Feed Additives with the Inclusion of Co and Mn Change Their Bioavailability and Digestibility of Substances in Bull Calves

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Abstract. In accordance with the scheme of the experiment animals in the control group received the basic diet (BD), Experimental Group I – BD + feed additive, replacing 30% of the concentrated part of the ration with the inclusion of salts of sulfates of cobalt and manganese, Group II – BD + feed additive, replacing 30% of the concentrated part of the ration with the inclusion of nano- and microparticles of cobalt and manganese.

In situ studies found that the dry matter digestibility of feed additives containing chemically pure metal particles of cobalt and manganese and inactivated feed yeast was 15.4-20.0% (P<0.05) lower than in additives with wheat bran. However, when simulating rennet digestion (in vitro), it was found that the dry matter digestibility of feed additives containing chemically pure metal particles of cobalt and manganese and inactivated feed yeast increased by 1.5-2.2% in relation to additives containing wheat bran. Comparing the availability of pure manganese, we note that in the mixture with feed yeast, it was higher than with bran by more than 2.6 times. Similar values were observed for its inorganic form.

1. Introduction
The development and implementation of reliable methods for correcting the physiological status of the animal body in order to optimize its productive functioning are important tasks for modern biological science [1-3]. Recently, various feed additives and biologically active substances have been widely used for breeding young cattle [4, 5]. Feed is the main source of vitamins and minerals for animals. At the same time, the mineral and vitamin composition of each type of feed is subject to significant fluctuations and depends on the type of soil, climatic conditions, plant species, vegetation phase, agrochemical measures carried out by farms, harvesting technology, storage and preparation of prepared feed for feeding, and other factors [6]. In the forage prepared by the farm, there is often a lack of some elements and an excess of others, which leads to diseases, reduced productivity, violations in reproduction, deterioration of the quality of milk and meat received from cows, and low efficiency of feed use.

It is known that minerals play a role in four types of animal body functions: structural, physiological, catalytic, and regulatory ones [7]. Thus, information concerning the requirements for these substances for maintaining the vital activity of the body is important for beef cattle, for example in order to achieve their maximum production potential [8]. Thus, according to the recommendations of the US National Research Council on cattle (NRC) [9], it is assumed that at least 17 minerals are required for these animals.
In modern conditions of animal husbandry, control by specialists over the supply of animals with minerals and vitamins is extremely important, since diseases associated with their insufficiency, imbalance and toxicity are now widespread [10-12].

The aim of the study was to study the effect of feed additives with the addition of essential chemical elements on their bioavailability and digestibility of the main nutrients in the diet of bull calves.

2. Materials and methods
The objects of the study were the ruminal fluid of young cattle (selection was carried out through a chronic fistula of the scar); young bulls of the red steppe breed at the age of 12 months. The research site is the physiological yard of the Pokrovsky Agricultural College, a branch of the Orenburg State Agrarian University.

Animal care and experimental studies were performed in accordance with the instructions and recommendations of Russian Regulations, 1987 (Order No. 755 on 12.08.1977 the USSR Ministry of Health) and "The Guide for Care and Use of Laboratory Animals (National Academy Press Washington, D.C. 1996)". When performing research, efforts were made to minimize animal suffering and reduce the number of samples used.

The basic diet (BD) included Sudan grass hay (2nd cutting), alfalfa hay, concentrates. The animals in the control group received BD, Experimental Group I – BD + feed additive, which replaced 30% of the concentrated part of the diet with the inclusion of salts of cobalt and manganese sulfates, Experimental Group II – BD + feed additive, which replaced 30% of the concentrated part of the diet with the inclusion of nano- and microparticles of cobalt and manganese.

