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# Effect of Nickel Supplementation on Growth Performance and Blood Metabolites of Growing Muzaffarnagari Lambs

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## ABSTRACT

The current study was designed to investigate the impact of nickel (Ni) supplementation on growth performance and blood metabolites of growing Muzaffarnagarilambs. Eighteen growing lambsaged10.3±0.67 months and weight 30.45±3.67 kgwere randomly distributed into 3 groups, with 6 lambs in each.The basal diet was similar in all groups except for Ni supplementation which were fed at rates of 0.0 (N0), 1.5 (N1.5), and 3.0 mg/kg DM (N3.0), respectively. Body weight (BW) and feed consumption (FC) were recorded at fortnightly intervals. The blood was sampled on 0, 15, 30, 45, 60, 75, and 90 d of the study period from the jugular vein in EDTA-containingtest tubes in the morning (07.30 h) before feeding and watering.Body weight and FCwere not influenced by the supplementation of any dose of Ni and were reported statistically similar ($P>0.05$) in all the groups. The BW gain (BWG) was statistically ($P<0.05$) higherin 3.0 mg/kg DM Ni fed group as compared to 1.5 mg/kg DM Ni fed and control groups. The BWG was significantly ($P<0.05$) higher in 3.0 mg Ni/kg DM received group of sheep on all days of the study period. Similarly, the feed conversion ratio (FCR) was statistically ($P<0.05$) lower 3.0 mg/kg DM Ni offered group as compared to the 1.5 mg/kg DM offered and control groups. The glucose, non-esterified fatty acid (NEFA), cholesterol, and HDL cholesterol did not vary significantly among the groups and remained statistically similar concentrations in all three groups. The results concluded that the growing lambs supplemented with 3.0 mg/kg DM Ni have better growth performance.

**KEYWORDS:** Blood metabolites,Growing lamb, Nickel,Growth performance.

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## INTRODUCTION

Minerals are required for growth and development and also act as essential components of several enzymes, vitamins, and hormones that help in metabolism, immunity, and reproduction. Minerals might havean adverse impact on animal performance even at moderate deficiencies.Ni is categorized as a newer essential trace element. The essentiality of the Ni has been examined in chicks (Nielsen and Sauberlich, 1970), rats (Spears et al., 1978a), and lambs (Spears et al., 1978b). It plays an important role in enzymes and hormone activation and is alsoinvolved in the regulation of the carbohydrate, protein, and lipid metabolism (La Bella et al., 1973). The Ni reduces the lipolysis by incorporating glucose into the fat cell membranes of rats (Alverez et al., 1993). The higher dose of Ni supplementation causes a rise in the serum cholesteroland triglycerides due to changes in the gene expression of hepatic HMG-CoA reductase or hydroxyl methyl-glutaryl-CoA (Kojima et al., 2004).The addition of Ni in the diet of ruminants have boosted growth performance and feed conversion ratio (Spears, 1984).The Ni-supplemented steers had higher feed efficiency compared to non-received steers (Spears et al., 1978b).Moreover,Chung et al. (1976) observed higher feed conversion efficiency in growingpigsfed diet containing Ni (27 ppm).The objective of the present study was to examine the effect of Ni supplementation on the growth and blood metabolites of growing Muzaffarnagari lambs.

## MATERIALS AND METHODS

The design of the experiment and protocols of this study was approved by the Institutional Animal Ethics Committee and CPCSEA, working under the Ministry of Fisheries, Animal Husbandry and Dairying, Government of India (V-11011 (13)/19/2020/CPCSEA-DADF).

