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Comparative Efficacy of Nano Zinc with Inorganic Zinc on Nutrient Digestibility and Mineral Availability in Barbari Goats

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ABSTRACT

Present experiment was conducted to compare the effect of nano and inorganic Zinc supplementation on feed intake, nutrient utilization, growth performance and mineral availability in Barbari bucks. Twenty four Barbari bucks (1.5-2 years, 35±2.84 kg) were randomly allocated into four groups (Gr 1, Gr 2, Gr 3 and Gr 4) having six bucks in each groups and fed for 90 days. Nutrients requirement of experimental bucks was met by feeding concentrate mixture, gram straw and corn silage as basal diet. Gr 1 fed on basal diet with no supplementation served as a control. Treatment groups (Gr 2, Gr 3 and Gr 4) were supplemented, 20.0 ppm inorganic zinc, 20.0 and 10.0 ppm of nano zinc respectively. Experimental bucks were monitored daily for DM intake (DMI) and fortnightly body weight change. At the end of the study, a digestion trial of 7 days was conducted to study the effect of inorganic and nano zinc supplementation on nutrients utilization. The results revealed no significant ($P>0.05$) difference in body weight change, feed intake and nutrient digestibility in the experimental groups fed on either inorganic or nano form of supplemental Zn. No significant ($P>0.05$) effect on Ca, P, Cu and Fe intake and absorption (%) was observed on dietary supplementation of nano and inorganic Zn. However, the retention of Zn (mg/d) was significantly higher in G3 than G4 and G2 groups indicating better bioavailability of zinc when supplemented in nano form at 20 ppm level in Barbari bucks.

KEY WORDS: Mineral Availability, Nano zinc, Nutrient Utilization

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INTRODUCTION

Zinc (Zn), is a second most prevalent trace mineral in the body and is essential for normal physical, physiological and disease-free status of an animal (Swain et al., 2016). It act as a co-factor for many metalloenzymes that have a vital role in nutrient metabolism, reproduction, growth, antioxidant ability and immunity of animals. Zn has critical role in various physiological functions of body and is directly or indirectly involved in growth, production and reproduction (Bhalakiya et al., 2019). In India, 25% of livestock are deficient in zinc (Goklaney et al., 2019) and it is well established that deficiency of Zn can affect growth, reproduction, immune system and gene expression in ruminants (Underwood and Suttle, 1999). However, lower storage, poor bioavailability and interaction with other minerals makes the deficiency more critical and hence a continuous supply of Zn through diet is required. Traditionally, Zn is commonly supplemented as inorganic salts but owing to its lower bioavailability higher levels of Zn are excreted from the animal that have raised concerns pertaining to environmental pollution (Feng et al., 2009). Thus, this problem opens a window for utilization of better bio- available Zn sources and if possible, to reduce the supplemental dose of Zn to the animal feed. Several technologies are adopted to increase the bioavailability of the minerals. Among all the probable approaches, use of nanotechnology to produce nano minerals is a novel and potential alternate to other conventional used sources. Nano particles exhibit unique properties in terms of chemical, physical, photo-electrochemical and electronic properties when compared to their respective bulk materials. Use of nanotechnology to produce nano sized Zn called as nano Zn (NZn) is a potential alternative to both organic and inorganic Zn sources. Nano minerals, owing to increased surface area showed improved bioavailability. The use of NZn has shown to produce better results as compared with conventional Zn sources and is also less toxic (Wang et al., 2006; Sahoo et al., 2014). Recently, some researchers have suggested positive effects of nano-ZnO (Swain et al., 2016; El Sabry et al., 2018; Abdollahi et al., 2020) on the performance and health of livestock. Due to its smaller size and higher surface-to-volume ratio (Swain et al., 2016) resulting in the high absorbability, surface activity and catalytic efficiency, it may be hypothesised that NZn can be more bioavailable and accordingly its lower dose can also be sufficient for posing beneficial effect on performance of bucks. Therefore, the present study was designed to evaluate the effects of NZn compared to inorganic Zn on the feed intake, nutrient utilization and minerals availability in Barbari bucks.

