EFFECT OF ADDING NATURAL AND NANO ZINC OXIDE TO THE DITE ON SOME GROWTH PARAMETERS OF *CYPRINUS CARPIO* L .

Ayat. A. Al-Noori **A. J. Abuelheni** **M. S. Al-Khshali**
Researcher **Chief Res.** **Prof.**
Coll. Agric. Engine. Sci, University of Baghdad / Iraq
Ayat.Alaa1201a@coagri.uobaghdad.edu.iq **Mohammed.shaker@coagri.uobaghdad.edu.iq**

ABSTRACT

The research aimed to study the effect of natural and nano zinc oxide on some growth parameters of common carp *Cyprinus carpio*. Seven treatments were used, including the control without any addition, the second, third and fourth treatments by adding nano-zinc oxide at concentrations of 25, 50 and 75 mg/kg feed, respectively, and the fifth, sixth and seventh treatments by adding natural zinc oxide in the same concentrations above. The results showed that the fourth treatment was superior to the traits of the total weight gain, daily growth rate, relative and specific growth rate over the other treatments, which is an indication that the fish benefit from feed enhanced with nanoparticles by 75%, and it differed significantly (p<0.01) from the rest of the treatments. T4 treatment was recorded the best daily growth was 0.29 g/day The best feed conversion ratio was recorded in the same treatment. It can be concluded from the study that the addition of zinc nanoparticles to the diet of fish improves the level of performance and growth without causing any harm to the fish.

Key words: carp , weight gain, relative and specific growth rate, feed conversion ratio

مجلة العلوم الزراعية العراقية- 2024:55(5):1612-1619 النوري وآخرون

تأثير اضافة أوكسيد الزنك الطبيعي والنانوي للعليقة في بعض مؤشرات النمو لأسماك الكارب الشائع *CYPRINUS CARPIO* L.

آيات علاء النوري عبد الكريم جاسم ابو الهني محمد شاكر الخشالي
باحث رئيس باحثين علميين استاذ
كلية علوم الهندسة الزراعية ، جامعة بغداد

المستخلص

هدف البحث إلى دراسة تأثير أوكسيد الزنك الطبيعي والنانوي في بعض مؤشرات النمو لأسماك الكارب الشائع *Cyprinus carpio*. استخدمت سبع معاملات تضمنت معاملة السيطرة بدون أي اضافة والمعاملة الثانية والثالثة والرابعة بإضافة مادة أوكسيد الزنك النانوي بتراكيز25 و50 و75 ملغم/كغم علف على التوالي والمعاملة الخامسة والسادسة والسابعة بإضافة أوكسيد الزنك الطبيعي بالتراكيز نفسها أعلاه. اظهرت النتائج تفوق المعاملة الرابعة في صفات الزيادة الوزنية الكلية ومعدل النمو اليومي ومعدلات النمو النسبي والنوعي على بقية المعاملات وهي إشارة الى إستفادة الأسماك من الغذاء المدعوم بالجسيمات النانوية بنسبة 75%، واختلفت معنوياً (p<0.01) عن بقية المعاملات، وسجلت المعاملة الرابعة افضل نمو يومي بلغ 0.29 غم/ يوم وسجلت أفضل نسبة تحويل غذائي في المعاملة نفسها. يستنتج من الدراسة ان اضافة اوكسيد الزنك النانوي الى العليقة الغذائية في الأسماك يحسن من مستوى الاداء والنمو دون ان يسبب اي ضرر للأسماك.

الكلمات المفتاحية: الكارب، الزيادة الوزنية، معدل النمو النسبي والنوعي، نسبة التحويل الغذائي

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INTRODUCTION

Aquaculture is the fastest growing agricultural sector and has become a global industry that provides a large percentage of animal protein, with nearly half of the amount of fish destined for human consumption, because of its great importance in the growth and building of the body (13), and to fill the shortage in food sources, which represents with high prices are the most important challenges facing human societies (10).Fish is considered one of the main food sources of proteins, vitamins and minerals, it is superior to other animal sources in terms of the percentage of protein in its meat (18). Fish occupies the forefront in many countries of the world, with rice as a good source of nutrition for many peoples, especially in Asia and Africa (1, 24). In addition to the importance of fish wealth, it contributes to reducing poverty by increasing employment opportunities in fisheries through appropriate investment, which results in an increase in income for both fishermen and food processors (15). For its great advantages of common carp, as it contributes 71.9% of fresh water production due to its rapid growth and its ability to withstand unfavorable environmental conditions and changes in temperature and oxygen (6). Nanotechnology represents the technology of manufacturing small particles at the atomic level from its basic materials, which are less than 100 nanometers in size, for use in many fields, and it has the enormous potential and capabilities to create and synthesize new and unique materials (21).The current study aims at the possibility of fortifying the feed ingredients with nanoparticles in order to know the extent to which fish benefit from them, and to identify the growth rates of fish after adding different levels of nano- and natural zinc oxide, and to determine the optimal ratio that achieves the best growth of fish.