The composition of the granulated feed additive included: feed yeast, TU 9291 – 003-12914410 – 03 (moisture content – no more than 8.3 %; protein content – no more than 46.4 %; ash content – no more than 8.9 %; activity – inactive); wheat bran. GOST 7169-66; metal salts or particles (cobalt (II) sulphate 7-water H (CoSO4*7H2O), mass fraction of the main substance – not less than 99.4, GOST 4462 – 78; manganese (II) sulphate, 5-water H mass fraction of the main substance – not less than 96.2 %, GOST 435 – 77; cobalt (Co), powder, 150 nm, 99.9 % (Sigma-Aldrich); manganese (MP), powder, 99.9%, (Acros).

Studies were performed using nylon pouches: in vitro using artificial SCAR KPL 01 - 24 and 12 hour exposure; in situ – chronic scar fistula on young cattle – 24 hour exposure.

Equipment and technical means: artificial SCAR KPL 01; thermostat (37°C); atomic absorption spectrophotometer, quadropulse mass spectrometer Nexion 300 D, atomic emission spectrometer Optima 2000 DV.

Mathematical processing of the obtained data was performed by the program "SPSS Statistics Version 20".

3. Research results and discussion
According to the results of the in situ studies, it was found (Fig.1) that the dry matter digestibility of feed additives containing chemically pure metal particles of cobalt and manganese and inactivated feed yeast was lower by 15.4-20.0% (P<0.05) than in additives with wheat bran.
Figure 1. Dry matter digestibility of feed additives in situ, % (24 hours. incubation in the rumen) a – inactivated feed yeast; b – wheat bran; C – feed yeast + bran.

In our opinion, this is primarily due to the fact that monnannoigosaccharides contained in large quantities in the cell walls of feed yeast have a sorption capacity, for example in relation to biologically active substances. Similar results were observed when using metal salts (sulfates).

At the same time, during further studies (imitation of rennet digestion in vitro), it was found (Fig. 2) that the dry matter digestibility of feed additives containing chemically pure metal particles of cobalt and manganese and inactivated feed yeast increased by 1.5-2.2% in relation to additives containing wheat bran.

Figure 2. Dry matter digestibility of feed additives in vitro (24 hours. incubation in artificial rumen with ruminal fluid and 12 h. with HCl and pepsin) a – inactivated feed yeast; b – wheat bran; C – feed yeast + bran.
The dry matter digestibility of feed additives containing trace element salts, depending on the origin of the main component, changed as follows: it decreased in the presence of feed yeast with respect to manganese (more than 6%) and slightly increased with respect to cobalt.

After incubation, the samples were analyzed for the composition of 25 trace elements (Table 1).

**Table 1.** Average values of the content of essential elements after incubation of sub-samples (in situ study), mg/kg.

| Composition of additives | feed yeast+Co | Fodder yeast+With neorg. Feed yeast+Mn | Feed yeast+Mn neorgan. Bran + Co | Bran + So non-organ. Bran + Mn inorganic Mn. |
|--------------------------|---------------|----------------------------------------|-------------------------------|------------------------------------------|
| Zn                       | 21.17±2.54    | 18.61±1.86                            | 59.73±4.15                   | 41.42±4.14                              |
| Mn                       | 123.0±0.15    | 350.8±3.51                            | 220.4±11.3                  | 158.1±1.43                              |
| Cu                       | 37±0.44       | 3.58±0.36                              | 3.84±0.38                   | 6.74±0.67                               |
| Fe                       | 246±0.30      | 198±0.20                               | 230±0.25                    | 421±0.42                                |
| Co                       | 3.18±0.38     | 0.425±0.20                             | 2.16±0.22                   | 2.4±0.24                                |
| Se                       | 0.128±0.019   | 0.089±0.042                            | 0.085±0.013                 | 0.125±0.015                             |
| I                        | 4.51±0.54     | 0.381±0.046                            | 0.741±0.076                 | 0.608±0.073                             |

After incubation in the rumen of the mixture of feed yeast (CD) + manganese (CP), the content of the latter was higher than in the mixture of bran (O) + manganese (CP) by 43.9% (P≤0.05), i.e. the availability of this element for the rumen microflora in the first variant was lower. As for cobalt, after incubation in the rumen of the CD + cobalt (CP) mixture, the content of the latter was higher than in the O + cobalt (CP) mixture by 32.5% (P≤0.05).

