

## Dawadawa and Kantong Additives Improve the Growth and Health of Nile Tilapia (*Oreochromis niloticus*)

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A feeding trial was conducted to evaluate the effects of condiment on the growth and

hematology of Nile tilapia (*Oreochromis niloticus*), stocked at 15 fish per tank with an initial average weight of 3.11±0.25 g for 8 weeks in twelve 55 m<sup>2</sup> circular tanks. Three experimental diets, such as Dawadawa, Kantong, and Dawakan additives, and a control

diet were studied in triplicate in this trial. The growth performance and feed utilization

showed significant differences (p<0.05) among the dietary treatments with SGR in the additives treatment, which ranged between  $3.77\pm0.61$  and  $4.31\pm0.09$ , which was higher than the control treatment ( $3.69\pm0.63$ ). The FCR of additive diets ranged

between 1.55±0.00 and 1.65±1.0 and was lower compared to the control diet

(1.67±0.00). The concentrations of white blood cells, red blood cells, hematocrit, and

hemoglobin (P≤0.05) observed in the control group were lower compared to the group

with an enhanced additives diet showing the best blood parameters. All in all, the fish-

fed Dawadawa additive showed the best growth and health among treatments. There

is, however, a need for further studies to observe the effect of the condiment on a

Abstract

challenged fish.

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#### Introduction

Aquaculture contributes significantly to a reduction in unemployment, poverty, and malnutrition globally (Julia et al., 2017). In Ghana, fish is the cheapest and most preferred source of animal protein, contributing about 60–75 percent to the total annual protein consumption (Lauria et al., 2018). Also, the annual mean per capita fish consumption is estimated to be about 22 kg, underscoring the significance of the role fish protein plays in the lives of Ghanaians (Akuffo & Quagrainie, 2019).

Tilapia is one of the most cultured fish species across the globe (Fitzsimmons, 2000), and it has the potential to feed on a variety of ingredients, which makes their culture easier and less costly due to the presence of this cheap source and easily accessible feedstuffs or plant products (Saikia and Das, 2015; Yue et al., 2016). Recently, animal feed production has been a focus on agriproducts, which have proven to influence growth performance and feed utilization positively due to their high protein-yielding characteristics (El-Sayed, 1999; Chaung et al., 2021; Manuelian et al., 2021; Magbanua & Ragaza, 2024). Identifying alternative sources of protein to be used as additives to improve the performance and functionality of feed for fish has gained favour in recent times as they contribute macromolecules to enhance efficient growth in Nile tilapia (Aragão et al., 2022; Youssef et al., 2023).

Plant-based feed additives come from roots, leaves, tubers, spices, fruits, and other plant parts that are mixed into animal feed to make it more digestible and help animals do better. They can also help fish feel less stressed (Felix & Francis, 2019; Encarnacúo, 2016). The use of them depends on the plant's part, the harvesting season, and its geographic origin (Steiner, 2006). When additives are added to aquaculture feed, they change how the feed works for aquatic organisms. The effects range from a low concentration of 100 g/ton feed (0.01% diet) to a higher concentration of 2-3 kg/ton feed (0.2–0.3%) of the active compounds in the product (Encarnação, 2016), which helps fish grow, gain weight, and have more lymphocytes (Ahmadifar et al., 2011). According to Sadik (2021), adding natural feed additives to feed helps to spur growth as well as improve the health and productivity of fish. These plant-based additives contain active ingredients or redox molecules like phenolics, flavonoids, terpenoids, phenol-derived aromatics, and aliphatic components (Rao, 2012; Orso et al., 2022). They have many effects, such as increasing appetite, boosting the immune system, killing microbes, reducing inflammation and swelling, stimulating gastric juice, and decreasing gut microbes (Chakraborty et al., 2014; Reverter et al., 2014). These chemicals are said to improve and support fish's overall health by having a big effect on their ability to grow faster, their immune systems, and how well they use their food (Firmino, 2021; Marimuthu et al., 2022; Vijayaram et al., 2022). The inclusion of feed additives in the diet of aquatic species has a functional effect on fish's weight gain and FCR, protein efficiency, fish health, and reduction of gut microbial effects (Encarnação, 2016; Hura et al., 2019; Liang et al., 2022). Feed additives work best when they come from a certain type of plant, are extracted in a certain way, and contain the right amount of extract for a standard protocol and the right amount of fish (Awad et al., 2021; EFSA et al., 2022; Orso et al., 2022).

