Introduction
Optimizing and updating nutrient requirements of poultry is a continuous process because of improved performance, variations in nutrient availability, interaction of different nutrients at sites of absorption and metabolism, and to achieve specialized designer products. Trace minerals are essential feed additives in the diets of broilers to ensure better health and productivity. Zinc (Zn) is an essential trace mineral for normal development of animal, and is involved in a variety of metabolic processes (Salim et al., 2008) \(^1\). It requires a regular supply in poultry diet for normal appetite, growth, skeletal developments, skin and feather integrity, reproduction, immune competence and many metabolic processes (O’Dell, 2000) \(^2\). It is an integral part of more than 300 enzyme systems that are involved in metabolism of energy, protein, and nucleic acids (Tabatabaie et al., 2007) \(^3\) and in numerous biological processes (Vallee and Auld, 1990) \(^4\). As it not stored in body, a continuous dietary intake is essential for the body’s optimum physiological functions (Zalewski et al., 2005) \(^5\). Zinc oxide (ZnO) and zinc sulphate (ZnSO\(_4\)) are two preferred sources of zinc as feed additives in broiler chicken diet (Wedekind et al., 1992) \(^6\). Zinc methionine (Zn-Met), an organic source of zinc is also widely preferred as feed additive (Richards et al., 2010) \(^7\). Nano zinc oxide (nZnO) is a new form of mineral presentation that has been produced and marketed using concepts of nano science and technologies (Song et al., 2010) \(^8\). The transition from micro particles to nano particles involves an increment of surface area, among other alterations in properties. Hence a larger surface area of the nano particles allows higher interactions with other organic and inorganic molecules (Nel et al., 2009) \(^9\). The nano Zn particles can thus effectively fulfill the requirement in poultry birds, promote growth rate and feed efficiency. With low use dosage, they can replace antibiotics as growth promoters, eliminate the residue of the antibiotics in the animal products, reduce the environmental contamination and produce pollution-free animal products.

Growth, feed conversion ratio and nutrient metabolizability of broiler chicks fed on inorganic, organic and nano zinc supplemented diets

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Abstract
The present study was planned to investigate the possible effects on the growth, feed conversion ratio and nutrient metabolizability of broiler chickens when supplemented with different sources of zinc. One hundred and sixty day-old broiler birds were equally and randomly distributed into four treatment (T\(_1\), T\(_2\), T\(_3\) and T\(_4\)) groups which subdivided into four replicates containing ten birds in each, reared on deep litter system up to 6 weeks of age. T\(_1\) group was kept as control and offered maize-soybean meal based diet without any source of zinc in mineral mixture. Treatment groups T\(_2\), T\(_3\) and T\(_4\) were supplemented with inorganic zinc, organic zinc and nano zinc, respectively. Nano zinc oxide fed groups had significantly (P< 0.05) higher average feed intake as compared with control group. Average body weight gain and feed conversion ratio in T\(_4\) group were significantly (P<0.05) higher as compared to other treatment groups. It was observed that the nutrient metabolizability in terms of dry matter metabolizability, nitrogen retention and gross energy metabolizability increased significantly (P<0.05) in the diets having nano zinc oxide compared to other groups. The results of the present study suggested that the dietary inclusion of nano zinc in broiler diets replacing either inorganic or organic zinc from mineral mixture was found to improve growth performance along with nutrient metabolizability in broiler chickens.

Keywords: Broiler, zinc supplementation, growth performance, metabolizability
Many studies have been undertaken in diversified animal species along with poultry and encouraging effects have been reported with nano Zinc supplementation. However, there is a need to optimize the dose and duration of Zinc Oxide nano particles supplementation in broiler chickens. Actual bioavailability of zinc oxide nano particles in poultry is still to be worked out. In view of the above the effect of supplementation of different zinc sources in relation to the growth performance and nutrient metabolizability in broiler chicken, the present study was planned.

Materials and Methods

Ethical approval

The animal experiment was conducted in accordance with guidelines approved by the Institutional Animal Ethics Committee, 12/CPCSEA Dated 17.10.2019 in the Department of Animal Nutrition, Lala Lajpat Rai University of Veterinary & Animal Sciences, Hisar.

Experimental design

A study of 6 weeks duration was conducted in which one hundred and sixty commercial broiler chicks were randomly distributed in four treatment groups with four replicates of ten birds each in a completely randomized design as presented in Table-1.

The control group (T1) was offered maize, soybean and fishmeal based diet without any source of zinc, which was formulated as per BIS (2007) to fulfill the metabolizable energy (ME) and crude protein requirements of broilers. Birds of second group (T2) were given the basal diet with inorganic zinc in mineral mixture. Third (T3) and fourth (T4) groups were supplemented with organic and nano zinc in mineral mixture, respectively in the diet.