MATERIALS AND METHODS

The required animal care procedures were approved (approval number, IAEC/21/22),and conducted under the established standard of the Institutional Animal Ethics Committee (IAEC), A total of 24 growing Barbari bucks were selected from the Buck Unit, maintained at Goat Farm Complex, DUVASU, Mathura. Experimental bucks were randomly assigned into four groups (N=6) on body weight (35±2.84) and age (1.5 to 2 year) basis, and the duration of the experiment lasted 90 days. The animals of each experimental group were maintained and fed individually on roughage and concentrate based ration to meet out requirement as per NRC (2007) feeding standard. The nutrient requirements of experimental bucks were met by feeding total mixed ration (TMR) consisted of concentrate and roughage in the proportion of 30:70 respectively. The roughage part is composed of Gram straw (46.66 %) and corn silage (23.34 %). Concentrate mixture was prepared by mixing barley grain, Maize grain, wheat bran, gram chunni, mustard oil cake, cotton seed cake and mineral mixture (without Zn) in 15, 15, 25, 10, 16.5, 16.5 and 2 parts, respectively. The animals were given an adaptation period of fifteen days, before the start of experiment. All the groups were kept on similar feeding regimen, except different sources and dose of Zn that was additionally supplemented to the treatment groups. Bucks either received a basal diet devoid of supplemental Zn(Gr 1) or were supplemented with 20 ppm of inorganic Zn (Gr 2) as zinc oxide (Thermo Fisher Scientific India Pvt Ltd, Powai, Mumbai, India); 20 ppm (G 3) and 10 ppm of nano Zn (G 4) as Zinc oxide Nanopowder Type- I (Sisco Research Laboratories Pvt Ltd (SRL), Taloja, MH, India). To ensure the required intake, the calculated amount of Zn was premixed in maize flour that differed in Zn source and dose and was offered to each buck prior to feeding. TMR was prepared daily by hand mixing and was offered at 09:00 h and 18:00 h during entire trial period. Clean and fresh drinking water was offered *ad libitum* twice to each animal daily. All bucks were housed in a well-ventilated individual sheds having the proper arrangement for feeding and watering. Feeds and fodders consumption was recorded daily by weighing feed offered and orts left, and dry matter intake (DMI) was calculated and recorded daily according to the DM content of the diet. Body weight of the experimental animals was recorded at the start of experiment, and then fortnightly using computerized weight management system (Leotronic Scales Pvt. Ltd., India) 2 consecutive days before feeding. The average of two days was considered as body weight for that fortnight and considered for body weight changes

The representative samples of feeds and fodders offered and orts left were dried in a hot air oven at 60°C till a constant weight was attained and then ground in a Wiley mill to pass a 1-mm sieve. The samples were analyzed for proximate composition using standard procedures (AOAC, 2005). Neutral detergent fiber (NDF) and acid detergent fiber (ADF) were determined according to the procedures described by Van Soest et al.(1991). Mineral content in samples of feeds and fodders, orts left, and fecal samples were analyzed by Inductively Coupled Plasma-Optical Emission Spectroscopy (5800 ICP-OES, Agilent, CA, USA). The wavelengths (nm) used were 213.857nm for Zinc, 238.204nm for Iron, 327.395nm for Copper, 422.673nm for Calcium and 213.618nm for phosphorus. The instrument conditions were 12 L/min plasma gas flow, 0.7 L/min nebulizer gas flow, 1 L/min Aux flow and the viewing mode was axial at 8 mm height for analysis of the minerals. All the samples were run in triplicate. Ingredients and chemical composition of the basal diet fed during the experimental period is depicted in Table 1. The Zn content of basal diet without supplemental zinc was estimated at 28.84 ppm.

Statistical Analysis.