MATERIALS AND METHODS

Study site and experimental fish: A total of 150 common carp *Cyprinus carpio*, with an average weight of 44 g In the Livestock and Fisheries Center of the Agricultural Research Department / Ministry of Science and Technology for the period from 3/15/0222 to 6/13/0222, were brought from fish farms in Babil province. They were acclimated for 15 days before starting the experiment, during which they were fed on a commercial diet. 84 fish were selected and distributed in 14 glass tanks with dimensions of 30 cm x 40 cm x 60 cm, with 6 fish in each tank, with two replications for each treatment.

Experimental design

The experiment was divided into 7 treatments, which included the control treatment without any addition, the second, third and fourth treatments by adding zinc nanoparticles at concentrations of 25, 50 and 75 mg/kg feed, respectively, while natural zinc oxide was added at the same concentrations above for each of the fifth, sixth and seventh treatments.

Preparation of the nanomaterial

Zinc oxide nanomaterial's were prepared in the Graduate Studies Laboratory - College of Agricultural Engineering Sciences / University of Baghdad by adopting the physical method using the Sol-gel technology, as 20 grams of aqueous zinc acetate was mixed with 120 ml of distilled water and stirred for 20 minutes. At a temperature of 35 °C to produce a solution of zinc acetate again, 80 g of sodium hydroxide powder was weighed and mixed with 80 ml of distilled water at a temperature of 35 °C and stirred for 20 minutes to produce a solution of sodium hydroxide, after which both solutions were mixed by adding 100 ml of ethanol in the form of drops accompanied by vigorous continuous stirring for 90 minutes to complete the reaction to obtain a gel-like solution (gel), then the gel was dried at a temperature of 80 °C for 16 hours and then calcined in the oven at a temperature of 250 °C for 4 hours, then we obtain on zinc nanoparticles (Fig. 3-2) (12).

Feed processing

The components of the diet were brought from the local markets in Baghdad governorate and consisted of animal protein (ready ground), soybean meal, yellow corn, local barley, wheat bran, vitamins and salts. The materials were mixed with each other and the required proportions of nano- and natural zinc oxide were added to it after dissolving it in 20 ml of edible oil. The ration was left for three days to homogenize. The fish were fed on the manufactured ration at a rate of 3% of the total weight of the fish. According to the treatments, this percentage was adopted after the feed was provided to the fish in gradual

proportions of 1, 2, 3 and 4%, and it stabilized at 3% during the acclimatization period.

The studied traits of fish

Total Weight Gain (T.W.G.): It was calculated from the equation mentioned by Al-Shiblawi and Al-Khshali. (8).

weight gain (g) = final weight (g) - starting weight (g)

Daily Growth Rate (D.G.R.).: It was calculated from the equation which was pointed out by Al-Hilali and M. Al-Khshali (4).

D.G.R. = (W2 – W1) / (T2 – T1)

Since:

W1 = first weight (g)

W2 = second weight (g)

T2 - T1 = the duration of the experiment or between the two weights in days.

Relative Growth Rate (R.G.R.): According to the equation mentioned by Alkafagy and Al-Khshali (7)

RGR= (W2-W1)/W1*100

Specific Growth Rate (S.G.R.): It was calculated from the equation mentioned by Al-Hassani and mustafa (3).

S.G.R. ={ (Ln W2 – L n W1) / (T2 – T1) }x 100

Ln W2 = natural logarithm of the second weight at time T2

Ln W1 = natural logarithm of the first weight at time T1

T2 - T1 = the time between the two

Weights

Feed Conversion Rate (F.C.R.): It was calculated from the equation mentioned by Al-Hassani and mustafa (3**)**

F.C.R =Weight of feed given to fish (g/fish)/ The wet weight gain of fish (g/fish)

Statistical analysis

The Complete Randomized Design (CRD) was used to analyze the effect of the experimental treatments on the studied traits using the ready statistical program (SAS) Statistical Analysis System (23). Significant differences between the mean of the studied traits were tested using Duncan´s Multiple Range Test (9) at a significant level 0.05 according to the following mathematical model equation:

Yij = μ + Ti + Eij

RESULTS AND DISCUSSION

Environmental factors: The water temperatures of the basins ranged between 17.5-29°C, at a rate of 23°C, the pH ranged between 7 -7.8, dissolved oxygen ranged between 6-8.2, and the total dissolved solids (T.D.S.) ranged from 515 to 690 mg/l. These ranges located within the safe levels for the survival and growth of common carp fish. Because common carp fish are warm water fish, the ideal temperatures for them lie between 23-28 °C. (20) mentioned that the ideal and appropriate degree for the growth of common carp fish is 25 ° C. As for (2) It has been shown that common carp fish have the ability to withstand wide ranges of temperatures ranging from 10-30 °C.

