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Dietary *Aloe vera* improves growth and hematology in Nile tilapia (*Oreochromis niloticus*)

Fatima Khan[*1,2]

[1]*Department of Marine Sciences, Ningbo University, Ningbo, Zhejiang, China*
[2]*Department of Zoology, Wildlife and Fisheries, University of Agriculture, Faisalabad, Punjab, Pakistan*

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ABSTRACT

This study evaluated the effects of dietary *Aloe vera* supplementation on growth performance and hematological parameters of Nile tilapia (*Oreochromis niloticus*). A total of 90 fish were randomly assigned to three dietary treatments: T0 (control, 0% Aloe vera), T1 (1%), and T2 (1.5%), with three replicate tanks per treatment. Fish were fed at 7% of body weight daily for eight weeks. Growth parameters were recorded weekly, while hematological indices were analyzed at the end of the trial. Results showed that fish fed the T1 diet exhibited significantly higher final body weight and specific growth rate (SGR), along with a lower feed conversion ratio (FCR), compared to T0 and T2 ($p < 0.05$). No significant differences were observed in length among treatments. Hematological analysis revealed significant increases in white blood cell count (WBC), hemoglobin (Hb), hematocrit (Hct), and mean corpuscular volume (MCV) in the T1 group, whereas red blood cell count (RBC), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) were not significantly affected ($p > 0.05$). Water quality parameters remained within acceptable ranges throughout the study. The findings indicate that 1% *Aloe vera* supplementation enhances growth performance and selected hematological parameters in Nile tilapia, suggesting its potential as a natural and effective feed additive in aquaculture. Further research is recommended to evaluate long-term effects and underlying mechanisms.

***Corresponding Author:** Fatima Khan ✉ Fatimakhan081996@gmail.com

INTRODUCTION

Aquaculture has become the rapidly emerging food production sector globally. For millions of people, it is vital to their livelihoods and food security (Aziza *et al.*, 2020). Because of its quick growth, ability to adapt to many environmental conditions, and beneficial nutritional qualities, tilapia is one of the most extensively farmed species (El-Sayed and Fitzsimmons, 2023). Tilapia are recognized as the second most significant category of cultivated finfish following Carps *Cyprinus* species in freshwater aquaculture (Abwao *et al.*, 2023). Around 140 countries are reportedly involved in the global cultivation and trade of tilapia (Kaleem and Sabi, 2021). Tilapia is a great source of the vital macro- and micronutrients, therefore, helps to lower the incidence of malnutrition. It contains significant concentrations of minerals, omega-3 fatty acids, and bioavailable micronutrients that are vital for the neurodevelopment and vision of the young children as well as it also helps to lower the risk of different cardiovascular disorders (Obiero *et al.*, 2019). Tilapia is marketed as "nature's superfood" in Kenya due to its high bioavailable micronutrient content (Obiero *et al.*, 2019). Farming Tilapia is economically viable and technically sufficient. However, the sustainability and profitability of Tilapia production depend on the development of cost-effective and eco-friendly feeding strategies that promote health and growth.

Aquaculture encompasses not only the necessary diets but also additional food additives that are helpful for the promotion of rapid growth and sustainability (Gule and Geremew, 2022).

Therefore, different antibiotics (Desbois *et al.*, 2021), hormones (Bardhan *et al.*, 2021), enzymes (Madhulika *et al.*, 2025) and supplements (Kabir *et al.*, 2024) are being introduced in the culturing medium for the rapid growth and disease prevention of the Tilapia. WHO advocates the utilization of eco-friendly and plant-based additive for eradication of use of hazardous compounds that adversely impact the environment. Therefore, now adays there has been an increasing interest of the researchers in phytogenic food additives which include: plant extracts, herbal compounds, and plant-derived phytochemicals for improvement in the growth and overall health of the fish (Gruber *et al.*, 2025).

