Asian Journal of
Biological Sciences

AJBS

OPEN ACCESS

Efficacy of *Vernonia amygdalina* on Columnaris Disease Management and Growth Enhancement in Nile Tilapia

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ABSTRACT

Background and Objective: Nile tilapia is vital for aquaculture but faces growth issues due to pollutants. Traditional antibiotic growth promoters pose risks like antibiotic resistance. This study evaluates the efficacy of *Vernonia amygdalina* leaf against columnaris disease and its impact on the growth of *Oreochromis niloticus* over eight weeks in Ghana, assessing its potential as a natural alternative. **Materials and Methods:** Different levels of powdered *V. amygdalina* leaf (300, 400 and 50 g) were added to 13.89 m^3 of water across four treatments (two replicates each). Treatments included leaf additions and a control (no additives). The study lasted for eight weeks, with samples taken to assess weight gain, feed conversion ratio, specific growth rate and condition factor. Microsoft Excel and Graph Pad Prism version 5.03 was used to analyze the data and one-way ANOVA was used to detect significant differences among treatments. **Results:** Groups with 300 and 400 g of *V. amygdalina* showed significant improvements in weight gain, feed conversion ratio, specific growth rate and condition factor without compromising water quality. The intestinal tract histomorphology exhibited larger and thicker surfaces. Mortality rates were low and survival rates were high (90.92±0.25 and 86.42±1.43 g). **Conclusion:** *Vernonia amygdalina* at 300 and 400 g in 13.89 m^3 of water effectively cures columnaris in *O. niloticus*, improves growth and enhances digestive tract morphology without harming the environment. As such, *V. amygdalina* can be a cost-effective, natural remedy that can benefit aquaculture by promoting healthier and more robust fish production.

KEYWORDS

Vernonia amygdalina, diseases, histomorphology, aquaculture sustainability, phyto-biotics

INTRODUCTION

Intensive fish production creates a stressful environment, which leads to immune system suppression and increased vulnerability to infectious disease in fish[1]. Diseases claim the lives of about a third to half of farm food fish before they reach marketable size. As such, diseases can also result in severe economic loss due to low growth rates, poor flesh quality or restricted commerce, resulting in lower profit margins[2]. Various parasites, fungi, viruses and bacteria are examples of infectious agents that can cause disease. In recent years, infections and other diseases caused by these agents and other unidentified and emerging pathogens have impacted farming areas in Asia and South America[3].

ISSN: xxxx-xxxx (Online)
ISSN: 1996-3351 (Print)
https://doi.org/10.3923/ajbs.2025.136.145

Received: 28 Oct. 2024
Accepted: 28 Nov. 2024
Published: 31 Mar. 2025

Freshwater fish are susceptible to bacterial infection called columnaris, which results in significant yearly material and financial losses at fish farms all over the world[4].

Columnaris disease is typically more severe and more frequent in waters that are warmer (>20°C), however, it can also affect salmonids raised in 12-15°C environments[5].

Although there are no known predisposing factors, stressful rearing conditions like low dissolved oxygen, high ammonia and nitrite concentrations, handling injuries and overcrowding are frequently linked to outbreaks of the columnaris disease in fish[5]. Depending on the specifics of the epizootic and the fish species involved, mortality patterns might be acute, subacute or chronic. Mortality rates are often very high, ranging from 60-90%[6]. Fish of all ages can contract columnaris disease, but young fish are more susceptible to it because their organs are not fully developed and as such not producing enough cells to resist such attack[4].

Tilapia is also harmed by biological and chemical pollutants that pollute the water and interfere with its growth during production[7]. This pollution and interference from the environment frequently lead to the usage of various antibiotic growth promoters, which have long-term impacts on fish consumers. It contributes to the development of antibiotic-resistant pathogens due to their continued or sub-optimal use[7]. For instance, according to Bekele *et al*.[8] the prevalence of bacterial infection in Nile tilapia was 75% within 14-17.9 cm length group, 52% within 18-21.9 cm length group and 33% within 22-26.9 cm length group.

