The growth performance and some physiological aspects for common carp *Cyprinus carpio* L. diets with different levels of chia seed powder *Salvia hispanica L.*

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Abstract

An experiment was conducted To evaluate the effect of adding different levels of chia seed powder *Salvia hispanica* to the diets On growth and some physiological aspects for common carp *Cyprinus carpio* Five diets with equal protein content 27.17% were used, included five treatments, including a control treatment, with a total of 50 fish. The fish were randomly distributed into 10 tanks 5 fish/ tank, with two replicates per treatment, and the experiment lasted for 90 days. Statistical analysis revealed a significant improvement in production performance with increasing levels of supplementation. Treatment T4, with the highest supplementation level (4%), recorded the highest final weight (338.50 g) and weight gain (177.26 g). This excelled continued in both Relative Growth Rate RGR and Specific Growth Rate SGR, as well as the best feed efficiency, represented by a lower feed conversion ratio (1.785) and a higher Feed Conversion Efficiency (55.99%) for Treatment T4. The results also showed improvement in some hematological indicators. The same treatment recorded the highest values for red blood cell count (1.26×10^6 cells/mm^3) and hemoglobin concentration (10.67 g/dL), while Treatment T3 (3%) recorded the highest white blood cell count, indicating stimulation of the immune response .Regarding liver enzymes, the results showed no significant differences in Alanine Transaminase ALT levels between treatments. However, a significant decrease in Aspartate Transaminase AST was observed at certain addition levels, along with a slight increase in Alkaline Phosphatase ALP in treatment T3 3%, without any apparent negative effects on liver function. The results indicated that adding 4% chia seed powder improved growth indicators and some hematological characteristics, without any clear signs of liver stress under the experimental conditions.

Keywords: Chia seeds, common carp, productive performance, hematological characteristics, liver enzymes.

Introduction:

Aquaculture development is a cornerstone of enhancing global food security, requiring the development of balanced diets based on high-value, sustainable feed ingredients. This need has led to increased interest in using plant-based sources rich in bioactive compounds, including chia seeds *Salvia hispanica L.*, which are considered important functional foods for aquatic organisms. Chia seeds possess a unique nutritional composition, including a high protein content, unsaturated fatty acids, particularly omega-3 fatty acids (α-linolenic acid), dietary fiber, phenolic compounds, and antioxidants, making them a suitable ingredient for improving fish productivity and health.Several studies have indicated that incorporating chia seeds or their derivatives into fish diets can enhance growth and feed utilization efficiency. One study demonstrated that using vegetable oils rich in

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unsaturated fatty acids, such as chia oil, improves fatty acid composition in fish tissues and increases omega-3 content, thus enhancing the nutritional value of the fish product [10]. Another study showed that replacing some traditional fat sources with functional vegetable oils improved growth indicators and reduced the accumulation of harmful fats in fish [3]. Regarding the functional properties of chia seeds, a recent study confirmed that chia seeds are a rich source of bioactive compounds, such as polyphenols, which play an important role in enhancing antioxidants and improving the health of living organisms [7]. Another review also showed that chia seed powder possesses technological and functional properties that contribute to improved digestion and increased nutrient absorption, which positively impacts productivity [11]. [13] demonstrated that regular consumption of chia seeds is associated with improved biochemical indicators and reduced oxidative stress due to their high antioxidant content. Similarly, [5] showed that chia oil is characterized by a high concentration of essential fatty acids, making it effective in supporting physiological functions and improving tissue quality in living organisms. Based on the above, it is clear that using chia seeds, whether in powder or oil form, represents a promising strategy in fish nutrition to improve growth and health. However, determining the optimal levels of supplementation and its effects under different rearing conditions still requires further applied studies, which this study aims to evaluate.

Materials and Methods

Common carp fingerlings (*Cyprinus carpio*) were obtained from a reputable and accredited fish hatchery, with an average initial weight of 32.2 ± 0.3 g. Upon arrival at the fish rearing laboratory of the Animal and Fisheries Department,Scientific Research Commission, Al-Zaafaraniya, Baghdad, the fish were sterilized using a 5 g/L saline solution and then acclimatized for 10 days in glass tanks designated for the experiments. The water source was the Tigris River. Environmental factors were monitored throughout the experiment. Water temperature ranged from 22–26 °C, dissolved oxygen concentration from 6-6.5 mg/L, pH from 7.2–8, and total ammonia concentrations from 0.02–0.05 mg/L. These values were within the limits suitable for fish growth.