Weight gain rates

The results of the statistical analysis of the characteristic of total weight gain and daily growth rate (Table 1) showed that the fourth treatment (75 mg of zinc nanoparticles) was significantly increase ($P<0.01$) compared to a treatments followed by the T3 and then T2 treatments, while no significant differences ($P>0.01$) were recorded. between T6 and T7, the lowest percentage was recorded for the control treatment, which reached to 4.77 g/ fish and 0.05 g/day/fish for each of the total weight gain and daily growth rate. The reason for the superiority of the fourth treatment may be due to several factors including the small size of the nanoparticles and their homogeneity with the diet, which led to the ease of absorption in the gastrointestinal tract and the use of nutrients. T4 achieved the highest rates of weight gain in the experimental fish, and this may be due to the fact that the nanomaterial adsorbs the nutrients necessary to improve performance and immune response (19). In addition to the health status of different fish species, ZnO supplements delivered innanoparticle form may be transported into cells more rapidly than organic forms (16).(Kishawy 14) confirmed that zinc nanoparticles showed significant improvements in body weight, feed conversion and specific growth rate when compared to the groups supplemented with natural zinc. Alishahi (5) indicated that the nanoform of zinc nanoparticles has a high efficiency compared with the natural zinc oxide particles, that supplementation of diets containing ZnO-NPs significantly improved the growth and metabolic functions of Indian carp *Labeo rohita* (24). Inclusion of zinc in the form of Nano-ZnO is a useful application for

improving fish performance, activating digestive enzymes, and improving the antioxidant capacity of enzymes. Feeding of common carp on a diet fortified with natural and nanoparticles of zinc oxide for 84 days increased final weight gain compared to the control treatment (Tabel 2), as the fourth treatment (75 mg nano-zinc oxide) was superior, followed by T3 and T2 in all periods, and treatment T7 recorded the lowest weight gain (75 mg of natural zinc oxide). It is noted that the body weight improved in treatments containing zinc oxide particles in both its nano and natural forms compared to the control treatment. On the other hand, it was noted that zinc nanoparticles are more effective in terms of bioavailability and intestinal absorption than other chemical forms (23). Ghazi (11) confirmed an increase in the rate of weight gain and final body weight when feeding *Oreochromis niloticus* on feed supplemented with nano-zinc.

Tabel 1. Average fish weight (SE) in different treatments during the experimental period

Duration / Treatment	Fish weight rates (g/fish every 14 days)							Total weight gain in average weights (g / fish)	Daily growth rate (g/day/fish)
	Initial weight rate	14 day	28 day	42 day	56 day	70 day	Final weight		
T1	44.67	43.30	45.53	46.24	47.485	48.58	49.44	4.77±0.35 e	0.055±0.01 e
T2	44.81	44.29	47.47	51.35	55.42	57.16	57.91	13.10±0.71 c	0.15±0.01 c
T3	44.55	44.37	47.51	51.83	56.74	60.65	61.55	17.01±0.31 b	0.20±0.01 b
T4	44.83	44.71	48.72	54.60	62.34	66.85	69.15	24.31±0.24 a	0.29±0.01 a
T5	44.78	43.66	46.45	49.64	50.44	53.37	53.92	9.13±0.53 d	0.11±0.10 d
T6	44.83	43.49	46.50	48.60	51.28	54.36	55.39	10.56±1.62 cd	0.13±0.02 cd
T7	44.88	43.60	46.480	49.64	52.37	52.37	54.76	11.32±0.99 cd	0.13±0.01 cd
significati on level								**	**

Vertically different letters indicate that there are significant differences among the averages, while similar letters indicate that there are no significant differences

