Squash (*Cucurbita maxima*) peel powder as a phytoadditive in diets of African catfish (*Clarias gariepinus*) fingerlings

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Abstract. Intensified aquaculture practices often rely on chemical additives to enhance production, which raises concerns regarding potential health risks and environmental impacts. As a sustainable alternative, the incorporation of phyto-additives has been explored to promote fish growth and survival without detrimental effects. This study aims to evaluate the effectiveness of squash (*Cucurbita maxima*) peel powder as a dietary phytoadditive on the growth, feed utilization, and survival of African catfish (*Clarias gariepinus*) fingerlings over a 60-day feeding trial. A total of 120 fingerlings were randomly assigned to four treatment groups: squash peel powder (SPP) at inclusion levels of 0%, 2%, 5%, and 10% (designated as SPP0, SPP2, SPP5, and SPP10, respectively), with three replications per treatment. The experimental design was a complete randomized design (CRD). Growth performance indicators, including weight gain and specific growth rate (SGR), feed performance measures such as feed conversion ratio (FCR) and feed conversion efficiency (FCE), and ammonia stress tolerance, and survival rates were assessed. The results indicated no statistically significant differences ($p > 0.05$) in growth, feed utilization, ammonia stress tolerance, or survival across the treatments. However, numerical trends suggested that the 5% inclusion level of squash peel powder may offer slight improvements in feed efficiency and growth performance. These findings suggest that squash peel powder can be safely included in catfish diets up to 10%, providing a sustainable and cost-effective alternative for feed formulation without compromising performance.
Key Words: agriculture by-products, circular economy, feed formulation, feed utilization, growth performance.

Introduction. Aquaculture, the cultivation of aquatic organisms, is rapidly expanding to meet the rising global demand for fish products (Zulhisyamet al 2020; Ahmad et al 2021; Purbomartono et al 2024). This growth is driven by population increases and declining wild fish catches. However, intensifying aquaculture practices to satisfy this demand often involve chemical additives, which raise concerns regarding potential health risks and environmental degradation (Purbomartono et al 2021; Jose Priya & Kappalli 2023). Phytoadditives, plant-based feed supplements that enhance palatability and improve performance, have been explored as a sustainable alternative to promote fish growth and survival while also supporting animal health and environmental protection (Hussain et al 2017; Gavris et al 2019; Rachmawati et al 2025).

One such promising phytoadditive is squash (*Cucurbita maxima*) peel, a by-product that, despite being considered waste, is a rich source of bioactive compounds and contains valuable nutrients that can be repurposed into high-value products (Kar et al 2023). Squash peels, which represent 10 to 12% of the total fruit mass (Kar et al 2023), are nutrient-dense, containing proteins, carbohydrates, vitamins (A, B-complex, C, E, and K), and essential minerals such as calcium, magnesium, sodium, potassium, phosphorus, iron, zinc, manganese, and copper (Dhiman et al 2009; Stryjecka 2025). Additionally, they contain phytochemicals like flavonoids and carotenoids (Hussain et al 2021), which may act as growth promoters and immune enhancers for fish, providing a sustainable and cost-effective alternative for aquaculture feed formulations.

In Southeast Asia, particularly in Thailand, Malaysia, and the Philippines, catfish (*Clarias* spp.) has become a highly profitable species in aquaculture (Coniza et al 2008). African catfish (*Clarias gariepinus* (Burchell, 1822)), a fast-growing, thermophilic species, is one of the most widely cultured freshwater fish (Nursyam et al 2017; Mustafa 2021; Kurniasih et al 2024) and can be grown at high stocking densities (Haylor 1991; Hengsawat et al 1997; Hossain et al 1998; van de Nieuwegiessen et al 2009; Abraham et al 2018; Pamula et al 2019; Mustafa 2021). *C. gariepinus* is a valuable source of fish protein, recognized worldwide as an essential food for both low-income and affluent families (Muchlisin et al 2014; Hastuti & Subandiyono 2018). The rising demand for catfish, driven by its good quality meat (Adamek et al 2011; Enzeline et al 2022), has spurred increased production, with the Philippines ranking as the fourth-largest producer in Southeast Asia during the early 2000s (Tan-Fermin 2003). However, despite its economic potential, catfish farming faces significant challenges, particularly high mortality rates among juvenile fish, leading to production losses of up to 50% (Coniza et al 2008; Obeda et al 2023). This pressing issue highlights the need for innovative strategies, such as the use of natural dietary additives, to enhance the health and survival of farmed catfish. This pressing issue highlights the need for innovative strategies, such as the use of natural dietary additives, to enhance the health and survival of farmed catfish.