The ingredients and chemical composition of the basal diet as analyzed according to the standards lay down by Association of Official Analytical Chemists (AOAC, 2013) and are presented in Table-2.

**Table 1: Experimental design**

| Treatment Groups | Particulars | Number of replicates | Number of birds/replicates | Total |
|------------------|-------------|----------------------|---------------------------|-------|
| T1               | Control- Standard broiler basal diet as per BIS (2007) specifications without zinc in mineral mixture | 4 | 10 | 40 |
| T2               | Basal diet + inorganic zinc in mineral mixture | 4 | 10 | 40 |
| T3               | Basal diet + organic zinc in mineral mixture | 4 | 10 | 40 |
| T4               | Basal diet + nano zinc in mineral mixture | 4 | 10 | 40 |
| **Total**        |             |                      |                           | **160** |

The birds were fed pre starter, starter and finisher diets for 1 to 7, 8 to 21 and 22 to 42 days of age, respectively. The birds were housed in deep litter pans using wheat straw as litter material and reared from day-old to 42 days of age following standard management practices. Feed and water were provided *ad-lib*. All the birds were vaccinated against Ranikhet disease on 4th day and IBD on 13th day of age. Weekly feed intakes and body weight gain were recorded. Feed conversion ratio (FCR) for each replicate was calculated as follows:

\[ \text{FCR} = \frac{\text{Total feed consumed (g)}}{\text{Total body weight gain (g)}} \]

To study the balance of nitrogen and energy, a metabolism trial was conducted during 6th week of growth period. One bird was randomly selected from each replicate and transferred to metabolic cages. Thus, the feed residue and excreta voided were weighed and properly recorded for final calculations of the total daily feed consumption and excreta voided. Gross energy of oven dried feed was determined by standard procedure using Bomb Calorimeter.

Nitrogen retention calculated as: \((\text{Nitrogen intake-Nitrogen excreted})/\text{Nitrogen intake} \times 100 \)

Similarly, body weight gain and gross energy metabolizability were also calculated. Protein efficiency ratio (PER) was calculated as:

\[ \text{PER} = \frac{\text{Body weight gain (g)}}{\text{Total protein intake (g)}} \times 100 \]

**Table 2: Ingredients and Chemical Composition (%DM Basis) of Experimental Diets in Different Growth Phases of Broiler Chicks**

| Ingredients (Kg/100kg) | Pre-starter | Starter | Finisher |
|------------------------|-------------|---------|----------|
| Maize                  | 52          | 56      | 61       |
| Soybean Meal           | 36          | 31      | 27       |
| Fish Meal              | 6           | 7       | 6        |
| Vegetable Oil          | 4           | 4       | 4        |
| Mineral Mixture        | 2           | 2       | 2        |

**Feed additives (g/100kg feed)**

| Spectromix | 10 | 10 | 10 |
| Spectromix BE | 20 | 20 | 20 |
| Coccidiostat | 65 | 65 | 65 |
| Choline chloride  | 100 | 100 | 100 |
| Lysine | 30 | 50 | - |
| DL-methionine | 150 | 160 | 130 |
| Antibiotic | 100 | 100 | 100 |

**Chemical Composition (%DM)**

| Moisture | 11.54 | 11.54 | 11.48 |
| Dry Matter | 88.46 | 88.46 | 88.52 |

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Results and Discussion

Growth performance: The average feed intake during pre-starter period differed significantly ($P<0.05$) in all treatment groups in comparison to control group (T1). A non-significant difference was observed among treatment groups supplemented with organic zinc (159.0 g/bird) and nano zinc (158.50 g/bird) but both differed significantly ($P<0.05$) in comparison to treatment group supplemented with inorganic zinc (154.50 g/bird). In starter period also treatment groups supplemented with organic zinc (871.0 g/bird) and nano zinc (884.75 g/bird) did not differ significantly ($P>0.05$), however group supplemented with nano zinc showed a significant difference ($P<0.05$) in comparison to treatment group supplemented with inorganic zinc (865.50 g/bird). During finisher phase, the average feed intake (g/bird) ranged from 2570.0 g/bird to 2761.25 g/bird showing highest feed consumption in nano zinc supplemented group (T4) and minimum in control (T1). No significant ($P<0.05$) difference was found between the feed intake of birds supplemented with zinc either organic (T3) or inorganic form (T4) however feed intake among these groups differed significantly ($P<0.05$) from the birds of control group (T1). The average feed intake (g/bird) during overall period ranged from 3577.50 g/bird to 3804.50 g/bird. The overall feed intake of different treatment groups differed significantly ($P<0.05$) with each other with a highest feed intake in treatment group supplemented with nano zinc (3804.5 g/bird) followed by groups supplemented with organic zinc (3617.0 g/bird), inorganic zinc (3601.5 g/bird) and control (3577.5 g/bird).

