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Milk trace minerals content in dairy cows supplemented with glycine chelated minerals

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Abstract. The paper presents data regarding the impact of some trace mineral supplementation on milk minerals content of glycine chelated mineral supplemented dairy cows. The study was made on thirty Holstein multiparous dairy cows, aging arround 62 ± 4.5 months that were divided randomly in two groups: C – Control receiving normal diet without glycine mineral supplementation and one experimental group (E) that received supplement of glycine chelated Cu, Zn, Mn and Fe as follows: 15 mg/kg Cu, 20 mg/kg Mn, 60 mg/kg Zn and, 100 mg/kg Fe, added in concentrate feed. The experiment was designed from day 30 of lactation until day 100 of lactation, and were assesed the milk levels of zinc (Zn), selenium (Se), iron (Fe), manganese (Mn) and copper (Cu). In the experimental group we observed significant ($P \leq 0.05$) increase of milk concentration of Se, Mn and Cu at day 65 of lactation and highly significant ($P \leq 0.01$) increase of Zn, Se, Mn and Cu in the day 100 of lactation. The Fe content was increased in all period of study, but the increase was not statisticaly significant ($P \geq 0{,}05$). We conclude that supplementing the dairy cows whit chelated minerals will be followed by a increase of milk mineral content that could be beneficial for health.
Keywords: milk minerals, dairy cows, supplements, minerals, trace elements
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Introduction.

Trace minerals are crucial for various physiological processes, including health maintenance, antioxidant protection, and enhancing the productivity of dairy cows (Faulkner MJ and Weiss WP, 2017; Wu G, 2018). Conversely, a deficiency in trace minerals can weaken the immune system of dairy cows (Spears JW and Weiss WP, 2008).

Ruminants often experience significant dietary deficiencies of trace elements such as cobalt, copper, iodine, selenium, zinc, and manganese (Hidiroglu M, 1979). These deficiencies can significantly impact their overall performance and production levels (Mackenzie AM et al., 2001).

Milk is a vital nutrient-rich food source that has been integral to human diets across various cultures for centuries. It is not only a significant source of macronutrients such as carbohydrates, proteins, and fats but also contains an array of bioactive compounds including vitamins and minerals. Among these, trace minerals are essential for many physiological functions and overall health. Trace minerals, including iron, zinc, copper, manganese, and selenium, play crucial roles in metabolic processes, immune function, and cellular integrity (Maret W, 2020).

While milk is often acknowledged for its calcium and vitamin D content, its profile of trace minerals is equally important. For example, studies indicate that the bioavailability of iron and zinc from dairy products is significant, contributing to the nutritional needs of populations worldwide (Hercberg S and Galan P, 2010). Additionally, the presence of trace minerals in milk has been associated with various health benefits, including enhanced bone health, immune support, and a reduced risk of certain chronic diseases (Silva BB et al., 2020).

Previous studies, including our own (Goilean GV et al., 2022), have reported positive effects of trace mineral supplementation or the use of different organic forms of trace minerals in dairy cows. These benefits include increased production of milk (Rabiee AR et al., 2010), reduced the number of somatic cells (Kellogg DW et al., 2004), decreased incidence of lameness and, improved general foot health in dairy cows (Nocek JE et al., 2000; Siciliano-Jones JL et al., 2008; Overton TR and Yasui T, 2014).

The aim of the research.

Study of the influence of glycine chelated minerals on the level of some selected trace minerals in the milk of supplemented dairy cows.

Materials and methods of the research.

Object of study. The study was conducted on 30 multiparous Holstein dairy cows, each aging approximately 62±4.5 months and weighing 462±23 kg. The cows were kept in individual tie stalls within a private agricultural facility in Bihor County, Romania.

The experimental procedures and the inclusion of the animals in the study were approved by the Scientific Committee (Decision no. 62 /15.11.2020). Furthermore, all procedures complied with the EU Directive on Animal Experiments (Directive 2010/63/EU…).

