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# Supplementation of Zinc Oxide and Zinc Methionine on Incidence of Non-Specific Scours in Piglets

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# Abstract

Pig production occupies an important place in modern agriculture. Piglets are far less demanding on nutrients as compared to poultry. The experiment was conducted to know the effect of feeding organic and inorganic sources of additional zinc on growth incidences of non-specific scours in piglets. Sixty graded (Large White Yorkshire) suckling piglets were randomly divided into three treatment group of twenty piglets each on the basis of litter size, parity and live birth weight. Treatment I ( $T_1$ ) served as control and these piglets were fed with deionized water daily as oral suspensions. Treatment II ( $T_2$ ) and Treatement III ( $T_3$ ) were fed with fed Zinc Oxide (ZnO) and Zinc Methionine (ZnM) daily (50 ppm) as oral suspension. The results showed that the incidence of diarrhoea in control (T1), zinc oxide ( $T_2$ ) and zinc methionine ( $T_3$ ) groups were (35%), (20%) and (25%) respectively. In the control group (T1), only seven piglets were seen with diarrhoea, whereas, in the zinc oxide supplemented group ( $T_2$ ) four piglets were observed with diarrhoea and with zinc methionine supplemented group (T3) five piglets had shown diarrhoea.

In the current study supplementation of zinc, reduced the incidence of diarrhoea in piglets. But the possible reason for the nonsignificance incidence of diarrhoea among the  $T_1$ ,  $T_2$  and  $T_3$  groups might be due to the feco-oral contamination of zinc, since there was no control over the piglets supplemented with and without zinc.

Keywords: Supplementation; Zinc Oxide; Zinc Methionine; Piglets

# Introduction

Pig production occupies an important place in modern agriculture. Piglets are far less demanding on nutrients as compared to poultry. The production cost of pork is comparatively not very high. However economical pig production is ahead of the hour for those involved in pig production activities. Among all the livestock production programmes, pig production is the most potential source of profit earning because of its prolificacy, fast growth rate, efficiency in feed conversion etc. Several *in vitro* studies have shown that zinc (Zn) has broad-spectrum antiviral activity against a variety of viruses, such as human immunodeficiency virus, transmissible gastroenteritis virus (TGEV), equine arteritis virus, and severe acute

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respiratory syndrome coronavirus [1,18]. Many potential mechanisms have been suggested to explain the potential beneficial effect of Zn against virus infections. For example, Zn mediates antiviral effects through the inhibition of nidovirus RNA-dependent RNA polymerases or other proteins essential for the different phases of the viral life cycle [2]. Zinc is known as gut conditioner which alters intestinal morphology and improves absorptive capacity, reduces scours and enhances the growth performance [22,25]. Zinc supplementation resulted in higher immoglobulin response to antigen challenge [24] and functions as antioxidant which destroys free radicals and prevents oxidative damage to the cells [10]. Zinc is needed for various physiological processes and has been found to be present in over 200 metalloenzymes [23]. Zinc is essential for reproduction, bone development and growth [26-28].

Late weaning in organic units, where piglets have to be over six weeks of age at weaning [5], is likely to decrease problems associated with post-weaning diarrhoea. The body weight and growth rate of non-descriptive kids improved by oral organic zinc supplementation. In addition to this there was no occurrence of gastrointestinal disturbances and other related health problems in zinc supplemented kids [15]. There are a range of causes of neonatal diarrhoea, of which *E.coli* infection is one of the most common, particularly in gilt litters [32]. The report of [13] reveals that high concentration as zinc from ZnO supplementation in the piglet diets increased the stability of the intestinal micro flora through a reduction in the diversity of coliforms. Therefore zinc oxide is currently included in many nursery diets at high concentration to aid the growth promotion [11] but there has been increasing environmental concerns recently regarding excretion of zinc in swine waste. Organic forms of zinc may exhibit greater bioavailability than commonly used inorganic forms of zinc and also have decreasing environmental concerns. Therefore, the present study was undertaken to with an objective of effect of supplementation of zinc at low levels on Incidences of nonspecific scours in pigs.