Over the years, different feed additives have been tried on fish to boost their physiological qualities. However, due to anti-nutritional factors found in some of the ingredients, some showed little effect in boosting the growth of fish. Feed additives such as oregano, carvacrol, and thymol are known to have antioxidative and antibacterial properties (Baser, 2008; Giannenas et al., 2012; Yilmaz et al., 2015; Aanyu et al., 2018; Novriadi et al., 2023); garlic (Allium sativum) has immunostimulation and antibacterial effects (Militz et al., 2013); ginger leaves (Zingiber officinale Roscoe) has enhance feed digestion and high antioxidant properties in cat fish (Wei et al., 2024); rosemary (Rosmarinus officinalis) has high antioxidant properties and mortality reduction in tilapia (Abutbul et al., 2004); peppermint (Mentha piperita L.); and cinnamon (Cinnamomum verum) improve growth performance, fish health, and reduction of microbial load in the gut of fish (Talpur, 2014; Ahmad et al., 2011). Similarly, Giannenas et al. (2012) reported improved feed efficiency with unaffected body weight gain in a study to evaluate the effect of two feed additives in trout. The above feed additives used in the various studies contain macro- and micronutrients that support the optimal growth and survival of fish (Kristiana et al., 2020; Tiamiyu et al., 2016).

Moreover, Dawadawa (locust bean meal) and Kantong (cotton seed meal) condiments have similar bioactive compounds to the above plant products, which can enhance the nutritional quality of fish feed (Olasupo & Okorie, 2019). Dawadawa has been known to be a good source of calcium, fats, protein, vitamin C, phosphorus, and potassium. According to Mante (2019), Dawadawa boosts the immune system and traditionally treats skin-related diseases such as wounds, skin ulcers, and ringworm. Kantong also has important nutrients and strong biologically active compounds that stop cancer cells from spreading, boost the immune system, and lower the risk of other long-term diseases in animals (Behera et al., 2020; Adadi et al., 2019). These products are accessible and affordable in sufficient quantity, which is a cheap source of protein added to feed to achieve enhanced growth performance and yield in fish. Medicinal plant inclusion in feed has significantly contributed to the growth and health of aquatic species due to the presence of bioactive compounds. These plant products used as additives in feed formulation function as appetite stimulators, growth promoters, stress resistance, and immune stimulators in animals and fish (Citarasu, 2010). Again, these plant products have the potential to ameliorate the cost effect of feed on farmers due to their easy accessibility, cheaper protein source, and presence of other essential nutrients such as minerals, energy, and essential vitamins, which have been found to improve animals' growth (Tadese et al., 2022). However, to the best of our knowledge, no work has been done on the effect of Dawadawa and Kantong condiments on the growth performance and immunity of Nile tilapia. Hence, the study examined the effect of the condiments (Dawadawa and Kantong) on the growth performance and hematology of Nile tilapia cultured in concrete tanks in the northern region of Ghana.

#### **Materials and Methods**

#### **Preparation of Condiments**

#### **Dawadawa Condiment**

Two kilograms of *Parkia biglobosa* seeds were separated from their pulp and cooked to obtain a sticky fermented combination, as presented in Figure 1. They were then covered in ash and pounded to remove the seed coat, after which they were washed with water, boiled again, and left to ferment for 3–4 days. After that, they were pounded and moulded into balls and then dried in the sun to remove excess moisture for seven days. About 1 kg of Raanan commercial feed was grinded with 100g/0.1% of Dawadawa into powder

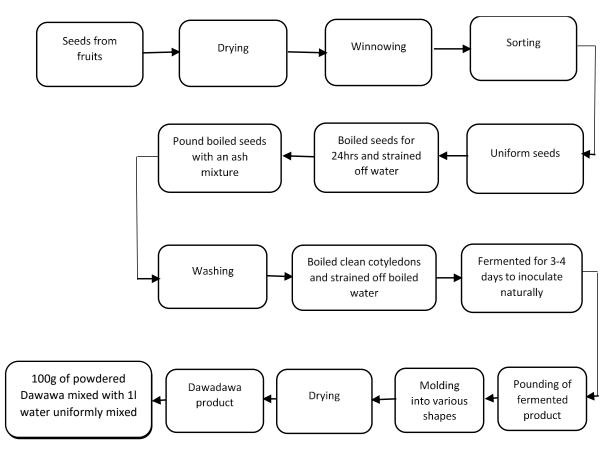


Figure 1: Schematics of stepwise Dawadawa preparation (Adadi et al., 2019)

form. Also, the combination of 50 g/0.05% each of Dawadawa and Kantong was ground with Raanan commercial feed into powder form. The Raanan feed and Dawadawa were grinded to a 3 mm particle size and then thoroughly mixed homogeneously with a Kitchener Beef Electric Meat Grinder (Model Number: 52021206) with 1 litre of water as presented in Table 1. The mashed product was then pressed through a Kitchener meat mincer with a 1.0 mm die plate. The pelleted feed was then air-dried between 27 and 30°C and stored in a moist-free environment for feed.

#### Kantong (Fermented Cotton Seed Meal) Condiment

One bowl of kapok tree (Ceiba pentandra) seeds was ground into powder, and 0.5 bowls of cassava flour and 1 liter of water were added and uniformly mixed as presented in Figure 2. The mixture was allowed to stand for a 48-hour period to ferment. Then the mixture was sun-dried for four days. About 1 kg of Raanan commercial feed was grinded with 100g/0.1% of Kantong into powder form. The Raanan feed and Kantong were grinded to a 3 mm particle size and then thoroughly mixed homogeneously with a Kitchener Beef Electric Meat Grinder (Model Number: 52021206) with 1 litre of water as presented in Table 1. The mashed product was then pressed through a Kitchener meat mincer with a 1.0 mm die plate. The pelleted feed was air dried between 27 and 30°C and stored in a moist-free environment for feeding.

