

KARAKTERISASI FITOKIMIA DAN EVALUASI NUTRISI *Ficus septica* UNTUK PENGEMBANGAN PAKAN AKUATIK PADA IKAN HERBIVORA

PHYTOCHEMICAL CHARACTERIZATION AND NUTRITIONAL EVALUATION OF *Ficus septica* FOR AQUAFEED DEVELOPMENT IN HERBIVOROUS FISH

R Adharyan Islamy^{1,*}, Najwa Lutfi Setyowati¹, Syakir Ni'matullah¹

¹ Aquaculture (Kedri City Campus), Department of Fisheries and Marine Resources Management, Faculty of Fisheries and Marine Science, Brawijaya University, Jl. Pringgodani, Mrican, District. Mojoroto, Kediri City, East Java 64111, Indonesia

*email penulis korespondensi: r.adharyan@ub.ac.id

Abstrak

Keberlanjutan akuakultur sangat bergantung pada pengembangan alternatif pakan yang bernutrisi seimbang dan hemat biaya yang mengurangi ketergantungan pada sumber protein konvensional. *F. septica*, tanaman obat yang kaya akan senyawa bioaktif, menawarkan potensi sebagai bahan pakan fungsional baru untuk ikan herbivora. Penelitian ini bertujuan untuk mengkarakterisasi profil fitokimia dan mengevaluasi komposisi nutrisi proksimat daun *F. septica*, dan untuk menilai efeknya pada kinerja pertumbuhan dan pemanfaatan pakan pada ikan herbivora. Skrining fitokimia mengungkapkan kadar fenolik yang tinggi ($33,5 \pm 2,1$ mg/g), flavonoid ($28,7 \pm 1,3$ mg/g), dan tanin ($16,1 \pm 0,9$ mg/g), yang menunjukkan potensi antioksidan dan antimikroba yang kuat. Analisis proksimat menunjukkan bahwa daun *F. septica* mengandung 18,7% protein kasar, 40,7% ekstrak bebas nitrogen, dan 16,3% serat kasar (berat kering). Uji coba pemberian pakan dengan diet yang mengandung 0% (kontrol), 5%, dan 10% bubuk daun *F. septica* menunjukkan peningkatan signifikan dalam penambahan berat badan, laju pertumbuhan spesifik, dan rasio konversi pakan pada kelompok 5% dan 10% ($p < 0,05$), tanpa mengorbankan kelangsungan hidup. Hasil ini menunjukkan bahwa *F. septica* dapat digunakan secara efektif sebagai aditif pakan akuatik yang kaya nutrisi dan bioaktif untuk ikan herbivora. Penelitian lebih lanjut tentang daya cerna, efek jangka panjang, dan keamanan residu direkomendasikan untuk memvalidasi aplikasi praktisnya dalam akuakultur komersial.

Kata Kunci: akuakultur berkelanjutan, aquafeed, *Ficus septica*, feed fungsional, ikan herbivora, kinerja pertumbuhan, fitokimia.

Abstract

The sustainability of aquaculture relies heavily on the development of nutritionally balanced and cost-effective feed alternatives that reduce dependency on conventional protein sources. *F. septica*, a medicinal plant rich in bioactive compounds, offers potential as a novel functional feed ingredient for herbivorous fish. This study aimed to characterize the phytochemical profile and evaluate the proximate nutritional composition of *F. septica* leaves, and to assess their effects on growth performance and feed utilization in herbivorous fish. Phytochemical screening revealed high levels of phenolics (33.5 ± 2.1 mg/g), flavonoids (28.7 ± 1.3 mg/g), and tannins (16.1 ± 0.9 mg/g), indicating strong antioxidant and antimicrobial potential. Proximate analysis showed that *F. septica* leaves contained 18.7% crude protein (dry weight basis), 40.7% nitrogen-free extract (dry weight basis), and 16.3% crude fiber. Feeding trials with diets containing 0% (control), 5%, and 10% *F. septica* leaf powder demonstrated significant improvements in weight gain, specific growth rate, and feed conversion ratio in the 5% and 10% groups ($p < 0.05$), without compromising survival. These results suggest that *F. septica* can be effectively utilized as a nutrient-rich, bioactive aquafeed additive for herbivorous fish. Further research on digestibility, long-term effects, and residue safety is recommended to validate its practical applications in commercial aquaculture.

