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ASSESSING THE EFFECT OF THE SELECTED FRUIT PEEL POWDERS AS A FEED ADDITIVE ON GROWTH PERFORMANCE, FEED UTILIZATION AND ANTIOXIDANT ACTIVITY OF THE WHITELEG SHRIMP, *Penaeus vannamei*

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ABSTRACT
A study was conducted to evaluate the effect of selected fruit peel powder as feed additive on growth performance and feed utilization efficiency of *Penaeus vannamei*. A 75-day feeding trial was conducted using with fruit peel powder inclusion level 5% of T0 (control), T1 (orange peel), T2 (apple peel), T3 (papaya peel) and T4 (mango peel). Results show that weight gain and specific growth rate of shrimp in T3 treatment are significantly higher ($p < 0.05$) compared to other treatments. Moreover, feed conversion ratio (FCR), protein efficiency ratio (PER) and feed efficiency ratio (FER) were significantly improved in treatment T3. Antioxidant activity and survival rate was significantly higher in T1 treatment. The findings of the present study demonstrated that dietary supplementation of fruit peel powders at 5% inclusion level significantly enhanced growth performance and feed utilization in *Penaeus vannamei.* Among the tested treatments, papaya peel supplementation showed superior growth promoting effect, while orange peel supplementation exhibited better on antioxidant activity survival rate.

KEYWORDS*: Penaeus vannamei*, fruit peel powders, growth performance, feed utilization, antioxidant activity.

1. INTRODUCTION

In 2024, total world aquaculture production reached 141.60 million tonnes, with the asia-pacific region contributing the largest share (FAO, 2026). India is second largest fish producing country and playing significant role in ensuring food security, employment generation and economic development. During a 2024-25, India's total fish production was estimated at 197.75 lakh tonnes, of which 147.37 lakh tonnes came from inland fisheries and aquaculture (PIB, 2025). Among crustaceans, whiteleg shrimp (*Penaeus vannamei*) is now the most widely cultured in various geographic locations due to its strong adaptability, delicious taste, rapid growth and high tolerance to various water salinities, high export value (Viet et al., 2022). The shrimp sector has changed dramatically in the past decade because of the high output potential of Pacific white shrimp (80% of global shrimp production) and increased disease resistance of the shrimp, a shortened time period for shrimp cultivation, lowered costs of production, and increased international demand for shrimp (ACAP, 2024). Its global production reaches 7.7 million tonnes in 2024, making it the most widely cultured crustacean worldwide (FAO, 2026).

High-quality seed, proper disease management, good water quality and appropriate aquafeed are the most critical factors for the success of aquaculture (Boyd et al., 2020; Kaleem and Sabi, 2020). In intensive culture systems, species are more vulnerable to diseases and stress; to mitigate intensifying stress, effective techniques must be used (Lumsangkul et al., 2022). Traditionally, antibiotics have been widely used in aquaculture to control disease. However, the use of antibiotics raises concern regarding overuse and misuse of antibiotics that may weaken the shrimp immune system and make them more vulnerable to infection. Additionally, antibiotic residue can accumulate in the aquatic environment, that poses risk of ecological balance and leading antibiotic resistance bacteria (Bondad-Reantaso et al., 2023). This challenge highlights a need of alternative approach that is beneficial for culture animal and also ensures the sustainability of aquaculture practices.

Alternative substance with low cost and high nutritional content is more viable and sustainable feed additive in aquaculture that promote organic approach in this sector (Dawood et al., 2022). Medicinal plants, exogenous enzymes such as pepsin, lipase, papain and other agriculture byproduct are the most appropriate supplements for aquafeed (Zheng et al., 2019). Many studies have demonstrated that using fruit waste and their bioactive compound in aquafeed enhance growth performance and feed utilization and also improves antioxidant activity

in aquatic animals (Dawood et al., 2022; Sulaiman et al., 2025). In this study orange, apple, papaya, and mango peel were used as feed additives because they are rich in bioactive compounds like polyphenols, flavonoids, pectin, dietary fibres, vitamins, minerals, essential oils, proteolytic enzymes and carotenoids that contribute to its nutritional value as aquafeed ingredient (Kang et al., 2010; Rafiq et al., 2018; Lebaka et al., 2021). Looking at the vast applications of fruit peels in several sectors such as pharmaceuticals, agriculture, and food processing, despite limited previous research having been done or reported in aquatic animals, given that research gap in aquaculture, a cost-effective feed additive study was conducted on orange, mango, apple, and papaya peel powder inclusion in the diet of *P. vannamei* to evaluate whether growth performance and feed utilization efficiency and antioxidant activity could be improved when *P. vannamei* is fed with selected fruit peel powder as a feed additive.

2. MATERIALS AND METHOD

2.1 Experimental animals

The experimental animals, *Penaeus vannamei,* were (post-larvae 12 specific pathogen free-SPF) were procured from a commercial hatchery - West Coast Shrimp Hatchery - Kotda, Kodinar, Gujarat, India, in 20 ppt saline water with sufficient oxygen level.