#### **Experimental Diet and Design**

Twenty (20) kilograms of Raanan commercial fish feed were purchased and grinded with the additives for the experimental feed preparation. Four experimental diets were tested in the feed trial, which were designated: Dawadawa (DA), Kantong (fermented cotton seed meal) (KA), Dawakan (DAKA), and the control diet. The treatments were randomly assigned to the experimental tanks in triplicate. The composition of the additives and commercial diet used to prepare the feed are presented in Table 2.

#### Fish and Stocking

All male Nile tilapia (*O. niloticus*) were obtained from the Tamale Water Research Institute in the Northern Region. The fish were conditioned in a  $55 \text{ m}^2$ tank for one week before stocking was done. The fish were stocked at densities of 15 fish per tank at an average weight of  $3.11\pm0.25$  g per  $55 \text{ m}^2$  in triplicate.

#### Feed and Feeding Frequency

The experimental diets were administered to the cultured fish twice daily, at 9:00 a.m. and 4:00 p.m. The feeding levels were adjusted every two weeks after sampling using a feeding chart. The fish were fed 2% of their body weight.

Table 1. The three diets with different condiment (additives) inclusions

Diet	Composition
Control	Raanan commercial diet 1kg only
100g DA	Raanan commercial diet 1kg with 100g DA
100g KA	Raanan commercial diet 1kg with 100g KA
100g DAKA	Raanan commercial diet 1kg with 50g each of DA and KA

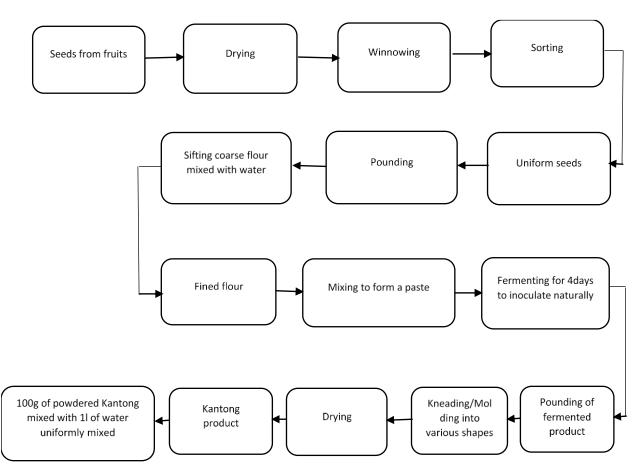


Figure 2. Schematics of stepwise Kantong preparation (Adadi et al, 2019)

Nutrients analyze	Dawadawa	Kantong
Carbohydrate (mg/100g)	16.25±0.65%	36.20±0.24
Crude protein (mg/100g)	41.50±0.77%	28.18±0.10
Lipid (mg/100g)	27.01±0.92%	3.22±0.77
Crude fiber (mg/100g)	3.75±0.86%	14.16±0.58
Total ash (mg/100g)	2.28±0.20%	4.23±0.71
Calcium (mg/g dry mass)	0.42±0.05	0.06±0.02
Magnesium (mg/g dry mass)	0.75±0.13	0.03±0.02
Potassium ((mg/g dry mass)	0.82±0.08	0.89±0.36
Sodium (mg/g dry mass)	0.27±0.05	0.85±0.39
Iron (mg/g dry mass)	1.43±0.21	0.01±0.02
Zinc (mg/g dry mass)	0.27±0.14	-
Copper (mg/g dry mass)	0.02±0.09	-

#### **Data Collection and Sampling**

All fish from the respective replicate tanks were sampled biweekly to monitor fish growth. The growth performance of fish and feed utilization in each tank was determined using the following growth parameters (Ridha, 2006; Lugert et al., 2018; Duodu et al., 2020).

Absolute growth rate (AGR) =  $\frac{\text{Final weight} - \text{Initial weight}}{\text{time(in days)}}$ Specific growth rate (SGR) In(Final Weight) – In(Initial Weight) Time interval (in days)  $\times 100$ Feed conversion ratio (FCR) =  $\frac{\text{Amount of feed fed(kg)}}{\text{Weight gained(kg)}}$  $Survival \ rate \ (SR) = \frac{\text{Number of fish at the end of the experiment}}{\text{number of fish at the beginning of the experiment}} \times 100$ Weight gain (WG) = Finalweight (FW) - Initial weight (IW)Feed efficiency ratio (FER) =  $\frac{\text{Weight gained by the fish}}{\text{amount of feed fed}}$ 

 $Feed intake = \frac{total feed quatity}{total number of fish}$ 

#### **Blood Sampling**

Blood samples were obtained after the 8-week feed trial. The fish were first anesthetized with benzocaine (100 mg/L). The fish blood was then collected by puncturing the caudal blood vessels using a 1 ml heparinized syringe (three samples/replicate) containing anticoagulants (0.1 ml of sodium citrate solution) and transported to the Tamale teaching hospital, where the full blood count was determined (Santos et al., 2009; Seibel et al., 2021; Esmaeili, 2021).