Higher feed intake obtained in nano zinc supplemented groups might be due to better growth and increased body weight gain. These results coincide with the significantly higher feed intake recorded on replacement of inorganic ‘zinc’ with organic form (Rao et al., 2016 and Mishra et al., 2013)\textsuperscript{[13,14]}. Ezzati et al. (2013)\textsuperscript{[15]} also concluded that dietary zinc increased the body weight and feed intake of broiler birds when used at different levels that is 50,75,100 and 125 mg/kg feed. Similarly, Ibrahim et al. (2017)\textsuperscript{[16]} found that there was no significant ($P>0.05$) change in feed intake between treatment groups supplemented with inorganic, organic, zinc-mix (organic zinc and nanoZnO) and nano zinc. Ramiah et al. (2019)\textsuperscript{[17]} found that regardless of heat stress ZnO nano particles supplementation at 100mg/kg significantly decreased feed intake without affecting the weight gain. Also in a study conducted by Asheer et al. (2018)\textsuperscript{[18]} it was observed that treatment groups given different doses of nano zinc had no significant ($P<0.05$) difference in weekly average feed intake with each other and in comparison with control group. Anil et al. (2012)\textsuperscript{[19]} stated that the supplementation of inorganic or organic zinc at 20, 40, 60 and 80 ppm did not influence the feed intake of broilers.

The average body weight gain (g/bird) differed significantly ($P<0.05$) in each treatment group in all three different growth phases during the experiment with a maximum body weight gain in treatment group supplemented with nano zinc (T4) followed by groups supplemented with organic (T3) and inorganic (T2) zinc and minimum body weight gain was observed in control group (T1). The overall average body weight gain (g/bird) ranged from minimum 1929.47 g/bird in control (T1) to 2175.90 g/bird in group supplemented with nano zinc (T4) showing maximum body weight gain. The findings of present study are in accordance with those of Rao et al. (2016)\textsuperscript{[10]} and Saleh et al. (2018)\textsuperscript{[20]} who concluded that growth performance in terms of increased body weight gain was significantly affected with supplementation of broiler’s diet with organic ‘Zn’. Similar findings were reported in study conducted by Ibrahim et al. (2017)\textsuperscript{[16]} that broiler group supplemented with nano zinc had significantly ($P<0.05$) higher body weight gain followed by the group supplemented with Zn-mix (organic and nano-ZnO) and finally organic zinc supplemented group when compared with the inorganic zinc supplemented group. Another findings of Sagar et al. (2018)\textsuperscript{[21]} were also in accordance with present study in which body weight gain of broilers fed diet with 80 ppm Zn was significantly higher followed by 60 ppm Zn in all the growth phases and it was concluded that nano zinc fed diet had the highest body weight gain followed by organic and inorganic Zn fed diets. ‘Zn’ positively affects feed utilization through participating in the metabolism of carbohydrates, lipids and proteins (Macdonald, 2000)\textsuperscript{[22]} which finally translated into improvements in growth performance. So increase in body weight gain can be attributed to better uptake of organic and nano zinc in the body of broilers.

Feed conversion ratio (FCR) of T4 group in pre-starter period differed significantly ($P<0.05$) from all other treatment groups while control (T1) and treatment group supplemented with inorganic zinc (T2) showed a non-significant ($P>0.05$) difference. The mean values of feed conversion ratio during starter phase of growth ranged from 1.86 (T1) to 1.79 (T4). A significant ($P<0.05$) decrease in mean FCR values from T1 to T4 was observed. The lowest FCR values were recorded in treatment group supplemented with nano zinc (T4). During finisher phase, it was observed that mean FCR value in group supplemented with nano zinc (1.77) was significantly ($P<0.05$) higher than mean FCR values of rest of the treatment groups. The mean values of FCR ranged from 1.88 in T1 to 1.77 in T4. The FCR values of overall growth period were recorded lowest in treatment group supplemented with nano zinc with a value of 1.74 while maximum in control group with a value of 1.85 thus showing better feed conversion in nano zinc supplemented group which implied better utilization of nano zinc particles over inorganic and organic zinc. FCR of chicks fed on diet containing inorganic zinc was inferior to that of organic and nano zinc fed group. This might be due to lower feed consumption as loss in appetite appears to be responsible for decline in feed intake in sulphate supplemented chicks resulting in lower growth rate of this group as compared with other (Sandovall et al., 1998 and Jahanian et al., 2008)\textsuperscript{[23, 24]}. The above findings are in agreement with Sahoo et al. (2016)\textsuperscript{[25]} who observed that FCR can be improved by supplementing the diet with either organic zinc @ 7.5ppm or nano zinc @0.06ppm in the broiler chicken. The results are also in accordance with the findings of Bao et al. (2007)\textsuperscript{[26]} that the organic supplements had positive effects on FCR. Improved FCR might be due to increased activity of digestive enzymes on addition of organic
‘zinc’. This may be because of the protective role of zinc on pancreatic tissue against oxidative stress, this might help pancreas to function properly including secretion of digestive enzymes thus improving the digestibility.

