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Research Paper

Dietary Impact of Sweet Orange (*Citrus sinensis*) Peel Meal on Growth, Wound Healing and Blood Components of *Clarias gariepinus* Juveniles (Burchell, 1822)

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Abstract

A twelve-week feeding trial was conducted to examine the dietary impact of air-dried sweet orange (Citrus sinensis) peel meal (OPM) on growth, wound healing and blood components of Clarias gariepinus juveniles. The study involved five dietary treatments comprising OPM included at 0 (control treatment), 10, 20, 30 and 40g/kg respectively in the experimental diets labelled T1 to T5 and had three replicates constituting fifteen plastic aquaria. A total of 210 fish were randomly distributed into the aquaria at the rate of 14 fish per aquarium and were fed at 3% of their body weight twice daily (8.00-9.00am and 4.00-5.00pm). During the feeding trial, growth and feed utilization parameters were determined while, blood indices were measured at the end of the feeding trial. Wound healing was monitored after the feeding trial on the surviving fish by cutting 1cm² area into the flesh of the fish selected from each treatment at its lateral line and tail regions. The study revealed that air-dried orange peel meal at 10g/kg optimized fish growth while 30g/kg improved the packed cell volume, haemoglobin, red blood cell, white blood cells, total protein, albumin, globulin, aspartate amino transferase, and alanine transaminase of C. gariepinus. Higher ammonia content was detected as the OPM amount increased. Thus, air-dried orange peel meal has promising potentials in enhancing growth, blood components and wound healing in C. gariepinus juveniles. Fish farmers may consider incorporating processed orange peel meal into fish feed for the production of fast-growing and healthy fish by monitoring amount of ammonia.

1. Introduction

The utilization of plants, plant extracts, by-products and wastes in aquaculture to improve the general wellbeing, blood components and fish growth rate is gaining acceptance recently, mainly because attention is being directed to the use of organic materials in fish production in an attempt to avert the problem of residual effects of the inorganic compounds on the tissues of the fish consumers. Bartley (2022) described aquaculture as one of the fastest growing food sectors globally. However, intensive fish farming operation has been reported to be faced with significant challenges such as disease outbreak and physical injuries among farmed fish and these have been described by Moreira et al. (2021) to have adversely affected fish health and yield. Thus, health management in aquaculture is very crucial in ensuring production of healthy fish. The treatment of

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diseases or other health-related issues in fish production has depended mainly on the use of synthetic chemicals whose misuse has led to the development of drug resistance, residual toxicity and environmental pollution (Mezmale et al., 2020). Consequently, the quest for natural, sustainable and eco-friendly means of addressing fish health becomes very crucial (Moreira et al., 2021).

The global war against the use of growth-promoting synthetic hormones or other inorganic compounds in fish production has successfully paved way for the use of bio-friendly materials which cause no negative effects on the fish and the consumers and could even perform better than synthetic or inorganic ones (Salem & Abdel-Ghany, 2018). This development has therefore led to the search for alternative organic materials that could enhance growth, health and wound healing rates of fish, especially those cultivated under intensive fish culture. Elferink et al. (2008) described investing on agro-industrial by-products as supplements in animal diets to be a suitable option that could alleviate the industrial impact on the environment and improve turnover.

Various fruit and vegetable by-products have been incorporated into animal feeds with positive results (Di Sotto et al., 2020; Williams et al., 2020). Orange peels are a by-product obtained from the processing of citrus or other orange species. *Citrus sinensis* is among the orange species commonly consumed in Nigeria and its peels are considered wastes whose disposal has been described to be posing environmental challenges (Ani et al., 2015). Sharma et al. (2017) stated that citrus-processing industries generate large quantities of orange peels, specifically about half of the fruit's weight and this constitutes a critical problem to the environment. However, orange peel is rich in nutrients which include carbohydrates, fibre, vitamins and bioactive compounds such as polyphenols, flavonoids and essential oils (Ghanem, et al., 2012; Russo et al., 2021). Furthermore, Ghanem et al. (2012) reported that these compounds possess antioxidant, antimicrobial and growth-promoting properties, thereby making orange peel an important ingredient to be studied for its growth-promoting and wound-healing properties as well as its effect on the blood components of *C. gariepinus* juveniles.