Comparing the availability of pure manganese, we note that in the mixture with feed yeast, it was higher than with bran by more than 2.6 times. Similar values were observed for its inorganic form. As for cobalt, the availability of a chemically pure substance did not differ depending on the carriers. When incubating feed yeast with its salts, there was an increase in availability for the body by 77% (P≤0.05).

Further studies evaluated the availability of chemical elements in the samples after in vitro incubation (Table 2).
Table 2. Average values of the content of essential elements in samples after incubation (in vitro study), mg/kg.

| Composition of feed additives | Zn  | Mn  | Cu  | Fe  | Co  | Se  | I   |
|------------------------------|-----|-----|-----|-----|-----|-----|-----|
| Feed yeast+Co                | 8.88| 243 | 3.66| 393 | 8.47| 0.102| 0.561|
| Fodder yeast+With non-organic| 3.34| 171 | 3.81| 245 | 8.36| 0.044| 0.422|
| Feed yeast+Mn non-organic    | 6.13| 267 | 4.79| 704 | 6.61| 0.221| 0.665|
| Bran + Co                    | 4.67| 277 | 4.9 | 291 | 8.32| 0.14 | 0.895|
| Bran + So                    | 6.33| 365 | 8.16| 867 | 14.82| 0.059| 0.965|
| Bran + Mn non-organic        | 9.59| 706 | 11.88| 879 | 13.14| 0.118| 1.1  |
| Bran + non-organic Mn        | 8.13| 308 | 9.42| 426 | 8.41| 0.096| 0.832|

After in vitro studies of the mixture of feed yeast and Co, the content of this element was 1.80% higher than in the bran + cobalt mixture, i.e. its availability in the first variant was lower. As for chemically pure manganese, the amount of it in the O+M mixture was higher (18.88%, P≤0.05), compared to the FY+Mn mixture. A limited number of similar studies have been found in the available literature, as it was noted [13] that there is a deficiency of cobalt and zinc (4.2% and 9.7%, respectively) in the diets of cattle, which confirms the relevance of our research.

Calves from cows that received a diet with added sulfate sources of Cu, Co, Mn, and Zn (INR) and with organic sources of the same elements (AAC) had higher (<0.01) concentrations of Co in the liver at birth compared to calves from control cows (CON). The concentration of Cu and Zn in the liver at birth was higher (≤ 0.05) in calves from AAC cows compared to analogues from CON cows. In a growing batch of calves from AAC cows, the incidence of bovine respiratory diseases was lower (<0.01) compared to the CON and INR groups. Taken together, these results suggest that feeding the AAC diet to late-aged nursing cows stimulated genetic effects on the growth and health of postnatal offspring compared to the CON diet [14, 15]. In other studies, it was noted that the sources of chemical elements did not affect the productivity of cattle and immune responses during rearing and fattening [15].

The fact that metal complexes exhibit higher antibacterial and antifungal efficacy in comparison with their corresponding ligands may also affect the digestibility of substances [16]. It was also noted that the consumption of organic trace elements in combination with amino acids during the transition period improved liver function and reduced inflammation and oxidative stress [17, 18].

4. Conclusions

Thus, in the course of research, it was proposed to prepare a feed additive for young cattle. It consists in mixing highly dispersed particles of cobalt (up to 150 nm in size) and manganese (up to 300 nm in size) in a dose 10% higher than the norm for young cattle on fattening. It is mixed with inactivated feed yeast, subjected to granulation at a temperature of 60-70 °C and under pressure up to 1.5 bar.
is further fed to animals at a dose of 30% of the concentrated part of the diet, taking into account its nutritional value, which contributes to increasing the productivity of farm animals.

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