The data was analyzed using the general linear model (GLM) procedure of statistical package for the social sciences (SPSS for windows, V21.0; Inc., Chicago, IL, USA) as a randomized block design with animal as the experimental unit. The effect of treatments on performance, nutrients digestibility and mineral metabolism was analyzed by using one-way ANOVA with significance defined at P<0.05. The pair-wise comparison of means was carried out using "Tukey's honest significant difference (HSD) test".

Table 1. Ingredient and chemical composition of total mixed ration fed to experimental bucks

Ingredients composition (%)	
Gram straw	46.66
Corn silage	23.34
Barley grains	4.5
Maize grain	4.5
Mustard oil cake	4.95
Cotton seed cake	4.95
Gram Chunni	3.0
Wheat Bran	7.5
Mineral mixture	0.6
Zinc supplement[a]	Variable
Chemical composition (% DM basis)	
Dry Matter	78.9
Crude Protein	12.1
Ether extract	2.87
Neutral detergent fiber	59.2
Acid detergent fiber	42.7
Calcium (%)	1.01
Phosphorus(%)	0.27
Zinc, ppm	28.8

aIn G 2 group- 20.0 mg/kg Zn as inorganic zinc oxide, G 3 and G 4 group 20.0 and 10.0 mg/kg Zn as nano zinc oxide, respectively

RESULTS AND DISCUSSION

The effect of inorganic and nano Zn supplementation on dry matter intake and body weight gain growth performance are presented in Table 2. Dietary supplementation of inorganic and nano zinc in Barbari bucks had no significant effect on DMI. Percent DMI (kg/100 kg BW) in different groups showed similar effect of inorganic and nano Zn supplementation. The difference between the groups for mean ADG (g) was also not significantly different (P>0.05). FCR, used as feed efficiency measures was not significantly different (P>0.05) between control and treatment group. The current results are in agreement with findings of Zaboli et al. (2013) who observed no significant differences in ADG, DMI and FCR on supplementation of 20 and 40 ppm of zinc oxide nanoparticles in Markhoz goat kids. Maan and Sihag (2014) also reported that

Zn supplementation through different sources (ZnO and $ZnSO_4$) at 45 ppm did not affect the feed (DM) intake in goat kids. Dhruw (2017) reported that feeding of nano-Zn and up to 40 ppm level in the diet had no effect on DMI in goat kids. Similarly, supplementation of Zn to a basal diet containing more than 25 mg Zn/kg DM had no effect on DMI in dairy goats (Salama et al., 2003), growing lambs (Garg et al., 2008) and beef steers (Mandal et al., 2007). Supplementation of inorganic zinc at 50 ppm and nano zinc at 50 and 25 ppm, had no significant effect on total dry matter intake in goats (Swain, 2017). However, the present results are not in agreement with the findings of Fadayifar et al. (2012) who reported that Zn supplementation at 20 mg Zn/kg DM improved performance in terms of average daily gain, body weight gain and feed efficiency of lambs.

Table 2. Effect of inorganic and nano zinc supplementation on dry matter intake and body weight gain

Parameters	Supplemental Zn (mg/kg DM)				SEM	P value
	Gr 1	Gr 2	Gr 3	Gr 4		
DMI (kg/day)	1.38	1.41	1.43	1.42	0.027	0.633
DMI (% BW)	3.36	3.40	3.39	3.37	0.029	0.720
ADG (g/day)	57.78	62.78	60.19	64.44	2.261	0.175
FCR	26.20	23.47	25.17	23.16	1.166	0.212