Extraction, fermentation, and drying have been commonly used in orange peel treatment. Extraction method is compound target specific (Li et al., 2022); it can target the essential oils or water soluble antioxidants. However, this method has the drawback of solvent, high temperature, and pressure requirement (Yogita, 2025). Fermentation method on the other hand is considered a green process because it uses microorganism in the breaking down process. However, it has been described to alter the chemical composition of the sample (Yogita, 2025) and reduce the functional value of other components (Ganiyu et al., 2024). Drying that involves high temperature has been reported to alter the phytochemical or the antioxidant properties of the peel (Oluba et al., 2021). However, air-drying orange peel has been reported to retain the fibre content, antioxidant capacity and the bioactive compounds. Drying reduces the moisture content and the bulkiness of the air dried sample.

The African catfish, *C. gariepinus*, is one of the widely cultured fish species in Nigeria, owing to its fast growth, high fecundity and resilience to unfavourable conditions (Popoola, 2022). Moreover, high protein content, palatable flesh and economic viability make it a staple fish in many African and global markets (Mbkaan, 2020; Popoola, 2022). Although, the inclusion of orange peel meal in animal diets has been associated with enhanced growth performance, improved feed utilization and appreciable health conditions in various livestock species (Vlaicu et al., 2020; Hosseini et al., 2020; See et al., 2024), there is lack of information on the specific impacts of air-dried orange peel meal on the health, blood indices and wound healing rate of *C. gariepinus*. Thus, this study is about investigating more about orange peel, which is an alternative organic material that could enhance growth, health and wound healing rates of fish, especially those under intensive culture.

2. Materials and Methods

2.1 Preparation of orange peel meal

Ten kilograms of *Citrus sinensis* peels, used for this study, were obtained from local orange merchants at Ejigbo market in Ejigbo, Osun State, Nigeria. The peels were soaked and washed in distilled water for thirty minutes, packed with colander for the water to drain off and later air-dried at room temperature (27.4 - 28.7°C) for three weeks. The air-dried orange peels were then milled into powder and stored in a dry container prior to use.

2.2 Experimental design and procedure

The study adopted a 5-by-3 factorial completely randomized experimental design, which was conducted in the Fish Nutrition Laboratory of the Department of Fisheries and Aquatic Resources Management, College of Agriculture and Renewable Natural Resources, Osun State University, Osogbo, Nigeria. Each treatment group contained an inclusion level of air-dried orange peel meal and was replicated thrice. Two hundred and thirty *C. gariepinus* post-fingerlings were obtained from the Departmental Fish Farm. A total of 210 fish were used for the experiment. The fish (initial mean weight: 5.55±0.16 g) were stocked in fifteen plastic, 45 L capacity rectangular aquaria at the rate of 14 fish per aquarium. Each aquarium was filled with 40 L of borehole water and covered with mosquito net to prevent the fish from escaping. Prior to the feeding trial, the fish were acclimatized for two weeks and fed twice daily with a commercial feed at 3% of their body weight.

2.3 Experimental diet formulation

Five iso-proteinous experimental diets were formulated with ingredients shown in Table 1. The air-dried powder orange peel meal was added as an additive to the other ingredients. The air-dried orange peel meal was not added to the control diet (Treatment T1), while treatments T2, T3, T4 and T5 contained air-dried orange peel meal of 10, 20, 30 and 40 g/kg, respectively, that is 0, 1, 2, 3 and 4 kg/100kg, respectively. The corresponding yellow maize added were 24.6, 23.6, 22.6, 21.6 and 20.6 kg/100kg to T1, T2, T3, T4 and T5, respectively. Five kilograms of each diet were prepared. The ingredients were thoroughly mixed by using one litre of warm distilled water. Each diet mixture was pressed through a 2-mm die of a pelletizing machine (Caps Feed Limited, Nigeria) to produce strands of pellets which were collected, air-dried for 6 h and then packed into air-tight containers.

Table 1: Gross ingredient composition of experimental diets, with graded levels of air-dried orange peel meal

Ingredients	Weight (kg) for each experiment
Yellow maize	24.6-20.6
Orange peel meal	0-4
Fishmeal	26
Soybean meal	23
Groundnut cake	22
Palm oil	2
Mineral/vitamin Fish premix	1
Di-calcium phosphate	0.5
Table salt	0.5
Lysine	0.2
Methionine	0.2
Total	100

2.4 Fish growth determination and blood samples analysis

Throughout the 12-week feeding trial, the diets were administered to the fish at 3% of their body weight at 9:00 am and 4:00 pm daily. Growth rate was monitored by determining the average weight change fortnightly using a digital pocket weighing scale (Sensitive Electronics, Rajkot, India). At the end of the growth period, growth parameters, namely mean weight gain, daily feed intake, specific growth rate, protein intake, protein efficiency ratio, feed conversion ratio and survival rate were determined, by using the following basic equations (Akanmu et al., 2024).

