

## The effect of Different Levels of effect of supplementing nano zinc oxide (nZnO) on the product traits Local goat Kids

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### Abstract

This experiment was conducted at the Animal Field of the College of Agriculture and Marshes, University of Thi-Qar, from September 15, 2024, to January 22, 2025, to investigate the effect of supplementing nano zinc oxide (nZnO) at three different levels (0, 20, and 30 mg/head/day) on the productive performance of local goat kids. A total of 12 local male kids, purchased from local markets in Thi-Qar Province, were randomly allocated to three experimental groups (n = 4 per group). All groups received the same concentrate diet at 3% of dry matter relative to live body weight, consisting of: barley (45%), wheat bran (25%), ground yellow corn (15%), soybean meal (12%), and a vitamin-mineral premix (3%). The main findings of the study were as follows:

There were no significant differences among treatments in terms of final body weight, total weight gain, or average daily gain, although a numerical improvement was observed in the second treatment group (T2: 20 mg/head/day), Feed intake and feed conversion efficiency were not significantly affected by the different levels of nano zinc oxide supplementation.

A significant increase ( $P \leq 0.05$ ) was observed in the mean total bacterial count and cellulolytic bacteria in favor of the second treatment group (T2) (20 mg nZnO/head/day), along with a numerical increase in the average count of lactic acid bacteria throughout the experimental period. Additionally, a significant improvement ( $P \leq 0.05$ ) in ruminal pH value was recorded after 45 days of the experiment in favor of the T2 treatment, with a numerical increase in pH also noted after 84 days for the same group.

**Key words:** nano zinc oxide, pH value, Local goat Kids

### I. Introduction

Minerals are vital components in animal nutrition across all stages of production, playing essential roles in digestion, reproduction, and growth processes. Although animals require minerals in much smaller quantities compared to proteins and carbohydrates, deficiencies in these micronutrients can lead to more severe physiological disturbances than deficiencies in macronutrients. In recent years, increasing attention has been directed toward nanotechnology, which has emerged as a promising approach to improving animal health and enhancing productivity, both quantitatively and qualitatively. Nanotechnology can be used to protect key enzymes and proteins that affect nutrient utilization in ruminant animals (Rizzello & Pompa, 2014; Raji et al., 2018).

Nanotechnology is an innovative scientific field that is now applied across various sectors, including medicine, chemical industries, biological research, disease diagnostics, and animal nutrition (Hassanein et al., 2021). Zinc is an essential



trace element required for numerous physiological functions in animals. It is considered a critical nutrient for living organisms (Youssef et al., 2019). However, in its conventional (bulk) form, zinc is not widely used in livestock diets due to its low bioavailability (Geetha et al., 2020).

The bioavailability of zinc oxide can be enhanced by reducing its particle size to the nanoscale (nano-ZnO), which improves zinc ionization and absorption in the digestive tract, effectively overcoming its deficiency in animal diets. Nano-sized particles (1–100 nm) provide a larger surface area, leading to improved solubility and absorption rates (Raji et al., 2018).

Overall, nanotechnology has significantly contributed to advancements in agriculture, food production, and energy by enhancing plant growth and productivity through the use of nano-fertilizers (Yanal et al., 2021). In the field of animal production, several studies have reported that the inclusion of nano zinc oxide in goat diets led to improved fiber digestibility and an increase in rumen protozoa population (Swain et al., 2018). Similarly, the use of nano-bentonite in local lamb diets resulted in significant increases in both daily and total body weight gain (Al-Sudani et al., 2021).

## II. Materials and Methods

### Experimental Design

This study was conducted at the Animal Field of the College of Agriculture and Marshes, University of Thi-Qar, during the period from September 15, 2024, to January 15, 2025. The objective was to evaluate the effect of nano zinc oxide (nZnO) supplementation at three different levels (0, 20, and 30 mg/head/day) as a dietary additive to improve economic traits in local male goat kids.

### Experimental Animals

A total of 12 local male goat kids, with an average initial body weight of  $20.77 \pm 1.78$  kg, were purchased from local markets in Thi-Qar Province. All animals were examined by a veterinarian prior to the study to ensure their health status and absence of disease.