The study was carried out at Instructional Livestock Farm Complex – 2, Sardar Vallabhbhai Patel University of Agriculture and Technology, Meerut, UP, India. Eighteen Muzaffarnagari lambs of similar age (0.33±0.67 months)and BW (30.45±3.67 kg) were used in this study and randomly assigned into three groups (*n* = 6). The diet and its composition were similar in all three groups except for Ni (Ni) offered additionally at 0.0 ($N_0$), 1.5 ($N_{1.5}$), and 3.0 mg/kg DM ($N_{3.0}$) in the respective groupsas in the form of Nisulfate heptahydrate ($NiSO_4.7H_2O$, Purity 98%, SRL, Pvt. Ltd., Mumbai, India) for 90 d of the study period. The experimental animals were kept in three separate groups. The diet was comprised of concentrate, straw, and green fodder in the ratio of 50:30:20, respectively, and prepared as per the recommendation given by ICAR (2012).Lambs were offered diet twice daily at 08.00 hand 17.00 h and allowed *ad libitum* consumption with free access to fresh drinking water.The calculated dose of Ni was weighed and incorporated intothe100 g concentrate and was offered separately to each experimental animal. The ingredients and nutritional composition of the diet are presented in Table 1.

The offered and refused feed was weighed daily and dried. The DMIwas calculated from subtraction of refused feedfrom offered feed. The lambs were weighed in the mourning (07.00 h) before feeding and watering at fortnightly intervals and WG was calculated by subtraction of body weight of previous fortnight from body weight current fortnights. The FCR was calculated as DMI (g)/WG (g).

Table 1. Ingredients and chemical composition of diet offered during the study period

| Ingredient composition of basal diet | %, as DM |
|---|---|
| Berseem | 15 |
| Oat fodder | 20 |
| Wheat straw | 20 |
| Ground maize | 20 |
| Wheat bran | 4 |
| Pulses chuni | 5 |
| Mustard cake | 15 |
| Mineral and vitamin premix[a] | 1 |
| Nickel sulfate heptahydrate | Variable* |
| Chemical Composition | |
| Dry matter | 58 |
| Crude protein | 14.5 |
| Ether extract | 2.90 |
| Total ash | 9.00 |
| Neutral detergent fiber | 47.0 |
| Acid detergent fiber | 29.0 |

[a]Composition of premix per kg: Vitamin A 7 lac IU, vitamin $D_3$ 3 lac IU, alpha-tocopherol 300 IU, Vitamin $B_{12}$ 6 mg, nicotinamide 1.0 g, Cu 1.2 g, Cobalt 40 g, Fe 8 g, Zn 15 g, Mg 6 g, Iodine 0.33 mg, Se 0.001 g, Mn 20 g, Ca 300 g, P 140 g, S 0.72 g, Na 20 g, and K 0.1 g

*1.5 and 3.0 ppm Ni were obtained by supplementing 1.5 and 3.0 mg Ni/kg DM in the basal diet in the f form of nickel sulfate heptahydrate (NiSO4. 7H2O, purity 98%, SRL Pvt. Ltd., Mumbai)

Blood samples were collectedfrom the jugular vein in the test tube containing EDTA as an anticoagulant at 07.30 h before feeding and watering on 0, 15, 30, 45, 60, 75, and 90 d of the study. Blood sampleswere taken to the laboratories in the chilled ice boxes and centrifuged at 3000 rpm for 30 min in the refrigerated centrifuge. Plasma samples were harvested and stored at -20°C till further analysis of glucose, NEFA, cholesterol, and HDL cholesterol.

The offered and residual feed and fodder were dried at 60°C in the hot air oven and then ground, sieve, and reserved for chemical analysis. The procedure of AOAC (2005) was used for the estimation of dry matter, crude protein, ether extract, and total ash of the feed samples. Feed samples were analyzed for the neutral detergent fiber and acid detergent fiber by using the method of Van Soest (1991). The circulating concentration of plasma NEFA was measured by the modified copper soap extraction method (Shipe et al., 1980). The plasma concentration of glucose was measured using the GOD-POD method by the kit (ERBA diagnostics Mannheim Germany). The CHOD- PAP method was used in the measurement of cholesterol in the plasma through the kit (ERBA diagnostics Mannheim Germany). The HDL- cholesterol level in the plasma was measured by the Phosphotungstic Acid method using the kit (ERBA diagnostics Mannheim Germany).