Mean weight gain (g) = Final weight (g) – Initial weight (g)

$$Specific\ growth\ rate\ (day^{-1}) = \frac{L_n(Final\ weight) - L_n(Intial\ weight)}{Duration\ of\ feeding\ trial\ (days)} * 100$$

$$Feed\ conversion\ ratio\ (FCR) = \frac{Mean\ dry\ feed\ fed\ (g)}{Mean\ weight\ gain\ (g)}$$

$$Protein\ intake\ (PI) = \frac{Feed\ intake * \%\ protein\ in\ the\ diet}{100}$$

$$Protein\ effeciency\ ratio\ (PER) = \frac{Mean\ weight\ gain\ (g)}{Protein\ intake}$$

$$Survival\ rate = \frac{Intial\ No\ fish\ stocked - final\ No\ fish\ cropped}{Intial\ No\ of\ fish\ stocked} * 100$$

At the end of the growth period, two juveniles were randomly picked from each aquarium for blood sample collection. Blood samples were collected between the ventral fin and the caudal fin, very close to the genital openings. The samples collected were divided into two sets of vials. The samples kept in the vials containing ethylene di-amine tetra-acetic acid (EDTA) were used for hematological analyses while those kept in the plain vials were used for serum biochemical analyses.

2.5 Wound healing test

At the completion of the growth trial, the wound healing study was conducted on the surviving fish in each treatment group. Each treatment group was allotted 24 juveniles (average weight: 63.28±0.42 g) at the rate of 8 juveniles per aquarium. The fish were conditioned for seven days before the wound healing study commenced. After the conditioning, an incision of 1cm long and 1 cm deep was made on each fish at the lateral line region (above the lateral line) and the tail areas of the fish. Each incision was made with a disinfected surgical blade and 1cm long broom stick was used as the template for this dimension and for uniformity. The wounds were not treated but the fish were returned into the aquaria and allowed to feed the allotted diets for fifteen days during which the wound closures on the fish in each group were monitored. Three fish per aquarium were randomly selected to examine the wound closure 3, 6, 9, 12 and 15 days of the study. The changes in wound areas, percentage healing rates and daily healing rates of the wounds were determined (Akanmu et al., 2016).

2.6 Statistical analysis

Data collected from the proximate composition of the experimental diets, growth parameters, wound healing rates, blood parameters, and water quality parameters were all subjected to one way analysis of variance using IBM SPSS version 21 software. The mean values and the standard error of the mean were determined using descriptive statistics. The data was subjected to test of homogeneity of variance and the individual mean values were separated using Duncan multiple range test at 0.05 significant level.

3. Results and Discussion

3.1 Proximate composition of the experimental diets

Table 2 shows the proximate composition of the diets containing graded levels of air-dried orange peel meal. There was no significant difference (p>0.05) in moisture, ash and crude fat among the treatments.

The insignificant difference in the moisture content of the diets suggests that the inclusion of orange peel meal did not influence the water retention properties of the diets. Maintaining consistent moisture levels is crucial for ensuring feed quality and stability during storage (Rathod & Kumawat, 2024). The ash content also showed no significant differences among the diets. Ash content is an indicator of the total mineral content in the diet and the present result suggests that the mineral composition of the diets remained stable despite varying levels of OPM inclusion.

Table 2: Proximate composition of experimental diets containing graded levels of air-dried orange peel (Mean ± SE)

Parameters	T1	T2	T3	T4	T5
Moisture content (%)	6.16±0.01	6.15±0.02	6.14±0.01	6.13±0.02	6.17±0.10
Ash content (%)	13.14±0.03	13.06±0.04	13.11±0.01	13.14±0.03	13.19±0.01
Crude protein (%)	40.43±0.12^{b}	40.60±0.06^{ab}	40.54±0.06^{ab}	40.72±0.06^{ab}	40.83±0.00^{a}
Crude lipid (%)	1.09±0.01	1.10±0.02	1.13±0.01	1.09±0.01	1.10±0.02
Crude fibre (%)	3.00±0.02^{a}	3.05±0.04^{a}	2.90±0.02^{b}	3.01±0.02^{a}	3.04±0.01^{a}
Carbohydrate (%)	36.18±0.08^{a}	36.04±0.09^{a}	36.18±0.05^{a}	35.91±0.05^{ab}	35.67±0.05^{b}
Energy (Kilocalories)	267.48±0.09^{ab}	267.44±0.23^{bc}	268.00±0.07^{a}	267.21±0.02^{b}	266.79±0.03^{c}

