



# Organic Trace Minerals Improved the Productive and Reproductive Performance of Friesian Cows Better Than Inorganic Minerals

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## Key words

Friesian cows, Inorganic and organic trace minerals, Digestibility, Production, Reproduction

## ABSTRACT

This study aimed to compare the influence of chelated organic (OTM) and inorganic trace minerals (ITM) on the digestibility coefficient, nutritive values, reproductive performance, and milk yield of Friesian cows. Cows (n=50) were divided into 5 groups; cows in the first group (control, G1) were given an untreated diet, whereas diets in G2 were supplemented with 100% ITM, G3 were received 50% ITM and 50% OTM, G4 were received 25% ITM and 75% OTM, and G5 were supplied with 100% OTM. G5 and G4 digested DM more efficiently than other groups. OM and NFE digestion were significantly larger in G5 and G4 compared to other groups. Cows supplemented with OTM and/or ITM showed significantly higher average daily milk, 4% fat corrected milk yield, and fat % than the control group. Protein % and total solid % significantly increased in G5 than in G1. The interval from calving to estrus was shorter in G5 than in other groups. G4 and G5 had a significantly lower number of days open and services per conception and a significantly greater conception rate than other groups. With these results, we conclude that supplementation of Friesian cows with a diet containing permissible limits of organic trace minerals could significantly improve the digestibility coefficient, nutritive values, reproductive performance, and milk yield compared to cows fed on inorganic trace elements.

## INTRODUCTION

Trace minerals account for less than 0.01% of an organism's total mass, however, they are critical for the production and health of dairy cows (NRC, 2001). Minerals in inorganic forms have traditionally been used most frequently in animal diets and additives. Inorganic trace elements (ITM) in fodder mixes are not always beneficial to animal health and have a low oral bioavailability. Because trace elements derived from sulphates, chlorides, and oxides are indigestible, they also increase the risk of heavy metal pollution in the environment, as they are

expelled from the body at a rate greater than the rate of absorption (Miller *et al.*, 1988). However, chelated (organic) minerals (OTM), which are bonded ligands like small peptides and are identical to the ones found in natural tissues for plants and animals, are absorbed selectively from the stomach (Webb *et al.*, 2005). Subsequently, organic minerals have more bioavailability and retention than inorganic minerals in the digestive tract (Henry *et al.*, 1992; Formigoni *et al.*, 1993). Pino and Heinrichs (2016) demonstrated reduced faecal Cu output when cows were fed Cu as a proteinate compared with a sulphate form across various dietary starch levels. The bioavailability of inorganic and organic forms of minerals has been compared in several species, where researchers have reported higher bioavailability for the latter (El-Ashry *et al.*, 2012; Zhao *et al.*, 2010) and have shown that lower levels of organic minerals can be used in total replacement of inorganic forms, without loss in animal performance (Perić *et al.*, 2010). Chelated minerals are preferred to minerals from inorganic sources to increase mineral bioavailability, absorption, and utilization and produce better results than traditional mineral supplementation

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(Nemec *et al.*, 2012; Bach *et al.*, 2015). Additionally, chelating decreases mineral excretion in feces and thus causes less environmental contamination (Flora and Pachauri, 2010). Trace minerals in chelated forms could supply an equivalent amount of mineral at lower dietary inclusion levels compared to inorganic forms (Cope *et al.*, 2009). Chelated complexes of minerals contain a central atom along with a ligand (proteins, carbohydrates lipids, or amino acids) containing a minimum of one ligand atom (sulfur, oxygen, or nitrogen) with a pair of free electrons. The ligand atom is bound with the metal atom by a coordinate bond by donating an electron pair from the ligand to the electron acceptor (Pacheco *et al.*, 2017).