The produced data of feed intake, growth performance, and blood metabolites were analyzed by the MIXED model with repeated measurements using SPSS statistical software version 20 (SPSS Inc., Chicago, IL, USA). The model was used as follows-

$$Yij = \mu + Ci + Gj + eij$$

Where, Yij is the dependent variable, ì is the overall mean of the population, Ci is the random effect of calves, Gjj is the fixed effect of treatment (j = 0, 1.5, and 3.0 mg/kg DM), and eij is the residual element. Tukey's multiple range test was used to compare the treatment means on different days of the study. Orthogonal polynomial contrasts were also analysed and it was two (no. of groups - 1) for the present experiment, first-order contrasts compute linear relationships and second-order contrasts compute quadratic relationships.

## RESULTS AND DISCUSSION

The effect of Ni supplementation on growth performance has been reported in Table 2. The BW and FC did not affect with any dose of Ni supplementation, however, mean BWG was higher in the N3.0 groupas compared to N1.5and control groups. The BWG increased linearly and was found higher in the N3.0 groupon 15, 30, and 75 d of the experimental period. The FCRwas statistically lower in the N3.0group than in all other groups.

Table 2. Effect of nickel supplementation on dry matter intake and growth performance of Muzaffarnagari growing lambs

| Variables | Days | Groups | | | SEM | *P values | | |
|---|---|---|---|---|---|---|---|---|
| | | N0 | N1.5 | N3.0 | | Contrast | Linear | Quadratic |
| Bodyweight, kg | 0 | 30.3 | 30.3 | 30.7 | 3.67 | 0.96 | 0.79 | 0.89 |
| | 15 | 30.9 | 30.9 | 31.4 | 3.68 | 0.94 | 0.75 | 0.89 |
| | 30 | 31.5 | 31.6 | 32.1 | 3.68 | 0.92 | 0.70 | 0.88 |
| | 45 | 32.1 | 32.2 | 32.8 | 3.68 | 0.89 | 0.67 | 0.87 |
| | 60 | 32.8 | 32.9 | 33.5 | 3.72 | 0.89 | 0.67 | 0.84 |
| | 75 | 33.4 | 33.5 | 34.2 | 3.72 | 0.86 | 0.62 | 0.84 |
| | 90 | 34.0 | 34.1 | 34.9 | 3.73 | 0.84 | 0.59 | 0.82 |
| | Mean | 32.1 | 32.2 | 32.8 | 3.70 | 0.51 | 0.29 | 0.65 |
| Body weight gain, kg | 15 | 0.04[a] | 0.043[b] | 0.047[c] | 0.001 | <0.001 | <0.001 | 0.21 |
| | 30 | 0.04[a] | 0.043[b] | 0.047[c] | 0.002 | <0.001 | <0.001 | 0.12 |
| | 45 | 0.04[a] | 0.04[a] | 0.047[b] | 0.002 | <0.001 | <0.001 | 0.18 |
| | 60 | 0.04[a] | 0.04[a] | 0.047[b] | 0.007 | 0.578 | 0.910 | 0.30 |
| | 75 | 0.04[a] | 0.044[b] | 0.047[c] | 0.002 | <0.001 | <0.001 | 0.85 |
| | 90 | 0.04[a] | 0.04[a] | 0.047[b] | 0.002 | <0.001 | <0.001 | 0.021 |
| | Mean | 0.04[a] | 0.04[a] | 0.047[b] | 0.002 | <0.001 | <0.001 | 0.05 |
| Dry matter intake, kg | 15 | 0.56 | 0.56 | 0.57 | 0.06 | 0.94 | 0.75 | 0.95 |
| | 30 | 0.57 | 0.57 | 0.58 | 0.06 | 0.92 | 0.71 | 0.87 |
| | 45 | 0.58 | 0.58 | 0.59 | 0.06 | 0.91 | 0.71 | 0.83 |
| | 60 | 0.59 | 0.59 | 0.60 | 0.07 | 0.88 | 0.69 | 0.77 |
| | 75 | 0.60 | 0.60 | 0.62 | 0.07 | 0.88 | 0.65 | 0.83 |
| | 90 | 0.61 | 0.62 | 0.63 | 0.07 | 0.84 | 0.58 | 0.83 |
| | Mean | 0.58 | 0.59 | 0.60 | 0.06 | 0.54 | 0.31 | 0.65 |
| Feed conversion ratio | 15 | 13.8 | 12.9 | 12.1 | 1.38 | 0.05 | 0.01 | 0.94 |
| | 30 | 13.9 | 13.3 | 12.2 | 1.62 | 0.09 | 0.03 | 0.74 |
| | 45 | 14.1 | 13.7 | 12.6 | 1.51 | 0.13 | 0.05 | 0.63 |
| | 60 | 13.4 | 14.0 | 12.8 | 2.06 | 0.53 | 0.57 | 0.34 |
| | 75 | 14.8 | 13.7 | 13.0 | 1.72 | 0.13 | 0.05 | 0.76 |
| | 90 | 15.1 | 14.6 | 13.4 | 1.42 | 0.05 | 0.021 | 0.52 |
| | Mean | 14.2[b] | 13.7[b] | 12.7[a] | 1.62 | <0.001 | <0.001 | 0.34 |