DMI, dry matter intake; ADG, average daily gain; FCR, feed conversion ratio

The data pertaining to nutrient intake and digestibility coefficient of Control and treatments group have been presented in Table 3. In present study, influence of Zn supplementation on DM intake and apparent digestible parameters showed no significant difference on DMI, nutrient digestibility and as well as total digestible nutrients (P>0.05). The mean daily DM, OM and CP intake was found to be similar among the control and Zn supplemented groups, which indicated that supplementation of inorganic and nano zinc had no effect on palatability and feed intake pattern of the bucks. The digestibility of organic nutrients and TDN was also found to be similar (P>0.05) among the treatment groups suggesting that supplementation of Zn through inorganic and nano sources had no effect on the digestibility of these nutrients. The results of most researchers are in consistent with the present findings, Swain, (2017) reported no significant differences (P>0.05) in intake of different nutrients (DM, OM, CP, DOMI, DCPI, TDNI (g/d and g/kg W0.75) due to supplementation of inorganic or nano form of zinc compared to control in goats. Singh et al. (2018) observed no significant difference in intake and digestibility coefficient of the nutrients between ZnO and nano-ZnO at 60 ppm supplemented lamb groups. Supplementation of organic Zn (ZnProt) at levels of 15, 30 or 45 ppm had no effect on digestibility of DM, OM, CP, EE, and fibre fractions in Nellore ram lambs as compared to inorganic Zn (ZnSO4) (Nagalaksmi and Himabindu, 2013). Dhruw, (2017) reported that supplementation of nano-Zn up to 40 ppm level in the diet had no effect on intake and digestibility of DM, OM, CP, EE, NDF and ADF in goats. Similar observations have been reported earlier (Mandal et al., 2007; Garg et al., 2008; Jia et al., 2008; Jia et al., 2009) in different species of animals, who reported no effect on intake and digestibility of nutrients on zinc supplementation. Also, Wenbin et al. (2008) found that digestibility of DM, CP, (NDF) and (ADF) did not differ (P>0.05) among treatments groups that supplemented with 30 and 45mg Zn/kg DM for goats. However, in contradiction to our observations, Habeeb et al. (2013) found that intake and digestibility of CP, CF, EE and NFE were significantly higher in goats fed diets containing 30 and 80 ppm Zn. Mann and Sihag (2014) also reported that Zn supplementation (ZnO and $ZnSO_4$) at 45 ppm improved the digestibility of CP, EE and NFE in goat kids.

Table 3. Effect of inorganic and nano zinc supplementation on nutrient intake and nutrient digestibility (% or as mentioned) during digestion trial

Attributes	Supplemental Zn (mg/kg DM)				SEM	P value
	Gr 1	Gr 2	Gr 3	Gr 4		
Initial Wt.(kg)	44.3	45.0	45.3	45.6	2.747	0.990
Final Wt. (kg)	44.8	45.5	45.8	46.1	2.717	0.988
Wt. gain(kg)	0.48	0.50	0.53	0.52	0.066	0.956
DM intake (kg/day)	1.29	1.31	1.33	1.35	0.023	0.792
DM intake (kg/kg $W^{0.75}$/ day)	0.62	0.63	0.65	0.67	0.021	0.872
DMI (% B.wt)	3.10	3.12	3.13	3.22	0.032	0.933
CP intake (kg/day)	0.16	0.15	0.15	0.16	0.011	0.916
DCP intake (kg/day)	0.10	0.10	0.10	0.11	0.009	0.762
DCP intake (g/kg $W^{0.75}$)	5.81	5.93	6.04	6.23	0.378	0.761
TDN intake (kg/day)	0.60	0.60	0.63	0.68	0.285	0.799
TDN intake (g/kg $W^{0.75}$)	33.0	33.7	36.2	38.9	2.766	0.442
Digestibility of nutrients (%)						
DM (%)	59.3	62.4	59.2	61.1	2.010	0.644
OM (%)	73.2	74.3	72.1	72.7	2.075	0.888
CP (%)	67.5	68.3	68.9	69.0	1.960	0.949
CF (%)	51.2	51.7	54.1	55.4	2.899	0.712
EE (%)	82.1	84.8	84.5	85.2	1.602	0.533
NFE (%)	58.	58.0	61.4	61.8	2.976	0.757
NDF (%)	60.1	62.7	62.6	62.6	2.800	0.893
ADF (%)	52.5	50.7	54.6	53.8	2.807	0.774