Aloe vera is a monocot that is a member of the Liliaceae/Asphodelaceae family (Behaiyn *et al.*, 2023). It has gained significant interest of the researchers because of various bio-active compounds present it which includes: flavonoids, saponins, anthraquinones, tannins, and alkaloids (Majumder *et al.*, 2022). Various researches have indicated the pharmacological properties of *Aloe vera* these include: anti-inflammatory (Salem and Moammer, 2024), antimicrobial (Li *et al.*, 2014; Yousaf *et al.*, 2025), anti-radiation (Zanuzzo *et al.*, 2017), and immunostimulatory effects (Mehrabi *et al.*, 2019).

Moreover, various studies report that *Aloe vera* acts as a growth enhancer in different fish species such as Catla catla (Yousaf *et al.*, 2025), *Clarias gariepinus* (Gabreil *et al.*, 2021) and in other animals as well such as broiler (Amber *et al.*, 2021) and rabbits (El-Kholy *et al.*, 2022).

Additionally, it has also been reported that it is also very crucial disease resistance agent in various animals like poultry (Khan *et al.*, 2022). In spite of all these promising results, minimum research has been done about the effects of selected inclusion levels (1% and 1.5%) for *Aloe vera* supplementation in Tilapia diets. Therefore, this study was designed for the assessment of the impact of incorporation of 1% and 1.5% *Aloe vera* into the diet on the growth performance and hematological parameter of Tilapia.

MATERIALS AND METHODS

The current study was carried out in the Fish Nutrition Laboratory within the Department of Zoology, Wildlife and Fisheries at the University of Agriculture Faisalabad. All experiments were conducted under the endorsement of ethical committee of the University of Agriculture Faisalabad. Additionally, every institutional

regulation and protocol established by the Institutional Animal Care and Use Committee (Protocol 18-051) concerning the care of the animals was also followed. Juvenile fish were collected from the fish farm of university of Agriculture, Faisalabad, Punjab, Pakistan. This experiment was conducted in a static aquaculture system. The system consisted of three light blue 100-liter tanks. The fish were cultivated in controlled aquaria within the laboratory setting for duration of eight weeks. The maximum average water temperature was 29.975 ± 0.47 while the pH was 7.79 ± 0.47. Photoperiod was consisting of 13h dark: 11h light. The fish were administered an experimental diet comprising the basal diet, which was further enhanced with the addition of 1% and 1.5% *Aloe vera*.

Experimental design

Ten fish were stocked in each aquarium, with three replicate tanks per treatment. All aquaria were maintained under identical environmental conditions. The experimental unit was considered to be the tank rather than individual fish. During sampling, three fish were randomly selected from each tank, and the mean values of the measured parameters were calculated. The experiment was conducted over a period of eight weeks.

Experimental diets were prepared by adding *Aloe vera* extract powder to the basal diet. Three diets were prepared with concentrations of 0% (T_0), 1% (T_1), and 1.5% (T_2) of *Aloe vera*. *Aloe vera* was added at 1 and 1.5 g per 100 g of fish feed to prepare 1% and 1.5% concentrations, respectively. Fish were fed twice a day, and extra feed was removed from the tanks to avoid toxicity. Sampling was performed weekly. Almost half of the total stocked fish were sampled, and data were recorded as variations in their length and weight. After the completion of the trial period, different growth parameters, such as FCR, SGR and condition factors, were calculated. In addition, hematological parameters were also analyzed at the end of the experiment. Moreover, water quality parameters were also calculated weekly during the trial period to assess weather water quality is maintain according to FAO/WHO criteria or not.

***Aloe vera* powder collection**

The *Aloe vera* powder was acquired from Insta Food Industries, which is a locally available product. The product was certified under ISO 9001:2008 and HACCP standards. The powder was composed of carbohydrates 19% and proteins 12%.