*Contrast effect of nickel supplementation; Linear, linear effect of nickel; Quadratic, the quadratic effect of nickel, SEM, Standard error mean;
Mean bearing different superscript (a, b) in a row showed a significant difference at P<0.05

The FCR on all days of the study period did not vary statistically among the groups but reported numerically lower in N3.0 group.Similar to present study, Anke et al. (1977) reported a 21% increase in weight gain in goats fedwith 10 mg Ni/kg DM than goats fed 1mg Ni/kg DM. The inclusion of 5 mg/kg DM Ni in the basal diet of steer improved BWG and feed efficiency (Oscar et al., 1987). Spears (1984) also reported a higher WG in lambs fed Ni in their basal diet. Similarly, Singh et al. (2019) reported a higher WG in the growing heifers supplemented with 3.0 mg Ni/kg DM.In the present study, Ni supplementation improved growth performance might be it increase the activity of the urease enzyme (Singh et al., 2019). Contrary to the present finding, Spear et al. (1986) reported that Ni did not affect growth performance of calves.The inclusion of Ni at the dose 6 or 12 ppm in the diet of broilers did not affect growth performance (Oscar et al., 1995).

The changes in the concentration of blood metabolites during different days of the study have been presented in Table 3. The mean plasma concentration of glucose, NEFA, cholesterol, and HDL-cholesterol did not vary statistically among the groups and was found statistically similar in all the groups indicated that Ni supplementation did not influence the plasma concentration of blood metabolites. Similarly, the glucose, NEFA, cholesterol, and HDL-cholesterol concentration on 0, 15, 30, 45, 60, 75, and 90 d of the study period did also not show a statistical difference among groups. In accord with the present finding, Singh et al. (2019) observed no significant effect of Ni supplementationon plasma concentration of glucose in growing calves.A non-significant effect of Ni (500 ppm) on glucose levels was observed by Yousuf (2002) in goats. However, Obone et al. (1999) reported lower glucose levels in the rats withNi (35 mg Ni as NiSO4/kg/day) supplementation.Plasma NEFA reflects body fat mobilization in response to negative energy balance or stress conditions. Plasma NEFA is formed from body fat reserve and acts as an important fuel in negative energy balance and stress conditions. The dietary Ni supplementation in the current study did not exert any statistical effect on the plasma concentration of NEFA. Similar to the present finding, Singh et al (2019) reported a non-significant effect of Ni supplementation on NEFA levels of growing calves.

In corroboration with the current study, Singh et al. (2019) reported that Ni supplementation at doses 1.5, and 3.0 in calves did not influence cholesterol concentration statistically. A significant increase in the cholesterol, however, a decrease in the HDL cholesterol concentration was reported by Das et al. (2001) in rats fed 2.0 mg Ni/100 g BW compared to control.