Keywords: *Ficus septica*, aquafeed, herbivorous fish, phytochemicals, growth performance, functional feed, sustainable aquaculture

INTRODUCTION

The growing global demand for sustainable aquaculture practices drives the exploration of alternative feed ingredients that are cost-effective and environmentally friendly, particularly for herbivorous and omnivorous fish species. Traditional feed ingredients such as fishmeal and soybean meal, although rich in nutrients, have become increasingly expensive and create significant environmental challenges due to overfishing and land-use changes (Ayyat et al., 2021; Shi et al., 2016). This situation highlights the necessity to investigate novel plant-based feed resources that are locally available and nutritionally adequate. Plant materials could potentially enhance fish health and performance, reducing reliance on conventional feeds that contribute to sustainability issues (Wang et al., 2013).

In this context, *F. septica*, a widely distributed tropical medicinal plant in Southeast Asia and the Pacific, has long been recognized in folk medicine for its antimicrobial, anti-inflammatory, and antioxidant properties (Sarker et al., 2018). Research indicates that *F. septica* leaves are abundant in bioactive compounds such as flavonoids, phenolics, saponins, alkaloids, and tannins, which may offer functional benefits when added to aquafeeds (Lozano-Muñoz et al., 2021). However, the exploration of *F. septica* as a feed ingredient in aquaculture remains largely uncharted territory, suggesting that more research is needed to fulfill its potential in sustainable aquaculture practices (Campbell et al., 2017).

The nutritional composition of plant-based feed ingredients is paramount in determining their suitability for aquacultural diets. Herbivorous fish species, including *Oreochromis niloticus* (Nile tilapia) and *Ctenopharyngodon idella* (grass carp), exhibit adaptations that favor diets rich in carbohydrates and fiber, making them ideal candidates for incorporating high-fiber leafy ingredients like *F. septica* (Xu et al., 2016). Evaluating the phytochemical and proximate nutritional profile of *F. septica* is essential for substantiating its incorporation into formulated feeds. Previous studies highlight the value of alternative plant ingredients in enhancing the performance of herbivorous fish (Liu et al., 2024).

Previous studies on plant-based feed additives, such as *Moringa oleifera*, *Azadirachta indica*, and *Carica papaya* leaves, have demonstrated improvements in fish growth, feed utilization, and immunity, although their effectiveness is sometimes limited by high fiber content or antinutritional factors (Webb et al., 2022; Somdare et al., 2023). Compared with these commonly studied plants, *F. septica* remains

largely unexplored despite its rich content of phenolics, flavonoids, and other bioactive compounds that may offer both nutritional and functional benefits. This highlights the novelty of the present study in evaluating *F. septica* as a potential functional feed ingredient for herbivorous fish.

The objective of the proposed study is centered on characterizing the phytochemical composition and analyzing the proximate nutritional content of *F. septica* leaves, as well as assessing their impact on growth performance and feed utilization efficiency in herbivorous fish species. Such research aims to contribute to the expanding body of knowledge that supports the development of sustainable, functional aquafeeds leveraging the natural bioactivity and nutritional properties of underutilized plant species.

MATERIAL AND METHODS

Collection and Preparation of *F. septica* Leaves

Fresh leaves of *F. septica* (Figure 1) were collected from wild plants in the Kediri region, East Java, Indonesia. Leaves were washed thoroughly with distilled water to remove dirt and contaminants, then air-dried at room temperature ($28 \pm 2^\circ\text{C}$) for 5–7 days until constant weight was achieved. The dried leaves were ground into fine powder using a laboratory blender, sieved through a 60-mesh screen, and stored in airtight containers at room temperature for further analysis.

The botanical identification of *F. septica* was confirmed based on its distinctive morphological and physiological characteristics. The plant is a shrub to small tree (2–6 m in height) with milky white latex exuding from stems and leaves when injured. Leaves are simple, opposite, elliptic to oblong, measuring 8–20 cm long and 4–10 cm wide, with entire margins, acute to acuminate tips, and a glossy green surface. The venation is pinnate with 6–10 pairs of lateral veins. The syconia are globose to pear-shaped, 1–2 cm in diameter, green when young and turning yellowish upon ripening, usually borne in pairs in the leaf axils. Physiologically, *F. septica* produces abundant latex rich in alkaloids and phenolic compounds, which has long been recognized in traditional medicine for antimicrobial and anti-inflammatory activity.