The amounts of trace minerals in the animal's bodies and products are influenced by the feeds they consume (Goldman, 2009). Trace minerals such as copper (Cu), zinc (Zn), and manganese (Mn), are essential for protein synthesis, connective tissue formation, immune function, and vitamin metabolism in dairy cows (Miller *et al.*, 1988). They also play a crucial role in antioxidant enzyme activities, maintenance of health, enhancement of nutritive value, and production efficiency in dairy cows, hence their deficiencies in cattle cause health problems (Griffiths *et al.*, 2007; Faulkner and Weiss, 2017; Wu, 2018). Steers given a dairy diet supplemented with Cu, Zn, and Mn as organic chelate or hydroxy chloride instead of sulphate exhibited an increase in NDF and ADF digestibility (Guimaraes *et al.*, 2020). Cortinhas *et al.* (2010) found that the use of a mix of organic carboaminochelates (Cu, Zn, and Se) for feeding led to a decrease in the mean somatic cell count (SCC) when compared to delivering the same amount of ITMs in the sulphate form. Apart from these results, (Formigoni *et al.*, 2011) observed that providing OTM to dairy cows instead of sulphate minerals had no influence on reproduction, production, or pathologic events. Furthermore, OTM sources have little influence on the milk production of dairy cows nutritionally (Zhao *et al.*, 2015; Faulkner *et al.*, 2017). Mn from organic (Mn-Met) or inorganic (MnSO<sub>4</sub>) sources exhibited similar apparent absorption while nourished during the dry period (Weiss and Socha, 2005). When the inorganic sources of Cu, Zn, and Mn were replaced by the biological ones, no significant changes in the DMI of cows were observed (Hackbart *et al.*, 2010; Bach *et al.*, 2015). Additionally Nemec *et al.* (2012) discovered that replacing Cu, Zn, and Mn sulphates with organic minerals reduced DMI in cows.

Thus, this study was aimed to compare the effect of inorganic or chelated organic trace minerals on the milk production and reproduction performance of Friesian cows.

## MATERIALS AND METHODS

### *Animals*

Friesian cows (n= 50, 547.6±32kg, 3-5 parity) were selected at prepartum (30 days at pre-calving) and the early postpartum period (120 days post-calving) from the Sakha Animal Production Research Station, Animal Production Research Institute, Agricultural Research Center, Ministry of Agriculture. Cows were divided into five groups of ten as follow: (i) cows in the first group (control, G1) were given an untreated baseline diet. (ii) G2 were given a baseline diet supplemented with 2.4g Zn, 0.78g Cu, and 2.4 g Mn (100% ITM) daily. (iii) G3 were received diet supplemented with the same minerals but in two forms which are 50%ITM and 50% OTM. (iv) G4 were received 25% ITM and 75% OTM. and (v) G5 were supplied with 100% OTM. The source of OTM was zinc chelate of glycine hydrate, cupric chelate of glycine hydrate, and manganese chelate of glycine hydrate. These supplementations were bought from Biochem (Germany). All supplementations were given for 150 days starting from day 30 of prepartum to 120 days of postpartum. All cows were disease-free and appeared healthy and were kept in separate groups beneath semi-open sheds.

### *A feeding system and management*

Cows in each group were fed an equal quantity of concentrate feed mixture (CFM), corn silage (CS), and rice straw (RS) throughout the trial period by the recommended requirements for live body weight and milk yield set by Animal Production Research Institute. The CFM is composed of wheat bran (22.5%), soybean meal (20%), corn gluten (15%), yellow maize (37.5%), molasses (3%), and common salt (0.5 and 1.5%). The chemical characterization of typical monthly food samples was used to determine the concentrations of crude protein (CP). Ether extract (EE), crude fiber (CF), nitrogen-free extract (NFE) and ash on a dry matter (DM), CP, EE, CF, NFE, and ash on a DM basis (AOAC, 2000). The chemical composition of CFM, RS, and CS as well as the chemical structure of the basal diets calculated for each group were provided in Table I.

### *Digestibility trials*

Three digestibility tests were performed on three cows from each group during the third month of the lactation period. The animals were maintained individually during the collection period and faces were extracted from the rectum in the morning before eating and in the evening after milking for 7 days. After the collecting period, representative samples (10% of fresh faces) were collected from each animal's faces and dried at 60 °C for 48 h. After

drying, samples were ground, and their size was reduced to 0.5 mm and stored in airtight plastic containers for chemical analysis. Based on the usage of silica materials as a marker, to measure the digestion coefficients of various nutrients in experimental diets, the acid-insoluble ash (AIA) method as previously described (Van Keulen and Young, 1977). Total digestible nutrients (TDN), and digestible crude protein (DCP) TDN and DCP values for various experimental diets were determined using the digestibility coefficients obtained. Representative samples from CFM, CS, RS, BH and feces were also taken and prepared for the chemical analysis according to the methods of (AOAC, 1995).