Basal feed preparation

The basal feed was purchased from the local market. It contained fish meal, rice polish, wheat flour, sunflower oil and soya bean meal. The feed was then crushed in the pestle and mortar to obtain a fine powder.

Preparation of experimental diet

The experimental diet for fish was prepared in laboratory of University of Agriculture Faisalabad. For preparation of the feed, basal fish feed purchased from local market was processed further. According to the experimental concentration, powdered basal feed was mixed with the *Aloe vera*. Three types of feed were prepared, one of which was a basal feed with 0% concentration of *Aloe vera*. This feed was used in the control treatment, denoted as T_0.

The other two concentrations were 1 g and 1.5 g of *Aloe vera* in 100 g of basal feed respectively, denoted as T_1 and T_2, respectively. The 1% and 1.5% concentrations of *Aloe vera* were used as experimental feed. A homogenizer and blender were used for mixing and homogenizing the feed.

The dried basal feed and *Aloe vera* feed were mixed for 5 minutes. Then sunflower oil was added in dried ingredients. Then the feed was stored in air tight jars and labelled. These air tight jars were stored at 4°C for preservation of the nutritional integrity of experimental diet. Throughout experimental period, test diet was provided to fish twice a day at feeding rate equivalent to 7% of their body weight.

Growth parameters

Various growth parameters were weekly measured. For this purpose, three of the fish were taken out of the tank and the parameters were studied in triplicate. These parameters include:

Weight gain (g)

The body weight of the fish from all the three aquariums was assessed weekly by using a digital weight balance. Following formula was used for calculation of weight gain.

Weight growth (g)= Final weight of the fish - Initial weight of the fish

Length gain (cm)

It was measured by using a scale. Fish was placed on the scale from the mouth to the caudal fin. The increase in length was assessed weekly.

Specific growth rate (SGR %)

Following formula was used for calculating the SGR:

SGR= (ln (final weight)- ln (initial weight) × 100)/T

Where T represents the duration in days.

Feed conversion ratio (FCR)

FCR was measured by using the following formula:

FCR = dry weight of feed administered (g)/weight increase of fish (g)"

Condition factor (g/cm³)

For the calculating the condition factor (K) following formula was used:

$K=(W/L^3) \times 100$

Blood collection and blood parameter

At the end of the experimental period different blood parameters were studied. Fish was fasted for 24 hours before collection of blood samples. Sterile syringes were used for the collection of blood from the caudal fin of the fish. Anesthesia was subjected to fish prior to sampling in order to mitigate any potential pain experienced during the procedure. Fish were immersed in the water bath containing the recommended concentration of tricaine methane-sulphonate (MS-222) i.e., 0.1mg/ml the fish were exposed to anesthesia for up to 2 minutes. At the end of the study, a comprehensive assessment of different blood parameters was conducted, this included the hemoglobin (Hb), total leukocyte count (TLC), mean corpuscular hemoglobin (MCH), total erythrocyte count (TEC), hematocrit value (Hct) and mean corpuscular volume (MCV).

For measuring RBCs in blood sample, total erythrocyte count (TEC) test was performed. Hemocytometer was used for counting the erythrocytes while the total RBCs were calculated by using the formula:

RBCs= (No. of cells counted × dilution × 400)/ (No. of small square count)

For measuring the number of WBCs in blood sample total leukocytes count (TLC) test was performed. Hemocytometer was used for calculating WBCs, while following formula was applied for calculation of the total White Blood Cell count (WBC).

WBC's= (No. of cells counted × dilution × 100)/(No. of square mm counted)

The cyanmethemoglobin method was employed for calculating the hemoglobin (Hb) in blood sample. 20µl of the blood sample was collected in a test tube them 5 ml of cyanmethemoglobin was added in it. The chemical was thoroughly mixed with the blood sample in test tube and set aside to stand. A jelly-like substance was appeared to be formed on the surface of the solution.

The tube was then centrifuged and the supernatant was eliminated. Then the measurement of optimal density of the sample was conducted by using a spectrophotometer.