Experimental scheme. The cows were randomly divided into two groups (n=15): a control group (C) receiving a normal diet without glycine mineral supplementation, and an experimental group (E) supplemented with glycine chelated Cu, Zn, Mn, and Fe (E.C.O. Trace®, Biochem, Germany) in the following amounts: 15 mg/kg Cu, 60 mg/kg Zn, 20 mg/kg Mn, and 100 mg/kg Fe, added to their concentrate feed.

The experiment starts from day 30 of lactation until day 100 of lactation. The milk was sampled in day 0 (day 30 of lactation) in the middle and end of experiment (day 65 and day 100 of lactation). The feeding was twice a day, in an individual front as was previously reported (Goilean G et al., 2022). The total chemical content of the ratio is presented in table 1.

Table 1. **Content of ratio for dairy cows used in experiment**

Parameter	Unit	Values
DM	%	48.2
Crude Protein (CP)	%	16.96
Soluble protein	% from CP	49.5
NE	Mcal/kg	1.65
Ca	%	1.02
P	%	0.31
Mg	%	0.38
K	%	1.07
Na	%	0.33
Cl	%	0.32
Vitamin A	IU/kg	11 400
Vitamin D	IU/kg	3100
Vitamin E	IU/kg	35200
Fe	ppm	208.33
Zn	ppm	26.91
Cu	ppm	9.14
Mn	ppm	24.27
Se	ppm	0.19

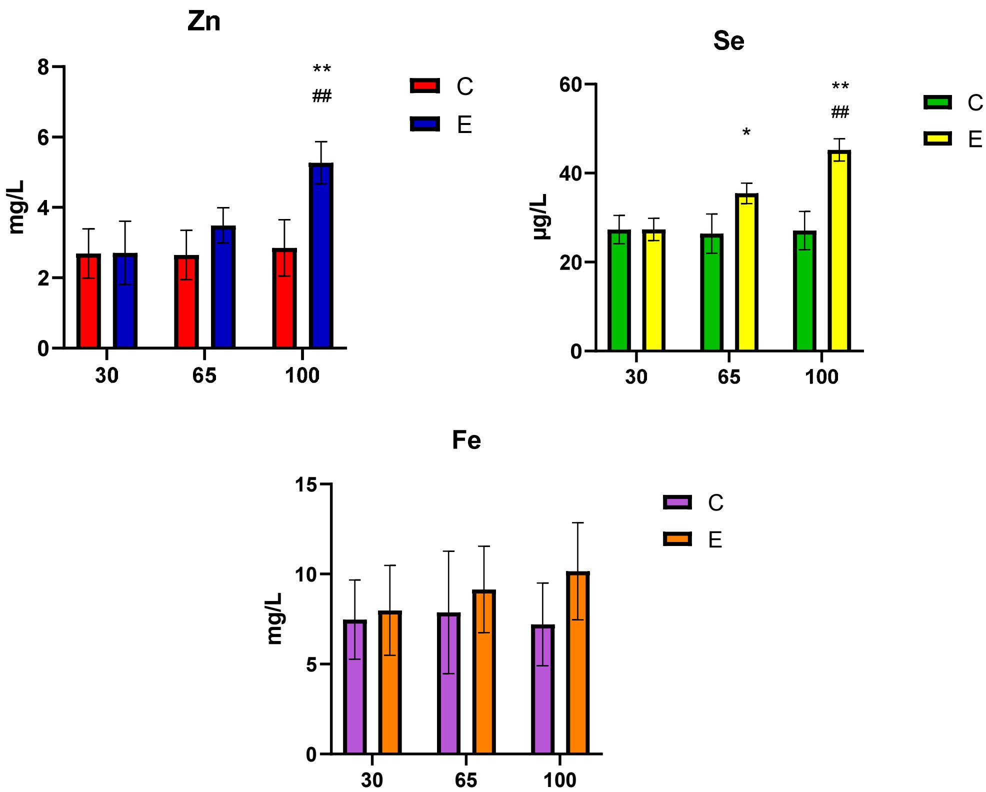
Equipment and technical facilities. The levels of the main bioelements manganese (Mn), selenium (Se), zinc (Zn), copper (Cu), and iron (Fe) in milk were analyzed using a furnace with pyrolitic tube for Mn, Se and Cu, and flame analisys for Zn and Fe by atomic absorption spectroscopy (AAS) using a ContrAA800 spectrophotometer (Analytic Jena, Germany). Preparation of the samples were by microwave digestion (Anton Paar, Austria) adding in the digestion vasels: 1 ml milk sample, 10 ml of concentrated nitric acid and 2 ml hydrogen peroxide. The microwave parameters were 10 min, 120^0C, 800 W.