As a result, scientists have stepped up their efforts to use natural goods like herbs to develop alternative dietary supplements that improve the growth, health and immune system of cultured fish. These products are inexpensive, safe, effective and can be easily prepared and are biodegradable with low toxicity[7,9,10]. Because of the existence of numerous active substances such as alkaloids, flavonoids, pigments, phenolics, terpenoids, steroids and essential oils, plants have a wide range of functions[11].

Bitter leaf (*V. amygdalina*) according to Gbolade[12] possess many qualities and as such, it is worth studying to enable the diverse harnessing of this product for efficient usage.

In the tropical regions of the world, *V. amygdalina* is cheap and widely available in many home gardens. In the humid climate, the plant grows abundantly in farmlands, bush fallows, forests and private dwellings. Bitter leaf (*V. amygdalina*) has been used in humans and animals as an antioxidant and growth booster. For example, including 7.2% of *V. amygdalina* in the diet of broodfish improves growth, survival and hematological parameters[13]. *Vernonia amygdalina* has anti-coccidiosis, anti-bacterial and anti-parasitic properties[12]. It also has as an anti-oxidant[14] and growth promoter which by improves the gastrointestinal enzymes and so increases feed conversion efficiency[15].

This study investigated the impact of the antibacterial properties of bitter leaf (*Vernonia amygdalina*) on diseased (columnaris) *O. niloticus* and its associated growth performance.

MATERIALS AND METHODS

Experimental site: The experiment was carried out from July to August, 2023 at the Pilot Aquaculture Center (P.A.C.) located 30 km from Kumasi along the Mampong/Agona road. The P.A.C. is a center under the Ministry of Fisheries and Aquaculture Development (MoFAD), Ghana, which was purposely established in the year 2005 to produce tilapia and catfish fingerlings for farmers in the Ashanti and its neighboring regions.

Experimental design: The entire experiment was run with four treatments in two replicates with Bitter leaf (*V. amygdalina*) [(BL300)-300 g, (BL400)-400 g, (BL50)-50 g of bitter leaf powder and control (CT) with no supplements][7,16].

A total of 240 diseased (columnaris) *O. niloticus* (18.34±0.25 g) were obtained from the Pilot Aquaculture Center (P.A.C.) along the Mampong/Agona road and conditioned for one-week prior to the experiment. Fish were randomly distributed at 30 fish/tank in 8 concrete tanks (4.5×2.2×1.4 m) and fed for eight weeks on satiation, thrice daily with commercial diets, 40% crude protein.

Treatment preparation: Fresh *V. amygdalina* leaves were plugged from Odumase, a community in the Ashanti Region. It was then dried under room temperature for several days while retaining the greenish coloration and milled into fine powder to form treatments BL300, BL400 and BL50. The CT was used as the control with no additives. Leaf powder were introduced to the water which is similar to Okukpe *et al.*[7] thus, different amount of bitter leaf powder was dissolved in 150 mL of water. Equal measures of the bitter leaf (powder/shred) were weighed with an electronic balance (Shimadzu AUW-120D-Shimadzu Corporation, Kyoto, Japan) and administered at the same time a week before the introduction of fishes.

An equal volume of water (13.89 m^3) in concrete tanks was ensured and the powdered leaves additions were mixed to obtain a uniform distribution and mixture.

Sample collection and growth parameters: Every 7 days, 15 fishes were randomly selected from each replicate by seining (passing through) and their body weight and length measured. This number was selected in other to prevent extra stress on all fish at a go and also to have a balance measure at a particular time interval across all treatments. The AP-2000-Aqua-probe (Aquaread Company, Broadstairs-Kent, United Kingdom) was used to measure physico-chemical parameters of the water (temperature, dissolved oxygen, pH, conductivity and transparency, ammonia and nitrate) once every week and the effluent water in tanks were discarded every two weeks and new bitter leaf solution per treatments was prepared.