The Mean Corpuscular Hemoglobin (MCH) is a parameter that quantifies the average quantity of hemoglobin present within an individual red blood cell. The value of Hct, represents percentage of RBCs within blood. Hematocrit value was measured by using the micro hematocrit capillary tube. The calculation is performed by taking the ratio of hemoglobin to hematocrit levels.

Statistical analysis

Upon the conclusion of the trial period, growth metrics and blood composition data were systematically compiled using Microsoft Excel (version 16). Mean values alongside their corresponding standard errors were calculated, and the resulting figures were presented using the same software application. Analysis of variance (ANOVA) with Statistix software (version 8.1) was applied for the analysis of the effect of *Aloe vera* on growth and hematological parameters of Tilapia. When significant differences were detected, treatment means were further compared using Tukey's Honestly Significant Difference (HSD) test at a significance level of $p < 0.05$.

RESULTS

Growth performance

The effect of dietary *Aloe vera* supplementation on growth performance of Nile tilapia is presented in Fig. 1.

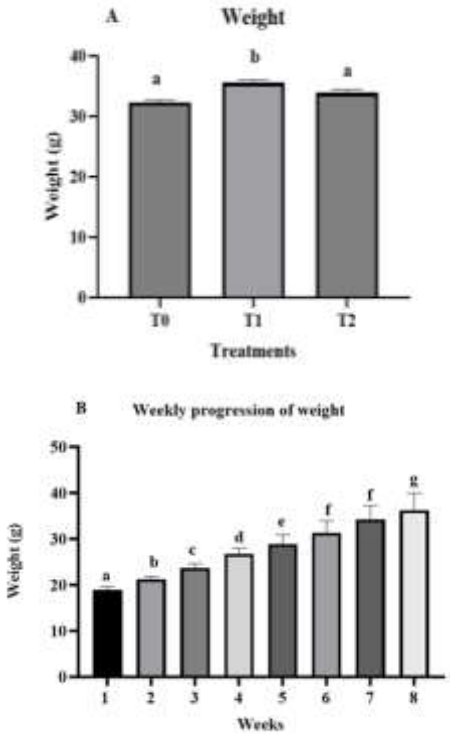

Fig. 1. Effect of *Aloe vera* supplementation on Nile tilapia growth over 8 weeks. (A) Final body weight (g) of fish fed different diets. T1 showed significantly higher weight than T0 and T2 ($p < 0.05$); T0 and T2 were not different. (B) Weekly body weight progression (g). Values are mean ± SD (n= 3). Different superscripts indicate significant differences among weeks (Tukey's HSD, $p < 0.05$).

Significant differences in final body weight were observed among treatments ($p < 0.05$). Fish fed the T1 diet attained the highest mean final weight (35.60 ± 0.74 g), followed by T2 (33.91 ± 0.73 g) and T0 (32.33 ± 0.67 g). Statistical analysis indicated that T1 differed significantly from both T0 and T2, whereas no significant difference was found between T0 and T2 (Fig. 1A).

Weekly body weight progression showed a consistent increase across the 8-week experimental period (Fig. 1B). Mean body weight increased from 18.93 ± 0.89 g in Week 1 to 36.18 ± 1.12 g in Week 8. Significant differences were observed among weeks ($p < 0.05$), with later weeks showing higher values than earlier weeks. Weeks 6 and 7 did not differ significantly from each other but were significantly lower than Week 8.