Table 3. Effect of nickel supplementation on blood metabolites of Muzaffernagari growing lambs

| Variables | Days | Groups | | | SEM | *P values | | |
|---|---|---|---|---|---|---|---|---|
| | | N0 | N1.5 | N3.0 | | Contrast | Linear | Quadratic |
| Glucose, mM | 0 | 2.57 | 2.59 | 2.62 | 0.08 | 0.37 | 0.17 | 0.83 |
| | 15 | 2.64 | 2.69 | 2.73 | 0.08 | 0.35 | 0.68 | 0.17 |
| | 30 | 2.73 | 2.76 | 2.78 | 0.13 | 0.12 | 0.12 | 0.15 |
| | 45 | 2.76 | 2.67 | 2.79 | 0.18 | 0.25 | 0.10 | 0.75 |
| | 60 | 2.77 | 2.76 | 2.78 | 0.13 | 0.04 | 0.02 | 0.21 |
| | 75 | 2.76 | 2.66 | 2.67 | 0.22 | 0.30 | 0.14 | 0.70 |
| | 90 | 2.75 | 2.72 | 2.69 | 0.24 | 0.36 | 0.17 | 0.69 |
| | Mean | 2.71 | 2.62 | 2.72 | 0.15 | 0.26 | 0.64 | 0.27 |
| Non-esterifies fatty acids, µM | 0 | 0.16 | 0.17 | 0.18 | 0.05 | 0.77 | 0.50 | 0.84 |
| | 15 | 0.18 | 0.18 | 0.17 | 0.04 | 0.77 | 0.53 | 0.72 |
| | 30 | 0.15 | 0.16 | 0.17 | 0.04 | 0.37 | 0.17 | 0.83 |
| | 45 | 0.16 | 0.15 | 0.16 | 0.03 | 0.83 | 0.97 | 0.55 |
| | 60 | 0.17 | 0.17 | 0.17 | 0.04 | 0.99 | 0.97 | 0.89 |
| | 75 | 0.16 | 0.16 | 0.18 | 0.03 | 0.30 | 0.14 | 0.62 |
| | 90 | 0.18 | 0.14 | 0.17 | 0.06 | 0.51 | 0.78 | 0.26 |
| | Mean | 0.16 | 0.16 | 0.17 | 0.04 | 0.54 | 0.39 | 0.47 |
| Cholesterol, mM | 0 | 2.85 | 2.81 | 2.87 | 0.23 | 0.87 | 0.85 | 0.63 |
| | 15 | 3.01 | 2.99 | 3.04 | 0.27 | 0.93 | 0.88 | 0.75 |
| | 30 | 3.06 | 3.00 | 3.07 | 0.28 | 0.87 | 0.94 | 0.60 |
| | 45 | 2.99 | 2.90 | 3.14 | 0.37 | 0.42 | 0.41 | 0.31 |
| | 60 | 3.14 | 3.13 | 3.16 | 0.35 | 0.99 | 0.93 | 0.91 |
| | 75 | 3.16 | 3.10 | 3.18 | 0.38 | 0.91 | 0.91 | 0.68 |
| | 90 | 3.15 | 3.12 | 3.15 | 0.21 | 0.96 | 0.97 | 0.78 |
| | Mean | 3.04 | 3.01 | 3.03 | 0.29 | 0.40 | 0.54 | 0.23 |
| HDL-cholesterol, mM | 0 | 1.53 | 1.55 | 1.58 | 0.11 | 0.64 | 0.35 | 0.98 |
| | 15 | 1.56 | 1.58 | 1.61 | 0.10 | 0.46 | 0.22 | 0.83 |
| | 30 | 1.59 | 1.60 | 1.57 | 0.13 | 0.87 | 0.83 | 0.64 |
| | 45 | 1.58 | 1.64 | 1.63 | 0.13 | 0.49 | 0.36 | 0.44 |
| | 60 | 1.62 | 1.55 | 1.62 | 0.17 | 0.65 | 0.96 | 0.36 |
| | 75 | 1.61 | 1.65 | 1.64 | 0.11 | 0.76 | 0.59 | 0.62 |
| | 90 | 1.60 | 1.64 | 1.59 | 0.12 | 0.56 | 0.86 | 0.29 |
| | Mean | 1.58 | 1.60 | 1.61 | 0.12 | 0.51 | 0.29 | 0.62 |

*Contrast effect of nickel supplementation; Linear, linear effect of nickel; Quadratic, quadratic effect of nickel, SEM, Standard error mean;

## CONCLUSION

The results of the present study revealed that Ni supplementation did not influence the blood metabolites,however, Ni supplementation at the dose of 3.0 mg/kg DM improved BW gain and feed conversion efficiency.

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