Hence, this study aims to evaluate the effectiveness of squash (*C. maxima*) peel powder as a dietary phytoadditive on the growth, feed utilization, survival, and ammonia stress tolerance of *C. gariepinus* fingerlings over a 60-day feeding trial.

Material and Method

Experimental setup and culture conditions. The 60-day feeding trial was conducted from February 13 to April 13, 2025, at the Aquaculture Laboratory of the Camarines Norte State College – College of Fisheries, Aquatic Sciences, and Technology, located in Mercedes, Camarines Norte. A completely randomized design (CRD) was employed to evaluate the effects of squash peel powder-enriched diets on the growth, feed utilization, survival, and ammonia stress tolerance of *C. gariepinus* fingerlings. The fingerlings were sourced from a commercial catfish hatchery, transported in oxygenated plastic bags, and acclimated to laboratory conditions for a period not exceeding one week, during which they were fed a commercial fish feed with 35% crude protein content. Four dietary treatments were tested: a control (SPP0, 0% squash peel powder), SPP2 (2% squash peel powder), SPP5 (5% squash peel powder), and SPP10 (10% squash peel powder), with three replicates for each treatment. A total of 120 fingerlings were stocked into 50-liter tanks, with 10 fish per tank, distributed across 12 tanks. Each tank was filled with filtered freshwater and aerated to maintain optimal water quality. To prevent escape and reduce the risk of injury, all tanks were securely covered with fine-meshed fish nets, thereby enhancing survival rates and ensuring the integrity of the experimental conditions.

Feed preparation and formulation. The compositions of the treatment diets are presented in Table 1. The basal diet formulation, adapted from Serrano et al (2022), comprised fish meal (sardines), soybean meal, corn starch, cod liver oil, vitamin mix, mineral mix, and carboxymethylcellulose (CMC). All ingredients were sourced from agricultural and feed supply stores in Daet, Camarines Norte. Squash peels were collected from a local market in Daet, washed thoroughly, sun-dried for one week, ground into a fine powder, and sieved to ensure uniform particle size. The powdered squash peel was incorporated into the basal diet at four inclusion levels: 0 g kg^{-1} (SPP0, 0%), 20 g kg^{-1} (SPP2, 2%), 50 g kg^{-1} (SPP5, 5%), and 100 g kg^{-1} (SPP10, 10%), based on the inclusion rates of Gonzales-Plasus et al (2022). All dry ingredients were pre-weighed and mixed thoroughly before gradually adding water to form a homogeneous dough. This dough was then steamed at 100°C for 10 minutes, pelletized into appropriate sizes, and air-dried for 2 to 3 days. The finished diets were stored in sealed containers at room temperature until use.

Table 1

Composition of experimental diets for squash peel (g)

Ingredients	*Treatments*			
	SPP0	*SPP2*	*SPP5*	*SPP10*
Fish meal	496	496	496	496
Soybean meal	267	267	267	267
Corn starch	125	105	75	25
Cod liver oil	22	22	22	22
Vitamin mix	25	25	25	25
Mineral mix	25	25	25	25
Carboxymethylcellulose	40	40	40	40
Squash peel powder (SPP)	-	20	50	100
Total	1,000	1,000	1,000	1,000

Feeding management. Fish in each tank were fed twice daily, at 8:00 a.m. and 3:00 p.m., with either the control diet or the corresponding phytoadditive-enriched diets. The daily feeding ration (DFR) was initially set at 5% of the total biomass and was adjusted every 6 days following sampling. The 60-day feeding trial included regular monitoring of fish health, feeding behavior, and tank conditions.

