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Effect of zinc sources on milk yield, milk composition and plasma concentration of metabolites in dairy cows

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Abstract

This study was conducted to evaluate the effects of different sources of zinc (Zn) on feed intake, milk yield, milk composition, and blood metabolites. Twenty-four dairy cows were randomly allocated to one of four treatments in a randomized complete block design. The treatments consisted of i) control diet (no zinc supplementation), ii) zinc oxide (ZnO), iii) zinc glycine (ZnGly), and iv) zinc nano (ZnN). The Zn sources were added to provide 60 mg of supplemental Zn per kg diet. There were no differences in dry matter intake, milk yield, bodyweight, and body condition score of the cows between treatments. Zinc supplementation in the form of ZnN and ZnGly decreased somatic cell count compared with the other treatments. The superoxide dismutase and plasma Zn concentrations in the cows provided ZnGly and ZnN were greater than those in the ZnO and control groups. No difference was detected between groups in biochemical and haematological parameters, except that blood urea nitrogen concentrations of cows supplemented with ZnGly and ZnN were less than for the ZnO supplemented and control cows. The results showed that nano and organic Zn sources in the diet of dairy cows were more suitable than inorganic Zn as supplements for dairy cows.

Keywords: milk composition, milk production, metabolism, somatic cell count
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Introduction

Zinc (Zn) is an essential trace mineral and plays an important role in DNA and RNA synthesis and replication and in cell proliferation (Cortinhas *et al*., 2012). NRC (2001) recommended that 40–60 mg of Zn/kg should be included in the diet of lactating dairy cows. The role of Zn in maintaining the status of the immune system has been documented (Nocek *et al*., 2006). It has been suggested that Zn deficiency could result in increased somatic cell count (SCC) and ultimately increase mastitis in dairy cows (Whitaker *et al*., 1997).

Krebs (1998) studied the effects of feeding organic sources of Zn, copper (Cu) and Selenium (Se) to dairy cows on antioxidant enzymes and SCC. They found that feeding these organic sources reduced the number of subclinical mastitis cases, but did not alter the concentrations of serum super oxide dismutase (SOD), glutathione peroxidase and ceruloplasmin. Zinc supplementation could also increase milk yield, live weight gain, and growth rate (Kellogg *et al*., 2004; Xu & Wang, 2001). Nocek *et al*. (2006) reported increased milk production in animals that received diets containing organically complexed minerals or a mixture of inorganic and organic complexed minerals. Zinc supplementation may also enhance the disease resistance and nutritional quality of livestock products (Wright & Spears, 2004; Tomlinson *et al*., 2004).

Supplemental Zn in rations for dairy cows is usually in the form of inorganic Zn (zinc oxide (ZnO) or zinc sulfate (ZnSO4)). Some studies reported that organic Zn is more readily absorbed by ruminants than inorganic Zn (Spears & Weiss, 2008; Qin *et al*., 2007). Thus, use of trace elements from organic sources (that is, complexed chelated amino acids, proteinates), which are more bioavailable (Spears *et al*., 2004) compared with those from inorganic sources, can be nutritionally important for maximizing milk production and maintaining health. Recently, a novel source of elemental Zn has been used in animal diets. The development of nanotechnology has led to the creation of Zn nanoparticles (ZnN) which have unique

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properties including great specific surface area, high surface activity, high catalytic efficiency, and strong absorbing ability (Kinal et al., 2005). The ZnN are a specially prepared mineral salt with a particle size of 1 to 100 nm. The feeding of ZnN has been shown to have greater efficacy and reduced toxicity compared to conventional Zn sources (Wang *et al*., 2006). The sudden rise in demand for ZnN has been attributed to its superior antibacterial properties when compared to Zn from conventional sources (Padmavathy & Vijayaraghavan, 2008). Dose-dependent effects of ZnN on growth performance have been observed in livestock and poultry (Hongfu, 2008; Mishra *et al*., 2014). It may also act as an antimicrobial and immune-modulatory agent in reducing the incidence of diarrhoea in piglets (Hongfu, 2008). Juncai & Zhisheng (2011) reported that using ZnN in the diet of dairy cows improved rumen bacteria growth and increased the efficiency of energy utilization.