**Table I. Chemical characteristics of various feed ingredients for cows feeding (based on a dry matter).**

Item	Chemical composition (%)			
	CFM	RS	Corn silage	BH
DM	91.30	90.14	30.67	90.25
OM	95.72	82.96	94.17	88.57
CP	16.80	3.02	9.40	13.16
CF	15.38	35.91	22.05	29.01
EE	2.77	2.24	2.31	2.83
NFE	63.77	41.63	60.41	43.57
Ash	4.28	17.02	5.83	11.43

CFM, concentrate feed mixture; RS, rice straw; BH, Berseem hay; DM, dry matter; OM, organic minerals; CP, crude protein; CF, crude fiber; EE, ether extract; NFE, nitrogen free extract.

#### Milk yield and composition

According to farm management standards, cows were milked twice daily by milking machine at 6:00 and 17:00 h. Separate daily milk outputs were recorded for morning and evening milking throughout the first four months (4 to 120 days) of lactation. Individual milk samples were obtained monthly to measure the content of milk using a Milko-Scan (Model 133B) and the number of somatic cells using a Fossomatic 360 (Foss Electronic, Slangerupgade, Denmark). The fat corrected milk (FCM) content was determined as follows: 4% FCM =  $0.4 \times \text{Kg milk} + 15 \times \text{Kg fat}$  (Mandal *et al.*, 2003).

#### Blood sampling

Blood samples were taken from each animal's jugular vein at 3- to 4-day intervals from 10 days after calving until conception and up to 120 days postpartum using an anticoagulant (heparin). To separate blood plasma, blood samples were centrifuged at 3000 rpm/10 min and the obtained plasma was then kept at -20 °C.

#### Detection of estrus and insemination

To determine the onset of the 1<sup>st</sup> estrus, an infertile bull (teaser) was presented to cows in each group for 20 min three times daily at 6 am, 12 pm, and 3 pm. Estrus was recorded in cows receptive to teaser and stood for mounting by the teaser. Cows in heat were inseminated. The number and duration of estrous cycles were monitored from calving until conception. Additionally, postpartum first ovulation, first estrus, and intervals post-treatment were recorded, as well as the number of services per conception (NS/C), conception rate (CR%), and days open (DO). On day 60 post insemination, conception was achieved through rectal palpation.

#### Statistical analysis

SAS software (SAS, 2004) was used to conduct the statistical analysis of the acquired data. Duncan's Multiple Range Test was performed to examine whether or not there were statistically significant changes between groups. The following statistical model was used:

$Y_{ij} = U + A_i + e_{ij}$ . Where:  $Y_{ij}$  = Observed values,  $U$  = Total mean,  $A_i$  = Experimental group,  $e_{ij}$  = Random error

## RESULTS AND DISCUSSION

#### Feed intake

The data in Table II describe the DCP, TDN, and daily feed intake as dry matter (DMI) from the experimental diets over the feeding period. DMI, DCP, and TDN were nearly similar for animals fed tested rations. Similarly, no significant differences were observed in DMI between the trace minerals received and control groups (Nunnery *et al.*, 2007; Vazquez-Anon *et al.*, 2007; Cope *et al.*, 2009; Hackbart *et al.*, 2010; Nemeč *et al.*, 2012; Zhao *et al.*, 2015). Furthermore, Cope *et al.* (2009) showed that neither the ITM nor the OTMs seemed to have a significant influence on the DMI. Numerous studies reported no obvious variation in the DMI of cows when organic copper, zinc, and manganese sources were replaced by inorganic sources (Bach *et al.*, 2015; Hackbart *et al.*, 2010). In contrast, it was observed that substituting organic minerals for zinc, manganese, and copper sulphates decreased DMI in cows (Nemeč *et al.*, 2012).

