Egyptian Journal of Veterinary Sciences

https://ejvs.journals.ekb.eg/

The Effects of Nano Zinc on Humoral Immunity, Blood Biochemical Indicators, Thyroid Hormones, and Carcass Characteristics of Broilers

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Abstract

THIS STUDY examined the effects of different concentrations of nano zinc oxide (ZnONPs) on the features of the carcass, weight of the bursa and thymus, kidney and liver functions, thyroid hormones, and humoral immunity in broilers that were reared from 1 to 35 days old. 360 male birds (arbor acres) one days old were comprised into six treatment groups. Each group consisted of six replicates. Six diets were designed to cover the nutritional needs of chicks, with the exception of zinc, which was given at three levels: 100% (of the strain guide recommended level), 50%, and 25% of traditional or nano zinc oxide was given. The results showed that the inclusion of ZnONPs significantly enhanced carcass weight in the source major effect when compared to the conventional source. Furthermore, the addition of ZnONPs at different levels resulted in the best carcass weight in the interaction (source x level). The percentages of the dressing, abdominal fat, bursa, thymus, and giblets (liver, gizzard, and heart) in the main source, levels, or interaction between treatments did not differ significantly. Additionally, birds fed diets containing ZnONPs showed enhanced immunity (humoral immunity). No significant differences between the different ZnONPs levels were detected. Thyroid hormone levels, kidney and liver functions did not significantly differ amongst the different treatments. According to the current findings, it could be concluded that adding ZnONPs had the best effects in terms of carcass weight and humoral immunity without having a negative effect on thyroid hormones, liver, or kidney function.

Keywords: Broiler, Zinc Oxide Nanoparticles, humoral immunity, thyroid hormones, blood biochemical parameters.

Introduction

The poultry industry, which is also the fastest growing farming sector and a major economic force, is leading the way in meeting consumer demand for meat as well as improving the nutritional value of human food. The sector has grown in prominence because of its quick results [1]. High animal productivity is ensured when poultry feed has readily available, high-quality ingredients that meet demand. A variety of types of minerals, including chelates, inorganic and organic compounds, and nanoparticles, could be fed to animals [2-3].

Zinc is an essential microelement that affects many biological processes, such as hormones and DNA synthesis. In addition to its role as a cofactor of several enzymes such as metabolism enzymes and antioxidant enzymes [3-5].

Since zinc in nano form has unique properties, which enable it to be used as a feed ingredient [6]. Utilizing nanotechnology in broiler nutrition may offer new methods for enhancing the birds' nutritional uptake, which would enhance their growth, digestibility of nutrients, and productivity [7]. In comparison to the control, the broiler diets supplemented with nano zinc showed improved body weight with no negative effects on urea, triglycerides, total protein, albumin, globulin, and liver function [8].

The addition of zinc oxide NPs (ZnO NPs) in broiler diets led to improved broiler productive performance, immunity and antioxidant state without any negative effect on thyroid hormone levels, liver function and kidney function [9]. Adding 40 mg/kg of ZnO nanoparticles to broiler diets increased productivity [10]. Thyroid function was improved in broilers given 40 mg ZnO NPs/kg as compared to the control group [11]. In comparison to the unchallenged control group fed basal diet, which comprises 75 mg/kg of inorganic zinc oxide, it was

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(Received 24 September 2024, accepted 02 December 2024)
DOI: 10.21608/EJVS.2024.323489.2388

demonstrated that adding 20 mg ZnO NPs/kg to the diet of challenged chicks with coccidian showed a substantial improvement in broiler productive performance [12].

Therefore, this study aims to compare the effect of zinc oxide in its conventional and nano forms on carcass characteristics, humoral immunity, thyroid hormones and blood biochemical parameters.

Material and Methods

The experiment site

The field experiment was conducted at the Poultry Nutrition Research Unit (PNRU), which is housed at Cairo University's Agriculture College in Giza, Egypt. Egypt's national research center also performed all laboratory analyses.