Thus, the objective of this study was to evaluate the effects of three sources of Zn on feed intake, milk yield, milk composition, plasma concentration of metabolites, and changes in body condition score (BCS) in lactating Holstein dairy cows.

Materials and Methods

The care and use of animals for this study was approved by the Animal Welfare Committee of the University of Mohaghegh-Ardabili. Twenty-four Holstein dairy cows (8 primiparous and 16 multiparous) were used in a 12-week study. Animals were blocked based on parity, and commenced the study at approximately 31 days (SD ± 10.3) into lactation. The cows were housed in individual stalls with continual access to water. The basal diet was formulated according to NRC (2001). All cows received the same basal diet, which was predicted to supply 34 mg/kg DM of Zn per cow per day (Table 1). This diet was supplemented at a rate of 60 mg Zn/kg DM with Zn in forms of zinc oxide (ZnO), zinc glycine (ZnGly) and ZnN (particle size >99% 15–21 nm). Therefore, the treatments consisted of i) the basal without further Zn supplementation; ii) the basal diet supplemented with ZnO; iii) the basal diet supplemented with ZnGly (B-TRAXIM 2C, Transchem, Sydney, Australia); and iv) the basal diet supplemented with ZnN.

Nanoparticles of Zn were prepared using the chemical co-precipitation method that was reported by Massart (1981). To prepare the Zn nano-composite, 3.21 g zinc nitrate ($Zn(NO_3)_2$) was dissolved in 100 mL of water and mechanically stirred for 30 min. An aqueous solution of 5 M sodium hydroxide was then added dropwise and stirred into the room-temperature solution until the pH of the solution reached 10. The suspension was then refluxed for 60 min at 96 °C. The suspension was centrifuged (3000 rpm, 5 min) to remove the precipitate, washed twice with double distilled water and ethanol to remove the unreacted reagents, and dried in an oven at 60 °C for 24 hours. Scanning electron microscopy) was employed to determine the structure and morphology of the Zn nano-prepared samples (Jeol JSM-6060LV, Tokyo. Japan). The average (n = 9) particle size was 18 to 22 nm.

Animals received the basal mix ration twice daily at 09h00 and 14h00 at the rate of 105% of the previous ad libitum daily intake. Orts were removed twice weekly and weighed. Feed samples were collected twice weekly and bulked every five weeks, with a representative subsample being sent to Direct Laboratories (Tabriz, Iran) for analysis.

Cows were milked twice daily at approximately 05h00 and 17h00. Milk yield was recorded automatically at each milking, and the composition of the milk from one 24-hour period was analysed weekly with additional samples being taken fortnightly for SCC. Samples for SCC determination were analysed within 24 hours (Direct Laboratories, Tabriz, IRAN). Body condition score was scored each week based on a 5-point scale with 0.25 intervals, where 1 = thin and 5 = fat (Lowman *et al*., 1976).

During weeks 0, 4, 8, and 12 of the study, blood samples were collected two hours after the morning feeding via venipuncture from the jugular vein. Samples were collected into vacutainer tubes containing either sodium heparin or EDTA. After centrifugation at 3500 × g for 20 min at 4 °C, the plasma was removed and stored at −20 °C for later determination of enzyme activity, Zn concentration and biochemical parameters. To determine the haematology profile of the whole blood, samples were collected in Zn-free no-additive tubes, allowed to clot at ambient temperature (15–21 °C), centrifuged at 3500 × g for 10 min, with the serum being analysed after two hours. White blood cell (WBC) count, red blood cell (RBC) count, haemoglobin concentration (Hb), packed cell volume (PCV), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), and number of platelets (PLT) were recorded with an automatic haematology cell counter (Celltak, NEK6108K, Istanbul, Turkey). Lactate dehydrogenase (LDH), alkaline phosphatase (ALP), super oxide dismutase (SOD), total protein (TP), glucose (GLU), blood urea nitrogen (BUN), total cholesterol (TC), triglyceride (TG), and albumin (ALB) contents of serum samples were determined, as were concentrations of high density lipoprotein (HDL) and low density lipoprotein (LDL) using commercial kits (Pars Azmoon kits, Pars Azmoon, Tehran, Iran) and the Chemistry analyser Dirui CS-400 (Changchun, China).