#### **Physiochemical Parameters**

Water quality variables such as dissolved oxygen, total ammonia, nitrite, nitrate, pH, TDS, and temperature were monitored weekly in each of the tanks between the hours of 6:00 a.m. and 7:30 a.m. The temperature and DO were measured using the EZDO DO/O<sub>2</sub>/Temp Meter PDO-408, while the pH and TDS were measured using the BANTE Portable Multi-Parameter Instrument 900P. The total ammonia, nitrite, and nitrate levels were measured using a Hach DR 2800 spectrophotometer with colorimetric assay kits (Hach Company, Loveland, Colorado).

#### **Statistical Analysis**

The data on growth performance and feed utilization of Oreochromis niloticus, as well as physicochemical parameters, were entered into Microsoft Excel 2016 version 16.0.4 and exported into GraphPad Prism version 9.4.1 software for analysis. The data was run through a normality test to ensure it was uniformly distributed, and a one-way ANOVA was conducted to ascertain the variation among treatments. Tukey multiple comparison tests were conducted to compare the differences in treatment means at the p<0.05 significant level. Correlation analyses were conducted on the nutrient factors of the water and the growth and feeding factors of fish. The results were presented in tables and figures as means ± SD.

#### Results

The growth performance and feed utilization of Nile tilapia fed with additive diets and control diets for eight weeks are presented in Table 3. In general, Nile tilapia fed with additives showed a significant increase in growth performance compared to fish fed with a control diet. Fish fed with a control diet, DA additive, KA additive, and DAKA additive witness a rise in weight from an initial average weight of 3.11±0.21 g to final mean weights of 19.85±0.15 g, 35.10±0.30 g, 28.85±0.25 g, and 32.70±0.10 g, respectively. The order of significance was as follows: DA > DAKA > KA additive > control diet (p<0.05). Overall, the average weight gain among fish fed with additives was significantly higher compared to those fed the control diet (p<0.05). The results of the SGR showed that the control diet had the lowest SGR (3.69±0.63 % day<sup>-1</sup>) compared with fish fed with KA additive, which recorded the best SGR (4.31±0.09 % day<sup>-1</sup>) followed by fish fed with DA (4.11±0.23 % day<sup>-1</sup>) and DAKA (3.77±0.61 % day<sup>-1</sup>). Survival rates among treatments were not significantly different, with the DA and KA treatments recording

Table 3: The growth performance and feed utilization of Nile tilapia fed with Dawadawa (locus bean meal) and Kantong (cottonseed meal) condiments cultured in tanks for 8 weeks

Treatments	Control	DA	KA	DAKA	P-Value
Initial Weight (g)	3.22±0.32	2.88±0.02	3.07±0.27	3.23±0.23	0.715
Final Weight (g)	19.85±0.15 <sup>d</sup>	35.10±0.30ª	28.85±0.25	32.70±0.10 <sup>b</sup>	0.001
WG (g)	16.5±0.37 <sup>d</sup>	31.94±0.14ª	25.95±0.25	29.85±0.15 <sup>b</sup>	0.001
SGR (%/day)	3.69±0.63 <sup>d</sup>	4.11±0.23 <sup>b</sup>	4.31±0.09ª	3.77±0.61 <sup>°</sup>	0.029
AG (g)	0.35±0.05 <sup>d</sup>	0.57±0.00ª	0.47±0.05	0.54±0.01 <sup>b</sup>	0.001
FCR	1.67±0.00	1.55±0.01	1.65±0.00	1.65±0.00	0.994
FI(g/day)	0.32±0.00	0.29±0.09	0.30±0.07	0.37±0.01	0.799
FER	11.57±0.27	22.24±0.13	15.86±4.95	14.76±7.01	0.450
Survival rate (%)	90.00±6.67°	90.00±3.33ª	90.00±3.33ª	83.33±3.33 <sup>b</sup>	0.898

90.00±3.33%, 83.33±3.33% for DAKA, and the control recording 90.00±6.67%.

Feed conversion ratio (FCR) of O. niloticus fed different additives had no significant difference between treatments (p>0.05). The results showed that fish fed with the control diet recorded the highest FCR (1.67±0.00) and were not significantly higher than fish fed with diets supplemented with DA (1.55±0.01), KA (1.65±0.00), and DAKA (1.65±0.00). Additionally, fish fed with DAKA had higher feed intake (0.37±0.01) than fish fed with the control diet (0.32±0.00), KA (0.30±0.07), and DA (0.29±0.09), which showed no difference between the dietary treatments at p>0.05 (p=0.799). Similarly, the feed efficiency ratio (FER) among treatments recorded was (22.24±0.13) for DA, (15.86±4.95) for KA, (14.76±7.01) for DAKA, and (11.57±0.27) for the control diet, respectively. There were no significant differences in feed efficiency ratio amongst the dietary treatments at p>0.05 (p=0.450), as presented in Table 3.