Proximate Analysis

The proximate composition of *F. septica* leaf powder was determined following AOAC (2005) procedures, including moisture content (oven-drying method at 105°C), crude protein (Kjeldahl method, $N \times 6.25$), crude lipid (Soxhlet extraction with petroleum ether), ash content (muffle furnace at 550°C for 6 h), crude fiber, and nitrogen-free extract (by difference). All analyses

were performed in triplicate ($n = 3$) to ensure reliability of the data.

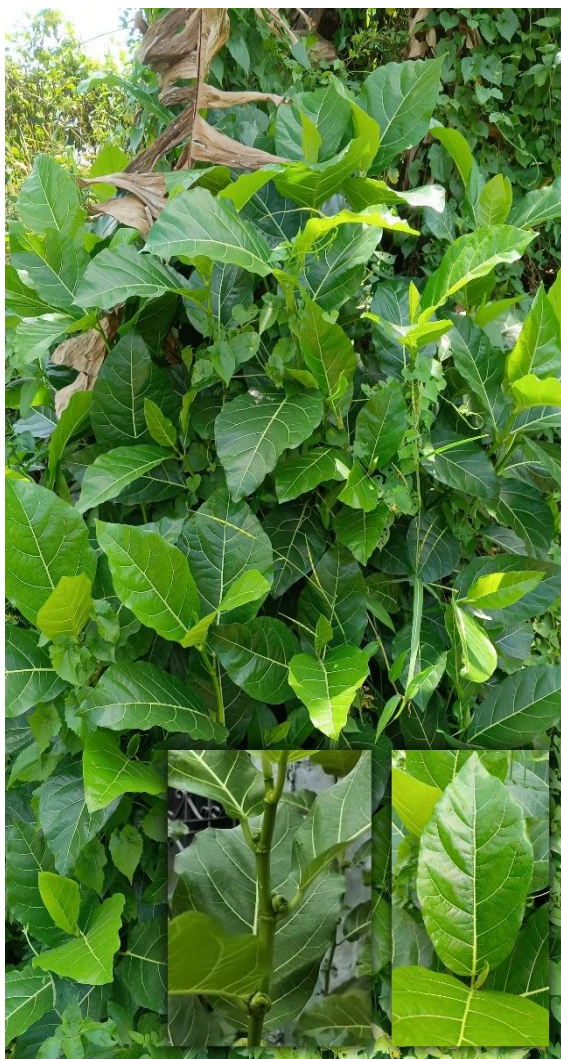


Figure 1. *Ficus septica* wild plant collected from Kediri, East Java, Indonesia, showing its characteristic opposite, elliptic-oblong leaves with entire margins and milky latex exudation. A voucher specimen (Code: FS-UBK25) was deposited in the Herbarium of Universitas Brawijaya (Kediri City Campus) for reference.

Phytochemical Screening

Qualitative phytochemical screening of *F. septica* leaf powder was conducted to detect the presence of major secondary metabolites including alkaloids, flavonoids, tannins, saponins, terpenoids, and phenolic compounds using standard protocols (Idris et al., 2023). Total phenolic content (TPC) was quantified using the Folin-Ciocalteu method and expressed as mg gallic acid equivalents (GAE) per gram of dry sample. Total flavonoid content (TFC) was measured using the aluminum chloride colorimetric assay and expressed as mg quercetin equivalents (QE) per gram. All analyses were performed in triplicate ($n = 3$) to ensure reliability of the data.

Experimental Diet Formulation

Three isonitrogenous and isolipidic diets were formulated: a control diet without *F. septica* (D0), and two experimental diets containing 5% (D1) and 10% (D2) of *F. septica* leaf powder, respectively. All diets were formulated to meet the nutritional requirements of herbivorous fish, using a combination of fish meal, soybean meal, rice bran, cornmeal, and vitamin-mineral premix. The ingredient composition of the diets was as follows (% dry weight basis): fish meal (15%), soybean meal (25%), rice bran (20%), cornmeal (20%), *F. septica* leaf powder (0%, 5%, or 10% depending on treatment), vitamin-mineral premix (2%), fish oil (3%), and wheat flour as binder (15%). The ingredients were mixed, pelleted (2 mm diameter), dried at 45 °C, and stored at 4 °C until use.

Fish and Rearing Conditions

Juvenile herbivorous fish (e.g., *Oreochromis niloticus*), with an average initial weight of 4.5 ± 0.2 g, were obtained from a local hatchery and acclimated in fiberglass tanks (200 L) for 7 days. Fish were randomly distributed into nine tanks (three treatments $\times$ three replicates), with 15 fish per tank. The feeding trial lasted 42 days. Fish were fed ad libitum three times daily (08:00, 12:00, 17:00). Water quality parameters were maintained within optimal ranges: temperature (26–28 °C), pH (6.8–7.5), and dissolved oxygen (>5 mg/L).