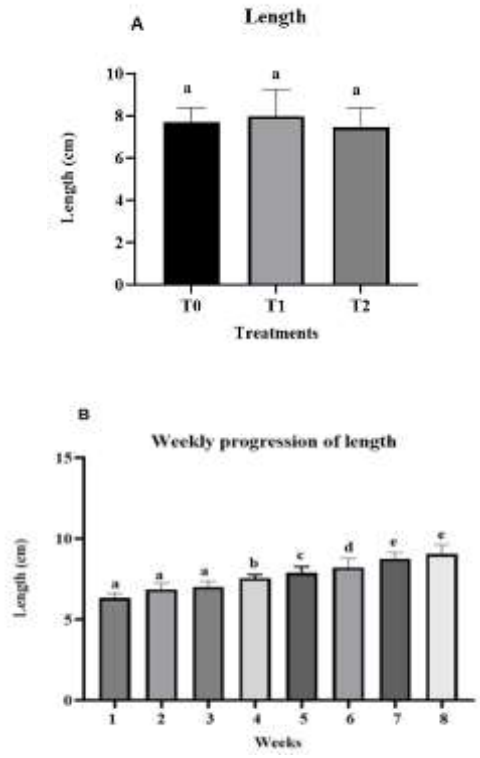

Fig. 2. Effect of dietary *Aloe vera* on Nile tilapia growth

(A) Mean length across treatments (To, T1, T2) over 8 weeks (ns, $p > 0.05$). (B) Weekly length progression; different superscripts indicate significant differences between weeks ($p < 0.05$, Tukey's test). Bars show mean ± SD.

Length growth

The influence of *Aloe vera* supplementation on fish length is shown in Fig. 2. Mean length values across treatments were not significantly different ($p > 0.05$), indicating no treatment effect on overall length gain (Fig. 2A).

However, weekly length progression demonstrated a significant increase over time ($p < 0.05$) (Fig. 2B). Fish in Weeks 4–8 showed significantly greater length compared to Weeks 1–3. The increase in length followed a progressive trend throughout the experimental period.

Hematological parameters

The hematological parameters of Nile tilapia in response to *Aloe vera* supplementation are presented in Fig. 3. Several parameters showed significant differences among treatments ($p < 0.05$).

White blood cell (WBC) count was significantly higher in T1 (15.75 ± 0.05 × 10^3/µL) and T2 (14.35 ± 0.05 × 10^3/µL) compared to To (11.51 ± 0.06 × 10^3/µL). Hemoglobin (Hb) concentration was highest in T1 (7.17 ± 0.28 g/dL), followed by T2 (5.74 ± 0.24 g/dL) and To (5.26 ± 0.39 g/dL), with T1 significantly different from the other treatments.

Hematocrit (Hct) values were also significantly higher in T1 (34.21 ± 0.55%) compared to T2 (31.83 ± 0.50%) and To (29.10 ± 0.33%). Mean corpuscular volume (MCV) showed a similar pattern, with T1 (260.5 ± 5.5 fL) significantly higher than T2 (228.28 ± 5.72 fL) and To (192.33 ± 2.66 fL).

Red blood cell (RBC) count did not differ significantly among treatments, although T1 showed the highest numerical value (1.275 × 10^6/µL). Similarly, mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) showed no statistically significant differences among treatments ($p > 0.05$), despite slight numerical variations.