The hematological parameters of O. niloticus fed with different additives such as hematocrit, erythrocytes, and leukocytes were significantly different among treatments (p<0.05), as presented in Table 4. The hematocrit ranged between 30.25±3.00 - 31.90±3.80% in the additives treatment, while the control group recorded 24.00±2.50%. The erythrocytes in the additive treatment ranged between 1.88±0.02 – 2.03±0.29  $106/\mu$ L, while the control recorded  $1.53\pm0.22$   $106/\mu$ L. There were significant differences in hemoglobin levels: the additive treatments had levels between 12.65±0.65 -14.20±0.35 compared to the control (12.15±1.75); the additive treatments had levels between 158.05±3.55 -162.2±0.00 for mean corpuscular volume (MCV) compared to the control ( $158.05\pm5.75$ ); and the additive treatments had levels between  $41.60\pm1.90-47.35\pm8.60$  compared to the control ( $51.95\pm12$ ). The mean corpuscular hemoglobin (MCH) was also different in the control group ( $82.80\pm23.10$ ) compared to the DA ( $68.58\pm0.08$ ) and DAKA ( $67.50\pm3.00$ ) treatments. The differential leukocyte cell counts were not observed between treatments for total leukocytes, neutrophils, or monocytes, and neither eosinophils nor basophils were observed.

The water quality variables monitored over the growth trial are shown in Table 5. Overall, the water quality variables such as temperature, total dissolved solids, dissolved oxygen, and pH were within the suitable range for *Oreochromis niloticus* and were not different among treatments. The dissolved oxygen recorded for all the experimental tanks was high, which was above the critical limit (2.3 mg/l) for tilapia culture. The lowest dissolved oxygen level was recorded in the control diet (8.69±0.57 mg/l), with the highest level recorded in the DAKA treatment (9.51±0.22 mg/l), and there were no differences among treatments (p=0.379 at p>0.05).

There were fluctuations in temperature over the study period for all the experimental ponds, which ranged from  $27.12\pm0.18$ °C, but there were no differences among treatments. Similarly, pH was not significantly different among treatments over the study period and ranged between  $6.59\pm0.02$  and  $7.01\pm0.11$  at p>0.05, as shown in Table 5. Moreover, total dissolved solids (TDS) were not significantly different among treatments over the study period and ranged from  $81.07\pm7.26$  mg/l to  $91.20\pm18.32$  mg/l at p>0.05.

 Table 4: Blood general parameters (mean ± SE) of Nile tilapia fed with Dawadawa (locus bean meal) and Kantong (cottonseed meal) condiments cultured in tanks for 8 weeks

Groups	Control	DA	KA	DAKA	P- value	
Hematocrit (%)	24.00±2.50 <sup>b</sup>	30.25±3.00ª	31.90±3.80 <sup>a</sup>	30.40±0.20 <sup>a</sup>	0.000	
Hemoglobin (g/dL)	12.15±1.75	12.83±1.43	14.20±0.35	12.65±0.65	0.089	
Erythrocytes (10 <sup>6</sup> /µL)	1.53±0.22 <sup>b</sup>	1.91±0.16ª	2.03±0.29 <sup>a</sup>	1.88±0.02ª	0.001	
Leukocytes (10³/µL)	91.05±4.21 <sup>b</sup>	95.40±0.32ª	97.17±1.03ª	94.13±3.89 <sup>a</sup>	0.002	
MCV (fL)	158.05±5.75 <sup>b</sup>	158.83±1.38 <sup>b</sup>	158.05±3.55 <sup>b</sup>	162.2±0.00 <sup>a</sup>	0.028	
MCH (g/dL)	82.80±23.10 <sup>c</sup>	68.58±0.08ª	75.10±15.25 <sup>b</sup>	67.50±3.00 <sup>a</sup>	0.010	
MCHC (g/dL)	51.95±12.75ª	43.03±0.18 <sup>c</sup>	47.35±8.60 <sup>b</sup>	41.60±1.90 <sup>c</sup>	0.018	
PLT (10 <sup>3</sup> /µl)	104.00±45.00	101.75±12.75	114.00±20.00	100.00±19.00	0.993	

 Table 5: The water quality parameters of Nile tilapia fed with Dawadawa (locus bean meal) and Kantong (cottonseed meal) condiments cultured in tanks for 8 weeks