Growth Performance and Feed Utilization

Growth performance was assessed at the end of the trial. Parameters calculated included:

- Weight Gain (WG) = Final weight – Initial weight
- Specific Growth Rate (SGR, %/day) = $[(\ln \text{Final weight} - \ln \text{Initial weight}) / \text{days}] \times 100$
- Feed Conversion Ratio (FCR) = Feed intake (g) / Weight gain (g)
- Survival Rate (%) = (Final number of fish / Initial number of fish) $\times 100$

Statistical Analysis

All data were analyzed using one-way analysis of variance (ANOVA) followed by Duncan's multiple range test to determine significant differences among treatments ($p < 0.05$). Statistical analyses were performed using SPSS version 25.0.

RESULT

The qualitative and quantitative phytochemical screening of *F. septica* leaf extract revealed the presence of several bioactive compounds, including alkaloids, flavonoids, tannins, saponins, terpenoids, and phenolic compounds (Table 1). Among these, phenolic compounds (33.5 ± 2.1 mg/g) and flavonoids (28.7 ± 1.3 mg/g) were the most abundant, followed by tannins (16.1 ± 0.9 mg/g) and

alkaloids (12.4 ± 0.6 mg/g). The presence of these compounds suggests that *F. septica* has significant potential as a natural functional ingredient in aquafeed formulations.

The proximate analysis of *F. septica* leaves indicated that this plant contains a balanced profile of macronutrients, making it a promising candidate for use in herbivorous fish diets (Table 2). The crude protein content was $18.7 \pm 0.6\%$ (dry weight basis), which falls within the recommended dietary protein range for many herbivorous freshwater fish such as tilapia

(*Oreochromis niloticus*) and carp (*Cyprinus carpio*) in the grow-out phase (Hassaan et al., 2019).

Growth Performance and Feed Utilization

The feeding trial demonstrated that inclusion of *F. septica* leaf powder in herbivorous fish diets had a positive impact on growth performance and feed efficiency (Table 3). Fish fed diets containing 5% (FS-5%) and 10% (FS-10%) *F. septica* showed significantly higher final weight, weight gain, and specific growth rate (SGR) compared to the control group ($p < 0.05$).

Table 1. Phytochemical Composition of *F. septica* Leaf Extract

Phytochemical Compound	Qualitative Presence (+/-)	Quantitative Content (mg/g extract)
Alkaloids	+	12.4 ± 0.6
Flavonoids	+	28.7 ± 1.3
Tannins	+	16.1 ± 0.9
Saponins	+	9.8 ± 0.4
Terpenoids	+	6.3 ± 0.3
Phenolic compounds	+	33.5 ± 2.1

Note: "+" indicates presence; values expressed as mean $\pm$ SD ($n = 3$).

Table 2. Proximate Composition of *F. septica* Leaves (Dry Weight Basis)

Nutrient Parameter	Content (% dry weight)
Moisture	9.6 ± 0.3
Crude Protein	18.7 ± 0.6
Crude Lipid	4.5 ± 0.2
Crude Fiber	16.3 ± 0.7
Ash Content	10.2 ± 0.4
Nitrogen-Free Extract (NFE)*	40.7 ± 1.1
Energy (kcal/100g)**	312.8 ± 5.4

* NFE = $100 - (\text{Protein} + \text{Lipid} + \text{Fiber} + \text{Ash})$

** Calculated using: 4 kcal/g for protein and carbohydrate, 9 kcal/g for lipid

Table 3. Growth Performance and Feed Utilization of Herbivorous Fish Fed Diets Containing *F. septica*

Parameter	Control (0%)	FS-5% Inclusion	FS-10% Inclusion
Initial Weight (g)	5.02 ± 0.14	5.03 ± 0.12	5.01 ± 0.15
Final Weight (g)	12.31 ± 0.45^a	14.02 ± 0.50^b	14.65 ± 0.39^b
Weight Gain (g)	7.29 ± 0.34^a	8.99 ± 0.42^b	9.64 ± 0.40^b
Specific Growth Rate (SGR, %/day)	2.41 ± 0.08^a	2.74 ± 0.09^b	2.88 ± 0.07^b
Feed Conversion Ratio (FCR)	1.87 ± 0.11^a	1.63 ± 0.07^b	1.59 ± 0.06^b
Survival Rate (%)	92.5 ± 2.9	95.0 ± 2.5	96.7 ± 1.8

Values are mean $\pm$ SD ($n = 3$); different superscripts (^a, ^b) in the same row indicate significant differences ($p < 0.05$).