Similarly, crude fat content did not exhibit significant differences among the diets. This indicates that the lipid composition of the diets was unaffected by the incorporation of OPM, which is essential for providing the necessary energy and supporting metabolic processes in the fish (NRC, 2011). Crude protein was highest in T5 and lowest in T1 (control). The increased protein content at higher OPM inclusion levels suggests that OPM can enhance the protein profile of the diets. Protein is a critical nutrient for fish growth and development, and the higher protein content at T5 (40 g/kg) could contribute to improved growth performance (Tacon & Metian, 2015). The values of crude fibre content (2.90 - 3.05%) indicate that OPM affects the fibre content variably. While fibre is essential for maintaining digestive health, excessive fibre can reduce feed digestibility and nutrient absorption (Hardy, 2010). Carbohydrates are a primary energy source and their varying levels across the diets suggest that higher OPM inclusion may reduce the carbohydrate content. This reduction could be attributed to the substitution of carbohydrate-rich ingredients with OPM, which contains higher fibre and protein (Sharma et al., 2017). The variation in caloric content is important as it influences the energy availability for metabolic processes and growth. The highest caloric content in T3 could indicate an optimal balance of nutrients for energy production while the lowest value in T5 suggests a possible reduction in energy density due to higher fibre and lower carbohydrate content.

3.2 Growth response in *C. gariepinus* juveniles

Table 3 shows the growth response indices in *C. gariepinus* juveniles fed diets containing graded levels of air-dried orange peel meal. The study revealed that air-dried orange peel meal inclusion had no significant differential effect on the quantity of feed fed, specific growth rate, feed conversion ratio, protein efficiency ratio, final number of fish cropped and % survival. However, the final weight was significantly highest ($p<0.05$) at T2 and lowest at T1 (control diet). Mean weight gain at T2 (10 g/kg) significantly exceeded ($p<0.05$) the values in the other treatments. Protein intake and protein efficiency ratio were also highest ($p<0.05$) at T2.

No significant variations occurred in the initial weights of the fish and the quantities of diets fed across the treatments. This indicates that the treatment groups were uniform at the start of the feeding trial and that the feed intake was consistent, ensuring a fair comparison of the effects of OPM inclusion levels on growth performance. The initial number of fish stocked and the final number of fish cropped did not differ significantly among the treatments. This suggests that OPM did not have a detrimental effect on fish survival, as the number of fish remained consistent throughout the study. Feed conversion ratio (FCR) and protein efficiency ratio (PER) showed no significant differences across the treatments. This implies that the efficiency of converting feed into body mass and the effectiveness of protein utilization were not adversely affected by the inclusion of OPM in the diets.

Table 3: Feed utilization and growth parameters of *C. gariepinus* juveniles fed diets containing graded levels of air-dried orange peel meal (Mean ± S.E)

Parameters	T1	T2	T3	T4	T5
Initial mean weight (g)	5.86±0.50	5.82±0.16	5.42±0.08	5.45±0.04	5.20±0.03
Final mean weight (g)	60.87±0.22^{b}	69.17±0.31^{a}	62.54±0.38^{ab}	61.52±0.63^{ab}	62.31±0.57^{ab}
Mean weight gain (g)	55.01±0.63^{b}	63.35±0.21^{a}	57.12±0.41^{b}	56.07±0.64^{b}	57.11±0.56^{b}
Specific growth rate(day^{-1})	2.79±0.21	2.95±0.06	2.91±0.04	2.89±0.03	2.96±0.02
Quantity of feed fed (g)	51.38±3.53	57.57±1.71	53.07±0.90	56.62±1.96	54.94±3.93
Feed conversion ratio	0.93±0.07	0.91±0.04	0.93±0.02	1.01±0.04	0.96±0.06
Protein intake	20.77±0.12^{c}	23.37±0.06^{bc}	21.51±0.06^{bc}	23.06±0.06^{ab}	22.43±0.00^{a}
Protein efficiency ratio	2.65±0.01	2.71±0.00	2.66±0.01	2.43±0.02	2.55±0.01
Initial № of fish stocked	14.00±0.00	14.00±0.00	14.00±0.00	14.00±0.00	14.00±0.00
Final № of fish cropped	8.00±0.58	9.00±0.58	10.00±0.88	9.00±0.67	11.00±0.58
Survival rate	57.14±4.13	64.29±4.13	71.43±6.30	64.29±4.76	78.57±4.12