Growth and feed efficiency parameters. Fish in each tank were measured for body weight and total length at the beginning of the experiment and subsequently every 6 days using a digital weighing scale and caliper. Growth performance and feed utilization efficiency were assessed through the following parameters: absolute growth (AG), specific growth rate (SGR), feed conversion ratio (FCR), feed conversion efficiency (FCE), and survival rate. These parameters were calculated using the formulas provided below:

$$AG\ (g) = FABW - IABW$$
$$SGR\ (\%\ day^{-1}) = 100 * (\ln FABW - \ln IABW) / D$$
$$FCR = FC\ (g) / AG\ (g)$$
$$FCE\ (\%) = WG\ (g) / FC\ (g) * 100$$
$$Survival\ (\%) = (Final\ count\ of\ fish / Initial\ count\ of\ fish) * 100$$

where: FABW = final average body weight (g) of individual fish;
IABW = initial average body weight (g) of individual fish;
FC = feed consumed;
D = days of culture.

Water quality monitoring and management. Water quality is a key determinant in predicting the success of an aquaculture system (Gagelonia et al 2024). Water in the culture system was exchanged after each weight sampling or as necessary to maintain optimal conditions. Continuous aeration was provided throughout the experimental period to ensure sufficient dissolved oxygen (DO) levels. Water quality parameters, including temperature, pH, and DO, were monitored twice daily, before the morning and afternoon feedings. Temperature was measured using a thermometer, pH with a pH meter, and DO with a DO meter. Nitrite and total ammonia-nitrogen concentrations were assessed weekly using commercially available test kits. Daily siphoning was performed to remove feces and uneaten feed, and any water loss during this process was immediately replenished with filtered freshwater. These management practices ensured the maintenance of suitable water quality throughout the 60-day feeding trial.

Ammonia stress test. Following the 60-day culture period, *C. gariepinus* fingerlings were continuously fed their respective experimental diets for an additional two weeks before the ammonia exposure test. Fish were exposed to ammonia at a concentration of 0.88 mg ammonia-N·L^{-1} for 24 hours, following the procedure of Wise et al (1989). The exposure medium was prepared by dissolving ammonium chloride (NH_4Cl) in freshwater.

Continuous aeration was provided, and no feeding occurred during the exposure. Fish survival was monitored at intervals: every 15 minutes for the first hour, every 30 minutes for the second hour, every hour after four hours, every four hours after sixteen hours, and every hour at 24 hours. Fish were deemed dead when they became immobile and unresponsive to tactile stimulation with a glass rod, as described by Baldove et al (2019).

Data analysis. Descriptive and inferential statistical methods were used to analyze the collected data. Parameters including final average weight gain, SGR, FCR, FCE, and survival rate were initially tested for normality and homogeneity of variances. Statistical analyses were performed using Jamovi software with a 95% confidence level. A one-way Analysis of Variance (ANOVA) was applied to assess significant differences among treatments. When significant differences were found, Tukey's Honest Significant Difference (HSD) test was applied to compare the treatment means.

Results

Growth performance. The inclusion of squash (*Cucurbita* spp.) peel powder in the formulated feeds for *C. gariepinus* fingerlings did not result in statistically significant differences ($p > 0.05$) in weight gain or SGR among the treatments (Table 2). The group receiving 5% squash peel powder inclusion (SPP5) achieved the highest mean weight gain (9.14±1.19 g) and SGR (5.20±0.28% day^{-1}), but these values were not significantly different from those of the control (SPP0) or other treatment groups (SPP2 and SPP10).