## **Materials and Methods**

The experiment was conducted at piggery Farm, Department of Instructional Livestock Farm Complex, Veterinary College, Bangalore. The experiment was conducted to know the effect of feeding organic and inorganic sources of additional zinc on growth performance of young piglets. Sixty graded (Large White Yorkshire) suckling piglets with a mean average weight of 1.313 kg were used in eight weeks growth trial. The piglets were randomly divided into three treatment group of twenty piglets each on the basis of litter size, parity and live birth weight, so that each group had comparative average initial weight (Table 1).

Treatment 1 (control)			Treatment 2 (ZnO)			Treatment 3 (ZnM)		
Piglets	Body weight	Sow No	Piglets num-	Body weight	Sow No	Piglets	Body weight	Sow No
number	(kg)		ber	(kg)		number	(kg)	
185	1.55	31	187	1.3	31	217	1.59	31
223	1.19	141	218	1.73	31	392	0.675	141
221	1.14	141	190	1.075	141	189	1.15	141
178	1.915	225	224	1.19	141	200	1.97	225
179	1.68	225	191	1.045	141	201	1.61	225
182	1.85	466	180	1.45	225	204	1.3	466
183	1.69	466	202	1.76	225	206	1.81	466
195	1.585	227	207	1.87	466	196	1.505	227
199	0.833	225	235	1.565	466	226	1.345	227
209	1.47	149	197	1.65	227	214	1.865	57
212	1.5	149	213	1.575	57	176	1.745	432
197	1.93	432	215	1.515	149	195	1.8	432
242	1.14	57	196	1.67	432	243	1.08	57
256	1.27	438	211	1.135	57	213	1.035	57
224	1.295	438	214	1.14	57	226	1.265	484
263	1.025	109	255	1.275	438	265	1.01	109
237	0.975	109	227	1.35	438	239	0.985	109
252	1.14	429	253	1.035	428	222	1.195	429
254	1.125	429	220	1.21	429	262	1.21	18
233	1.2	429	234	1.145	223	236	1.105	223
SE	0.073			0.059			0.079	

Table 1: Particulars of the piglets used in the experiment.

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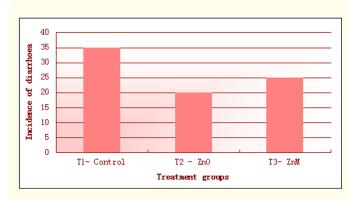
Treatment I ( $T_1$ ) served as control and these piglets were fed with deionized water daily as oral suspensions. Treatment II ( $T_2$ ) and Treatement III ( $T_3$ ) were fed with fed Zinc Oxide (ZnO) and Zinc Methionine (ZnM) (BIOPLEX) supplied by (VetCare India Pvt Ltd, Bengaluru daily (50 ppm) as oral suspension. Piglets were allowed to suckle its mother's milk *ad libitum* during the growth trial, till the completion of duration of study.

- Housing and management: The experimental animals (piglets) were maintained in three groups along with their respective sows in separate concrete pens of size 4.2 x 2.8 meter. Pigs were allowed in open yard in the morning for exercise and had access to grass and sunlight. Plenty of fresh water was made to available all the time. During the whole experimental period the sows along with their piglets were kept under hygienic condition. Pens were daily washed and kept dry and clean. Fifteen days prior to farrowing deworming was carried out using pipeparzine adipate at the rate of 100mg per kg body weight. Sows were allowed *ad libitum* access to feed i.e., the kitchen waste was fed in concrete feeders provided in the pens and for the entire duration of the study the piglets were allowed to suckle milk from the respective mothers without any extra supplementation
- Incidence of nonspecific scours: Fecal consistency was measured daily using a macroscopic score from 1-4 (firm to watery) (Figure 2). If daily feces could not be assigned unequally to one of those classes, a mid-score was appointed. Diarrhoea was defined as liquid consistency (score 4 and greater) over a minimum of two consecutive days [16]. Diarrhoea incidence (%) was calculated as a quotient from the number of newly affected piglets during the first eight weeks before. In order to verify whether diarrhoea is associated with the dietary zinc treatments, scours occurrence was recorded every day throughout the study period.
- **Preparation of media**: All the media employed for *E coli* counts were prepared as per the guidelines of [4].