FCR and PER are critical indicators of feed quality and fish health, and the results suggest that OPM can be included in fish diets without compromising these efficiency metrics (NRC, 2011). The specific growth rate (SGR) and percentage survival also showed no significant variations among the treatments. SGR is a measure of the daily growth rate of the fish, and the lack of significant differences indicates that the growth rate remained stable across the different levels of OPM inclusion.

The high survival rates further supported the assertion that OPM does not negatively impact fish health. Final weight was highest at T2 and lowest at T1 (control). This finding suggests that a moderate inclusion level of OPM can enhance the final weight of *C. gariepinus* juveniles. The increase in final weight at T2 could be attributed to the balanced nutritional profile provided by the moderate level of OPM, which might have optimized growth conditions (Tacon & Metian, 2015). Mean weight gain and feed conversion efficiency were significantly high at T2 compared to the other treatments. This further reinforces the assertion that a moderate inclusion of OPM is beneficial for growth performance. Higher weight gain and feed conversion efficiency at T2 indicate that the fish were able to effectively utilize the nutrients in the diet, resulting in better growth outcomes. Protein intake was significantly highest at T5 and lowest at T1. This result is consistent with the proximate composition of diet T5, where crude protein content was highest at T5. High protein intake suggests that the fish placed on this diet consumed more protein, which is crucial for growth and development. However, the highest protein intake did not result in the highest growth performance, indicating that other factors such as nutrient balance and digestibility might play a role (Hardy, 2010).

3.3 Hematological and serum biochemical indices of *C. gariepinus* juveniles

Table 4 indicates the hematological indices of *C. gariepinus* juveniles fed air-dried orange peel meal-supplemented diets. No significant differences ($p>0.05$) existed in the values obtained for packed cell volume, hemoglobin count, red blood cells, platelets, mean corpuscular volume, mean corpuscular hemoglobin, mean corpuscular hemoglobin concentration, lymphocytes, eosinophil and monocytes. However, white blood cells were high ($p<0.05$) at T4 and T5 but lowest at T1. Neutrophils were significantly higher ($p<0.05$) in T1 and T4 than in the other treatments.

The hematological indices were not significantly different among the treatments. This consistency suggests that the dietary inclusion of OPM did not adversely impair the general hematological health of the fish.

Table 4: Hematological indices of *C. gariepinus* juveniles fed diets containing graded levels of air-dried orange peel meal (Mean ± SE)

Parameters	T1	T2	T3	T4	T5
PCV	15.00±4.04	16.33±3.84	14.67±3.67	17.00±2.08	14.00±2.31
HB	4.90±1.33	5.33±1.29	4.77±1.22	5.57±0.66	4.03±0.37
RBCs	3.53±1.33	3.78±0.69	4.81±0.38	5.42±0.66	4.55±0.20
WBCs	5.90±0.55[c]	8.07±0.87[b]	8.67±0.70[b]	10.20±0.35[a]	10.33±0.68[a]
PLATELETS	4.00±0.58	4.00±0.58	3.67±0.67	4.67±0.33	3.67±0.33
MCV	46.67±11.67	42.67±4.84	30.00±6.25	30.67±0.88	30.33±4.10
MCH	15.00±4.00	12.67±1.45	9.00±2.08	9.67±0.33	8.33±0.67
MCHC	33.00±0.00	33.00±0.00	33.00±0.00	33.00±0.00	33.00±0.00
LYMP	38.00±0.58	40.00±1.16	38.33±0.33	40.33±0.67	41.00±0.58
NEUT	61.33±0.88[a]	58.67±0.33[b]	61.00±0.58[a]	58.67±0.88[b]	57.67±0.33[b]
EOS	0.00±0.00	0.67±0.67	0.33±0.33	0.33±0.33	1.00±0.58
MONO	0.67±0.33	0.67±0.33	0.33±0.33	0.67±0.33	0.33±0.33

PCV: Packed Cell Volume; HB: Hemoglobin; RBCs: Red Blood Cells; WBCs: White Blood Cells; MCV: Mean Corpuscular Volume; MCH: Mean Corpuscular Hemoglobin; MCHC: Mean Corpuscular Hemoglobin Concentration; LYMP: Lymphocytes; NEUT: Neutrophils; EOS: Eosinophils; MONO – Monocytes