Composition of the feedstuffs was determined using AOAC (1990) methods, namely dry matter (DM) (method 934.01), organic matter (OM) (method 942.01), Zn (method 969.32), and crude protein (CP) (method 954.01). Neutral detergent fibre (NDF) and acid detergent fibre (ADF) analyses were performed according to the methodology of Van Soest & Mason (1991), using α-amylase without the addition of sodium sulfite when determining NDF. Milk samples were analysed for Zn content following Cope *et al*. (2009). Milk constituents were analysed using a Milko Scan (Ekomilk Total, Stara Zagora, Bulgaria).

Table 1 Composition of the basal diet that was fed to dairy cows to assess supplementation of zinc in various forms

Ingredient (g/kg of DM)	Amount
Alfalfa hay	143.9
Corn silage	281.2
Wheat straw	9.5
Barley grain	111.7
Corn grain	180.4
Wheat bran	31.3
Sugar beet pulp	36
Molasses beet sugar	12.7
Fish meal	26.9
Extruded linseed	25.2
Soybean, meal	88.4
Soybean seeds, whole heated	24.7
Di-calcium phosphate	2.1
Calcium carbonate	4.2
Magnesium oxide	2.9
Salt	4.2
Sodium bicarbonate	8.4
Mineral and vitamin premix[a]	6.3
Chemical composition (g/kg of DM)	
Dry matter (DM)	520
Organic matter (OM)	912
Crude protein	155
Acid detergent fibre (ADF)	19
Neutral detergent fibre (NDF)	315
Net energy lactation (Mcal/kg of DM)	1.61
Zinc (mg/kg DM)	34

[a]Premix composition per kg: Ca: 100 g, P: 50 g, Na: 61 g, Mg: 17 g, Cu: 330 mg, Fe: 235 mg, Mn: 720 mg, I: 13 mg, Co: 6 mg, Se: 5 mg, vitamin A: 86,000 IU, vitamin D3: 20,000 IU, vitamin E: 770 IU

The serum Zn concentration was determined according to the method reported by Fick *et al*. (1979). Briefly, 10 ml nitric acid was added to 4 ml of plasma and the mixture was heated for 15 minutes at 45 °C, then 5 ml of concentrated nitric acid was added, and the mixture was heated to 45 °C for an additional 30 minutes. This step was repeated twice. Then 5 ml hydrochloric and 10 ml distilled water were added. This mixture was increased in volume to 50 mL by adding more distilled water filtered through a Whatman No 42 paper. The concentration of Zn in the solution was measured with a flame atomic absorption spectrophotometer (spectrAA220Variant, Perth, Australia).

The data were analysed using PROC MIXED of SAS (2002) for a randomized complete block design with repeated measurements. The variance among animals within treatment was regarded as a random error to test treatment effects. Initial values for recorded traits were considered covariates in the repeated measures model. Thus, the linear model was:

$$y_{ijkl} = \mu + b_i + t_j + cow_{k(ij)} + p_l + tp_{jl} + e_{ijkl},$$

where: y_{ijkl} = an observation from the from the k[th] cow in the i[th] block (b_i) that was subjected to the j[th] treatment (t_j); μ = the mean which was common to all observations; p_l = the l[th] time at which the observation was recorded; tp_{jl}= the interaction of treatment and time; and e_{ijkl}= the residual variation within animals, which served as error for the effects of time and the interaction of treatment with time. Differences between means were tested using the LSMEANS statement in SAS.