Table 2

Feed performance indices of *C. gariepinus* fingerlings fed diets with varying squash peel powder levels. Values are mean±SD

Growth performance indices	*Treatment*			
	SPP0	*SPP2*	*SPP5*	*SPP10*
Weight gain (g)	7.03±0.07^{a}	7.29±0.50^{a}	9.14±1.19^{a}	7.80±1.73^{a}
Specific growth rate (% day^{-1})	4.82±0.35^{a}	4.81±0.18^{a}	5.20±0.28^{a}	4.81±0.64^{a}

Note: Superscripts indicate significant differences between treatments. Same letters within a row indicate no significant difference, while different letters denote significant differences.

Although no statistically significant differences were observed, the higher weight gain and SGR in the SPP5 group suggest that moderate levels of squash peel powder may offer growth-enhancing benefits. This trend may be attributed to the bioactive compounds and fiber content in squash peels, which could improve digestion and nutrient utilization. However, the variability in results, particularly in the SPP5 and SPP10 groups, may have obscured the treatment effects. These findings are aligned with those of Mounes et al (2024), who observed improved growth in Nile tilapia (*Oreochromis niloticus*) fed pumpkin seed cake, another Cucurbitaceae-derived additive. While differences in species and plant parts may explain the contrasting outcomes, the data supports the potential of squash peel as a functional ingredient in aquafeeds.

Feed performance. Feed performance, assessed by FCR and FCE, showed no statistically significant differences ($p > 0.05$) among dietary treatments supplemented with squash peel powder (Table 3). FCR values ranged from 1.37±0.10 in the SPP5 treatment to 1.55±0.06 in SPP2, while FCE values ranged from 64.62±2.55% (SPP2) to 73.38±12.68% (SPP0). Although no significant differences were found, the 5% inclusion level (SPP5) showed improved feed utilization.

The SPP5 group recorded the lowest FCR and one of the highest FCE values, reflecting its superior growth performance. These results suggest that moderate inclusion of squash peel powder may enhance feed utilization, likely due to digestible fiber, bioactive compounds, or essential micronutrients that promote gut health and nutrient absorption.

Table 3
Feed performance indices of *C. gariepinus* fingerlings fed diets with varying squash peel powder levels. Values are mean±SD

Feed performance indices	*Treatment*			
	SPP0	*SPP2*	*SPP5*	*SPP10*
FCR	1.39±0.22[a]	1.55±0.06[a]	1.37±0.10[a]	1.46±0.12[a]
FCE	73.38±12.68[a]	64.62±2.55[a]	73.17±5.69[a]	68.96±5.83[a]

Note: Superscripts indicate significant differences between treatments. Same letters within a row indicate no significant difference, while different letters denote significant differences.

Survival. Survival rates of *C. gariepinus* fingerlings fed diets supplemented with varying levels of squash peel powder showed no statistically significant differences among treatments ($p > 0.05$) (Figure 1). Survival rates ranged from 76.67% in the SPP10 group to 93.33% in the SPP2 group, with both the control (SPP0) and SPP5 treatments recording 83.33%.