Statistical processing. The results were statisticaly analyzed by one-way ANOVA using the Bonferroni correction, considering the differences statistically provided when P≤0.05 or lower, all walues being expressed as mean ± SD. The used software was GraphPad Prism 6.0 for Windows (GraphPad Software, San Diego, USA).

Results and discussions

Cow milk is a widely consumed dairy product rich in essential nutrients, including various minerals that will provide energy and support growth across all age groups and species (Gaucheron F, 2005). It contains minerals at levels of lower than 1% (w/w), which are valuable sources of electrolytes and vital trace elements necessary for healthy growth and development (Reilly C, 2004). Research has shown that the mineral composition of milk varies significantly among different cattle breeds, influenced by physical and environmental factors (Raynal-Ljutovac K et al., 2008a). Variations in the electrolytes and essential trace elements found in cattle milk may result from several factors, including the species of cattle, their health, nutritional diet, stage of lactation, and the ecological and geographical conditions (Raynal-Ljutovac K et al., 2008a; Navarro-Alarcon M et al., 2011; Chen L et al., 2020).

As is presented in figure 1, in our study we observed that the Zn milk content was almost at the same level at the beginning of milking period (day 30) in both groups (E/C: +0.74%) but increased during the lactation period in cows supplemented with chelated minerals , not significantly (P≥0.05) at day 65 0f lactation (E/C: +31.69%) but strongly significant (P≤0.01) in the day 100 of lactation (E/C: +84.91%). Comparative to the start of milking period the Zn level was increased in group received chelated minerals (E_{65}/E_{30}: +28.78%, E_{100}/E_{65}: +51.01%) the highly significant (P≤0.01) increase being observed in day 100 comparative to day 30 (E_{100}/E_{30}: +94.46%).



Comparision E/C: * - P≤0.05, ** - P≤0.01   Comparision 100 days with days 30 and 65: ## - P≤0.01

Figure 1. **Milk levels of Zn, Se and Fe in dairy cows supplemented with glycine chelated minerals**