**Nutrient metabolizability:** Mean values of dry matter (DM) metabolizability (%) ranged from 62.77% (T1) to 64.70% (T4). Nutrients metabolizability data revealed that birds supplemented with nano zinc resulted into higher DM metabolizability and (P<0.05) from the organic (64.09%), inorganic (63.73%) and control group (62.77%). Highest nitrogen metabolizability (%) was recorded in T4 (64.70%) supplemented with nano zinc and showed a significant (P<0.05) difference from T1 (61.29%) and T2 (61.60%) while non-significant (P>0.05) difference from T3 (62.48%). Mean values of gross energy metabolizability were recorded highest in T4 (65.55%) group supplemented with nano zinc followed by T3 (62.66%) and T2 (61.86%) groups supplemented with organic and inorganic zinc, respectively and all these differed significantly with each other and control (T1) which had the lowest value of 61.16%. Efficient utilization of nutrients was observed when diet was supplemented with nano zinc in comparison to inorganic and organic source of zinc in the mineral mixture. The above findings are in support of data documented by Saleh et al. (2018) [20], Sahin and Kucuk (2003) [27] who noted that organic ‘Zn’ increased digestibility of dry matter, organic matter, crude protein and ether extract by increasing the secretions and activity of digestive enzymes. ‘Zn’ is also required for the activity of many enzymes and digestive functions including metabolism of carbohydrates, lipids and proteins in the body.

Reference Table 3: Average feed intake, body weight gain and feed conversion ratio under different dietary treatments in experimental period

| Treatments | Pre-Starter | Starter | Finisher | Overall |
|------------|-------------|---------|----------|---------|
| Average feed intake (g/bird) | | | | |
| T1 | 150.50±0.81 | 857.50±1.44 | 2570.00±2.85 | 3577.50±1.25 |
| T2 | 154.30±0.64 | 865.50±2.75 | 2581.50±1.04 | 3601.50±3.66 |
| T3 | 159.00±1.47 | 871.00±1.47 | 2587.00±1.08 | 3617.00±2.67 |
| T4 | 158.50±0.64 | 884.75±1.54 | 2761.25±1.37 | 3804.50±3.22 |
| Average weight gain (g/bird) | | | | |
| T1 | 110.72±0.51 | 458.87±0.85 | 1359.87±1.84 | 1929.47±1.92 |
| T2 | 115.12±0.65 | 468.72±1.43 | 1378.67±4.25 | 1962.52±3.69 |
| T3 | 120.20±0.73 | 476.90±2.01 | 1410.50±4.44 | 2007.60±2.72 |
| T4 | 122.30±0.60 | 493.72±3.92 | 1559.87±1.27 | 2175.90±3.46 |
| Feed conversion ratio | | | | |
| T1 | 1.35±0.002 | 1.86±0.005 | 1.87±0.003 | 1.85±0.002 |
| T2 | 1.34±0.005 | 1.84±0.005 | 1.87±0.005 | 1.83±0.003 |
| T3 | 1.32±0.004 | 1.82±0.004 | 1.83±0.006 | 1.80±0.003 |
| T4 | 1.29±0.003 | 1.79±0.004 | 1.77±0.002 | 1.74±0.001 |

Means bearing different superscripts in a column differ significantly (P<0.05)

**Conclusion**

All the growth parameters like average feed intake, average body weight gain, feed conversion ratio were found to be improved in the organic and nano zinc supplemented groups. The best results were obtained in nano zinc supplemented group in regards to growth parameters and nutrient metabolizability. So it can be concluded that nano zinc supplementation @ 42 mg/kg can improve body growth parameters and can also improve nutrient absorption and metabolizability.

**Table 4:** Mean values of Dry Matter Metabolizability, Nitrogen Metabolizability and Gross Energy Metabolizability under different dietary treatments

| Treatment | DM Metabolizability (%) | Nitrogen Metabolizability (%) | GE Metabolizability (%) |
|-----------|-------------------------|-------------------------------|-------------------------|
| T1 | 62.77±0.161 | 61.29±0.188 | 61.16±0.162 |
| T2 | 63.73±0.115 | 61.60±0.171 | 61.86±0.241 |
| T3 | 64.09±0.142 | 62.48±0.187 | 62.66±0.131 |
| T4 | 64.70±0.063 | 62.66±0.092 | 63.55±0.252 |

Means bearing different superscripts in a column differ significantly (P<0.05)
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