#### Digestibility coefficient

Results in Table III show that the rate of DM, OM, CP, and NFE digestion was significantly ( $P < 0.05$ ) greater in G5 and G4 than in other groups (70.97 and 69.96 vs. 67.22, 66.91, and 65.89%). However, the differences between G2, G3 or control were insignificant. EE digestion was significantly ( $P < 0.05$ ) greater in G3, G5,

and G4 than in other groups. Digestibility coefficients of CF were not changed significantly by dietary supplementation. However, the digestibility coefficient of NFE improved in cows fed diets containing 75 and 100% organic compared to the control diet. Nutritive value of TDN was significantly higher in all dietary supplemented, with highest value in the OM group, than in the control group. Faulkner *et al.* (2017) also reported that cows fed organic minerals digested NDF more efficiently than cows fed sulphate-containing diets. Moreover, lambs treated with Zn methionine (organic Zn) exhibited an enhanced ability to digest acid detergent fiber (ADF) (Garg *et al.*, 2008). Guimaraes *et al.* (2020) also observed an increase in the nutritional value of ADF and NDF when steers fed a dairy diet was supplemented with organic chelates of copper, zinc, and manganese in the form of organic chelate or hydroxy chloride rather than sulphate. In contrast, organic Zn had lower NDF and DM digestibility than hydroxy chloride Zn or Zn sulphate in lambs (VanValin *et al.*, 2018). In another study using growing heifers, no effect was observed on apparent total-tract digestibility when organic and sulfate trace minerals were compared at various levels of starch (Pino and Heinrichs, 2016).

#### Milk yield and composition

Supplementation with inorganic and organic trace

elements Zn, Cu, and Mn significantly enhanced average daily milk production ( $P<0.05$ ) over 120 days of lactation compared to the control group (Table IV). Cows fed varying levels of organic and inorganic trace elements produced significantly greater average daily milk (AMY) and fat corrected milk yield (FCMY) at 4% during the 120 days of lactation, with highest levels in G4 and G5, than G1. Moreover, treatment with ITM (G2 and G3) and OTM (G4 and G5) substantially increased fat percentages ( $P<0.05$ ) compared to G1. Protein and total solids percentages rose considerably ( $P<0.05$ ) in G5 compared to G1. All treatments had no significant effects on fat, protein, and total solid percentage (Table IV). The results are consistent with Horchanok *et al.* (2019), who observed that organic forms of Mn, Cu, and Zn significantly increased total milk yield in dairy cow diets compared to inorganic trace elements. Cows fed organic minerals rather than inorganic minerals had significantly higher milk production (Ashmead and Samford, 2004; Kinal *et al.*, 2005; Pomport *et al.* 2021). Additionally, there was an increase in milk output when organic Zn replaced inorganic Zn (ZnO) in the diet (Cope *et al.*, 2009). On the other hand, no difference in milk yield was observed between cows that received full doses of inorganic and organic minerals (Nocek *et al.*, 2006; Petrović *et al.*, 2010).

**Table II. Feed intake of cows in different experimental groups.**

Item	Experimental groups				
	G1	G2	G3	G4	G5
Total DM intake	16.40±1.61	15.55±1.47	16.30±1.52	16.30±1.37	15.17±1.52
TDN intake	10.28±0.81	9.73±0.75	10.24±0.68	10.25±0.84	9.49±0.74
DCP intake	1.78±0.25	1.69±0.21	1.78±0.24	1.78±0.21	1.64±0.22

DM, dry matter; TDN, total digestible nutrients; DCP, digestible crude protein; G1, control; G2, 100% ITM; G3, 50% ITM±5%OTM; G4, 25% TTM±75% OTM; G5, 100% OTM.