Parameters/Treatments	Control	DA	KA	DAKA	P-values	
Temperature (ºC)	28.15±0.09	27.12±0.18	27.13±0.69	27.81±0.86	0.430	
Total dissolved solids (mg/L)	91.17±10.96	84.45±14.37	81.07±7.26	91.20±18.32	0.835	
рН	6.59±0.02	7.01±0.11	6.99±0.15	6.77±0.35	0.195	
Dissolve oxygen (mg/L)	8.69±0.57	9.51±0.22	8.78±0.36	9.12±0.67	0.379	
TA-N (mg/L)	0.02±0.00	0.02±0.20	0.02±0.50	0.02±0.40	0.995	
NO <sub>2</sub> -N (mg/L)	0.01±0.00	0.01±0.02	0.02±0.05	0.02±0.04	0.046	
NO <sub>3</sub> -N (mg/L)	0.20 ±0.00	0.22±0.10	0.23±0.55	0.24±0.48	0.608	

Additionally, there was no significant difference in total ammonia  $(0.02\pm0.00 - 0.02\pm0.50)$  or nitrate  $(0.20\pm0.00 - 0.24\pm0.48)$  among treatments; however, there was a difference in nitrite  $(0.01\pm0.00 - 0.02\pm0.05)$  among treatments, with p=0.046 at p<0.05.

The effect of the condiment on the water quality and the growth of the tilapia were analyzed over the period. As shown in Table 6, the weight gain was linked to higher levels of total ammonia (r=0.784; p=0.117 at p>0.05), nitrite (r=0.948\*; p=0.014 at p<0.05), and nitrate (r=0.966\*\*; p=0.007 at p>0.001). As shown in Table 6, weight gain was linked to higher levels of total ammonia (r=0.926\*, 0.913\*, 0.907\*; p=0.024, 0.030, 0.034 at p<0.05), nitrite (r=0.801, 0.818, 0.832 p=0.103, 0.091, 0.081 at p>0.05), and nitrate (r=0.880\*, 0.918\*, 0.897\* p=0.049, 0.028, 0.039 at p<0.05) of all three condiment treatments. FCR also had a correlation with total ammonia (r=0.761; p=0.135 at p>0.05), nitrite (r=0.818; p=0.090 at p>0.05), and nitrate (r=0.988\*\*; p=0.002 at p>0.001) in the control treatment. Again, FCR was linked to total ammonia (r=0.703, 0.760, 0.799; p=0.185, 0.136, 0.105 at p>0.05), nitrite (r=0.807, 0.847, 0.798; p=0.099, 0.070, 0.106 at p>0.05), and nitrate (r=0.989\*\*, 0.985\*\*, 0.979\*\* p=0.001, 0.002, 0.004 at p=0.001) in the DA, KA, and DAKA treatments, in that order. Furthermore, dissolved oxygen had a positive correlation with total ammonia (r=0.197; p=0.751 at p>0.05) and nitrate (r=0.332; p=0.586 at p>0.05), respectively.

#### Discussion

Broadly, fish fed with the condiment had a higher growth rate compared to fish fed without additives. Fish fed with DA and DAKA exhibited the best weight gains compared to their counterparts, KA and the control diet. The reason for significant increases in weight gain can be attributed to an increase in protein, total free amino acid levels, vitamins, fatty acids, increased antioxidant activity, and reduced levels of anti-nutrients, as have been explained earlier (Akabanda et al., 2018; Ndukwe & Solomon, 2017; Oboh et al., 2008). Dawadawa by itself has a great potential for enhancing fish weight gain; however, when combined with KA, it performed better than KA alone, which is unable to cause the necessary change in weight because the anti-nutrients oppose the full functionality of the DA nutrients and some KA nutrients. It's possible that KA alone makes fish gain less weight because it has some anti-nutritional ingredients, like phytic acid and gossypol that stop fish from using protein and minerals properly, which is why the fish gain less weight (Mbahinzireki et al., 2001; Li et al., 2006; Agbo et al., 2011; Duodu et al., 2018). Before fermentation, soaking, adding water, and cooking the Parkia biglobosa plant helps lower the amount of harmful chemicals like tannins, alkaloids, and glycosides (Rosemond et al., 2022; Ndekwe & Solomon, 2017). During the fermentation of Dawadawa, certain fungi species (Saccharomyces spp.) help break down their anti-nutritional factors and make essential secondary metabolites easier to digest and more available (Garrido-Galand et al., 2021). The use of commercial additives such as Aqua Superliv, Aqua Booster, Aqua Pro, and Noni has been reported to significantly improve weight gains in fish (Dada, 2015). It's possible that the higher weight gain in this study was due to a lot more nutrients, like proteins, fatty acids, vitamins, and minerals, in the processed cotyledon. Other factors could include good feeding methods, better feed metabolism, and fish of the same size in the tank (Cadorin et al., 2022; Reverter et al., 2021; Adadi et al., 2019; Van Hai, 2015). The average weight gain recorded in this study was higher in the additive treatment compared to the control, which was lower than the results obtained by Kord et al. (2021), which had a higher average weight gain in the additive treatment compared to the control. These growth improvements could be attributed to stress, voluntary appetite suppression, competition for food and space, and a short period of culture (Rahman and Arifuzzaman, 2021). However, the weight gain in this study's additive treatment was similar to what Hassaan et al. (2019) found. They found that the Silybum marianum seed additive treatment led to more weight gain than the control.