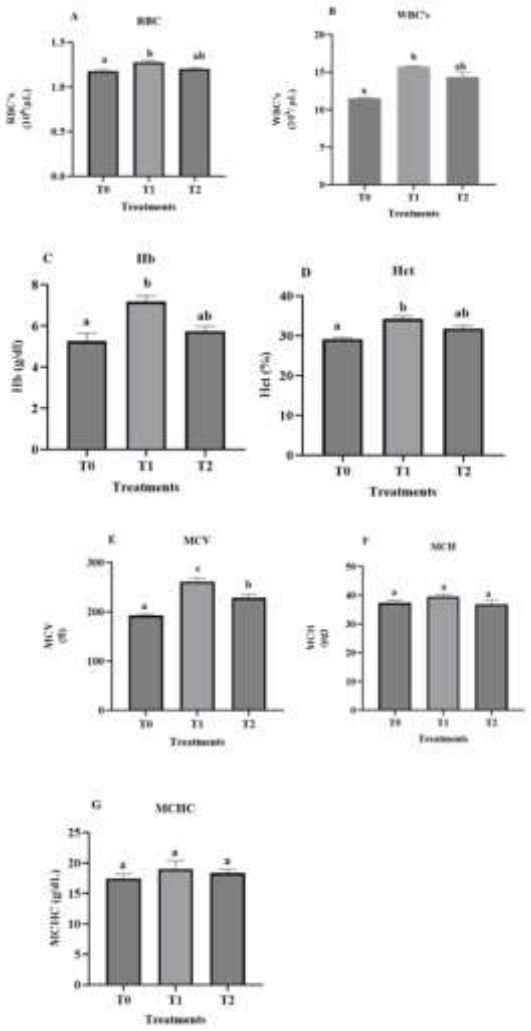

Fig. 3. Impact of varying doses of *Aloe vera* on certain blood parameters of Nile Tilapia after an eight weeks experiment

Treatment groups are as follows: To = control, T1 = 1% *Aloe vera*, and T2 = 1.5% *Aloe vera*. Measured parameters include: (A) red blood cell count (RBC) [10^6/µL], (B) white blood cell count (WBC) [10^3/µL], (C) hemoglobin (Hb) [g/dL], (D) hematocrit (Hct) [%], (E) mean corpuscular volume (MCV) [fL], and (F) mean corpuscular hemoglobin (MCH) [pg] (G) mean corpuscular hemoglobin concentration (MCHC)

DISCUSSION

Aquaculture plays a critical role in global food security by providing essential nutrients such as proteins, amino acids, vitamins, and minerals (Kwasek *et al.*, 2020; Ali *et al.*, 2022). However, the increasing use of synthetic additives, including antibiotics, hormones, and chemical supplements, raises concerns due to their potential negative impacts on human health and the environment (Salma *et al.*, 2022; Hossain *et al.*, 2018). Consequently, attention has shifted toward phytogenic feed additives as sustainable alternatives. Plant-derived compounds have been widely investigated for their capacity to enhance growth performance and physiological health in fish (Reverter *et al.*, 2021; Gabriel *et al.*, 2021). In this context, the present study evaluated the effects of *Aloe vera* supplementation at 1% and 1.5% on the growth and hematological responses of Nile tilapia.

The results demonstrated that dietary inclusion of *Aloe vera* significantly influenced growth performance, particularly weight-related parameters. Fish fed the T1 diet (1% *Aloe vera*) exhibited significantly higher final body weight and specific growth rate compared to the control (T0) and higher-dose group (T2). In contrast, no significant differences were observed in length among treatments, indicating that *Aloe vera* primarily affected weight gain rather than linear growth. These findings are consistent with previous studies reporting enhanced growth performance in fish fed *Aloe vera*-supplemented diets (Khan *et al.*, 2018). Similarly, Gabriel *et al.* (2021) and Roy *et al.* (2021) reported that moderate inclusion levels of *Aloe vera* resulted in improved growth responses in African catfish and Nile tilapia, suggesting that optimal dosage is critical for achieving maximum benefits.

The improved growth performance observed in T1 is further supported by feed utilization efficiency. Lower feed conversion ratio (FCR) and higher specific growth rate (SGR) in T1 indicate more efficient nutrient utilization compared to T0 and T2. Comparable findings have been reported in tilapia and other aquaculture species, where *Aloe vera* supplementation enhanced feed efficiency and growth performance (Gabriel *et al.*, 2015). In addition, herbal feed additives, including *Aloe vera*-based mixtures, have been shown to promote growth and physiological performance in fish (Manaf *et al.*, 2016). Similar growth-promoting effects have also been observed with other phytogenic additives such as garlic (*Allium sativum*), which improves digestion and metabolic efficiency in aquatic species (Hassaan and Soltan, 2016; Büyükdeveci *et al.*, 2018; Labrador *et al.*, 2016). These findings collectively suggest that phytogenic additives can serve as effective alternatives to synthetic growth promoters.