Intestine samples from three fish per replicate was collected from each experimental group to show and explain the pattern of longitudinal sections of the epithelium of the intestinal sections. Intestine samples were fixed in 10% buffered formalin, dehydrated in graded ethanol series and embedded in paraffin. Approximately 5 μm thick sections of the intestinal sections (anterior, middle and posterior) were stained with Hematoxylin and Eosin and the sections were then observed under Olympus BX53 microscope, Olympus Corporation, Tokyo, Japan[1].

The specific growth rate (SGR), feed conversion ratio (FCR), mean weight gain (MWG), condition factor (K) and survival/mortality rate (SR) of sampled fish were calculated for the determination of growth in each treatment[17] over the experimental period as described below.

Specific growth rate (SGR): The SGR for each treatment group of the study was estimated as[17]:

$$SGR = \frac{\text{Final weight (g) - Initial weight(g)}}{\text{Number of experimental days}} \times 100$$

Mean weight gain (MWG): The MWG was estimated as[17]:

$$MWG = \text{Final mean weight (g)-Initial mean weight (g)}$$

where, W is the mean weight (g) of the fish and SL is the mean standard length (cm) of the fish.

Condition factor (K) was computed as[17]:

$$K = \frac{W}{SL3} \times 100$$

Feed conversion ratio (FCR) was estimated as[17]:

$$FCR = \frac{\text{Feed intake, FI(g)}}{\text{Weight gain (g)}}$$

Survival rate was computed as[17]:

$$SR\ (\%) = \text{Total number of fish stocked-Total number of fish at termination} \times 100$$

Statistical analysis: No significant interactions were obtained when multivariate and univariate GLM were employed in the data analysis and so only One-way Analysis of Variance (ANOVA) was used to compare treatments. Tukey's multiple tests was used to compare all pairs of columns where significant. Significant levels were set at 0.05 probability ($p<0.05$). Homogeneity of variance and data normality were tested using Bartlett's test and Shapiro-Wilk normality test, respectively. All analysis was also performed using the GraphPad Prism V.5.03 and results presented as Mean±Standard Error of the Mean (SEM).

RESULTS

Effects of *Vernonia amygdalina* leaf powder/shreds on growth performance of *O. niloticus*: Group BL400 had the highest final weight (73.66±9.93 g), followed by BL300 (62.46±99.10 g) and BL50 (40.32±3.82 g) as indicated in Table 1. A huge significant difference was observed between groups BL400 and BL50 and BL400 and CT. A significant difference ($p<0.05$) was observed in the mean weight gain of fish in the various treatments at the end of the experiment. The BL300 had the highest mean weight gain (2.49±0.51 g) followed by BL400 (2.47±0.47 g) and BL50 (071±0.16 g). However, there was a significant difference ($p<0.05$) between BL300 and BL50, BL300 and CT, BL400 and BL50, BL400 and CT.

There was a significant difference ($p<0.05$) in the amount of feed consumed by fish in all the treatments. The CT group consumed the highest amount of feed (194.35±2.25 g) whereas, BL50 consumed the least feed (143.28±1.41 g). Significant differences between BL300 and BL400, BL300 and BL50, BL300 and CT, BL400 and BL50, BL400 and CT, BL50 and CT were also noted.

Feed conversion ratio was significantly lower in the BL300 group. However, no significant difference ($p<0.05$) was noted between BL300 and BL400 and BL50 and CT.

Group BL400 recorded a higher significant specific growth rate (0.17±0.02 g). However, no significant difference ($p>0.05$) was observed between BL300 and BL400, BL50 and CT. But significant difference ($p<0.05$) was observed between BL300 and BL50, BL300 and CT, BL400 and BL50, BL400 and CT. Showing recovery of fish with the additions of the *V. amygdalina* in Fig. 1a and b significant differences ($p<0.05$) in K-value were obtained among the treatments. The BL400 recorded the highest value (2.24±0.17), while the CT recorded the least value (1.23±0.17).