Results

No effects of Zn source or the interaction of Zn source and time on DMI, milk yield, and milk composition were detected (Table 2). However, there were significant effects of time on DMI and milk yield, with an increase in milk yield being observed from the third to the eighth week of lactation. The concentration of Zn in milk did not differ between dietary treatments. Cows that received ZnGly and ZnN had reduced (*P* <0.05) SCC compared with those fed the control and ZnO diets. Somatic cell count decreased as lactation progressed (*P* <0.05). Effects of zinc source, time, and the interaction of Zn source and time on BW and BCS were not detected.

Table 2 Effects of the form of zinc in supplements provided to Holstein dairy cows on feed intake and milk production

Traits[2]	Treatment[1]				SEM	Probability		
	C	ZnO	ZnGly	ZnN		Source	Time	Source × Time
DMI (kg/d)	22.8	22.8	23.2	23.6	0.2	0.39	0.01	0.63
Milk yield, (kg/d)	37.9	37.3	38.7	38.2	0.4	0.07	<0.01	0.76
Fat (g/kg)	30.1	30.5	30.1	30.1	0.3	0.76	0.87	0.57
Fat (kg/d)	1.14	1.14	1.162	1.15	1.41	0.60	0.13	0.45
Protein (g/kg)	30.6	31.0	31.9	31.1	0.6	0.09	0.95	0.28
Protein (kg/d)	1.16	1.16	1.23	1.19	1.56	0.49	0.17	0.29
Lactose (g/kg)	45.7	46.3	45.9	45.5	0.38	0.49	0.08	0.29
Lactose (kg/d)	1.73	1.73	1.78	1.74	2.30	0.41	0.37	0.23
Zn (mg/kg)	3.07	3.07	3.13	3.18	0.08	0.70	0.24	0.34
SCC (10^3/ml)	153.7[b]	150.0[b]	142.5[a]	140.2[a]	1.5	0.01	<0.01	0.12
SNF (g/kg)	80.3	79.5	78.8	78.1	0.5	0.04	0.75	0.87
SNF (kg/d)	3.05	3.00	2.81	3.05	2.99	0.32	0.13	0.29
BW (kg)	632	642	641	638	6	0.67	0.12	0. 23
[1]BCS	3.31	3.32	3.24	3.26	0.42	0.73	0.10	0.37

[1]C: control; ZnO: zinc oxide supplement, ZnGly: zinc glycine supplement; ZnN: zinc nanoparticle supplement
[2]DMI: dry matter intake; Zn: zinc; SCC: somatic cell count; SNF: solids not fat; BW: bodyweight; BCS: body condition score, recorded on a 5-point scale (Lowman *et al*., 1976) with 0.25 intervals, where 1 = thin and 5 = fat
[a,b] Within a row, means without a common superscript letter differ with *P* <0.05

The results of the haematological characterization are shown in Table 3. There was no significant effect of time, treatment, or time and treatment interaction on red and white blood cell-related variables. The activity of SOD was greater for cows that were supplemented with ZnN and ZnGly than for those fed the basal diet alone or supplemented with ZnO (*P* <0.05). Differences between treatments were not detected for ALP, TP, GLU, ALB, TG, TC, HDL, and LDL. However, BUN concentration in plasma decreased with ZnN and ZnGly supplementation (*P* <0.05). The Zn serum concentrations of Zn were greater for cows that were supplemented with ZnN and ZnGly compared with those that were supplemented with ZnO and those fed the basal diet (*P* <0.01). Highly significant effects of time, which affected Zn concentration in plasma, were also observed, as were similarly significant effects on all of the indicators of metabolic status.