Preparation of the Experimental Diets

Chia seeds were ground using a dedicated laboratory grinder. Five balanced diets were then prepared, each with an equal protein content of 27.17%. The diets consisted of basic ingredients including animal protein, soybean meal, yellow corn, wheat bran, barley, and wheat, in addition to a vitamin and mineral supplement to meet the fish's nutritional requirements.

Chia seed powder was added to four treatments at rates of 1%, 2%, 3%, and 4%, replacing the other ingredients in the diet. The fifth diet served as the control, without the addition of chia seeds.

Experimental Design and System Management

The experimental fish were randomly distributed into 10 glass tanks (90 liters per tank), with two replicates per treatment, and five fish were placed in each tank. A static rearing system was implemented, with a 50% partial water change daily to maintain water quality.

The fish were fed twice daily (morning and evening) until apparent satiety. The experiment lasted for 90 days. Statistical Analysis

The data were statistically analyzed using SPSS software. One-way ANOVA was used to examine the differences between treatments. Initial and final weights were recorded based on the total live mass of fish within each tank (5 fish), not on the weight of individual fish.

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Therefore, each tank was considered the experimental unit. Means were compared using the least significant difference (LSD) test at a significance level of $P \leq 0.05$. Results were expressed as mean ± standard error (ME).

Results and Discussion

table1: Effect of adding different levels of chia seed powder to the diet on the productive performance of common carp *Cyprinus carpio*.

Treatment	InitialWeight (g)	Final Weight (g)	Weight Gain (g)	RGR (%)	SGR	FCR	FCE (%)
Control	161.44±0.11a	298.61±1.86c	137.17±1.98c	84.97±1.28c	0.73±0.01c	2.135±0.05a	46.82±0.0
T1 (%1)	161.26±0.04a	299.59 ± 0.79c	138.33±0.83c	85.78±0.54c	0.735±0.05c	2.155±0.05a	46.40±0.0
T2 (%2)	161.29±0.11a	305.90±0.22bc	144.61±0.11bc	89.66±0.01bc	0.76±0.01bc	2.085±0.05b	48.00±0.1
T3 (%3)	161.27±0.16a	321.78±4.03b	160.51±3.87b	99.53±2.30b	0.825±0.15b	1.93 ± 0.05c	51.79±1.2
T4 (%4)	161.24±0.04a	338.50±4.22a	177.26±4.18a	109.94±2.56a	0.885±0.015a	1.785±0.25d	55.99±0.7

Means with different letters within the same column differed significantly at $P \leq 0.05$.

At the end of the rearing period, a significant advantage was observed with increasing levels of chia seed powder. Treatment T4 resulted in the highest final weight (338.5 g), followed by treatment T3 (321.78 g). The control and T1 treatments did not differ significantly and recorded the lowest values. This trend reflects a gradual positive response to increasing levels of chia seed powder supplementation in the diet. Weight gain also showed similar results, with treatment T4 recording the highest weight gain (177.26 g), followed by treatment T3 (160.51 g). The control and T1 treatments recorded the lowest weight gain. This indicates improved nutrient utilization efficiency with increasing levels of supplementation. This is consistent with what was shown in the study by [1] on tilapia fish, where the use of chia seeds led to a significant improvement in final weight and weight gain with increasing level of supplementation. Similarly, [9] demonstrated that chia seed powder led to a marked increase in final weight and growth indicators. Furthermore, [6] showed that chia seed oil enhances fish productivity. This is attributed to the high nutritional value of chia seeds, particularly their protein and essential fatty acid content [8]Regarding growth rates (RGR) and specific growth rates (SGR), treatment T4 recorded the highest values for both RGR and SGR, indicating enhanced metabolic processes and increased protein deposition efficiency. In contrast, the control and T1 treatments recorded the lowest values, reflecting limited utilization of the feed in the absence or low levels of supplementation.

This aligns with the findings of [6] showed a significant improvement in SGR with the use of chia seed oil. Similarly, [1] demonstrated improved growth indicators with chia supplementation. [9] also showed improved relative and qualitative growth rates. This can be attributed to the role of fatty acids and active compounds in enhancing metabolic efficiency. Regarding the feed conversion ratio (FCR), treatment T4 achieved the best

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feed conversion efficiency at 1.78 (the lowest value), followed by treatment T3 at 1.93. The control and T1 treatments recorded the highest values, indicating a low feed conversion efficiency in these treatments. Feed conversion efficiency (FCE) also showed a significant improvement in treatment T4, reaching 55.99%, followed by treatment T3 at 51.79. This confirms the inverse relationship between FCE and FCE and supports the hypothesis that adding chia seed powder contributes to improved feed utilization.