A significant difference ($p<0.05$) was observed in the survival rate of fish in the treatment groups. The BL300 recorded the highest number of fishes that survived (90.92%), followed by BL300 (86.42%), BL50 (81.34%) and CT had the least survival value (73.33%). Significant differences ($p<0.05$) were obtained between BL300 and BL400, BL300 and BL50, BL300 and CT, BL400 and BL50, BL400 and CT, BL50 and CT.

(a) (b)

Fig. 1(a-b): Graphical representation of diseased fish before experiment and recovered fish after eight weeks trials, (a) Pictorial view of the diseased fish before study and (b) Recovered fish after *V. amygdalina* inputs

Table 1: Effects of *V. amygdalina* on growth performance of *Oreochromis niloticus* for 8 weeks during recovery

Parameter	BL300	BL400	BL50	CT	p-value
Initial weight (g)	18.72±0.75[a]	17.76±0.50[a]	18.34±0.25[a]	17.81±0.00[a]	0.4941
Final weight (g)	62.46±9.10[a]	73.66±9.93[a]	40.33±3.82[a,b]	34.69±3.76[b]	0.0019
Mean weight gain (g)	2.50±0.51[a]	2.47±0.47[a]	0.71±0.16[b]	0.56±0.13[b]	0.0003
Feed intake (g)	171.40±0.63[a]	183.80±1.47[b]	143.30±1.41[c]	194. 40±2.25[d]	0.0001
FCR[a]	0.003±0.001[a]	0.01±0.001[a]	0.013±0.003[b]	0.02±0.003[b]	0.0005
SGR[b] (%)	0.16±0.03[a]	0.17±0.02[a]	0.08±0.01[b]	0.06±0.01[b]	0.0003
SR[c] (%)	90.92±0.25[a]	86.42±1.43[b]	81.35±0.42[c]	73.33±0.0[d]	0.0003
K[d]	2.21±0.06[a]	2.24±0.17[a]	1.60±0.149[b]	1.23±0.178[b]	0.0001

All values are Mean±SEM. Different superscript in each row represent significant differences ($p<0.05$) determined by one-way ANOVA. [a]FCR: Feed conversion ratio, [b]SGR: Specific growth weight, [c]SR: Survival Rate, [d]K: Condition factor, BL300: Bitter leaf group with 300 g, BL400: Bitter leaf group with 400 g, BL50: Bitter leaf group with 50 g and CT: Control group with no additives/supplement

Table 2: Impact of *V. amygdalina* on water quality parameters in *Oreochromis niloticus* environment

Parameters Indices	Temperature (°C)	Dissolved oxygen (mg/L)	pH	Electrical conductivity (S/m)	Turbidity (NTU)	Ammonia (mg/L)	Nitrate (mg/L)
Initial BL300	26.53±0.02	7.58±0.15	6.69±0.07	189.00±0.15	122.50±0.12	0.21±0.00	0.06±0.05
Final BL300	26.04±0.38	7.70±0.24	6.54±0.48	141.00±0.23	90.00±0.20	0.24±0.02	0.09±0.05
Initial BL400	26.68±0.26	8.22±0.36	6.62±0.17	194.00±0.18	125.50±0.21	0.20±0.02	0.08±0.05
Final BL400	25.95±0.40	7.07±0.33	6.39±0.40	150.00±0.31	98.00±0.31	0.26±0.02	0.09±0.05
Initial BL50	26.25±0.24	7.65±0.39	6.53±0.30	207.00±0.20	134.00±0.13	0.18±0.001	0.06±0.02
Final BL50	25.91±0.35	7.70±0.41	6.46±0.38	161.00±0.26	106.00±0.28	0.25±0.06	0.08±0.03
Initial control	26.25±0.33	7.77±0.15	6.54±0.04	207.00±0.11	131.00±0.16	0.22±0.07	0.03±0.01
Final control	26.07±0.37	7.07±0.25	6.40±0.22	168.00±0.21	108.00±0.23	0.22±0.03	0.06±0.07