Table 3 Effects of the form of zinc in supplements provided to Holstein dairy cows on haematology, serum enzyme activities and indicators of metabolic status in dairy cows

Trait[2]	Treatments[1]				SEM	Probability		
	C	ZnO	ZnGly	ZnN		source	Time	Source × Time
WBC (10^3/ml)	8.59	8.77	9.09	8.85	0.17	0.22	0. 32	0.07
Lymphocytes (10^3/ml)	2.36	2.33	2.32	2.23	0.06	0.40	0.25	0.06
Monocytes (10^3/ml)	0.51	0.62	0.74	0.78	0.06	0.22	0.69	0.99
Granulocytes(10^3/ml)	5.74	5.78	5.95	5.72	0.12	0.56	0.48	0.33
RBC (10^6/mm^3)	6.13	6.14	6.09	6.11	0.03	0.94	0.22	0.79
HGB (g/dl)	9.18	9.21	9.16	9.16	0.03	0.44	0.06	0.72
HCT (%)	28.1	28.9	28.1	28.1	0.1	0.81	0.33	0.79
PLT (10^3/mm^3)	452	434	430	442	12	0.59	0.34	0. 31
MCV (fl)	46.0	46.0	46.1	46.0	0.1	0.98	0.07	0.65
MCH (pg)	15.0	15.1	15.0	15.0	0.1	0.75	0.27	0.85
MCHC (g/dl)	32.6	32.8	32.6	32.7	0.1	0.36	0.20	0.56
Lactatedehydrogenas (IU/l)	1796	1812	1835	1827	15	0.99	0.02	0.81
Super oxide dismutase (IU/l)	2146[b]	2149[b]	2170[a]	2187[a]	13	0.02	0.01	0.08
Alkaline phosphatase (IU/l)	107.3	108.1	109.2	108.3	0.82	0.44	0.001	0.74
Cholesterol (mg/dl)	259.8	258.4	258.4	258.1	1.1	0.68	<0.001	0.79
Triglyceride (mg/dl)	16.21	16.48	16.04	15.98	0.52	0.34	<0.001	0.002
High density lipoprotein (mg/dl)	135.4	134.4	134.9	133.9	0.7	0.46	<0.001	0.38
Low density lipoprotein (mg/dl)	130.1	126.8	128.1	129.7	1.5	0.40	<0.001	0.0398
Blood urea nitrogen (mg/dl)	17.0[a]	16.5[a]	15.9[b]	15.5[b]	0.3	0.006	<0.001	0.15
Albumin (mg/dl)	3.53	3.48	3.76	3.83	0.15	0.32	<0.001	0.16
Glucose (mg/dl)	57.8	57.6	57.8	56.9	0.5	0.59	<0.001	0.08
Total protein (mg/dl)	6.48	6.66	6.62	6.59	0.12	0.73	<0.001	0.57
zinc (µg/ml)	1.21[b]	1.83[b]	2.45[a]	2.33[a]	1.05	0.002	0.006	0.88

[1]C: control; ZnO: zinc oxide supplement, ZnGly: zinc glycine supplement; ZnN: zinc nanoparticle supplement
[2]HGB: haemoglobin concentration; HCT: haematocrit; MCH: mean corpuscular haemoglobin; MCHC: mean corpuscular haemoglobin concentration; MCV: mean corpuscular volume; PCV: packed cell volume; PLT: platelet concentration; RBC: red blood cell count; WBC: white blood cell count
[a,b]Within a row, means without a common superscript letter differ at P <0.05

The interaction of Zn sources and time affected concentrations of LDL and TG in the plasma (Table 4). The concentration of LDL in plasma was reduced (P <0.05) in weeks 8 and 12 in the cows that received ZnGly and ZnN compared with those that were fed the C diet. In comparison with the cows fed the basal diet, plasma TG concentration were significantly decreased in cows that received the ZnGly and ZnN supplements from week 4 to week 12. Cows that were supplemented with ZnO had reduced plasma triglyceride levels only at weeks 8 and 12, compared with the C diet.