2.2 Experimental site and study period

The study was conducted in the wet laboratory of the Department of Aquaculture, College of Fisheries Science, Kamdhenu University, Veraval, Gujarat, India (20°56′21.6″N 70°21′09.2″E). A feeding trial of *P. vannamei* was carried out for 75 days (01st May to 14th July, 2025), followed by the assessment of various biological parameters of the experimental shrimp.

2.3 Preparation of fruit peels

All fruits were purchased from the local market and washed thoroughly with clean water and the peel was removed carefully by using a vegetable peeler. After that, the peels were cut in small pieces and dried it in a dryer at 50 ℃. Once it was dried, the peel was grounded in fine powder and stored in airtight containers.

2.4 Experimental design

Experimental *P. vannamei* post-larvae were reared in a 50 L plastic tank after an acclimation period. The experimental tank was filled with filtered 20 ppt saline water with optimum water parameters. A total of five treatments with four replications in a Completely Randomized Design (CRD) to achieve random allocation of treatment to tanks (2×1×1 feet). Shrimps were fed with their respective experimental diet at 07:30 hrs, 11:30 hrs and 18:30 hrs, three times a day. The feeding rate was set at 10% body weight of shrimp as initially (Tanjung et al., 2022).

2.5 Preparation of experimental diets

Five experimental diets were prepared, including the control (T0) diet without fruit peel powder inclusion and T1, T2, T3 and T4 diets containing 5% orange, apple, papaya and mango peel powders, respectively. All diets were prepared using the same basal formulation and processing conditions for comparative evaluation. Diets were formulated using the Pearson's square method to achieve the isonitrogenous composition with 35% crude protein. Basal ingredients were fish meal, wheat flour, soybean flour, tapioca powder, fish oil, sunflower oil and vitamin & mineral used. Based on the proximate analysis of the ingredients, they were weighed, thoroughly mixed, steam-cooked at 121°C and 15 psi for 10-15 min and then supplemented with the respective additives. After mixing, additives were pelletized, spread on a metallic tray and dried at 40-50°C for 5-6 h to obtain the final experimental diets.

2.6 Growth performance and feed utilization parameters

Growth measures like body weight gain, specific growth rate, feed conversion ratio, protein efficiency ratio, and feed efficiency ratio were recorded. Initial body weight of shrimp was calculated and at the end of feeding trial shrimps body weight was measured. Growth parameters were subsequently analyzed using the following formula:

Body weight gain = Final body weight – Initial body weight

$$\text{SGR (\%/day)} = \frac{\text{Ln(final body weight} - \text{initial body weight)}}{\text{Time of Experiment (75days)}} \times 100$$

Feed Conversion Ratio (FCR) = Feed intake / Weight gain

Protein Efficiency Ratio (PER) = Body weight gain (g) / Protein intake (g)

Feed Efficiency Ratio (FER) = Body weight gain (g) / Total feed consumption

$$\text{Survival rate (\%)} = \frac{\text{Total no. of survived shrimp (N)}}{\text{Total no. of initial shrimp (N)}} \times 100$$

2.7 Assessment of the antioxidant parameters

After the feeding trial completion, antioxidant parameters superoxide dismutase (SOD), catalase (CAT), and lipid peroxidation (malondialdehyde, MDA) were analyzed in the hepatopancreas tissues. Fresh tissues were homogenized in chilled 0.25 M sucrose solution at 8,000 to 10,000 rpm for 10-12 min at 4°C. The supernatant was collected and used for biochemical assays.

Parrilla-Taylor and Zenteno-Savin (2011) described the xanthine-xanthine oxidase method of Superoxide dismutase (SOD). Briefly, 1 g of tissue was homogenized in 2 mL potassium phosphate buffer and centrifuged at 10,000 rpm for 15 min at 4°C. The supernatant was mixed with sodium carbonate buffer, xanthine, NBT, EDTA, and xanthine oxidase. Absorbance was recorded at 560 nm at 30-s intervals for 5 min. SOD activity was expressed

as U/mg protein. Catalase activity was determined as follows, according to Aebi (1984). Tissue homogenate was prepared in potassium phosphate buffer and centrifuged at 10,000 rpm for 15 min at 4°C. Phosphate buffer, H_2O_2, and enzyme extract were the reaction mixture. The decrease in absorbance due to H_2O_2 decomposition was measured at 240 nm at 15-s intervals for 1-2 min. CAT activity was expressed as U/mg protein.

Ohkawa et al. (1979) TBARS method was followed for the lipid peroxidation determination. 1 g tissue was homogenized in crustacean isotonic solution and centrifuged at 3,000 rpm for 10 min at 4°C. The supernatant was reacted with HCl, trichloroacetic acid, and thiobarbituric acid, heated at 90°C for 10 min, cooled, and centrifuged. Absorbance was measured at 532 nm, and MDA level was calculated from a standard curve and expressed as nmol/mg protein.