All values are Mean±SEM. Different superscripts in each row represent significant differences ($p<0.05$) determined by one-way ANOVA and BL: Bitter leaf and groupings

From Table 2, no significant difference ($p>0.05$) was observed in both the initial and final values obtained at the termination of the study among water quality parameters measured. The physico-chemical parameters measured were not significant from each treatment and the ranges following water quality standards were not affected by the supplement. The ranges in parameter were noted as; temperature (25.91±0.35-26.58±0.02°C), dissolved oxygen (7.07±0.33-8.22±0.36 mg/L), pH (6.39±0.40-6.69±0.07), electrical conductivity (141.00±0.23-207.00±0.20 S/m), turbidity (90.00±0.20-134±0.13 NTU), ammonia (0.18±0.001-0.26±0.02 mg/L) and nitrate (0.03±0.01-0.09±0.05 mg/L).

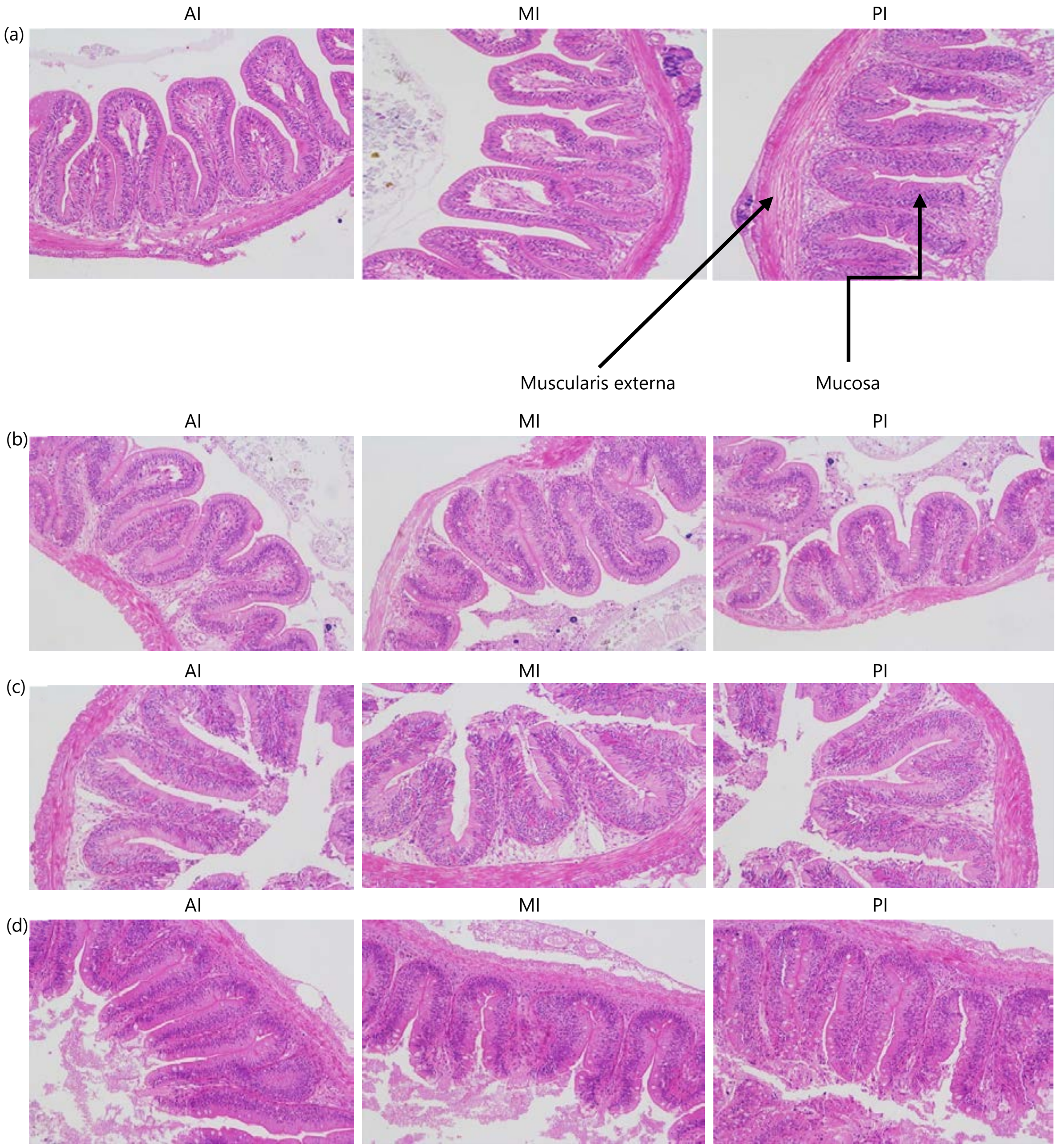

Fig. 2(a-d): Histo-morphological representation of 5 µm thick section of *O. niloticus* intestine in different sections, (a) Morphology of intestine in the control group (CT) from the anterior to the posterior ends, (b) Intestinal morphology of groups in the 300 g bitter leaf treatment (BL300), (c) Fish intestinal tract morphology in treatments of 400 g bitter leaf (BL400) and (d) Morphology of fish intestines in 50 g bitter leaf treatments (BL50)

AI: Anterior intestine, MI: Middle intestine, PI: Posterior intestine, CT: Control group, BL300: 300 g of bitter leaf group, BL400: 400 g of bitter leaf group and BL50: 50 g of bitter leaf group

Histomorphology of intestine of fish in different environments of *V. amygdalina* leaf powder solution: In Fig. 2a-d, the intestinal mucosal folds were observed in Hematoxylin and Eosin-stained sections of the intestinal sections (Anterior Intestine (AI), Middle Intestine (MI) and Posterior Intestine (PI)).

The folds in the control group (A) have similar shapes and arrangements in the AI and MI, whereas the PI was different in shape. However, the muscularis externa in PI was thicker and larger than in AI and MI in control group A. Again, the muscularis externa in PI of the control group (A) can be observed to have similar size and thickness with all the intestines (AI, MI and PI) in BL400 (C) and BL50 (D) groups whereas, the muscularis externa in AI and MI of the A can be observed to have similar size with the MI and PI in BL300 (B) group.