The data regarding Ca, P, Cu, Zn and Fe intake, absorption and absorption (%) observed during digestion trial has been presented in Table 4. The Zn intake(mg/d) differ significantly ($P<0.05$) due to additional supplementation of Zn in treatment groups. Zn intake was significantly ($P<0.05$) higher in G 2 and G 3 group than G 4 and control group. Intake of Zn also varied significantly ($P<0.05$) in G 4 and control. The absorption of Zn was significantly higher in G 3 than control and other treatment groups whereas; the absorption (%) of Zn was significantly higher in control than other treatment groups. However, the net absorption of Zn was highest in G 3 group compared to other groups. The present findings are in agreement with the findings of Jia et al. (2009) who reported higher absorption of Zn in control group compared to the treatment group in Cashmere goats. The deficiency of Zn in control group leads to higher absorption of Zinc by altering the homeostatic mechanism. Highest values of net absorbed Zn in G 3 compared to G 4 and G 2 groups indicates better bioavailability of nano zinc than inorganic zinc. Half the dose of nano Zn is comparable to inorganic Zn. However, the net absorbed Zn was higher in G 3 but the absorption (%) did not differ significantly in inorganic and nano supplemented groups. The present results showed no significant effect of inorganic and nano Zn supplementation on absorption of Ca, P, Cu and Fe. In consistent with our results Jadhav et al.(2008) also reported that the intake of Ca and P, their excretion through faeces and urine and balances were also similar ($P>0.05$) among the three groups supplemented with 0, 35 and 70 ppm of Zinc sulphate in male buffalo calves. Also, Swain, (2017) reported no significant variation in Ca, P and Mg intake (g), absorbed (g/d) and absorption (%) in goats supplemented with inorganic and nano zinc.

Table 4. Effect of inorganic and nano zinc supplementation on mineral absorption

Attributes	Supplemental Zn (mg/kg DM)				SEM	P value
	Control (Zn0)	IZn20	NZn20	NZn10		
Calcium						
Intake (g/day)	1.33	1.30	1.35	1.32	0.11	0.766
Absorbed (g/day)	0.43	0.41	0.43	0.42	0.27	0.174
Absorption (%)	32.4	31.3	32.0	32.3	2.14	0.417
Phosphorus						
Intake (g/day)	0.42	0.45	0.48	0.47	0.19	0.690
Absorbed (g/day)	0.23	0.23	0.24	0.24	0.43	0.571
Absorption (%)	51.7	52.3	53.4	52.7	1.70	0.778
Zinc						
Intake (mg/day)	38.1^{c}	88.3^{a}	89.5^{a}	53.5^{b}	9.11	0.060
Absorbed (mg/day)	12.2^{b}	16.5^{b}	23.5^{a}	13.3^{b}	2.53	0.020
Absorption (%)	30.1^{a}	20.1^{b}	26.6^{b}	23.0^{b}	2.82	0.014
Copper						
Intake (mg/day)	11.2	12.4	11.51	12.2	2.19	0.520
Absorbed (mg/day)	1.13	1.34	1.23	1.32	0.32	0.954
Absorption (%)	10.1	11.1	10.64	11.1	1.78	0.253
Iron						
Intake (mg/day)	421	415	428	424	11.3	0.219
Absorbed (mg/day)	115	117	116	117	2.13	0.109
Absorption (%)	27.6	28.1	29.1	29.4	2.6	0.150

$^{a, b, c}$means with different superscript in a row differ significantly. ($P<0.05$)

CONCLUSION

On the basis of the present study, it may be concluded that dietary supplementation of Zn either from inorganic or nano source did not exert any adverse effect on, nutrient intake, digestibility and the absorption of minerals *viz.* Ca, P, Fe and Cu except Zn. Significantly higher bioavailability from nano source of zinc was observed as compared to inorganic source.

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