Table 4 Effects of the interaction between the form of zinc in supplements provided to Holstein dairy cows and week of lactation on serum triglyceride and serum low density lipoprotein concentrations (mg/dl)

Diet[1] Time	Triglyceride[2]			Low density lipoprotein[3]		
	Week 4	Week 8	Week 12	Week 4	Week 8	Week 12
C	16.12[a]	16.11[a]	16.11[a]	129.15	131.19[a]	130.25[a]
ZnO	16.11[ab]	16.09[b]	16.08[b]	129.56	128.11[ab]	127.11[ab]
ZnGly	16.05[c]	16.03[c]	16.01[c]	128.73	125.5[b]	123.35[b]
ZnN	16.06[c]	16.05[c]	16.02[c]	129.84	127.8[b]	125.64[b]

[1]C: control; ZnO: zinc oxide supplement, ZnGly: zinc glycine supplement; ZnN: zinc nanoparticle supplement
[2]SEM = 0.11, *P* = 0.002
[3]SEM = 0.908, *P* = 0.040
[a,b,c]Within a variable means without a common superscript letter differ at *P* <0.05

Discussion

The failure of supplemental Zn to affect DMI that was observed in the current study is in contrast with the results of Miller *et al*. (1985), who reported that increasing Zn concentrations reduced the DMI of lactating dairy cows. As in previous studies, DMI increased through early lactation. After calving, voluntary feed intake increases rapidly due to the rise in demand for nutrients that are targeted for lactation (Cortinhas *et al*., 2012). Grant *et al*. (1995) also found that the DMI increased approximately 1.5 to 2.5 kg/week during the first three weeks of lactation.

Previously reported effects of types of trace mineral supplementation on milk yield are variable. Kinal *et al*. (2005) and Cope *et al*. (2009) reported an increase in milk production of dairy cows by feeding diets that contained organically complexed minerals or a mixture of inorganic and organically complexed minerals. Rajerden *et al*. (2013) supplemented Holstein Friesian crossbred cows with nano-zinc oxide and observed that milk yield increased compared with control. However, some studies (Campbell *et al*., 1999; Uchida *et al*., 2001) found that the form of supplemental Zn had no effect on milk yield, in agreement with the results of the current study. In the present study, milk composition was not affected by various forms of Zn in the diet, verifying the reports of Cope *et al*. (2009) and Nocek *et al*. (2006). The form of Zn had no effect on the concentration of Zn in milk, either, which is consistent with the results of Nocek *et al*. (2006). The concentration of Zn in milk was also found to be constant with varying concentrations of dietary Zn (Cope *et al*., 2009). It has been suggested that Zn flux in the mammary gland of dairy cows is strictly regulated to maintain an adequate supply of Zn to the neonate (Kelleher & Lönnerdal, 2005).

The increase in the SCC for cows that received low levels of Zn may be attributed to a decrease in leukocyte function, which can lead to an increase in the susceptibility of the mammary gland to bacterial infection (Song et al., 2010). Here, cows that were fed the ZnGly and ZnN diets had reduced SCC concentrations compared with those fed control and ZnO diets. The antimicrobial properties of ZnN nanoparticles are associated with their specific high surface, area to volume ratio, and shape and size of particles (Arabi *et al*., 2012; Auffan *et al*., 2009). Tong *et al*. (2013) suggested that the size of 1–100 nm Zn nanoparticles could increase their antimicrobial activity owing to exposure of the polar surface plane of ZnO to the negatively charged bacterial cell membrane. Given the size of Zn nanoparticles that were used in present study (15–21 nm), low SCC was expected for cows that received the ZnN treatment. The addition of Zn nanoparticles has been reported to generate reactive oxygen species causing oxidative stress to bacterial cells (Feris *et al*., 2010; Kumar *et al*., 2011). Additionally, Zn nanoparticles may cause electrostatic disruption of the outer membrane and interruption of normal cell membrane functionality, resulting in the leakage of intracellular materials (Liu *et al*., 2009). The solubility of Zn may also affect metabolic activity and inhibit the growth of bacterial cells (Song *et al*., 2010). The results of the current study agreed with the findings of Rajerden *et al*. (2013) and Kinal *et al*. (2005), who recorded decreased SCC when cows were fed nano and organic chelated Zn, respectively. In contrast, Cope *et al*. (2009) observed no effect of Zn form on the SCC of dairy cows. However, Cope *et al*. (2009) had lower initial SCC, (means ≈ 130000 cells/mL) compared with the present study (mean = 152,000 cells/mL). Thus, it can be speculated that the initial level of SCC may be an important factor in beneficial effects from supplemental inorganic or organic Zn.