The anterior intestine (AI) in fish of BL300 (B) group can be observed to have a different structure as compared to the middle intestine (MI) and posterior intestine (PI) which have similar structures. However, the muscularis externa of MI and PI of BL300 (B) group can be observed to have relatively the same structure which is observed to be thinner than in AI.

The mucosal folds in the BL400 (C) group can be observed to have similar structure and size in AI and PI. Unlike the structure and size of MI which can be observed to be different and larger than the AI and PI. Again, the arrangements of mucosal folds of all the intestines (AI, MI and PI) in BL400 (C) group can be observed to be loosely arranged as compared to the mucosal folds arranged in the intestines (AI, MI and PI) of the BL50 (D), CT (A) and BL300 (B) groups.

It can be observed that the mucosal folds in all the intestines (AI, MI and PI) of the BL50 (D) group are compactly arranged.

DISCUSSION

The addition of *V. amygdalina* had no negative impact on the overall growth performance of *O. niloticus* and its environment. This indicates the imperative viability of herbs to replace antibiotics and enhance physiological functioning. In agreement with the results on final weight, other findings[7,13,16], indicated that treatments with the *V. amygdalina* have an increase in final weight compared to the control group.

Also, studies by Nwogwugwu *et al.*[18], supported this study with an indication that final weight was more significant in treated bitter leaf groups than untreated group (control) in an experiment that determined the effect of *V. amygdalina* (bitter Leaf) extract on growth performance, carcass quality and economics of production of broiler chickens which is in agreement with the results in this study. This confirmed that, bitter leaf has a positive correlation with growth.