Experimental Materials

Sources of zinc: zinc oxide was bought from a chemical company, whereas according to Ismail *et al.* [13], Samy *et al.* [14], and Menazea *et al.* [15] nano zinc oxide was chemically manufactured using Sol-Gel techniques.

Experimental design and diets

The purpose of the chick growth experiment was to examine the effects of zinc oxide nanoparticles on the carcass characteristics (carcass weight, dressing %, giblets, bursa and thymus), blood biochemical parameters, liver and kidney functions, thyroid hormones and humoral immunity (hemagglutination inhibition test) of broiler chicks from 1 to 35 days of age. 360-day-old male chicks (Arbor Acres) were kept in pens, and the birds were divided into six treatment groups (6 groups x 6 replicates × 10 chicks each).

The diets were balanced in elements and covers all the nutritional needs required according to the strain guide, except for zinc, which is added at three levels of 100%, 50% and 25% of the strain required needs, whether in its traditional form or in nano form.

All of the replicates' initial chick weights were within a reasonable range of 42g. The chicks had unrestricted access to feed and water. During the experimental study, 16 hours a day of light were supplied.

Measured items

Features of the carcass and the slaughter

Six chicks per treatment group were slaughtered at 35 days of age to evaluate the carcass, the weight of immune organs. Prior to slaughter, calm the birds by minimizing their stress levels during handling and transportation. After the birds were slaughtered and their feathers removed, measurements were made of their live weights and the weight of their carcass to calculate dressing percentage, also weight of giblets which contained the liver, heart, and gizzard in addition to the weight of immune organs (bursa and thymus were be recorded. The percentages of each organ to the live body weight in each bird were calculated independently.

Blood biochemical parameters

The blood samples were collected during slaughtering in plain tubes. The tubes were centrifuged for 15 min at 2000 g to get the serum, which was stored for later analysis at -20 °C.

Serum was examined using commercially available kits (Bio Diagnostic, Cairo, Egypt) in an automatic spectrophotometer with a high-performance readout (FlexorEL200 Biochemical Analyzer) to quantify AST, ALT, albumin, creatinine, and alkaline phosphatase (ALP).

Thyroid hormones

T3/T4 ratio was calculated after total T3 and T4 determination using ELISA Kits from My Bio Source Inc. in San Diego, California.

Immune state

To find out how it affects immunity. Immunization levels against the Newcastle disease (ND) and Avian flu (H5) vaccines were determined using blood serum samples that were subjected to the hemagglutination inhibition (HI) test. hemagglutination was indicated by expressing all titers as the logarithm of the reciprocal of the highest dilution [16].

Statistical analysis

Two-way analysis of variance was employed to statistically assess the data using SAS [17] General Liner Model. The new multiple range test developed by Duncan [18] was utilized to identify the difference in means at ($P<0.05$).

Results

Carcass characteristics

The effect of feed manipulations on carcass characteristics is shown in Table 1. When compared to the conventional source, the addition of nano zinc greatly increased ($P\leq0.05$) carcass weight in the source main effect. Furthermore, the addition of nano zinc at various levels produced the best ($P\leq0.05$) carcass weight in the interaction (source X level); however, no significant ($P>0.05$) variations were seen in the level main effect of carcass weight.

No significant differences ($P>0.05$) were observed in the percentages of the carcass, abdominal fat, bursa, thymus, and giblets (liver, gizzard, and heart) in main source, levels, or interaction (source X level) between treatments during all of the periods. The addition of conventional or nano zinc sources at varying levels

showed no effect on the carcass features of broiler chicks, while gave the superior carcass weight.

Thyroid hormones

The main effect of source, levels and the interaction (Source X Level) of treatments on thyroid hormones are shown in Table 2. No significant differences (P>0.05) recorded among all treatments compared to the control in T3, T4 and the ratio between them. Which indicated that the addition of nano zinc with different levels to broiler diets had no negative effect on thyroid hormones.

Blood biochemical parameters

The main effect of source, levels and the interaction (source X level) of treatments on blood biochemical parameters are shown in Table 3.

No significant differences (P>0.05) observed between all treatments in the liver function (ALT and AST) which indicated that the addition of nano zinc at different levels had no adverse effect on the liver function. In addition to no significant differences (P>0.05) in creatinine level among all treatments, which indicated that no negative effects on the kidney function.