**Table III. Coefficient of digestion and nutritive value of experimental rations.**

Item	Experimental groups				
	G1	G2	G3	G4	G5
<b>Digestibility coefficient</b>					
DM (%)	65.89±1.07 <sup>c</sup>	66.91±1.05 <sup>bc</sup>	67.22±1.08 <sup>bc</sup>	69.96±1.04 <sup>ab</sup>	70.97±1.08 <sup>a</sup>
OM (%)	66.59±0.87 <sup>b</sup>	68.04±0.88 <sup>ab</sup>	68.58±0.91 <sup>ab</sup>	70.96±0.81 <sup>a</sup>	71.78±0.98 <sup>a</sup>
CP (%)	69.91±0.52 <sup>b</sup>	70.40±0.47 <sup>b</sup>	70.95±0.52 <sup>ab</sup>	70.60±0.50 <sup>ab</sup>	72.26±0.58 <sup>a</sup>
CF (%)	66.90±1.30	67.01±1.11	67.69±1.27	69.04±1.22	69.15±1.26
EE (%)	66.69±1.15 <sup>b</sup>	66.68±1.12 <sup>b</sup>	71.99±1.12 <sup>a</sup>	71.11±0.13 <sup>a</sup>	71.56±1.18 <sup>a</sup>
NFE (%)	65.86±1.51 <sup>c</sup>	67.95±1.41 <sup>bc</sup>	68.77±1.37 <sup>abc</sup>	71.90±1.45 <sup>ab</sup>	72.37±1.38 <sup>a</sup>
<b>Nutritive values</b>					
TDN	63.72±0.98 <sup>c</sup>	64.88±0.95 <sup>bc</sup>	65.97±1.02 <sup>ab</sup>	67.82±0.87 <sup>ab</sup>	68.36±0.96 <sup>a</sup>
DCP	8.88±0.14	8.97±0.14	9.04±0.12	8.92±0.11	9.11±0.11

<sup>a</sup> and <sup>b</sup>: Within a single row, the means denoted by several superscripts vary significantly at ( $P<0.05$ ). For abbreviations, see Tables I and II.



**Table IV. Milk yield and composition of cows in different experimental groups.**

Item	G1	Experimental groups			
		G2	G3	G4	G5
<b>Average daily milk yield (kg/day)</b>					
Actual milk yield	10.6±0.21 <sup>c</sup>	11.6±0.22 <sup>b</sup>	11.73±0.22 <sup>b</sup>	11.95±0.24 <sup>xb</sup>	12.5±0.25 <sup>a</sup>
4% fat corrected milk yield	10.01±0.28 <sup>c</sup>	11.03±0.30 <sup>b</sup>	11.34±0.27 <sup>ab</sup>	11.5±0.28 <sup>a</sup>	12.15±0.32 <sup>a</sup>
<b>Milk composition (%)</b>					
Fat	3.63±0.03 <sup>b</sup>	3.76±0.02 <sup>a</sup>	3.78±0.03 <sup>a</sup>	3.75±0.02 <sup>a</sup>	3.81±0.03 <sup>a</sup>
Protein	2.75±0.07	2.85±0.05	2.86±0.06	2.84±0.07	2.89±0.05
Lactose	4.32±0.05	4.40±0.03	4.34±0.03	4.42±0.04	4.45±0.05
Total solid	11.41±0.08 <sup>b</sup>	11.71±0.07 <sup>ab</sup>	11.76±0.09 <sup>ab</sup>	11.71±0.07 <sup>ab</sup>	11.84±0.08 <sup>a</sup>
<b>Component yields (g/day)</b>					
Fat	0.385± <sup>b</sup>	0.436± <sup>ab</sup>	0.443± <sup>ab</sup>	0.448± <sup>a</sup>	0.476±0.021 <sup>a</sup>
Protein	0.291± <sup>b</sup>	0.331± <sup>ab</sup>	0.335± <sup>ab</sup>	0.339± <sup>ab</sup>	0.362±0.024 <sup>a</sup>
Lactose	0.458±	0.510±	0.509±	0.528±	0.556±0.032
Total solid	1.209± <sup>b</sup>	1.358± <sup>ab</sup>	1.379± <sup>ab</sup>	1.376± <sup>ab</sup>	1.480±0.06 <sup>a</sup>

For abbreviations, see Table III.