Hematological analysis further revealed that *Aloe vera* supplementation significantly influenced several blood parameters. The increase in white blood cell (WBC) count in T1 and T2 indicates enhanced immune responsiveness in treated groups compared to the control. Elevated hemoglobin (Hb) and hematocrit (Hct) values in T1 suggest improved oxygen transport capacity and overall physiological status. Similarly, the increase in mean corpuscular volume (MCV) in T1 indicates changes in erythrocyte characteristics associated with improved hematological function. In contrast, red blood cell (RBC) count, mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) did not differ significantly among treatments, suggesting that *Aloe vera* supplementation selectively influences specific hematological indices rather than uniformly affecting all parameters.

These findings are consistent with previous studies reporting that *Aloe vera* enhances hematological and immune parameters in fish. Yousaf *et al.* (2025) and Gabriel *et al.* (2021) demonstrated that *Aloe vera* supplementation promotes erythropoiesis and improves immune-related markers. The observed increase in hemoglobin and hematocrit levels may be associated with improved physiological condition and metabolic activity, although the exact mechanisms remain unclear. Kumar *et al.* (2018) suggested that bioactive compounds in *Aloe vera* may stimulate hemoglobin synthesis and red blood cell production, contributing to improved hematological profiles.

The present findings also align with studies showing that moderate levels of *Aloe vera* supplementation yield optimal physiological responses. Syed *et al.* (2022) reported improved hematological parameters in Nile tilapia at inclusion levels ranging from 0.5% to 3%. However, higher concentrations may not necessarily produce proportional benefits, as observed in the current study where T2 (1.5%) did not outperform T1. Similar variability has been reported in other studies, where excessive supplementation resulted in reduced or inconsistent hematological responses (Gabriel *et al.*, 2021). In contrast, Alishahi *et al.* (2017) reported significant increases in RBC and hematocrit values in common carp at lower inclusion levels, indicating species-specific and dose-dependent effects.

Overall, the findings of this study demonstrate that *Aloe vera* supplementation, particularly at 1% inclusion, enhances growth performance and selected hematological parameters in Nile tilapia. The results support the potential of *Aloe vera* as a natural feed additive in aquaculture. However, the underlying physiological and biochemical mechanisms remain to be fully elucidated, and further research is required to determine optimal dosage levels and long-term effects under commercial production conditions.

CONCLUSION

The present study demonstrates that dietary supplementation of 1% *Aloe vera* (T1) significantly improves growth performance and selected hematological parameters in Nile tilapia (*Oreochromis niloticus*). Fish in the T1 group exhibited the highest final body weight, improved feed conversion efficiency, and enhanced specific growth rate compared to the control and higher supplementation level. No significant differences were observed in length among treatments, indicating that the effects of *Aloe vera* were more pronounced in weight-related growth parameters.

Hematological analysis revealed significant increases in white blood cell count (WBC), hemoglobin (Hb), hematocrit (Hct), and mean corpuscular volume (MCV) in the T1 group, while red blood cell count (RBC), mean corpuscular hemoglobin (MCH), and mean corpuscular hemoglobin concentration (MCHC) remained unaffected. These findings suggest that *Aloe vera* supplementation exerts selective effects on hematological indices associated with immune response and physiological status.

Water quality parameters remained within optimal ranges throughout the experimental period, indicating that the observed effects were attributable to dietary treatments rather than environmental variation.

Overall, the results indicate that 1% *Aloe vera* supplementation can be considered a promising natural feed additive to enhance growth performance and physiological health in Nile tilapia. However, the underlying biological mechanisms were not investigated in this study. Further research is required to evaluate long-term effects, optimize dosage levels, and validate the applicability of *Aloe vera* under commercial aquaculture conditions.

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