the content of inorganic substances in the blood of calves, it was found that the animals of the experimental groups probably had more zinc by 34.96 %; copper – by 35.72 %; iron – by 92.29 %; manganese – by 41.13 %; selenium – by 3.22 % and cobalt – by 98.33 % compared to the control (p≤0.05). The results achieved indicate a higher level of absorption of biogenic metal nicotinates compared to the conventional sulfate form in standard premixes.

4. Discussion of research results

The valence state of a mineral and its molecular form (inorganic or organic) in the diet play an important role in the bioavailability of trace elements. These specific characteristics of the mineral may be responsible for the complexes they form with other ingredients in the intestine, which can interfere with or promote mucosal absorption, transport and / or metabolism of the mineral in target tissues [11]. With common inorganic oxides and sulfates, during digestion of food in the stomach, the released ions can either remain soluble in the intestine, interacting with components such as amino acids and carbohydrates, or interact with water molecules to form an insoluble hydroxyl metal, or bind to ligands. form poorly soluble salts [12]. Chelated mineral complexes, which are minerals bound to an organic ligand such as proteins or a specific amino acid, are more stable in the gastrointestinal tract and are protected from complexing with other components. Therefore, using the processes of absorption of amino acids in the intestine, organic Zn, Cu, Fe, Co, Mn, which are structurally and functionally important for calves, are more easily absorbed by the intestinal epithelium in comparison with inorganic ones.

It should be noted that the use of the premix with metal sulfates and the experimental group – biogenic metal nicotinates in the diet of calves of the control group did not have a negative effect on lipid metabolism in the animal body. The level of total protein in the body of calves from the experimental groups was probably higher by 16.12 % compared to the control (p≤0.05). Therefore, it is possible to state the fact that in animals that received a premix based on nicotinates of biogenic metals, the protein was better absorbed. In addition, in research animals, the activity of the enzymes alanine aminotransferase (ALT) and aspartate aminotransferase (AST) was higher than the physiological norm, which shows the insignificant effect of nicotinates of biogenic metals on internal organs and systems. Studies have found that in the research group the level of magnesium was probably higher by 52.38 % and potassium – by 14.94 % compared to the control (p≤0.05). There was also a high content of Ca and P in the blood of calves from the experimental groups, which reduces the risk of rickets. Also, when determining the content of inorganic substances in the blood of calves, it was found that the animals of the experimental groups probably had more zinc by 34.96 %; copper – by 35.72 %; iron – by 92.29 %; manganese – by 41.13 %; selenium – by 3.22 % and cobalt – by 98.33 % compared to the control (p≤0.05).

The introduction of various premixes into the diet of calves did not affect the content of vitamins A and E in the blood serum.

Study limitation. The limitation of the study consists in the insufficient number of animals involved in the experiment, for a more reliable difference in the results obtained. However, the results show a difference between the use of organic and inorganic metals as a feed additive for weaning calves.

Prospects for further research. The prospect for further research is to determine the effect of biogenic metal nicotinates on the growth and development of calves after weaning.

6. Conclusions

The positive effect of the use of biogenic metal nicotinates on the metabolism of calves at weaning has been proven. It was found that the level of total protein in the body of calves of the experimental groups was probably higher by 16.12 %, magnesium – by 52.38 %; potassium – by 14.94 %. When determining the content of inorganic substances, it was found that the animals of the experimental groups probably had more zinc by 34.96 %; copper – by 35.72 %; iron – by 92.29 %; manganese – by 41.13 %; selenium – by 3.22 % and cobalt – by 98.33 % compared to the control (p≤0.05).

Conflict of interest

The authors declare that they have no conflicts of interest.

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Oksana Shkromada*, Doctor of Veterinary Sciences, Professor, Department of Obstetrics and Surgery, Sumy National Agrarian University, Herasyma Kondrateva str., 160, Sumy, Ukraine, 40021

Tatiana Fotina, Doctor of Veterinary Sciences, Professor, Department of Veterinary Sanitary Examination, Microbiology, Zoohygiene and Safety and Quality of Livestock Products, Sumy National Agrarian University, Herasyma Kondrateva str., 160, Sumy, Ukraine, 40021

Roman Petrov, Doctor of Veterinary Sciences, Professor, Department of Virology, Pathoanatomy and Avian Diseases, Sumy National Agrarian University, Herasyma Kondrateva str., 160, Sumy, Ukraine, 40021

***Corresponding author:** Oksana Shkromada, e-mail: oshkromada@gmail.com; Oksana.Shkromada@snau.edu.ua*