Table 6. Correlation between nutrient factors and weight gain and feed conversion ratio of Nile tilapia fed with Dawadawa (locus bean meal) and Kantong (cottonseed meal) condiments cultured in tanks for 8 weeks

	C-WG	C-FCR	DA-WG	DA-FCR	KA-WG	KA-FCR	DAKA-WG	DAKA-FCR	AMMONIA	NITRITE	NITRATE	DO
C-WG	1	.951*	.925*	.931*	.945	.968**	.945*	.945*	.784	.948*	.966**	.109
C-FCR	.951*	1	.899*	.993**	.934*	.997**	.912*	.997**	.761	.818	.988**	.409
DA-WG	.925*	.899*	1	.844	.996**	.914*	.998**	.922*	.926*	.801	.880*	.188
DA-FCR	.931*	.993**	.844	1	.888*	.984**	.860	.983**	.703	.807	.989**	.442
KA-WG	.945*	.934*	.996**	.888*	1	.945*	.997**	.953*	.913*	.818	.918*	.238
KA-FCR	.968**	.997**	.914*	.984**	.945*	1	.929*	.993**	.760	.847	.985**	.346
DAKA-WG	.945*	.912*	.998**	.860	.997**	.929*	1	.931*	.907*	.832	.897*	.167
DAKA-FCR	.945*	.997**	.922*	.983**	.953*	.993*	.931*	1	.799	.798	.979**	.424
AMMONIA	.784	.761	.926*	.703	.913*	.760	.907*	.799	1	.667	.764	.197
NITRITE	.948*	.818	.801	.807	.818	.847	.832	.798	.667	1	.873	169
NITRATE	.966**	.988**	.880*	.989**	.918*	.985**	.897*	.979**	.764	.873	1	.332
DO	.109	.409	.188	.442	.238	.346	.167	.424	.197	169	.332	1

More so, the specific growth rate of this study was the best among the additive diets compared to the control diets, showing their enhancement of the growth performance of tilapia. This was similar to the results reported by Hassaan et al. (2019), who obtained a higher specific growth rate in the additive treatment compared to the control treatment. This could be as a result of higher nutrient composition, a good feeding regime, good water quality, and better metabolism, with the highest protein retention rate in both additives (Cadorin et al., 2022; Craig et al., 2017). Also, the Dawadawa additive treatment had more crude protein (41.50±0.77%) than the Kantong additives. The combination of these two was much more effective than commercial additives or probiotics (Saccharomyces, Lactobacillus sp., and Bacillus strains), as reported by Kord et al. (2021). The Dawadawa and Kantong additives also have biochemicals in them that act as feeding stimulants, pre/probiotics, enzymes, and hormones. These substances improve the metabolism and absorption of feed, which leads to higher SGR in fish and better growth (Gule & Geremew, 2022). It's similar to what other studies have found: higher SGRs were found in fish that were fed water spinach and dietary ALE supplementation than in fish that were fed the control (Chepkirui et al., 2022; Syed et al., 2022).

The survival rate of this study was between 83 -90% in all additive treatments, indicating a less limiting effect of the DA and KA additives on tilapia culture compared to the control (90%). The additive treatment had some leaching effects on the diet. It was observed that the DA and KA treatments fed to the fish are not instantly consumed by the fish, and if they stay in the water for some minutes, some of the additives dissolve into the bottom of the tanks, which may likely affect dissolved oxygen levels. Syed et al. (2022), who reported 92.88 - 97.82% in additive treatment, found that the survival rate of this study was lower. Again, the survival rate (100%) of feeding the red tilapia with Noni additives was also lower than the results obtained from this study (Kristiana & Mukti, 2020). The survival of tilapia as reported could arise based on environmental factors, predators, and social factors, which were all minimal during the cultural period. The high survival recorded could be related to good environmental factors, less social aggression, less density depending on factors in the culture system, as well as no predation activities, which were consistent with the studies by Kord et al. (2021); Hassaan et al. (2019); Ahmed & Abdel-Tawwab (2011), who obtained 92.66-100%, respectively, in the additives diet.

The feed utilization of fish-fed Dawadawa and Kantong additives showed better efficiency than that of the control and was found to be below the recommended FCR for tilapia culture (1.5–2.0) (Rahman & Arifuzzaman, 2021; Watanabe et al., 2002; Gule & Geremew, 2022). With this high efficiency, with the feed additives used in this study, it is envisaged that waste generation would be less when Dawadawa and Kantong

feed additives are used in feed. High FCR has negative impacts on water environments, and these could be detrimental to fish growth (Boyd, 2019). More so, the relatively low FCRs in this study could be a result of the preparatory methods employed in the feed formulation process. Prepared feeds could have less leaching, improve appetite stimulation, good metabolism, digestibility, and protein retention in fish (Akabanda et al., 2018; Li et al., 2020).