DISCUSSION

Phytochemical Composition of *F. septica* Leaf Extract

Phenolic and flavonoid compounds are well-documented for their antioxidant properties, which are beneficial in protecting fish against oxidative stress and enhancing immunity. Such antioxidants can mitigate the damaging effects of free radicals, thereby improving overall health and resilience in aquaculture settings (S. Chakraborty et al., 2014; Islamy et al., 2024; Kilawati & Islamy, 2019; Reverter et al., 2017). Additionally, tannins and saponins, while sometimes regarded as antinutritional factors at

high levels, can exhibit favorable antimicrobial and immunomodulatory effects when used judiciously in aquafeeds (S. B. Chakraborty & Hancz, 2011; Shojaemehr et al., 2020). This nuanced understanding emphasizes that the effects of these compounds are concentration-dependent and their application must be strategically managed to optimize benefits in fish health.

Furthermore, terpenoids present in *F. septica*, quantified at 6.3 ± 0.3 mg/g, have been linked to anti-inflammatory and hepatoprotective activities, which are essential for maintaining gut health and promoting

nutrient metabolism in herbivorous fish (Lee et al., 2020; Muffler et al., 2011). The varied bioactive profile of *F. septica* suggests its potential utility in aquafeeds not just as a nutritional supplement, but also as a functional additive that can enhance fish health, improve immune responses, and increase feed efficiency (Reverter et al., 2017). Previous studies have corroborated the medicinal benefits of various *Ficus* species in aquaculture, showcasing their phytochemical constituents that act as immunostimulants and possess antimicrobial properties (Ahmad et al., 2024; Liu et al., 2024).

When compared with other species in the same genus, the phenolic and flavonoid contents of *F. septica* (33.5 and 28.7 mg/g, respectively) are higher than those reported in *F. carica* leaves (phenolics 21–25 mg/g; flavonoids 15–20 mg/g) and *F. hispida* (phenolics 18–22 mg/g) (Osowe et al., 2021). These values also exceed the levels typically found in commonly used feed additives such as *Moringa oleifera* leaves (phenolics 12–18 mg/g; flavonoids 10–15 mg/g). This indicates that *F. septica* has a richer profile of bioactive compounds that may contribute to stronger antioxidant and immunomodulatory effects in aquafeeds.

It should be noted that *F. septica* also contains tannins (16.1 mg/g) and saponins (9.8 mg/g), which are considered antinutritional factors when present at high concentrations. Tannins can reduce protein digestibility, and saponins may cause hemolytic effects in excessive doses. However, at the inclusion levels tested in this study, these compounds likely acted within beneficial ranges, contributing antimicrobial and immunostimulatory properties rather than impairing growth. The balanced presence of these metabolites suggests that controlled use of *F. septica* can maximize health-promoting benefits while minimizing antinutritional risks.

However, it is crucial to consider that the efficacy and safety of these bioactive compounds hinge on their concentration and bioavailability within formulated feeds. Ongoing research is warranted to investigate the stability of these compounds during feed processing, as well as their in vivo effects on fish physiology, to fully harness the benefits of *F. septica* in aquafeed applications.

Proximate and Nutritional Composition of *F. septica*

The proximate analysis of *F. septica* leaves indicates a balanced profile of macronutrients, making it a promising candidate for inclusion in the diets of herbivorous fish. Specifically, the crude protein content was measured at $18.7 \pm 0.6\%$ (dry weight basis), which aligns with the dietary protein requirements for various

herbivorous freshwater fish species, including tilapia (*Oreochromis niloticus*) and carp (*Cyprinus carpio*), particularly during their grow-out phase (Nabit et al., 2001). The nitrogen-free extract (NFE), representing carbohydrates, was determined to be $40.7 \pm 1.1\%$ (dry weight basis), supporting its role as a significant energy source conducive to the natural diets of herbivorous species that thrive on carbohydrate-rich plant materials (Nguyen et al., 2009).