### Feeding Dite

The animals were fed a concentrate diet at 3% of dry matter relative to their live body weight. The feed was offered in two equal meals per day: the first at 7:00 a.m., and the second at 3:00 p.m. The feed amount was adjusted biweekly based on the updated body weights of the animals. Daily feed intake was calculated by subtracting the feed refusal from the total feed offered. Barley straw was offered ad libitum to all animals throughout the trial period. All feed ingredients were ground and mixed thoroughly to ensure uniformity. The concentrate diet was composed of (Barley: 45% , Wheat bran: 25% , Ground yellow corn: 15% , Soybean meal: 12% , Vitamin-mineral premix: 3% )

### Experimental Treatments

The goat kids were randomly assigned to three treatment groups, with four animals per group, as follows:

- **T1 (Control):** 0 nano zinc oxide supplementation
- **T2:** 20 mg/head/day of nano zinc oxide
- **T3:** 30 mg/head/day of nano zinc oxide



## Measurements and Sampling

### 1. Body Weight and Weight Gain:

Initial body weights were recorded after a two-week adaptation period. Subsequently, body weights were measured biweekly until the end of the trial.

- $\text{Daily weight gain (g/day)} = (\text{Final weight} - \text{Previous weight}) / \text{Number of days between measurements}$

### 2. Feed Intake and Feed Conversion Ratio (FCR):

Daily feed consumption was recorded, and feed conversion efficiency was calculated as follows:

- $\text{FCR} = \text{Total feed intake during a specific period (g)} / \text{Weight gain during the same period (g)}$

## Collection of Rumen Fluid:

Rumen fluid samples were collected using a stomach tube inserted orally into the rumen. The extracted fluid was transferred into tightly sealed plastic containers on days 45 and 84 of the experiment, for each animal, at two time points: before feeding and 3 hours after feeding. These samples were used for bacterial enumeration and pH measurement.

## Rumen Variables:

### Ph Measurement:

Immediately after sampling the rumen fluid from the animal, the pH was measured using a digital pH meter (PW Philips 9909 PH meter).

## Enumeration of Cellulolytic Bacteria:

Samples stored at  $-20^{\circ}\text{C}$  were prepared to determine the count of cellulolytic bacteria. One milliliter of each sample was taken and diluted with 9 mL of sterile 0.1% peptone solution to obtain a 1:10 dilution. From this, a serial dilution (up to six-fold) was performed. A 1 mL aliquot from the final dilution was plated onto a pre-prepared selective culture medium (Mast Diagnostica), following the manufacturer's instructions.

The medium contained plant tissue extract as a cellulose source to promote the growth of cellulolytic microorganisms. The inoculated plates were incubated for 48 hours at  $37^{\circ}\text{C}$ . The total bacterial count was calculated using the following formula as described by APHA (1992):

**Total Bacterial Count /  $\text{cm}^3$  = Number of colonies on the plate  $\times$  reciprocal of the dilution factor**



### III. Results and Discussion

#### Productive Traits

#### Body Weight of Kids

Table 1 shows the body weights of goat kids across experimental treatments supplemented with different levels of nano zinc oxide (nZnO). The results indicated no significant differences ( $P > 0.05$ ) in average body weights among treatment groups throughout the experimental period. However, a numerical increase in average body weight was observed in the second treatment group (T2: 20 mg nZnO/head/day) compared to other treatments. This increase appeared notably at weeks 2, 4, 6, and 10, with average weights of 21.45, 23.62, 24.50, and 25.75 kg, respectively.

In contrast, the third treatment group (T3: 30 mg nZnO/head/day) consistently recorded the lowest average weights throughout the trial, with means of 20.57, 20.40, 22.17, 22.75, 21.00, 22.85, and 24.15 kg at the corresponding weeks.

These findings align with those of Al-Ghazali (2022), who reported that supplementation with nano zinc oxide (0, 15, and 30 mg/head/day) in local lamb diets did not significantly affect lamb body weights.

However, the current results differ from those of Yusuf et al. (2022), who observed a significant increase ( $P \leq 0.05$ ) in average body weight in West African Dwarf goats fed 600 mg/kg nano zinc oxide compared to other treatment groups.

The improvement in body weights may be attributed to enhanced rumen environment. Zinc, uniquely among dietary minerals, binds with feed particles and microorganisms, contributing to rumen stability (Caldera, 2015). This stabilization enhances fiber digestion by increasing total rumen microbial populations, thereby improving nutrient utilization and weight gain (Al-Zubaedi et al., 2021; Ghazi & Al-Galiby, 2022).