Haematology parameters are essential in evaluating nutritional suitability, concentrations, and toxicity, as well as their impact on aquatic animals' circulatory systems (Dash et al., 2015). It is an important metric to judge the health of fish and evaluate the effectiveness of fish feed (Dawood et al., 2016; 2019; Burgos-Aceves et al., 2019; Doan et al., 2019). In this study, the Dawadawa and Kantong additives improved the delivery of oxygen to cells by increasing WBCs and lowering HCT, Hb, and RBCs. These changes were important for fish's immune systems. This was consistent with the findings of Dawood et al. (2020) and Wei et al. (2024), who reported that the administration of immunobiotics and ginger leaves powder in the diet of Nile tilapia, rainbow trout and cat fish respectively, had significant effects on WBC, RBC, hematocrit, and hemoglobin compared to the control group. The results show that Dawadawa and Kantong can be safely added to O. niloticus's food to boost the fish's immune system. This was seen in the rise in the fish's white blood cells, which helps them fight off diseases. Again, the study showed that the condiment additives improved blood health compared to the control diet (Ramesh et al., 2015; Kord et al., 2021; Suphoronski et al., 2021). The results were evident that there was an increase in hemoglobin and hematocrit in Nile tilapia (i.e., improving oxygen delivery), which fed on the condiment, and were similar to the study where Bacillus amyloliquefaciens was used as additives in feeding the same Nile tilapia (Selim & Reda, 2015; Reda et al., 2016). The Dawadawa and Kantong additives might help the white blood cells multiply, which can lead to more antibodies, higher lysozyme activity, a faster phagocytic rate, and changes in the immune system. This can make Nile tilapia better at fighting off disease (Suphoronski et al., 2021). The white blood cells are key components of the innate immune response that regulates immune function and disease resistance in teleost fish (Smith et al., 2019). The severity of infection has a direct correlation to the WBC count level in fish (Seibel et al., 2021). In this study, white blood cell levels were enhanced in condiment-treated groups, but standardized doses of condiment were needed to improve the lymphocyte and neutrophil counts. The condiment increases the immune cells, especially lymphocytes that produce antibodies, and promotes fish defences against infection (Sahoo et al., 2021).

Feed is the primary source of waste in the culture system; diet formulation or feeding strategies should be managed properly to minimize waste generation (Gule & Geremew, 2022; Amirkolaie, 2011). The dietary treatments did not have any negative impact on the quality of water in the experimental tanks, and most of the recorded values, such as DO and temperature, were within suitable ranges for *O. niloticus* (Swann, 2007). The suitable water quality observed in the experimental tanks over the culture period could be attributed to the prevention of feed waste during the feeding process and also to the frequent topping up of the tank water with fresh water.

Even though the effect of the nutrient factors was not seen to impact the water quality during the study due to the proper feeding regime adopted, they could have some level of limitation on the total functioning of the water at certain periods. This is clear from the study that found links between nutrient factors, growth performance, and feed utilization. Dissolved oxygen, total ammonia, and nitrate were all linked in a good way. The levels of ammonia and nitrite in the different treatments were 0.02 mg/l and 0.01-0.02 mg/l, respectively. These levels were below the level that could affect fish performance. They were similar to the levels found by Putra et al. (2020) but lower than the levels found by Pedersen et al. (2012) and Davidson et al. (2017). The nitrate levels were 0.02 mg/l, which was within the optimal levels for good tilapia growth performance (Chapman & Kimstach, 1996); however, this was lower than the levels reported by Putra et al. (2020). This could be attributed to good water exchange practices that were adopted at regular intervals.

#### Conclusion

The study demonstrates that Dawadawa additives in the Nile tilapia diet enhance their growth performance, feed utilization, and immune responses (health outcomes). This additive may have a positive effect because it leads to better optimal growth (WG and SGR), good blood parameters (WBC, RBC, hematocrit, and hemoglobin), and less impact on water quality. Therefore, farmers could include 100g/0.1% of Dawadawa additive in Nile tilapia feed to improve farm productivity, as it has a potential cost-effective protein source that can boost tilapia immunity and overall health. The study recommends further trials of these additives at dose-response levels to obtain the optimum dose level for inclusion in the Nile tilapia diet. Additionally, the study suggested testing the fish with bacterial isolate to assess the immune response to the additives.

#### **Ethical Statement**

Approval for the conduct of the study was obtained from the Ethical and Research Committee of the University for Development Studies (UDS/RB/0122/22)

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#### **Author Contribution**

F.T. Iddrisu proposed the idea (study) as well as drafted the manuscript. T. Mohammed, S.O. Dandi, and M. Wumbedow Jamal Kadiru executed the experiment by following the methodology given in setting up the experiment, feeding the fish, monitoring water quality as well as recording the necessary data for the manuscript. E.D. Abarike, F. T. Iddrisu, D. N. Akongyuure, C. L. Ayisi computed the data, analyzed the data as well as presented the results and corrected writing errors in the manuscript, edited the manuscript for final submission, checking for the accurateness of data analysis and data presentation.

#### **Conflict of Interest**

The authors declare that they have no competing interests.

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