**Table V. Cow reproductive characteristics in various experimental groups.**

Item	G1	Experimental groups			
		G2	G3	G4	G5
Postpartum 1 <sup>st</sup> oestrus interval (day)	44.5±3.2 <sup>a</sup>	47.0±3.0 <sup>a</sup>	45.0±2.9 <sup>a</sup>	39.5±3.2 <sup>ab</sup>	32.5±2.8 <sup>b</sup>
Postpartum 1 <sup>st</sup> service interval (day)	53.5±3.1 <sup>a</sup>	62.0±2.8 <sup>a</sup>	58.0±2.6 <sup>a</sup>	47.5±3.0 <sup>ab</sup>	42.7±2.8 <sup>b</sup>
Services period	55.8±5.9 <sup>a</sup>	39.0±6.2 <sup>ab</sup>	53.0±5.4 <sup>a</sup>	28.3±5.2 <sup>bc</sup>	20.0±4.5 <sup>c</sup>
Number of services per conception	3.0±0.15 <sup>a</sup>	2.20±0.12 <sup>b</sup>	2.67±0.12 <sup>a</sup>	1.75±0.09 <sup>b</sup>	2.000.14± <sup>b</sup>
Days open (day)	109.3±7.1 <sup>a</sup>	101.0±6.7 <sup>a</sup>	112±7.2 <sup>a</sup>	75.8±6.4 <sup>b</sup>	62.7±6.6 <sup>b</sup>
Total conception rate (%)	50 <sup>b</sup>	66 <sup>b</sup>	66 <sup>b</sup>	100 <sup>a</sup>	100 <sup>a</sup>

For abbreviations, see Table III.

#### Reproductive traits

The interval from calving to estrus was significantly shorter in G5 than in other groups (Table V). G1, G2, G3, and G4 had similar estrous activity, while the postpartum first service interval (PPFEI) was significantly shorter in G5. Despite these findings, treatment had a beneficial effect on the number of services given per conception (NS/C) and the days open (DO). Organic supplementation of cows in G4 and G5 significantly ( $P<0.05$ ) reduced NS/C to 2.0 and 1.75 services and DO to 75.8 and 62.7 days, respectively as compared to 2.2, 2.67, and 3.0 services and DO of 101.0, 112.0 and 105.3 days in G2, G3, and G1. However, G4 and G5 showed a similar pattern, and the differences between G2, G3, and G1 were not significant. Treatment was beneficial in terms of conception rate (CR), which was significantly ( $P<0.05$ ) higher in G4 and G5 than in other groups (Table V). Consistent with our results, dairy

cows fed OTM had a 44-day short interval from calving to estrus, decreased service per conception, shorter days open, and a higher conception rate than those fed ITM (Rabiee *et al.*, 2010; Horchanok *et al.*, 2019; Daniel *et al.*, 2020; Uchida *et al.*, 2001). In addition, supplementing amino acid chelates minerals improved the CR of first-calving beef heifers compared to the control group fed inorganic minerals (Kropp, 1990). Supplementation with organic zinc, copper, and manganese has been found to improve fertility, and improve mammary gland health (Uchida *et al.*, 2001; Kellogg *et al.*, 2003, 2004; Machado *et al.*, 2014). As a result, the livestock's economic production increases. Chelated trace metals in amino acids reduced open days and services per conception as they could raise the concentration of certain minerals in the uterine tissue (Campbell *et al.*, 1999; Nocek *et al.*, 2006). Mineral supplementation from organic sources is effective in the

treatment of reproductive diseases (Hassan *et al.*, 2011). By supplementing organic mineral sources, it is possible to improve udder health by lowering the herd's somatic cell count (Cao *et al.*, 2000).

## CONCLUSIONS

The current study demonstrated the favorable benefits of supplemented organic trace minerals of levels 75 or 100% (zinc chelate of glycine hydrate, cupric chelate of glycine hydrate, and manganese chelate of glycine hydrate) on milk yield and composition, as well as digestibility coefficient and nutritive values and reproductive performance throughout the first 120 days of lactation in Friesian cows. Organic trace minerals improved the productive and reproductive performance of Friesian cows better than inorganic minerals.

### Statement of conflict of interest

The authors have declared no conflict of interest.

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