Immune state

At 35 days of age, Figure 1 shows the effects of various treatments on the hemagglutination inhibition test (HI) against avian flu (H5) and Newcastle disease (ND).

The addition of zinc oxide in nano form led to significantly (P<0.05) enhanced in the immune state of birds against H5 and ND compared to the control group.

Discussion

In comparison to the control group, broilers fed 90 mg ZnO NPs/kg at 35 days showed a significant improvement in body weight gain and carcass weight [19]. When compared to the control group fed conventional zinc oxide, broilers fed 40 mg ZnO NPs/kg showed a substantial (P<0.05) improvement in carcass, thymus and spleen weight but did not differ significantly in bursa weight [10, 16]. El-Maddawy *et al*. [12] demonstrated that there were no statistically significant variations in the relative weights of the spleen, liver, kidney, gizzard, and heart between the control group and the coccidially challenged chicks given 20 mg Zno NPs/kg. While there were no significant variations in the relative weight of thymus, broilers fed deferent levels of ZNO NPs for 35 days had significantly higher carcass, liver, spleen, and bursa weight in compared to the control group [5].

The addition of ZnO-NPs in broiler diets led to improved performance and reduced occurrences of oxidative stress and inflammation [21].

Zinc oxide NPs is more effective in Zn utilization when compared with conventional ZnO, so used low quantity of ZnO NPs compared to traditional ZnO, while producing comparable or even superior results [22].

Triiodothyronine (T3) and thyroxine (T4) levels in serum of birds fed nano zinc containing meals were not affected [23]. When compared to broiler diets fed traditional sources of zinc oxide or sulphate, there were no discernible changes in T3, T4, or T3/T4 ratio and no negative effects on functions of kidney and liver in birds fed diets containing nano zinc, whether from chemical or green synthesis [9].

The obtained results of the superior effect of nano zinc to enhanced humoral immunity were agreed with the previous studies of Eskandani *et al*. [24] who found that the addition of zinc oxide in low levels led to improved humoral immunity in broiler chicks. Hatab *et al*. [3] concluded that all immunological organs showed histological alteration and increased both types of immunity.

Conclusion

According to our findings, the inclusion of nano zinc oxide provided the best carcass weight, and humoral immunity (Hemagglutination inhibition test) while having no adverse impacts on thyroid hormones, liver, or kidney functions. As a result, the appropriate amount of nano zinc is 25%.

Acknowledgments

Not applicable.

Funding statement

This study didn't receive any funding support

Declaration of Conflict of Interest

The authors declare that there is no conflict of interest.

Ethical of approval

The Agricultural Research Center's institutional animal care and use committee (ARC-IACUC) granted this project ethical approval with number ARC-APRI-25-24.

TABLE 1. Effect of dietary treatments on carcass characteristics.

Item							Giblets		
		Carcass weight (g)	dressing %	Abdominal fat %	Bursa %	Spleen %	Liver %	Gizzard %	Heart %
Zinc oxide level %	Zinc oxide Source								
100%	CZnO	1295[b]	74.66	1.39	0.07	0.12	2.87	1.65	0.47
50%	CZnO	1169[c]	73.80	0.86	0.08	0.10	2.21	1.66	0.44
25%	CZnO	1155[c]	74.73	0.71	0.09	0.09	2.35	1.66	0.51
100%	NZnO	1450[a]	75.15	1.03	0.09	0.13	2.22	1.59	0.43
50%	NZnO	1471[a]	74.48	0.84	0.07	0.13	2.53	1.52	0.45
25%	NZnO	1393[a]	74.50	0.86	0.08	0.06	1.89	1.47	0.49
SE of means		±36.70	±0.98	±0.15	±0.02	±0.03	±0.16	±0.15	±0.04
Main effects									
Source									
CZnO		1206[b]	74.43	1.02	0.08	0.10	2.50	1.60	0.48
NZnO		1438[a]	74.71	0.91	0.08	0.11	2.22	1.53	0.46
SE of means		±27.29	±0.56	±0.09	±0.01	±0.02	±0.09	±0.09	±0.03
Level %									
100%		1372	74.91	1.21	0.08	0.12	2.54	1.62	0.45
50%		1320	74.14	0.87	0.07	0.12	2.39	1.48	0.45
25%		1274	74.62	0.79	0.09	0.07	2.12	1.57	0.50
SE of means		±24.38	±0.69	±0.11	±0.02	±0.02	±0.11	±0.11	±0.03
Significances									
Source of variation			NS	NS	NS				
Source effect		***	NS	NS	NS	NS	NS	NS	NS
Level effect		NS	NS	NS	NS	NS	NS	NS	NS
Source × Level		***				NS	NS	NS	NS