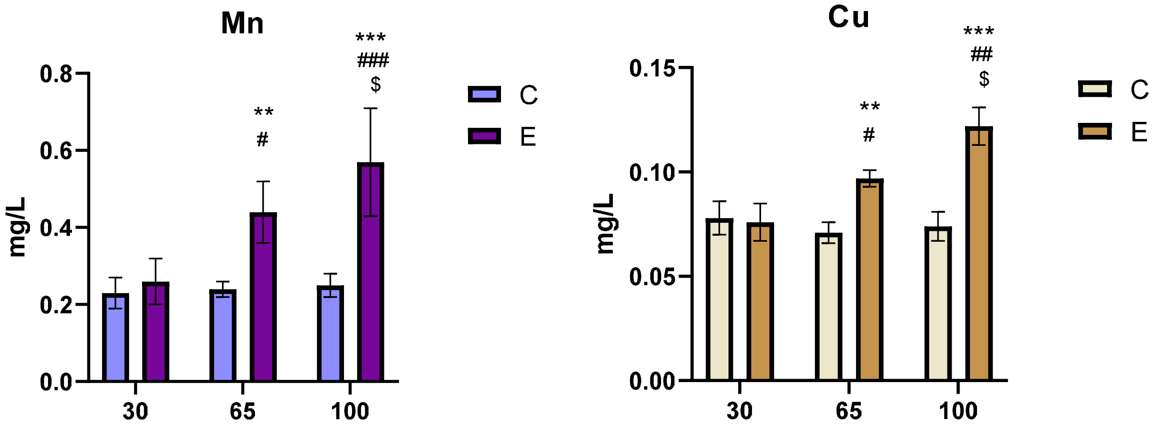
Zinc is an essential trace element that supports immune function and is involved in numerous enzymatic processes. According to Raynal-Ljutovac K et al. (2008b), the concentration of zinc in cow milk can vary among different breeds, with some breeds exhibiting higher levels due to genetic factors and dietary intake. In this aim, Proskira N et al. (2016) presented a Zn concentration of 3.7 µg/dm^3 in Jersey cow's milk, and Pilarczyk R et al. (2013) a concentration of 3.02 µg/mL in Simmental and 3.27 µg/mL in Holstein-Friesian cows, concentration that is above that we found in our study in control cows but lower that we observed in cows supplemented with chelated minerals in the day 100 of lactation period.

The Se milk content presented the same level at the beginning of milking period (day 30) in both groups (E/C: +0.14%) and increased during the lactation period in cows supplemented with chelated minerals, significantly (P≤0.05) at day 65 0f lactation (E/C: +34.19%) and strongly significant (p<0.01) in the day 100 of lactation (E/C: +66.80%). Comparative to the start of milking period the Se level was increased in group received chelated minerals (E_{65}/E_{30}: +29.72%, E_{100}/E_{65}: +27.45%) but the highly significant (P≤0.01) increase was recorded in day 100 comparative to day 30 (E_{100}/E_{30}: +65.33%).

Selenium is an essential trace mineral that plays a key role in antioxidant defense and thyroid function. Chen L et al. (2020) found that selenium levels in cow milk can be influenced by the selenium content in the cows' diet and the geographical region in which they are raised. The selenium content in cow milk is reported to be around 0.01 mg per 100 grams according to the (USDA, 2021). However, research by Haug W et al. (2020) indicates that selenium levels can vary significantly, with some milk samples containing up to 0.03 mg per 100 grams, largely influenced by the selenium content in the cows' diet.

Analyzing the Fe milk content, in our study we observed that at the beginning of milking period (day 30) there were not significant differences between groups (E/C: +6.82%). These differences were increased during the lactation period in cows supplemented with chelated minerals, but all differences were not statistically significant (P≥0.05) at day 65 of lactation (E/C: +16.13%) and in the day 100 of lactation (E/C: +40.91%). Comparative to the start of milking period the Fe level was increased in group received chelated minerals (E_{65}/E_{30}: +14.53%, E_{100}/E_{30}: +27.31%, E_{100}/E_{65}: +11.15%) but all these differences were not significant (P≥0.05).

Iron is crucial for oxygen transport in the blood and overall metabolic processes. However, the iron content in cow milk is relatively low compared to other food sources. Reilly C (2004) noted that while cow milk is not a significant source of iron, it can still contribute to the overall dietary intake, especially in combination with other iron-rich foods. Cow milk is generally low in iron, with the USDA reporting about 0.03 mg per 100 grams (USDA, 2021). A study by Hurrell RF et al. (2019) supports this finding, noting that cow milk is not a significant source of iron, which can be a concern for populations relying heavily on dairy for nutrition. Gorska A и Oprzadek K (2011) presented a concentration of 144 µg/kg Fe in the cow's milk in period lower that 100 day of lactation which are much lower that our findings.