Okukpe *et al.*[7] reported that proximate composition of *V. amygdalina* leaf meal (VALM) shows a chemical composition of 527.83 metabolic energy (ME) kcal/kg, 86.40% dry matter (DM), 21.50% crude protein (CP), 13.10% crude fat (CF), 6.80% ether extracts (EE), 11.05% ash and the result on mineral composition indicate that *V. amygdalina* has 3.85% calcium, 0.40% magnesium, 0.03% phosphorus, 0.006% iron, 0.33% potassium and 0.05% sodium. Most of the minerals including calcium, magnesium and phosphorus play basic roles in countless biochemical reactions at the cellular level. They are also the main components of the skeleton and without magnesium many metabolic functions could not take place[19]. Therefore, it can be suggested that the increase in final weight in the *Vernonia amygdalina* group is influenced by the nutrient composition of the leaf, in agreement with Okukpe *et al.*[7].

Also, insignificant differences ($p>0.05$) between some of the treatments can be attributed to be due to their close relationship in concentrations. For instance, BL300 and BL400 seem to have no significant difference which is by other results where no significant difference was observed between 200 and 300 and 400 g[7]. Again, the differences in mean weight gain are by other literatures[7,18,20] where there was significant weight gain in bitter leaf treatments than the control group. The daily body weight gain of the birds on the test ingredients was better than those on the control diet.

The CT group consumed more of the feed which might be a result of no inclusion level of *V. amygdalina* leaf powder which did not reflect in their body weight gain, unlike the other groups with bitter leaf inclusions. The anti-nutritional factors indicated may be suspected to be the cause of why fish in *V. amygdalina* leaf powder treatment groups did not consume much feed but converted enough for WG.

However, Okukpe *et al.*[7] cited some nutrient qualities in the bitter leaf which positively improves growth rate and can be related to the performance of the groups with the bitter leaf. This study also indicates the efficiency of the bitter leaf to enhance increase in body weight gain while less feed is consumed and as such, serves the purpose of reducing cost of production.

Similar to other findings, *V. amygdalina* leaf meal (VALM) and other feed utilization in photobiotics such as garlic and ginger equally show significant differences in FCR. Shalaby *et al.*[21], in their study showed significant decreases in feed conversion ratio (FCR); 1.77±0.04 and 1.65±0.25 in *O. niloticus* fed 30 g *Allium sativum*/kg diet and 15 mg chloramphenicol/kg diet, respectively comparing to the control group (2.27±0.09) as also reported in a study by Banjoko *et al.*[16]. Suggestively, this may be a result of *V. amygdalina* leaf meal (VALM) with anti-bacterial properties which is in agreement with other study results.

The significant difference ($p<0.05$) in the specific growth rate of fish in the various treatments and the control may be as a result of an increase in feed intake, feed conversion ratio and condition factor because, generally, growth parameters affect each other. In many literatures[7,13,16,21], an increase or decrease in one of the growth parameters such as final weight, weight gain and feed intake affects others such as specific growth rate and feed conversion ratio.

From the data obtained, fish in *V. amygdalina* leaf powder/shreds groups showed higher survival rate (90.92 and 86.42%, respectively) which might be the effect of the antibacterial properties as indicated by Erasto *et al.*[14], which have a great potential of fighting against microbial infection in fish. From this study, the initial condition factor of the fish in the treatments and the control were all below 1.0. According to Datta *et al.*[22], k-values below 1 is an indication that, fish physiological well-being is at risk hence, it implies that the fish used for the experiment were not in good condition.