Furthermore, the numerical weight gain observed in the T2 group may be due to increased efficiency of nano zinc absorption during growth, when tissues are more receptive to minerals and metabolic activity peaks, facilitating better nutrient utilization and growth rate.

**Table 1.** Mean body weights ( $\pm$  standard deviation) of control and treatment goat kids receiving different levels of nano zinc oxide throughout the study period

Treatment	Week 0	Week 2	Week 4	Week 6	Week 8	Week 10	Week 12
Control (T1)	20.87 $\pm$ 1.28	21.07 $\pm$ 0.88	22.72 $\pm$ 1.72	23.95 $\pm$ 2.18	23.72 $\pm$ 0.70	25.25 $\pm$ 1.19	27.90 $\pm$ 1.76
T2 (20 mg nZnO)	20.87 $\pm$ 2.23	21.45 $\pm$ 2.91	23.62 $\pm$ 3.39	24.50 $\pm$ 2.57	23.55 $\pm$ 4.13	25.75 $\pm$ 5.37	27.67 $\pm$ 5.45
T3 (30 mg nZnO)	20.57 $\pm$ 1.83	20.40 $\pm$ 2.13	22.17 $\pm$ 1.98	22.75 $\pm$ 1.95	21.00 $\pm$ 2.64	22.85 $\pm$ 3.00	24.15 $\pm$ 4.18
Significance	N.S	N.S	N.S	N.S	N.S	N.S	N.S



- N.S: Not significant ( $P > 0.05$ )
- T1: Control (0 mg nano zinc oxide/head/day)
- T2: 20 mg nano zinc oxide/head/day
- T3: 30 mg nano zinc oxide/head/day

### Total and Daily Weight Gain

Table 2 presents the total weight gain (kg) and daily weight gain (g/day) for the control group and treatment groups receiving different levels of nano zinc oxide (nZnO). The results showed no significant differences ( $P > 0.05$ ) among the experimental treatments in both total weight gain and daily weight gain throughout the study period.

These findings are consistent with those of Al-Ghazali (2022), who reported that supplementing local lamb diets with different levels of nano zinc oxide (0, 15, and 30 mg/head/day) did not significantly affect either daily or total weight gain.

In contrast, the current results differ from those reported by Elsayed (2018), who studied the effects of various zinc sources on digestibility and weight gain in lambs. He found significant differences ( $P \leq 0.05$ ) in daily weight gain when lambs were supplemented with zinc methionine or zinc sulfate compared to control and other zinc treatment groups.

Similarly, Anil et al. (2019) investigated the effect of nano zinc oxide (nZnO) supplementation on growth performance in crossbred calves. Their study included three treatments: T1 with 25 mg zinc sulfate ( $\text{ZnSO}_4$ ), T2 with 5 mg nZnO, and T3 with 10 mg nZnO. They reported highly significant differences ( $P \leq 0.01$ ) in both daily and total weight gain, with T3 showing the highest total gain of 39.67 kg compared to 37.54, 35.25, and 18.75 kg for T2, T1, and control groups, respectively.

**Table 2.** Total and daily weight gain ( $\pm$  standard deviation) of control and treatment goat kids receiving different levels of nano zinc oxide during the study period

Treatment	Total Weight Gain (kg)	Daily Weight Gain (g/day)
Control (T1)	$7.02 \pm 1.09$	$83.63 \pm 13.09$
T2 (20 mg nZnO)	$6.80 \pm 3.37$	$80.95 \pm 40.13$
T3 (30 mg nZnO)	$4.75 \pm 1.87$	$56.54 \pm 22.30$
Significance	N.S	N.S

- N.S: Not significant ( $P > 0.05$ )
- T1: Control (0 mg nano zinc oxide/head/day)
- T2: 20 mg nano zinc oxide/head/day
- T3: 30 mg nano zinc oxide/head/day



### Feed Intake and Feed Conversion Efficiency

Table (3) shows the feed intake (kg) and feed conversion efficiency (kg feed/kg weight gain) of the control group and groups given different levels of nano zinc oxide (nZnO). The results indicate no significant differences ( $P > 0.05$ ) between the experimental treatments in feed intake and feed conversion efficiency throughout the 90-day trial period.