The parameters are crucial indicators of the fish's ability to transport oxygen and maintain immune function, and their stability across treatments indicates that OPM is a safe feed additive ingredient in terms of these health markers (Blaxhall & Daisley, 1973). However, the white blood cell (WBC) count was highest at T4 and T5 and lowest at T1 (control). The elevated WBC counts at higher levels of OPM inclusion suggest an enhanced immune response, possibly due to the bioactive compounds in orange peels that may stimulate immune function (Ghanem et al., 2012). Neutrophils were significantly high in T1 and T4 than in the other treatments, indicating a variable response in neutrophil count that could be linked to stress or infection response mechanisms modulated by the dietary components.

Table 5 presents the serum biochemical indices of *C. gariepinus* juveniles fed graded levels of orange peel meal-supplemented diets. The parameters measured showed no significant differences ($p>0.05$) across all the treatments.

The serum biochemical indices were not significantly different across the treatments. This observation specifies that the metabolic and liver functions of the fish were not affected by the varied inclusion levels of OPM. Serum biochemical indices are critical for evaluating the dietary and health condition of fish, and the lack of significant changes suggests that OPM does not induce metabolic disturbances or liver dysfunction (Hrubec et al., 2000). The stability of serum biochemical parameters across the treatments underscores the potential of OPM as a safe and effective dietary component. The absence of significant changes in these indices further supports the findings from the hematological data, indicating that OPM can be included in fish diets without compromising metabolic health.

3.4 Water quality parameters in experimental aquaria

Table 6 presents the water quality parameters in the experimental aquaria. Ammonia significantly increased ($p<0.05$) as the concentration of the air-dried orange peel meal in the diets increased across the treatments. Dissolved oxygen was highest ($p<0.05$) in T1 than in the other treatments. The air-dried orange peel meal had no significant differences ($p>0.05$) in the pH readings (6.47 - 6.50) of the culture water medium. Nitrate and nitrite were not detected in the culture water medium.

Water quality parameters are crucial factors which determine the welfare and the environmental conditions of the cultured fish. The result of this study showed that the values of all the water quality parameters measured were within the optimal limits for *C. gariepinus* (Boyd, 1990). However, elevated ammonia levels can be toxic to fish as well as cause stress and gill damage (Randall & Tsui, 2002).

Table 5: Serum biochemical indices of *C. gariepinus* juveniles fed diets containing graded levels of air-dried orange peel meal (Mean ±SE)

Parameters	T1	T2	T3	T4	T5
TP	15.00±4.04	16.33±3.84	14.47±3.67	17.00±2.08	14.00±2.31
Albumin	4.90±1.33	5.33±1.29	4.77±1.22	5.57±0.66	4.03±0.37
Globulin	3.53±1.33	3.78±0.69	4.81±0.38	5.42±0.66	4.55±0.20
AST	5.90±0.55	8.07±0.87	8.67±0.70	10.20±0.35	10.33±0.68
ALT	4.00±0.58	4.00±0.58	3.67±0.67	4.67±0.33	3.67±0.33
ALP	46.67±11.67	42.67±4.84	30.00±6.25	30.67±0.88	30.33±4.10

TP – Total Protein; AST – Aspartate Amino Transferase; ALT – Alanine Transaminase; ALP – Alkaline Phosphate

Table 6: Water quality parameters in experimental aquaria (Mean ±SE)

Parameters	T1	T2	T3	T4	T5
Ammonia (ppm)	0.25 ± 0.04^{d}	0.35 ± 0.03^{c}	0.52 ± 0.00^{b}	0.52 ± 0.00^{b}	0.55 ± 0.00^{a}
Dissolved oxygen (ppm)	5.53 ± 0.00^{a}	5.33 ± 0.00^{b}	5.30 ± 0.00^{b}	5.33 ± 0.00^{b}	5.33 ± 0.00^{b}
Temperature (^{O}C)	23.67 ± 0.56^{ab}	23.33 ± 0.58^{ab}	23.00 ± 0.00^{b}	24.00 ± 0.00^{a}	23.67 ± 0.58^{ab}
pH	6.47±0.02	6.48±0.02	6.48±0.02	6.48±0.02	6.50±0.02

The addition of air-dried orange peel meal might have led to an increased nitrogen load in the culture water, which could possibly be due to the metabolic breakdown of the diets. The higher ammonia concentration could suggest a potential increase in waste production or reduced efficiency in ammonia assimilation or nitrification processes. This could be due to the specific dietary composition or the metabolic response of the fish to the particular feed formulation. Proper management of ammonia levels is crucial, as prolonged exposure to high ammonia can lead to ammonia toxicity, affecting the respiratory efficiency and overall health of the fish (Francis-Floyd et al., 2003). The result from this study revealed that all the treatments maintained DO levels sufficient for the growth and survival of *C. gariepinus*, suggesting that the addition of air-dried OPM did not critically compromise oxygen availability in the experimental water (Boyd & Tucker, 1998).