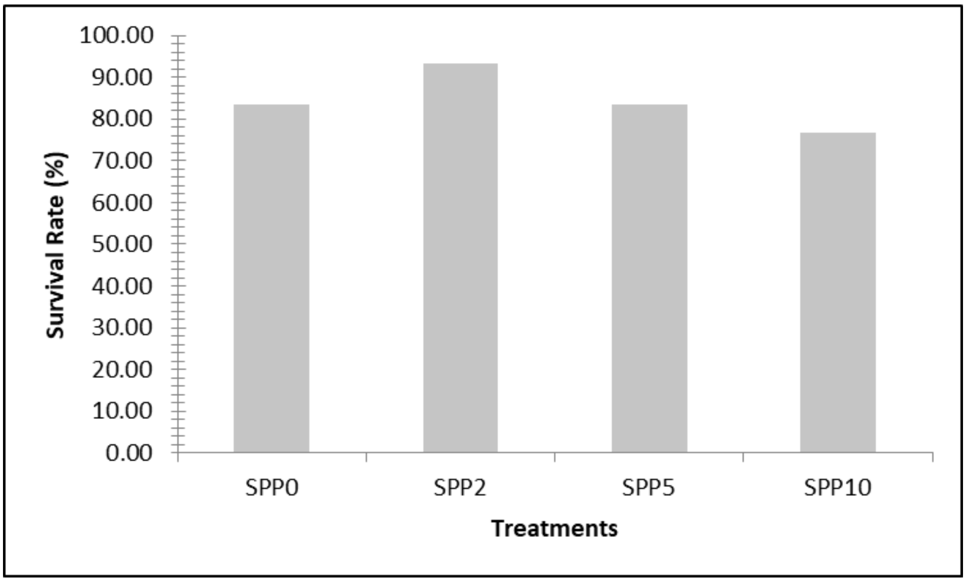

Figure 1. Survival rate of *C. gariepinus* fingerlings fed diets supplemented with varying levels of squash peel powder.

Ammonia stress test. The ammonia stress test on *C. gariepinus* fingerlings revealed no mortality in any of the treatment groups, including the control (SPP0) and all squash peel-enriched diets (SPP2, SPP5, and SPP10), during the 24-hour exposure at a sub-lethal ammonia concentration of 0.88 mg NH_3-N/L. This finding indicates that the inclusion of squash peel powder, even at levels up to 10%, did not compromise the fish's resistance to ammonia-induced stress.

Discussion. In the present study, the inclusion of squash peel powder in the diets of *C. gariepinus* fingerlings resulted in comparable growth performance, survival, and feed utilization across treatments, indicating that this agricultural by-product can be used as a phytoadditive without compromising key production parameters.

Growth performance. Although no statistically significant differences were observed, the increased mean weight gain and SGR in the SPP5 group suggest a potential growth-promoting effect at moderate inclusion levels. This trend is consistent with findings by Mounes et al (2024), who observed improved growth in Nile tilapia fed pumpkin seed cake, another Cucurbitaceae-derived additive. While species and plant part variations may explain differing outcomes, the general pattern supports the inclusion of squash peel powder as a functional aquafeed ingredient.

The positive growth and feed utilization may be attributed to the beneficial phytochemicals present in squash peel. As noted by Hussain et al (2022), squash peel is rich in fiber, protein, β-carotene, essential minerals (calcium, iron, zinc), and bioactive compounds such as phenolics, flavonoids, and carotenoids. These components are linked to improved gut health, enhanced antioxidant defense, and better nutrient absorption, potentially supporting the observed growth in *C. gariepinus*. This underscores squash peel's potential as a cost-effective and sustainable phytoadditive for aquaculture.

At higher inclusion levels (10%), excess fiber or anti-nutritional factors may have limited nutrient absorption, potentially neutralizing the benefits seen at moderate levels. However, the lack of negative growth responses across all treatments suggests that squash peel powder can be safely included in diets up to 10% without adverse effects on growth performance.

Feed performance. Moderate dietary fiber levels, such as those from squash peel, can improve digestive efficiency in fish. Research on species like largemouth bass (*Micropterus salmoides*) and channel catfish (*Ictalurus punctatus*) has shown that optimal fiber levels reduce FCR and enhance protein efficiency without affecting growth (Li et al 2012; Zhong et al 2020). In this study, the 5% squash peel inclusion (SPP5) likely provided a beneficial fiber level that aided digestion and nutrient absorption.

Squash peel contains bioactive compounds like phenolics, carotenoids, and minerals, which support gut health and antioxidant defense, further enhancing nutrient utilization (Hussain et al 2022). Similar improvements in feed efficiency have been observed when fishmeal was supplemented with functional plant additives such as yeast or fruit peel extracts. Therefore, the improved feed efficiency in SPP5 may be attributed to a synergistic effect of fiber and phytonutrients improving gut function.