However, a numerical increase in feed intake was observed in favor of treatment T2, which recorded an average of ( $70.31 \pm 10.76$  kg), and a numerical increase in feed conversion efficiency was noted for treatment T3 with an average of ( $15.24 \pm 4.46$  kg feed/kg weight gain).

These results agree with the study by Zaboli et al. (2013), which used different sources of natural zinc oxide (ZnO) and nano zinc oxide (nZnO) at different levels (0, 20, 40 mg/kg feed) in goat rations, showing no significant differences in feed intake.

Chang et al. (2020) also reported no significant effect of zinc supplementation on feed intake in Holstein calves.

Similarly, Al-Ghazali (2022) found no significant differences in feed intake (roughage, concentrate, and total feed) and feed conversion efficiency when using different levels of nano zinc oxide (0, 15, 30 mg/head/day) in feeding local lambs throughout the study period.

**Table (3).** Feed intake (kg) and feed conversion efficiency (kg feed/kg weight gain) of the control group and groups given different levels of nano zinc oxide during the study period ( $\pm$  standard deviation)

Treatments	Feed Intake (kg)	Feed Conversion Efficiency (kg feed/kg weight gain)
Control (T1)	$69.51 \pm 3.78$	$10.03 \pm 1.30$
Treatment 2 (T2)	$70.31 \pm 10.76$	$12.58 \pm 6.42$
Treatment 3 (T3)	$66.63 \pm 5.52$	$15.24 \pm 4.46$
Significance	N.S	N.S

- N.S: No significant differences ( $P > 0.05$ )
- T1: Control treatment (0 mg nano zinc oxide/head/day)
- T2: 20 mg nano zinc oxide/head/day
- T3: 30 mg nano zinc oxide/head/day



**Rumen Fluid Ph:**

Table (4) presents the pH values of rumen fluid after 45 and 84 days of the experiment, measured at 0 and 3 hours post-feeding for the experimental groups that received different levels of nano zinc oxide (nZnO) throughout the study period.

The results indicate a significant difference ( $P \leq 0.05$ ) in rumen fluid pH values on day 45, 3 hours after feeding, among the treatment groups that received nano zinc oxide. Treatment group T2 (20 mg nZnO/head/day) recorded a significantly higher mean pH value (6.29) compared to T3 (30 mg nZnO/head/day), which had a mean value of (5.77). Meanwhile, the control group (T1) did not show any significant differences compared to the other treatments, with a mean value of (6.02).

After 84 days of the experiment, no significant differences were observed in rumen pH values among the treatments. However, a numerical increase was recorded in T2 (6.16), remaining within the normal physiological range of rumen fluid, when compared to T1 (6.02) and T3 (5.94).

These findings are consistent with those of **Elsayed et al. (2018)**, who reported that supplementing diets with 20 mg of various zinc sources (zinc sulfate or zinc methionine) helped maintain rumen pH levels and prevented excessive decline, especially compared to zinc sulfate and control treatments.

Conversely, **Sarker et al. (2018)** found that supplementing diets with varying levels of nano zinc oxide had no significant effect on rumen pH. Similarly, **Petrič et al. (2021)** reported no significant differences in rumen pH in lambs fed diets supplemented with nano zinc oxide, medicinal herbs, or their combination.

Overall, treatment T2 (20 mg nZnO/head/day) appeared to help stabilize rumen pH within the optimal physiological range (5.5 to 6.5), particularly near the ideal value of 6.0, which is essential for the optimal activity of rumen microflora (Boomker, 2000). This stability suggests a balanced rumen environment and improved fermentation efficiency. Maintaining a stable rumen pH is particularly crucial for the growth and function of cellulolytic bacteria (Stewart, 1992). The observed improvement may be attributed to nano zinc oxide's role in modulating fermentation patterns and enhancing pH balance in the rumen, contributing to improved volatile fatty acid production, as also reported by **Zhao et al. (2014)**.