Tabel 2. Changes in average weights of common carp during different periods

Duration / Treatment	Weight gain rate (g / fish)					
	14 day	28 day	42 day	56 day	70 day	84 day
T1	-1.36±0.14 b	2.22±0.20 d	0.71±0.06 e	1.24±0.08 d	1.10±0.05 c	0.86±0.22 b
T2	-0.51±0.15 a	3.18±0.12 b	3.87±0.02 bc	3.89±0.55 bc	1.925±0.26 c	0.74±0.16 b
T3	-0.18±0.16 a	3.14±0.09 b	4.32±0.06 b	4.91±0.22 b	3.905±0.53 ab	0.90±0.44 b
T4	-0.12±0.08 a	4.01±0.25 a	5.880±0.420 a	7.73±1.06 a	4.510±0.16 a	2.30±0.40 a
T5	-1.12±0.07 b	2.79±0.13 cb	3.19±0.32 cd	0.80±0.69 d	2.930±0.37 abc	0.54±0.41 b
T6	-1.34±0.02 b	2.51±0.12 cd	2.60±0.02 d	2.67±0.32 cd	3.080±1.13 abc	1.03±0.28 b
T7	-1.285±0.285 b	2.88±0.08 bc	3.16±0.17 cd	2.73±0.19 cd	2.39±1.25 abc	1.44±0.11 ab
signification level	**	**	**	**	*	**

Vertically different letters indicate that there are significant differences among the averages, while similar letters indicate that there are no significant differences

Relative and specific growth rate: Fish weights are taken every two weeks to know the response of the fish to the diet and to identify the growth characteristics more accurately. Tabel 3 showed the Relative growth rate of decreased for a period of 14 day in all treatments due to the low temperature, With the gradual rise in temperatures after the first two weeks of the experiment, the average fish weights began to rise and gradually increase and the fourth treatment recorded a significant difference ($P<0.01$) for a period of 28 days, as it recorded of 8.96% compared to the rest treatments, and for the same period it was not recorded significant differences ($p<0.01$) among the treatments T2, T3,T5 and T7. Fourth treatment was significantly difference in the period of 42 day compared with treatments T1, T2, T3, T4, 5T, T6 and T7, while no significant differences were recorded between treatments T2 and T3, treatment T3 also outperformed treatments T1, T5, T6 and T7 for the same period. The superiority of the fourth treatment continued for a period of 56 day over all treatments, and for a period of 70 day, also, treatment T4 excelled over all treatments, while no significant differences $p>0.05$ were recorded among treatments T1, T2, T3, 5, T6 and T7 within the same period. The results of the statistical analysis showed that the highest relative growth rate was for the fourth treatment in the period of 84 days, was significantly increase $p< 0.01$ compared to all treatments. Specific growth rate, (Tabel 4) showed that the fourth treatment is superior in all periods over the treatments T1, T2, T3, 5T, T6 and T7 for the period of 28, 42 and 56 day, there is a highly significant difference $p<0.01$ of the treatment T4 followed by the treatment T3, T2, for the period 70 day, and 84 days no significant differences were $p>0,01$ recorded among the treatments T1, T2, T3, 5T, T6 and T7. The current results indicate that the decrease in the relative growth rate for the period of 14 day may be attributed to the decrease in temperature to 17.2 °C, which led to lethargy, lack of appetite, and lack of feed intake, then the relative and specific growth rate improved when the temperature gradually increased, which led to the possibility of improving growth performance of fish fed on diets supplemented with zinc nanoparticles. Mohammady (18) found that a diet containing ZnO-NPs enhanced growth performance, health, immunity, and biomarkers of oxidative stress in Nile tilapia *Oreochromis niloticus*. Mahmoud and Al Khshali. (17) reported when feeding fish on a diet supplemented with zinc oxide nanoparticles at concentrations of 20 and 30 mg / kg, the results showed a significant increase in body weight and the rate of relative and specific growth rate. It is clear from the results of the statistical analysis of the feed conversion ratio (Table 5) in the period of 14 days that all treatments recorded negative values due to the low temperature. Significant ($P\leq0.01$) on treatments 1T, 5T, 6T and T7, while no significant differences were recorded between treatments T2, 3T, T5 and T7, The fourth treatment continued to be significantly superior ($P\leq0.01$) in all periods The positive results obtained in the treatments fortified with nano- and natural zinc oxide compared with the control treatment for growth standards (daily weight gain, feed conversion ratio and efficiency, relative and specific growth rates), may be attributed to the role of zinc supplementation in increasing the surface area for nutrient absorption and thus increasing the efficiency of Benefiting from the food compared to the control diet, and the superiority of the fourth treatment may be due to several reasons, including the acceptance and palatability of the fish to the diet containing fine particles of zinc oxide nanoparticles. It was concluded that the addition of zinc nanoparticles to common carp fish diets by 75% led to an increase in body weight, as well as an improvement in growth parameters (relative and specific growth rate, feed conversion ratio).