Means designated with the same letter in the same column are not significantly different at 0.05 level of probability.

TABLE 2. Effect of dietary treatments on thyroid hormones of broiler.

Item		T3	T4	T4/T3
Zinc oxide level %	Zinc oxide Source			
100%	CZnO	4.38	17.94	4.16
50%	CZnO	4.37	22.21	5.21
25%	CZnO	3.88	15.03	4.13
100%	NZnO	4.26	17.17	4.14
50%	NZnO	4.39	22.36	5.16
25%	NZnO	4.26	16.65	4.00
SE of means		±0.41	±1.74	±0.74
Main effects				
Source				
CZnO		4.21	18.40	4.50
NZnO		4.30	18.73	4.43
SE of means		±0.23	±1.00	±0.43
Level %				
100%		4.32	27.55	4.15
50%		4.38	22.29	5.19
25%		4.07	15.84	4.07
SE of means		±0.29	±1.23	±0.52
Significances				
Source of variation				
Source effect		NS	NS	NS
Level effect		NS	NS	NS
Source × Level		NS	NS	NS

Means designated with the same letter in the same column are not significantly different at 0.05 level of probability.

Table 3. Effect of dietary treatments on blood parameters of broiler.

Item		ALT (U/L)	AST (U/L)	Creatinine (mg/dl)
Zinc oxide level %	**Zinc oxide Source**			
100%	**CZnO**	14.53	186.63	0.51
50%	**CZnO**	14.30	174.50	0.41
25%	**CZnO**	15.57	177.03	0.17
100%	**NZnO**	13.53	172.63	0.44
50%	**NZnO**	14.03	178.47	0.51
25%	**NZnO**	13.93	184.10	0.66
SE of means		±0.90	±9.97	±0.13
Main effects				
Source				
CZnO		14.80	179.39	0.37
NZnO		13.83	178.40	0.54
SE of means		±0.52	±5.75	±0.07
Level %				
100%		14.03	179.63	0.48
50%		14.17	176.48	0.46
25%		14.75	180.57	0.41
SE of means		±0.64	±7.05	±0.09
Significances				
Source of variation				
Source effect		NS	NS	NS
Level effect		NS	NS	NS
Source × Level		NS	NS	NS

Means designated with the same letter in the same column are not significantly different at 0.05 level of probability.