Temperature is a vital environmental factor influencing metabolic rates, growth, and reproductive performance in fish (Jobling, 1993). The stable and narrow temperature range observed in this study suggests that orange peel meal inclusion did not significantly affect the culture water temperature and equally indicates a stable thermal condition across all the experimental groups. This stability is important for ensuring that any observed differences in growth performance and health are primarily due to dietary treatments rather than temperature fluctuations (Brett, 1979). The temperature range falls within the suitable limits for *C. gariepinus*, promoting normal physiological processes and minimizing thermal stress (Hecht et al., 1996).

The consistent pH values indicate that the inclusion of OPM did not alter the acidity or alkalinity of the culture water medium, providing a stable environment for the fish (Wedemeyer, 1996). Nitrate and nitrite were not detected in the culture medium. The absence of these nitrogenous compounds suggests effective management of nitrogen waste in the culture system, preventing the accumulation of potentially harmful substances.

3.5 Rates of healing at the tail and lateral line wound regions of C. gariepinus juveniles

Table 7 shows the rates of healing at the tail and lateral line wound areas of *C. gariepinus* juveniles. Though no significant differences (p>0.05) existed in the healing rates of the tail wound area for days 3, 6, 9, 12 and 15, at day 15, the healing rate was 100%. For the lateral line wound area, no significant differences (p>0.05) occurred in the healing rates for days 3, 6, 9 and 15. However, at day 12, T5 had the highest (p<0.05) healing rate. Again, at day 15, the healing rate was 100% across all the treatments.

The result of wound healing in this study suggested that the inclusion of air-dried OPM significantly accelerated the wound healing process at the caudal and lateral wound areas. The OPM included in the experimental diets possibly contributed to the complete wound closure recorded by all the experimental fish on the 15th day of the study. The better healing rates observed in the fish placed on diets containing higher inclusion of OPM could be ascribed to the orange peel meal included in the experimental diets. This suggests that higher levels of OPM could enhance wound healing, possibly due to the presence of bioactive compounds in the orange peels that possess antimicrobial and anti-inflammatory properties, which are beneficial for wound repair (Hernandez et al., 2021).

The higher white blood cell counts recorded in T4 and T5 suggested that higher levels of OPM might enhance immune response. Elevated white blood cell counts are indicative of a stimulated immune system, which could be beneficial in improving disease resistance (Hernandez et al., 2021). Additionally, higher neutrophil counts in T1 and T4 could indicate some variations in the immune cell distribution among the treatments. This variability could reflect differential responses to dietary inclusion levels of OPM, which necessitate further investigation to fully understand the immunomodulatory effects of OPM. The serum biochemical indices of *C. gariepinus* juveniles fed with graded levels of air-dried OPM suggest that its inclusion does not disrupt the biochemical health markers of the fish, maintaining stable levels of serum biochemical indices across the treatments. This finding aligns with previous research reports indicating that plant-based additives can support fish's immune function and overall health without adversely affecting normal physiological processes (Bampidis & Robinson, 2006).

Table 7: Rates of healing at the tail and lateral line region wound areas of *C. gariepinus* juveniles fed diets containing graded levels of air-dried orange peel meal