In agreement with Cope *et al*. (2009), who supplemented Zn at 66% and 100% of the recommended level (NRC, 2006), the form of dietary Zn had no effect on BW or BCS. Whitaker *et al*. (1997) and Uchida *et al*., 2001 found that the form of Zn supplemented to the diet did not appear to affect BW or BCS in dairy

cows, respectively. Similarly, Nocek *et al*. (2006) reported no significant effects on BW or BCS when inorganic Zn or organically chelated Zn was supplemented to cows.

In the present study, the form of dietary Zn had no effect on blood haematology. However, the activity of SOD in cows supplemented with ZnGly and ZnN was greater than was found in the cows that were fed the basal diet or were supplemented with ZnO. Similarly, when Xu & Wang (2001) supplemented pigs with 3000 mg Zn oxide/kg diet activity of the SOD enzyme increased compared with pigs that were fed a control diet.

In this study, plasma BUN concentration decreased with ZnGly and ZnN supplementation. These results are consistent with Shakweer *et al*. (2010) and Shakweer *et al*. (2005), who found that Friesen calves whose diets were supplemented with zinc sulphate or zinc methionine at 45 mg/kg DMI had decreased serum urea concentration. There were no significant effects of dietary treatment on plasma concentrations of GLU, TP, ALB, TG, TC, HDL, and LDL. Sobhanirad *et al*. (2009) did not find any effects of organic and inorganic Zn supplementation on indicators of metabolic status. The absence of treatment effects on these indicators implies that Zn supplements do not influence synthesis of protein and fat metabolism in animals (Cortinhas *et al*., 2012). However, the observed time effects that showed increased concentrations of TC, HDL, GL, and TP, and gradual reductions in concentrations of TG, BUN, and ALB during lactation may be associated with physiological processes that were concurrent with the experiment.

Kinal *et al*. (2005) and Spears *et al*. (2008) reported an increase in blood plasma Zn when animals were supplemented with organically chelated minerals compared with those fed inorganic (sulphate) minerals. Their results are consistent with data obtained in the current study. However, Cope *et al*. (2009) reported no change in plasma Zn concentration in dairy cows that were fed diets supplemented with organic and inorganic Zn. Therefore, since blood Zn concentration is a common Zn index in the clinical field, increased concentration in the blood may be deemed a suitable tool to evaluate the Zn status of the body.

Conclusion

In general, supplementation of Zn in organic, inorganic and nano forms had no significant effect on DMI, milk yield, milk composition and blood haematology in early lactation cows. These data indicate reduced SCC in milk of animals that were supplemented with ZnGly and ZnN. This decreased in the SCC of milk confirms the antimicrobial properties organic and nano treatments.

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Authors' Contributions

SB collected the data and FM drafted the manuscript. AT designed the study, JS and BN analysed the data. All authors approved the final manuscript.

Conflict of Interest Declaration

The authors have no financial, personal or other relationships with other people or organizations that could inappropriately influence or perceived to influence their work.

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