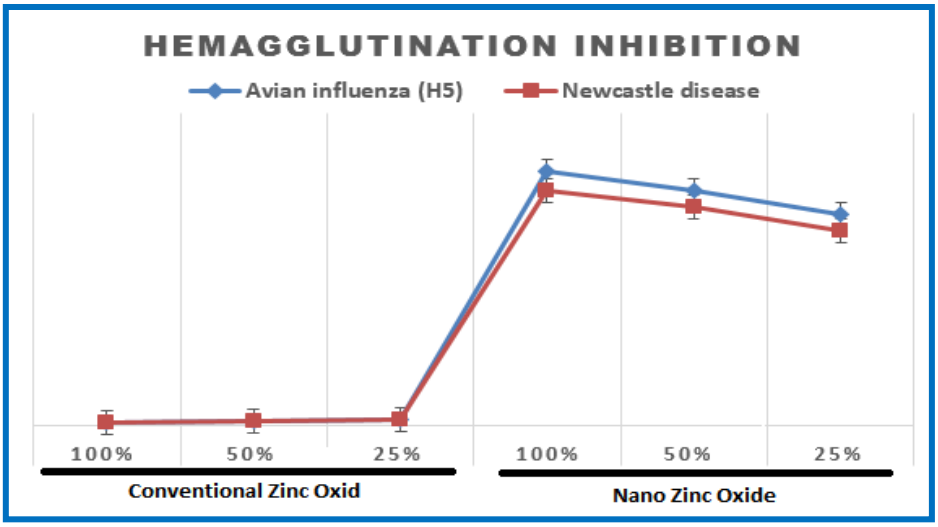

Fig. 1. The effect of different treatments on hemagglutination inhibition (HI) at 35 days of age.

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تأثير الزنك النانوي على المناعة الخلطية، والمقاييس الكيميائية الحيوية في الدم، وهرمونات الغدة الدرقية، وخصائص الذبيحة

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الملخص

تناولت هذه الدراسة تأثير تركيزات مختلفة من أكسيد الزنك النانوي على خصائص الذبيحة ووزن الأعضاء المناعية ووظائف الكلى والكبد وهرمونات الغدة الدرقية والمناعة الخلطية في دجاج التسمين الذي تم تربيته من عمر 1 إلى 35 يومًا. تم تقسيم 360 طائرًا ذكرًا (Arbor Acres) بعمر يوم واحد إلى ست مجموعات تجريبية. تتكون كل مجموعة من ستة مكررات. تم تصميم ستة علائق لتناسب الاحتياجات الغذائية للكتاكيت، باستثناء الزنك، الذي تم إعطاؤه على ثلاثة مستويات: 100٪ (من مستوى دليل السلالة الموصى به)، و 50٪، و 25٪ من أكسيد الزنك في صورتة التقليدية أو النانوية. أظهرت النتائج أن إدراج أكسيد الزنك النانوي عزز بشكل كبير ($P \leq 0.05$) وزن الذبيحة في التأثير الرئيسي للمصدر عند مقارنته بالمصدر التقليدي. علاوة على ذلك، لم يُظهر التأثير الرئيسي لنسب الإضافة المختلفة أي تأثير معنوي علي وزن الذبيحة ($P> 0.05$)؛ ومع ذلك، فإن إضافة الزنك النانوي بمستويات مختلفة أدي الي أفضل وزن للذبيحة ($P \leq 0.05$) في تأثير التداخل بين المصدر x مستوي الإضافة. بينما لم تختلف النسب المئوية لنسبة التصافي ودهون البطن والأعضاء المناعية والأحشاء الداخلية (الكبد والقوانص والقلب) سواء في المقياس الرئيس لمصدر الزنك المستخدم أو مستويات الإضافة أو التداخل بين المصدر x مستوي الإضافة المعالجات ($P> 0.05$) . بالإضافة إلى ذلك، أظهرت الطيور التي تتغذى على علائق تحتوي على أكسيد الزنك النانوي مناعة محسنة (المناعة الخلطية). لم يتم ملاحظة أي فروق ذات دلالة إحصائية ($P> 0.05$) بين التركيزات المختلفة لأكسيد الزنك النانوي. كما لم تختلف مستويات هرمون الغدة الدرقية ووظائف الكلى كما يتضح من مستويات الكرياتينين ووظائف الكبد ($P> 0.05$) بين المعاملات المختلفة. ونتيجة لذلك، لم يتم ملاحظة أي آثار سلبية على هرمونات الغدة الدرقية ووظائف الكبد والكلى.

ووفقا للنتائج الحالية، يمكن الاستنتاج أن إضافة أكسيد الزنك النانوي كان له أفضل التأثيرات من حيث وزن الذبيحة والمناعة الخلطية دون أن يكون له تأثير سلبي على هرمونات الغدة الدرقية أو وظائف الكبد و الكلى.

الكلمات الدالة: دجاج التسمين، أكسيد الزنك، الجسيمات النانوية، خصائص الذبيحة، المناعة الخلطية، هرمونات الغدة الدرقية، المقاييس الكيميائية الحيوية للدم.