Day	Parameters	Tail region wound area					Lateral line region wound area				
		T1	T2	T3	T4	T5	T1	T2	T3	T4	T5
3	Wound area change (cm)	0.80±0.04	0.77±0.04	0.80±0.04	0.85±0.02	0.80±0.04	0.79±0.03	0.77±0.03	0.77±0.04	0.80±0.04	0.82±0.03
	Rate of healing (%)	20.00±3.50[b]	23.33±4.22[a]	20.00±3.25[b]	15.00±2.24[c]	20.00±3.65[b]	21.67±3.07	23.33±4.22	23.33±4.22	20.00±3.65	21.67±3.07
	Rate of healing (day)	6.67±1.10[b]	7.78±1.41[a]	6.67±1.22[b]	5.00±0.75[c]	6.67±1.02[b]	7.22±1.03	7.78±1.41	7.78±1.41	6.67±1.22	7..22±1.03
6	Wound area change (cm)	0.65±0.02	0.60±0.04	0.65±0.03	0.67±0.02	0.65±0.03	0.63±0.02	0.53±0.03	0.60±0.05	0.58±0.03	0.63±0.03
	Rate of healing (%)	35.00±2.24	40.00±3.65	35.00±3.42	33.33±2.11	35.00±3.42	36.67±2.11	46.67±3.33	40.00±4.47	41.67±3.07	36.67±3.33
	Rate of healing (day)	5.84±0.37	6.67±0.61	5.83±0.57	5.56±0.35	5.83±0.57	6.11±0.35	7.78±0.56	6.67±0.75	6.95±0.51	6.95±0.80
9	Wound area change (cm)	0.53±0.02	0.47±0.03	0.53±0.03	0.50±0.03	0.52±0.03	0.45±0.02	0.43±0.03	0.45±0.04	0.48±0.03	0.45±0.02
	Rate of healing (%)	46.67±2.11	53.33±3.33	46.67±8.17	50.00±2.58	48.33±3.07	55.00±2.24	56.67±2.24	55.00±4.28	51.67±2.34	55.00±2.24
	Rate of healing (day)	5.10±0.24	5.93±0.37	5.19±0.37	5.74±0.34	5.37±0.34	6.12±0.25	6.30±0.37	6.11±0.48	5.74±0.34	6.12±0.25
12	Wound area change (cm)	0.25±0.02[b]	0.25±0.02[b]	0.27±0.02[a]	0.23±0.02[ab]	0.25±0.02[b]	0.25±0.02[ab]	0.25±0.02[ab]	0.28±0.02[a]	0.27±0.02[a]	0.22±0.02[b]
	Rate of healing (%)	75.00±2.24[a]	75.00±2.24[a]	73.00±2.11[b]	76.67±2.11[a]	75.00±2.24[a]	75.00±2.24[ab]	75.00±2.24[ab]	71.67±1.67[c]	73.33±2.11[ab]	78.33±1.67[a]
	Rate of healing (day)	6.25±0.19[b]	6.25±0.19[b]	6.11±0.18[ab]	6.39±0.18[a]	6.25±0.19[b]	6.25±0.19[ab]	6.25±0.19[ab]	5.97±0.14[c]	6.11±0.18[ab]	6.53±0.14[a]
15	Wound area change (cm)	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00	0.00±0.00
	Rate of healing (%)	100±0.00	100±0.00	100±0.00	100±0.00	100±0.00	100±0.00	100±0.00	100±0.00	100±0.00	100±0.00
	Rate of healing (day)	8.33±0.00	8.33±0.00	8.33±0.00	8.33±0.00	8.33±0.00	8.33±0.00	8.33±0.00	8.33±0.00	8.33±0.00	8.33±0.00

4. Conclusions

The findings showed that dietary inclusion of air-dried orange peel meal) has promising potential in enhancing the growth, blood components and wound healing process in *C. gariepinus* juveniles. Thus, it can be effectively combined into the diets with no adverse effect on growth, hematological and serum biochemical parameters. Moderate OPM level (10 g/kg) significantly improved growth and feed utilization indices, indicating its potential as a beneficial feed supplement. Elevated WBC counts at higher OPM levels suggest enhanced immune response while stable water quality parameters indicate no environmental impact.

Overall, air-dried OPM offers a sustainable alternative feed supplement, promoting growth and health in *C. gariepinus* juveniles. The orange peel meal can be an effective dietary supplement in aquaculture, providing a natural and sustainable alternative to synthetic additives. By enhancing the health and resilience of *C. gariepinus*, orange peel meal can potentially improve productivity, leading to an efficient aquaculture practices. Based on the findings of this study, it is recommended that aquaculture practitioners consider incorporating processed orange peel meal into the diets of *C. gariepinus* to promote wound healing and improve blood parameters. However, the inclusion levels should be optimized through further research to determine the most effective dosage. Additional studies should also be conducted to explore the long-term effects of orange peel meal utilisation on the growth and overall health of *C. gariepinus*. Further research should also focus on understanding the specific mechanisms through which the bioactive compounds in orange peels exert their beneficial effects.

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