# Media, Reagent, and Chemicals

- Media: Violet Red Bile Agar
- Reagents: Phosphate buffer Saline (PBS, pH 7.4)

• **Composition**: 0.8 g sodium chloride. 0.2 g potassium chloride, 1.44 g disodium hydrogen phosphate, 0.24 g potassium dihydrogen phosphate and 1000 ml sterile distilled water, pH 7.5



**Figure 1:** Incidence of nonspecific scours (diarrhoea) (%) of the experimental piglets during the eight weeks trial period.

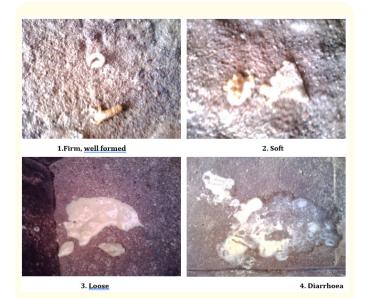


Figure 2: Fecal Consistency score.

# **Methods**

• **Collection of fecal samples:** Samples were collected once a week from day three to day sixty postpartum, by using a

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sampling spoon, rectal fecal samples were collected from piglets, placed in a sterile jar and transferred to the laboratory where analysis was started on the same day of collection.

- **Isolation of organisms:** The following different media was used in the present study for isolation of E coli
  - Violet Red Bile Agar
  - Phosphate Buffer Saline (PBS) agar.

The collected material was plated on to the above-mentioned different media following standard procedures. All the plates were incubated aerobically at 37°C for twenty-four hours. The lactose fermenting colonies were transferred to nutrient agar slants and incubated at 37°C fortwenty four hours and stored at 4°C for further identification (Figure 4).

#### Viable counts

The spread plate method described by [21] was employed to enumerate the surface viable counts (Figure 3). Prior to inoculation, nutrient agar plates were dried for at least two hr at 37°C. Viable counts were estimated using tenfold serial dilution of the bacterial suspensions directly obtained from broth culture and free cells and after vigorous vortexing of bentonite clay in test tube for the biofilm cells were made in sterile PBS. Agar plates in five replicates were seeded with one ml of each dilution once spread widely with a sterile glass spreader and incubated at 37°C.After twenty hour the colony were carried out in all three plates. The dilutions that were giving the counts between one and hundred per plates were considered. The average number of colonies per plate was multiplied by the dilution factor to obtain in the original suspension and expressed as cfu/g of feces.



Figure 3: Enumeration of colony forming unit.



Figure 4: Culture plate showing growth of E.coli on VRB agar.

#### **Statistical analysis**

Data on growth performance, incidence of nonspecific scours, piglet mortality, skin coat condition and gut microbial status were analyzed by ONE WAY ANOVA using statistical software (SPSS, Version 16) for windows (2008).

### **Results and Discussion**

# **Incidence of nonspecific scours**

The incidence of diarrhoea in control (T1), zinc oxide ( $T_2$ ) and zinc methionine ( $T_3$ ) groups were (35%), (20%) and (25%) respectively (Table 2 and Figure 1). In the control group (T1), only seven piglets were seen with diarrhoea, whereas in the zinc oxide supplemented group ( $T_2$ ) four piglets were observed with diarrhoea and with zinc methionine supplemented group (T3) five piglets had shown diarrhoea. The incidence of diarrhoea was seen more inT1 than  $T_2$  and  $T_3$  groups. In almost all the three groups the incidence of diarrhoea were seen more commonly during the first, second and fifth and sixth week of the trial period. Statistical analysis revealed that there were no significant differences between T1,  $T_2$  and  $T_3$  respectively (Table 3).