Moreover, the crude fiber content was found to be 16.3% crude fiber, a typical range for leafy plants. While excessive fiber can impede digestibility, moderate levels are beneficial for enhancing gut motility and promoting microbial fermentation in herbivorous fish (Abdelhamid & Soliman, 2012). The lipid content was relatively low at $4.5 \pm 0.2\%$, which is appropriate for herbivorous fish that generally require lower dietary lipid levels compared to carnivorous fish (Goda et al., 2007). An ash content of $10.2 \pm 0.4\%$ indicates the presence of essential minerals important for processes such as bone development and osmoregulation in fish (Wang et al., 2013).

The calculated energy content of the leaves was approximately 312.8 ± 5.4 kcal/100g, which could significantly contribute to the overall dietary energy when *F. septica* is included partially in feeds (Xiao et al., 2017). These nutritional attributes suggest that *F. septica* can function both as a nutrient source and as a potential functional additive, making it suitable for sustainable aquafeed development targeted at herbivorous fish (Goda et al., 2007). Nevertheless, further studies to assess digestibility, palatability, and any anti-nutritional factors in practical feed trials will be essential to maximize its utilization in fish diets (El-Saidy & Gaber, 2005; Yang et al., 2021).

Growth Performance and Feed Utilization

The inclusion of *F. septica* leaf powder in herbivorous fish diets has demonstrated noteworthy positive impacts on growth performance and feed efficiency during feeding trials. Fish that were fed diets incorporating 5% (FS-5%) and 10% (FS-10%) of *F. septica* exhibited significantly enhanced final weight, weight gain, and specific growth rate (SGR) when compared to control groups ($p < 0.05$). Notably, the FS-10% group achieved the highest weight gain and SGR, implying that *F. septica* may bolster nutrient utilization and growth when included at appropriate levels (Osowe et al., 2021).

The bioactive constituents of *F. septica*, particularly flavonoids and phenolics, are key players in promoting digestive enzyme activity and modulating gut microbiota. (Gewaily et al., 2021) highlight the widespread benefits of these compounds, which enhance metabolic efficiency

and overall health in fish species (Somdare et al., 2023). The improvement in growth performance can thus be attributed to these bioactive compounds acting as immunostimulants and facilitating better nutrient absorption and digestion.

Moreover, the analysis of the Feed Conversion Ratio (FCR) revealed significant improvements in both FS-5% and FS-10% groups. A lower FCR signifies more efficient feed utilization, an important factor in aquaculture for mitigating production costs and reducing environmental waste. This aspect is crucial for sustainable aquaculture practices, as reduced feed waste not only enhances fish growth but also abates the ecological footprint associated with fish farming (Zare et al., 2021).

High survival rates across all treatment groups further affirm that *F. septica* does not detrimentally affect fish health, with the FS-10% group reporting the highest survival rate. This indicates that incorporating such plant-based feed ingredients may not only support growth and efficiency but may also enhance immune resilience, a critical factor in aquaculture where health management is essential (Webb et al., 2022).

These findings collectively support the notion that *F. septica* can serve as a functional feed ingredient that provides both nutritional and health benefits to herbivorous fish. However, it is essential to conduct long-term trials and dose optimizations, alongside mechanistic studies, to elucidate the exact pathways through which *F. septica* enhances growth and to evaluate any possible chronic effects or bioactive residues in fish tissue (Jung, 2014). This rigor will inform best practices for its application in aquafeed formulations, ensuring that both efficacy and safety are accounted for in aquaculture nutrition.

The inclusion levels of 5% and 10% were selected based on preliminary feeding trials and literature reports on other plant-based additives in herbivorous fish diets, which often show optimal performance improvements within this range (El-Saidy & Gaber, 2005; Zare et al., 2021). Lower levels (<5%) may provide insufficient bioactive compounds, whereas higher levels (>10%) increase the risk of excessive fiber and antinutrients that could compromise digestibility. The improved growth and feed utilization observed at both 5% and 10% in the present study confirm that these levels are effective and safe for practical application.

CONCLUSION

This study demonstrated that *F. septica* leaves contain a diverse array of bioactive phytochemicals—most notably phenolics, flavonoids, tannins, and saponins—which are

known for their antioxidant, antimicrobial, and immunomodulatory properties. The proximate composition of the leaves also showed promising nutritional value, with adequate levels of protein, carbohydrates, and fiber that meet the dietary needs of herbivorous fish.

Incorporating *F. septica* leaf powder into fish diets at 5% and 10% inclusion levels significantly improved growth performance, feed conversion efficiency, and survival rates compared to the control group. These results suggest that *F. septica* can serve not only as a source of nutrients but also as a functional feed additive for sustainable aquafeed development.

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