The study by [1] showed improved feed conversion efficiency when chia seeds were added to tilapia fish diets. Furthermore, the study by [9] demonstrated that chia seeds enhance feed utilization efficiency. This is attributed to improved digestibility resulting from their bioavailable nutrients [8] which aligns with the findings of [6]that showed improved feed conversion efficiency

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Table2: Effect of chia seed powder addition levels on hematological characteristics of common carp.

Treatment	RBC*10^6 (Cell/mm³)	Hb (g/dl)	WBC*10^3 (Cell/mm³)	PCV (%)
Control	1.14 ± 0.02^{c}	9.65 ± 0.35^{b}	10.75 ± 1.25^{ab}	29.00 ± 1.00^{a}
T1	1.11 ± 0.01^{c}	9.97 ± 0.64^{b}	9.50 ± 0.50^{b}	30.00 ± 2.00^{a}
T2	1.12 ± 0.02^{c}	9.66 ± 0.33^{b}	9.25 ± 0.75^{b}	29.00 ± 1.00^{a}
T3	1.22 ± 0.09^{b}	9.98 ± 0.68^{b}	11.50 ± 0.50^{a}	30.00 ± 2.00^{a}
T4	1.26 ± 0.04^{a}	10.67 ± 0.67^{a}	10.00 ± 1.00^{b}	32.00 ± 2.00^{a}

Means with different letters within the same column differ significantly at P≤0.05.

The results in Table (2) indicate a significant effect of chia seed powder supplementation levels on some hematological traits in common carp. Red blood cell (RBC) count showed a significant increase in treatment T4 compared to the other treatments, followed by treatment T3. No significant differences were found between the Control, T1, and T2 treatments. This excelled may be attributed to the physiological role of chia seed powder in promoting red blood cell formation due to its content of unsaturated fatty acids (omega-3) and antioxidants, which contribute to improved blood cell production and increased oxygen transport capacity in fish. These results were consistent with the hemoglobin (Hb) concentration characteristic, as treatment T4 showed a significant advantage over the other treatments. This aligns with the red blood cell (RBC) results, reflecting improved oxygen transport capacity due to stimulated hemoglobin production. This may be due to the high nutritional content of chia seed powder, which supports hematopoiesis.

The significant improvement in RBC and Hb in treatment T4 is consistent with previous studies on the use of chia seeds in fish feed. [1] demonstrated that adding chia seeds to Nile tilapia diets led to a clear improvement in hematological and immunological indicators, including increased RBC and hemoglobin. [9] also showed that increasing chia powder levels in tilapia diets resulted in a significant increase in RBC and Hb, reflecting improved oxygen transport efficiency and the physiological

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status of the fish. This improvement may be attributed to the nutritional composition of chia seeds, which contain high levels of polyunsaturated fatty acids, particularly α-linolenic acid (ALA), in addition to phenolic compounds and antioxidants. These contribute to reducing oxidative stress and improving the efficiency of physiological processes related to hematopoiesis, a finding also noted by [2] in their study on the role of dietary supplements in improving the health and immunity of fish. Regarding white blood cell (WBC) count, treatment T3 showed a significant improvement compared to treatments T1, T2, and T4, while it did not differ significantly from the control treatment. This suggests the role of chia seed powder in stimulating the immune system. Chia seed powder contains bioactive compounds such as phenols and antioxidants that enhance the immune response and increase the efficiency of the fish's biological defenses. The significant increase in WBC count in treatment T3 may be an indicator of immune system stimulation, which aligns with the findings of [2] that plant supplements act as immune stimulants by promoting the production of white blood cells. The difference in the level of response between T3 and T4 indicates that the immune effect is not always directly proportional to the level of supplementation, as some studies have shown that there is an optimal level of supplementation that gives the best immune response, which can also be explained in light of the results of [9].

These findings support other studies on plant supplements. [4] demonstrated that the use of moringa leaves in fish diets significantly increased red blood cell count (RBC), white blood cell count (WBC), and hemoglobin (Hb), reflecting the positive role of plant compounds in improving hematological parameters. These compounds, such as phenols and flavonoids, also play a significant role in enhancing immune system efficiency and improving the physiological response of fish.