Tabel 3. Relative growth rate of common carp in different treatments

Duration / Treatment	Relative growth rate (%) 14 day	28 day	42 day	56 day	70 day	84 day
T1	-3.05 ±0.30 b	5.13 ±0.46 d	1.57 ±0.15 e	2.68 ±0.18 d	2.31 ±0.11 b	1.77 ±0.46 ab
T2	-1.14 ±0.34 a	7.18 ±0.29 b	8.16 ±0.05 bc	7.57 ±1.06 bc	3.48 ±0.44 ab	1.31 ±0.31 b
T3	– 0.40 ±0.36 a	7.09 ±0.23 b	9.09 ±0.12 b	9.47 ±0.43 b	6.88 ±0.92 ab	1.50 ±0.75 ab
T4	– 0.27 ±0.18 a	8.96 ±0.54 a	12.06 ±0.78 a	14.19 ±2.14 a	7.23 ±0.29 a	3.44 ±0.60 a
T5	-2.51 ±0.17 b	6.39 ±0.26 bc	6.87 ±0.74 cd	1.62 ±1.40 d	5.80 ±0.65 ab	1.04 ±0.80 b
T6	- 2.99 ±0.05 b	5.77 ±0.28 cd	5.65 ±0.03 d	5.50 ±0.67 cd	5.99 ±2.17 ab	1.89 ±0.47 ab
T7	-2.86 ±0.63 b	6.60 ±0.21 bc	6.80 ±0.38 cd	5.50 ±0.38 cd	4.58 ±2.41 ab	2.63 ±0.25 ab
signification level	**	**	**	**	*	*

Vertically different letters indicate that there are significant differences among the averages, while similar letters indicate that there are no significant differences

Tabel 4. Specific growth rate of common carp in different Treatments

Duration / Treatment	Specific growth rate (%/day) 14 day	28 day	42 day	56 day	70 day	84 day
T1	-0.22 ±0.02 b	0.36 ±0.03 d	0.11 ±0.01 e	0.19 ±0.01 ed	0.16 ±0.01 b	0.12 ±0.03 ab
T2	-0.08 ±0.02 a	0.49 ±0.01 b	0.56 ±0 bc	0.52 ±0.07 bc	0.24 ±0.03 ab	0.09 ±0.02 b
T3	-0.02 ±0.02 a	0.48 ±0.01 b	0.62 ±0.01 b	0.64 ±0.02 b	0.47 ±0.06 ab	0.10 ±0.05 ab
T4	-0.02 ±0.01 a	0.61 ±0.03 a	0.81 ±0.05 a	0.94 ±0.13 a	0.50 ±0.02 a	0.24 ±0.04 a
T5	-0.18 ±0.01 b	0.44 ±0.02 bc	0.47 ±0.05 cd	0.11 ±0.09 e	0.40 ±0.04 ab	0.07 ±0.05 b
T6	-0.21 ±0.01 b	0.40 ±0.02 cd	0.39 ±0.01 d	0.38 ±0.04 dc	0.41 ±0.14 ab	0.13 ±0.03 ab
T7	-0.20 ±0.04 b	0.45 ±0.01 bc	0.47 ±0.03 cd	0.38 ±0.02 cd	0.31 ±0.16 ab	0.18 ±0.01 ab
signification level	**	**	**	**	*	*

Vertically different letters indicate that there are significant differences among the averages, while similar letters indicate that there are no significant differences

Tabel 5 . Feed conversion efficiency of common carp in different treatments

Duration / Treatment	Feed conversion efficiency(%)					
	14 day	28 day	42 day	56day	70day	84day
T1	152.53-16.05± c	34.29 3.119± d	7.83 0.79± e	13.39 0.93± d	11.60 0.54± a	17.63 4.55± b
T2	56.94-17.02± b	47.90 1.96± b	39.97 0.54 ± bc	38.72 6.19± bc	17.40 2.22± a	13.08 3.01± b
T3	1.51-1.12± a	47.37 1.50± b	44.61 0.21± b	48.24 1.30± b	34.39 4.65± a	20.50 2.10± b
T4	13.66-9.19± ab	59.88 3.58± a	60.36 3.94± a	70.96 10.69± a	36.18 1.51± a	34.40 6.04± a
T5	125.71-8.55± c	42.64 1.80± bc	34.30 3.74 ± cd	13.97 1.12± d	23.13 9.12± a	16.55 1.88± b
T6	149.34-2.32± c	38.52 1.92± cd	28.18 0.17± d	27.60 3.39± cd	29.99 10.90± a	18.90 4.75± b
T7	143.69-32.13± c	44.10 1.49± bc	34.01 1.86 ± cd	27.49 1.87± cd	22.93 12.09± a	26.40 2.43± ab
signification level	**	**	**	**	NS	*

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