However, the significant differences ($p<0.05$) observed in the final k-values is also a clear the indication that some of the fish recovered from the columnaris disease as shown in the Fig. 1. The BL300 and BL400 treatments which had higher k-values according to the results mean that, *V. amygdalina* leaf powder/shreds significantly affected the health of the fish. Erasto *et al.*[14] reported the phytochemical analysis of ethanol extract from the leaves of *V. amygdalina* afforded two bioactive sesquiterpene lactones, vernolide and vernodalol. The two compounds were tested by agar dilution method against 10 bacteria strains and 5 fungi species. Both compounds exhibited a significant bactericidal activity against five Gram-positive bacteria however, lacking efficacy against the Gram-negative strains. However, the antimicrobial results of this study correspond positively with the claimed ethno-medical uses of the leaves of *Vernonia amygdalina* in the treatment of various infectious diseases.

The results showed that the water quality parameters (temperature, dissolved oxygen, pH, conductivity, turbidity, ammonia and nitrate) were not significantly different from each other. This informs that fish in different treatments were given equal environmental conditions. Also, *Vernonia amygdalina* leaf powder/shreds have no effect on any of the water quality parameters measured. This might be a result of the small amount of *Vernonia amygdalina* leaf powder/shreds used to form the solution.

Fish in the *V. amygdalina* treatments took the colour of the leaf powder (black) which showed or indicated recovery of fish from its original diseased pale color. This can mean that the concentrations were within limits that would not compromise the water quality in the systems.

The structure of the intestine is crucial for digestive processes in the intestinal tract. These factors are essential for fish growth and are widely acknowledged as key indicators of intestinal health and function[23,24]. The study revealed that in the groups treated with bitter leaf powder/shreds (B, C, D), the surface area of certain intestines became broader, thicker and more compact. Enhanced intestinal morphology is believed to facilitate improved digestion and nutrient absorption, leading to better feed utilization and growth performance[17]. This justifies why there is higher weight gain, feed conversion ratio, specific growth rate, survival rate and condition factor in the bitter leaf powder/shreds groups than in the control group. It can be suggested that bitter leaf powder/shreds have promoted intestinal digestion/absorption by activating digestive enzymes. This was in line with Apraku *et al.*[17] where alternative dietary plant lipid sources improved feed absorption and digestion. Again, this is also in line with Zhu *et al.*[25], where Gln (glutamine) promoted intestinal digestion and absorption. The disintegration of some of the intestines may suggestively be attributed to the functionality of some unknown nutrients and acids coupled with other factors of the biter leaf powder/shreds. Knudsen *et al.*[26] and Novriadi *et al.*[27], argued that gut inflammation in Atlantic salmon might be induced by saponins alone or in combination with other factors, such as the intestinal microflora. The antinutritional factors mentioned are no different since some of them are recorded in this current study.

CONCLUSION

Vernonia amygdalina leaf powder/shreds enhanced growth despite anti-nutritional factors. Fish in the BL300 and BL400 groups converted feed more efficiently, improving final weight, weight gain and growth rate. Economically, supplementing water with bitter leaf powder benefits feed conversion. *Vernonia amygdalina* enriched environments improve fish growth and disease management, with the highest survival rates and enhanced stress tolerance. The leaf increases the intestinal surface area, aiding nutrient absorption and digestion, resulting in significant weight gain. Recommended dosages of 300 and 400 g per 13.9 m^3 water treat columnaris effectively. Further studies should explore its antibacterial properties, impact on fish taste, skin texture and higher dosages.

SIGNIFICANCE STATEMENT

The study highlights the potential of bitter leaf (*Vernonia amygdalina*) as an effective antibacterial agent and growth enhancer for Nile tilapia (*Oreochromis niloticus*). Traditionally, antibiotics are used, but they pose long-term risks, like antibiotic resistance and health impacts. This research shows that *V. amygdalina* significantly improves growth metrics (feed intake, condition factor, final weight and weight gain) without harming water quality. The leaf's antibacterial properties effectively combat columnaris disease, highlighting its dual benefits for fish health and growth. By using *V. amygdalina*, the study suggests a sustainable, cost-effective and safer alternative to chemical treatments, promoting natural, biodegradable herbs in aquaculture for healthier and more efficient fish production.

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