Comparision E/C: * - P≤0.05, *** - P≤0.001 Comparision 100 days with 30 days: # - P≤0.05, ## - P≤0.01, ### - P≤0.001 Comparision 100 days with 65 days: $ - P≤0.05

Figure 2**. Milk levels of Mn and Cu in dairy cows supplemented with glycine chelated minerals**

When we analyzed the Mn milk levels, in the start of experiment (day 30) in there were not significant differences between groups (E/C: +13.04%). Advancing in the lactation period we recorded a highly significant ($P \leq 0.01$) increase of Mn content during the lactation period in cows supplemented with chelated minerals at day 65 of lactation (E/C: +83.33%) and even more in the day 100 of lactation (E/C: +128.00%). Comparative to the start of milking period the Mn level was increased in group received chelated minerals (E_{65}/E_{30}: +69.23%, E_{100}/E_{65}: +29.54%) and highly significant ($P \leq 0.01$) increase in day 100 comparative to day 30 (E_{100}/E_{30}: +119.23%).

Manganese is another important mineral found in cow milk, albeit in lower concentrations compared to other minerals. It plays a role in bone formation, metabolism, and antioxidant defense. Research by Navarro-Alarcon M et al. (2011) indicates that the manganese content in milk can be influenced by the cows' diet, particularly the mineral composition of the feed they consume. The manganese content in cow milk is relatively low, with the USDA indicating levels around 0.003 mg per 100 grams (USDA, 2021). Research by Kahn SA et al. (2019) corroborates this, suggesting that cow milk is not a significant source of manganese, as its levels are much lower compared to other food sources. Pilarczyk R et al (2013) a concentration of 0.02 µg/mL in Simmental and Holstein-Friesian cow's milk, concentration that is almost similar to our findings in control cows but lower that we observed in cows supplemented with chelated minerals in the day 65 and 100 of lactation period.

For copper, we found the same dynamic as were for Mn, respectively, in the start of experiment (day 30) in there were not significant differences between groups (E/C: -2.56%). Advancing in the lactation period we recorded a highly significant ($P \leq 0.01$) increase of Cu content during the lactation period when supplementation with chelated minerals was used at day 65 of lactation (E/C: +36.61%) and even more in the day 100 of lactation (E/C: +64.86%). Comparative to the start of milking period the Cu level was increased in group received chelated minerals (E_{65}/E_{30}: +27.63%, E_{100}/E_{65}: +25.77%) and highly significant ($P \leq 0.01$) increase in day 100 comparative to day 30 (E_{100}/E_{30}: +60.52%).

Copper is vital for the formation of red blood cells and maintaining healthy connective tissues. The copper content in cow milk has been shown to vary, with studies indicating that dietary supplementation can enhance copper levels in milk (Gaucheron F, 2011). This highlights the importance of a balanced diet for lactating cows to ensure optimal mineral levels in their milk. The USDA reports that cow milk contains about 0.02 mg of copper per 100 grams (USDA, 2021). However, a study by Kelleher SL et al. (2020) found that the copper content can vary, with some samples showing levels as high as 0.05 mg per 100 grams, indicating that environmental factors and feed can influence copper levels in milk. Proskura N et al. (2016) presented a Zn concentration of 18 µg/dm^3 in Jersey cow's milk, and Pilarczyk R et al (2013) a concentration of 0.03 µg/mL in Simmental and 0.04 µg/mL in Holstein-Friesian cows' milk, concentration that are lower that we found in our study in control cows and. Significantly much lower that we observed in cows supplemented with chelated minerals in the day 65 and 100 of lactation period.

Conclusions.

In summary, cow milk provides essential minerals, but the content of zinc, copper, selenium, iron, and manganese can vary based on several factors, including the breed of the cows, their diet, and environmental conditions. Administration of supplements with chelated minerals was followed by a significant increase of milk concentration of studied trace minerals, especially Mn, Zn and Se.

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