Even modest reductions in FCR offer significant benefits in terms of cost and environmental sustainability. A lower FCR indicates more efficient feed utilization, leading to lower production costs (Besson et al 2014; Omasaki et al 2017) and reduced nutrient excretion, as more nutrients are retained by the fish (White 2013). If applied in large-scale catfish farming, the 5% squash peel powder inclusion in the SPP5 treatment could help reduce feed costs and decrease nitrogen and phosphorus discharge into the environment.

Survival. Although survival rates did not differ significantly, the highest survival rate in the SPP2 group suggests that a low-level inclusion of squash peel powder (~2%) is well tolerated and may enhance stress resilience or immune function. In contrast, the slightly lower survival in the SPP10 group may indicate a threshold where increased fiber or anti-nutritional factors (ANFs) in the squash peel - such as phytates, tannins, or oxalates - could negatively affect palatability or gut health (Gopan et al 2020).

The high survival rates across all treatments (> 75%) suggest that squash peel powder, up to 10%, does not cause lethal or toxic effects in *C. gariepinus* fingerlings. This finding aligns with Mohaammed et al (2014), who reported that *C. maxima* fruit parts, including the peel, contain low levels of ANFs, such as phytate ($0.68\ mg \cdot 100\ g^{-1}$) and oxalate ($0.23\ mg \cdot 100\ g^{-1}$), which are within safe limits and unlikely to cause mineral deficiencies or toxicity. Furthermore, the nutritional benefits of squash peel, including fiber, protein, and essential minerals (calcium, iron, and potassium), likely supported the physiological resilience and adaptation of the fish to the formulated diets, reinforcing the safety of squash peel powder as a feed additive for aquaculture.

Ammonia stress test. The complete absence of mortality across all treatments suggests that the fingerlings maintained physiological stability under ammonia stress. This result may be attributed to the bioactive compounds in squash peel, such as phenolics, flavonoids, carotenoids, and essential minerals like potassium, calcium, and iron, which are known to support antioxidant defense, immune modulation, and cellular homeostasis (Mohaammed et al 2014; Hussain et al 2022).

Ammonia toxicity in fish is often linked to oxidative stress, particularly in organs responsible for detoxification, such as the gills and hepatopancreas. Menon et al (2023) highlighted that elevated ammonia concentrations can induce the generation of reactive oxygen species (ROS), disrupting cellular functions and causing tissue damage. The survival of *C. gariepinus* under such conditions suggests that this species possesses inherent mechanisms to mitigate oxidative stress at moderate ammonia levels.

Furthermore, the phytoactive compounds in squash peel may have enhanced the fish's resistance to oxidative stress, though further studies are needed for conclusive confirmation. The consistent survival observed across all treatments suggests that squash peel is not only a safe phytoadditive but may also provide a supportive role in maintaining fish health under environmental stress. These findings suggest the suitability of *C. gariepinus* for aquaculture systems where ammonia buildup may occur, particularly in high-density tank or pond environments.

In the absence of mortality under ammonia exposure underscores the potential of squash peel as a functional ingredient in aquafeeds. Its dual benefits - contributing to both nutritional performance and resilience under environmental stress - support its application in sustainable aquaculture practices.

Conclusions. This study demonstrates that squash (*Cucurbita maxima*) peel powder can be incorporated into the diets of African catfish (*Clarias gariepinus*) fingerlings up to 10% without adverse effects on growth, feed utilization, or survival. While no significant differences were observed, the 5% inclusion level showed slight improvements in feed conversion ratio and feed efficiency, suggesting its potential as a growth-promoting and cost-effective phytoadditive. Furthermore, the high survival rates and resilience to ammonia stress across all treatments highlight the safety and potential of squash peel powder as a sustainable alternative in aquaculture feed. These findings support its use in aquafeed formulations, contributing to both fish health and the sustainability of aquaculture practices. Further studies are needed to explore long-term effects and the underlying mechanisms of squash peel's benefits.

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Conflict of interest. The authors declare that there is no conflicts of interest.

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