Particulars	T1	<b>T</b> <sub>2</sub>	<b>T</b> <sub>3</sub>
Number pigs with diarrhoea		4	5
% of pigs with diarrhoea	35	20	25

**Table 2:** Effect of feeding T1, T2 and T3 on incidence of non-specific scours expressed as percentage.

F value is not significant.

ANOVA Table	SS	df	MS	
Treatment (between columns)	6.667	7	0.9524	
Residual (within columns)	4.667	16	0.2917	
Total	11.33	23		

**Table 3:** Analysis of variance for incidence of nonspecific scours in response to different zinc sources in piglets (n = 20).

The results of the present study are in accordance with earlier observations made by [19,28]. But, these authors conducted experiments with higher doses zinc (2500 to 3000 ppm of Zn) to piglets and were found successful in reducing the incidence of diarrhoea. However, the results of the present experiment are in contrary with the findings of [12] in which they observed that supplementation of 2500 ppm of ZnO didn't reduce the incidence of diarrhoea.

Diarrhoea in piglets is common and the occurrence is affected by many factors such as climate, age at weaning, nutrition, exposure to different environment and management conditions [17]. It causes considerable economic loss to the pig industry. The study by [30] revealed that high dietary Zn could provide enhanced protection in the intestinal tract and stimulate the systemic humoral immune response against TGEV infection.

According to [17] the etiology of the diarrhoea is often complex and includes interactions between sow (litter size, parity, postpartum dysgalactia syndrome), piglet (birth weight, birth order, genotype), environmental conditions (temperature, crate/pen, season) and management system(stockman ship, attendance at farrowing, vaccination), and also can be related to numerous changes in the early-weaned piglet, such as morphological [9] and functional alterations of the small intestine [7], changes in intestinal colonization with predominance of *E. coli* [13] and weakening of the immune system [30].

#### Conclusion

In the current study, diarrhoea was prevalent in piglets during the first five weeks of age, although the incidence was more in  $T_1$ compared to the other zinc supplemented groups  $T_2$  and  $T_3$  but, the results were not significant statistically. The high frequency of diarrhoea during first week especially, during the first three days after birth is mainly due to limited ability to cope with environmental stressors (cold, disease and limited nutrition) which predisposes the piglets to relatively high rates of neonatal morbidity and mortality [14]. From four to fourteen days of age, piglet diarrhoea may be associated with various enteric pathogens. The agents include: E coli, Clostridium perfringens type C, Transmissible gastro enteritis virus (TGE), Corona virus, Isospora suis, rotavirus, and occasionally Serpulina byodysenteriae, usually there will be one primary causative agent, but piglets suffer with mixed infections [6]. During third, fifth and sixth week the incidence of diarrhoea is mainly due to decrease in the milk production in the sows at the same time piglets start to eat the solid food. This could be due to a lack in the capacity to produce sufficient amounts of gastric acid and digestive enzymes [3] in GIT, which makes piglets more susceptible for dietary changes. In an experiment in pigs [9,20] on change of of nutrition from twenty one to thirty five days may lead to changes in the histology and biochemistry of the small intestine, which will lead to a reduction in digestive and absorptive capacity. Indigestible food in the lower gut allows pathogenic strains to multiply, which may lead to diarrhea [8].

In the current study supplementation of zinc, reduced the incidence of diarrhea in piglets. But the possible reason for the non-significance incidence of diarrhea among the  $T_1$ ,  $T_2$  and  $T_3$  groups might be due to the feco-oral contamination of zinc, since there was no control over the piglets supplemented with and without zinc.

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