Regarding packed cell volume PCV The results showed no significant differences between all treatments, indicating the stability of this trait and its lack of effect from chia seed supplementation levels. The absence of significant differences in PCV between treatments suggests that the effect of chia seed powder on this trait was limited, which aligns with [14] finding that some hematological parameters in fish may remain stable despite improvements in others, depending on the nature of the physiological response and the type of dietary treatment. In general, the results of this study indicate that chia seed powder is an effective feed additive in improving certain hematological characteristics of common carp. High levels (T4) enhanced oxygen transport efficiency by increasing red blood cell count (RBC) and hemoglobin (Hb), while intermediate levels (T3) were more effective in stimulating the immune response. This suggests that the effect of chia seed powder is level-dependent and requires determining the optimal level to achieve the best physiological response.

Table 3: Effect of adding different levels of chia seed powder on the liver enzyme activity of common carp.

Treatment	ALT (U/L)	AST (U/L)	Alkaline Phosphatase (IU/L)
Control	57.00 ± 5.00^{a}	66.00 ± 1.00^{a}	12.07 ± 8.83^{b}
T1	50.50 ± 11.50^{a}	63.00 ± 4.00^{a}	14.95 ± 8.45^{b}
T2	52.00 ± 0.00^{a}	58.00 ± 1.00^{b}	11.75 ± 7.35^{b}

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T3	54.50 ± 2.50^{a}	61.00 ± 1.00^{ab}	25.15 ± 0.35^{a}
T4	62.00 ± 0.00^{a}	57.00 ± 2.00^{b}	20.70 ± 13.33^{ab}

Means with different letters within the same column differ significantly at $P \leq 0.05$.

Table 3 shows that adding chia seed powder to common carp diets did not significantly affect ALT enzyme activity, as no significant differences were recorded between all treatments, indicating the stability of this indicator across different levels of supplementation.

Furthermore, the results showed significant differences in AST enzyme activity. The control and T1 treatments recorded the highest values compared to treatments T2 and T4, while they did not differ significantly from treatment T3. As for Alkaline Phosphatase (ALP) activity, the results showed a significant advantage for treatment T3 compared to all other treatments, except for treatment T4, which did not differ significantly from it. The stable ALT enzyme activity across all treatments indicates that the addition of chia seed powder did not negatively affect liver cell integrity. This enzyme is a sensitive biomarker of liver damage, and its unchanged level reflects stable physiological conditions and the absence of stress or damage to the liver. This aligns with [14], who suggested that some physiological indicators may remain within normal levels despite dietary changes, provided no direct tissue damage has occurred.The relative decrease in AST enzyme activity in treatments T2 and T4 compared to the control is a positive indicator of improved liver function and reduced oxidative stress, as elevated AST levels are associated with liver or muscle tissue damage. This improvement can be attributed to the protective role of chia seeds, which contain unsaturated fatty acids and antioxidants that contribute to reducing oxidative damage and protecting liver cells. This finding is consistent with those of [1] and [9]. This finding also supports the findings of [12] who indicated that plant-based supplements rich in bioactive compounds and antioxidants improve liver function and reduce the activity of liver enzymes such as ALT and AST by mitigating oxidative stress and protecting cells from damage. Regarding the ALP enzyme, the significant increase in the T3 treatment may reflect an increase in metabolic activity or the stimulation of certain vital processes associated with liver function, where this enzyme is linked to metabolism and tissue growth. Furthermore, the lack of significant differences between T3 and T4 suggests that the increased supplementation level did not have a negative effect but rather maintained the balance of enzyme activity within normal limits. This aligns with [2], who suggested that natural food supplements may selectively influence certain physiological parameters without affecting overall homeostasis. In general, the results of this study indicate that the addition of chia seed powder, especially at medium and high levels, contributed to maintaining liver health and improving some of its functions by reducing oxidative stress, without causing negative effects on overall physiological balance. The response of liver enzymes was also variable, indicating that the effect of chia was selective and dependent on the type of enzyme and the level of addition.

Conclusion

The results showed that adding chia seed powder to common carp diets, particularly at a 4% level, significantly improved production performance, feed utilization efficiency, and some hematological characteristics. The results also showed no clear negative effects on liver enzymes, suggesting the potential of chia seed powder as a promising natural feed additive for fish.

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