**Table (4):Rumen pH values (mean  $\pm$  standard deviation) for control and treatment groups supplemented with varying levels of nano zinc oxide throughout the study period.**

Treatments	pH after 45 days		pH after 84 days	
	0 hour	3 hours	0 hour	3 hours
<b>T1 (Control)</b>	5.50 $\pm$ 0.31	6.02 $\pm$ 0.15 ab	5.50 $\pm$ 0.16	6.02 $\pm$ 0.22
<b>T2 (20 mg nZnO)</b>	5.66 $\pm$ 0.24	<b>6.29 <math>\pm</math> 0.23 a</b>	5.59 $\pm$ 0.31	6.16 $\pm$ 0.18
<b>T3 (30 mg nZnO)</b>	5.42 $\pm$ 0.31	<b>5.77 <math>\pm</math> 0.14 b</b>	5.76 $\pm$ 0.30	5.94 $\pm$ 0.14
<b>Significance</b>	N.S	<b>P <math>\leq</math> 0.05</b>	N.S	N.S



- Different superscript letters within the same column indicate significant differences at  $P \leq 0.05$ .
- **N.S:** No significant differences among means.
- **T1:** Control group (0 mg nano zinc oxide/head/day)
- **T2:** 20 mg nano zinc oxide/head/day
- **T3:** 30 mg nano zinc oxide/head/day

### Rumen Bacterial Counts in Goat Kids

Table (5) illustrates the mean counts of cellulolytic bacteria in the rumen fluid of control and treated groups that received varying levels of nano zinc oxide (nZnO) throughout the study period.

The results show a significant increase ( $P \leq 0.05$ ) in the average counts of cellulolytic bacteria in the rumen fluid of goat kids in treatment groups T2 and T3, which were supplemented with 20 and 30 mg nZnO/head/day, respectively, compared to the control group (T1), particularly 3 hours post-feeding at both sampling times (day 45 and day 84).

Treatment T2 recorded the highest mean values:

**Day 45:**  $5.63 \times 10^8$  CFU/ml (3 hours post-feeding)

**Day 84:**  $6.77 \times 10^8$  CFU/ml (3 hours post-feeding)

These findings reflect the positive impact of nano zinc oxide supplementation on the microbial environment of the rumen. The significant rise in cellulolytic bacterial counts indicates improved microbial activity, thereby enhancing the animal's digestive efficiency. This aligns with the observations of Zhao et al. (2014), who reported improved rumen fermentation and microbial balance with zinc supplementation.

Additionally, Wang et al. (2013) highlighted the role of zinc in promoting bacterial enzyme activity, which in turn stimulates the growth of beneficial rumen bacteria, particularly cellulolytic species like *Fibrobacter succinogenes*.

**Table (5): Mean cellulolytic bacterial counts ( $\times 10^8$  CFU/ml  $\pm$  standard deviation) in the rumen fluid of goat kids from the control and nano zinc oxide treatment groups over the experimental period.**

Treatments	Cellulolytic Bacteria after 45 Days		Cellulolytic Bacteria after 84 Days	
	0 hour	3 hours	0 hour	3 hours
<b>T1 (Control)</b>	$0.69 \pm 0.15^a$	$2.30 \pm 0.79^b$	$0.80 \pm 0.15^a$	$2.98 \pm 0.71^b$
<b>T2 (20 mg nZnO)</b>	$0.52 \pm 0.09^{ab}$	<b><math>5.63 \pm 0.96^a</math></b>	$0.51 \pm 0.77^{ab}$	<b><math>6.77 \pm 0.62^a</math></b>
<b>T3 (30 mg nZnO)</b>	$0.42 \pm 0.09^b$	<b><math>4.32 \pm 0.77^a</math></b>	$0.53 \pm 0.08^b$	<b><math>5.48 \pm 1.79^a</math></b>





Treatments	Cellulolytic Bacteria after 45 Days		Cellulolytic Bacteria after 84 Days	
Significance	$P \leq 0.05$	$P \leq 0.05$	$P \leq 0.05$	$P \leq 0.05$

- Different superscript letters in the same column denote **significant differences ( $P \leq 0.05$ )** between treatment means.
- **T1:** Control group (0 mg nano zinc oxide/head/day)
- **T2:** 20 mg nano zinc oxide/head/day
- **T3:** 30 mg nano zinc oxide/head/day

#### IV. Conclusions

The results of the current study indicate that supplementation with varying levels of nano zinc oxide (nZnO) contributed to improvements in the weight gain rate of local goat kids. Although no statistically significant differences were observed among treatments, numerical increases in growth performance suggest potential benefits of nano zinc oxide supplementation in enhancing productive traits in local goats.

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