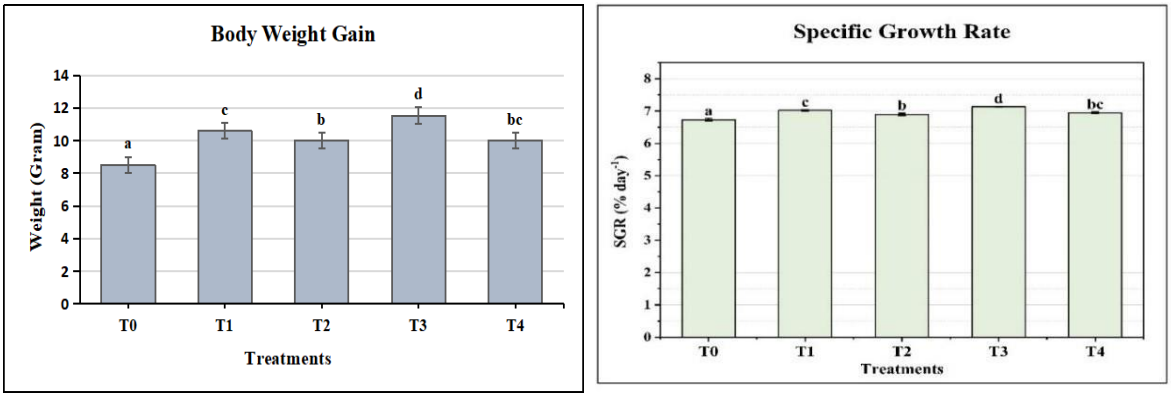
2.8 Data Analysis

Statistical analysis of the data was carried out using a Statistical Package for the Social Science (SPSS v.27.0). One-way ANOVA was applied to evaluate the effect of various experimental diets supplemented on growth performance, feed utilization efficiency and antioxidant activity, at a 0.05 level of significance, followed by Duncan's multiple comparisons to determine significant differences among the treatments (Snedecor and Cochran, 2014).

3. RESULTS

3.1 Growth performance

Growth performance was evaluated through body weight gain and specific growth rate recorded for 75-days. The results obtained were illustrated in Figure 1. At initiation of the experiment (0 days) the mean initial weight of shrimp was similar across all treatments (0.055 ± 0.004 g), indicating the homogeneity of experimental animals. After 75 days of feeding trial, shrimp in T3 treatment exhibited a significantly higher mean weight gain (11.537 ± 0.113 g) followed by T1 (10.587 ± 0.172 g) and T4 (10.015 ± 0.192 g), while T2 (9.645 ± 0.280 g) exhibited moderate growth. The lowest final mean weight gain was observed in the control group T0 (8.521 ± 0.232 g). Dietary supplementation of fruit peel powder significantly improved the specific growth rate of *Penaeus vannamei*. Results revealed that T3 (7.134 ± 0.013 % day^{-1}) had the maximum specific growth rate (SGR) followed by T1 (7.020 ± 0.022 % day^{-1}). Treatment T4 shows intermediate growth (6.946 ± 0.026 % day^{-1}) between T1 and T2. The SGR in T2 had moderate growth in shrimp 6.895 ± 0.039 % day^{-1} and the lowest was recorded in control treatment T0 (6.731 ± 0.037 % day^{-1}).



(a) **(b)**

FIGURE 1: Growth performance of *Penaeus vannamei* (a) Body weight gain (b) Specific growth rate

3.2 Feed utilization

Dietary supplementation with fruit peel powder significantly improved the feed utilization efficiency in *Penaeus vannamei*. The feed conversion ratio (FCR), feed efficiency ratio (FER), protein efficiency ratio (PER) are presented in Figure 2. Significant differences were observed in FCR among treatments ($p < 0.05$), with the lowest FCR recorded in the T3 treatment (1.294 ± 0.015), indicating superior feed utilization compared to other treatments. Then T1 with FCR of 1.572 ± 0.060. Treatments T4 and T2 had intermediate FCR of 1.787 ± 0.063 and 1.932 ± 0.092, respectively which were statistically similar to each other. Treatment T0 had the highest FCR value (2.170 ± 0.085), indicating the least feed utilization. The highest of FER was recorded in treatment T3 (0.773 ± 0.009), indicating better feed utilization efficiency by T1 (0.639 ± 0.023) among all the treatments. Treatment T4 shows a moderate FER value (0.562 ± 0.020) and treatment T2 exhibited 0.521 ± 0.026 and remained statistically comparable with T4 and T0. The lowest FER was observed in treatment T0 (0.463 ± 0.018). Protein efficiency ratio (PER), which indicates the efficiency of dietary protein efficiency into body mass, differed significantly among treatments ($p < 0.05$). T3 treatment (2.209 ± 0.02) exhibited the highest Protein efficiency ratio, followed by T1 treatment (1.825 ± 0.065). T4 treatment (1.605 ± 0.057) shows a moderate PER value;

treatment T2 exhibited 1.490 ± 0.073 and remained statistically comparable with T4 and T0. The lowest Protein efficiency ratio was observed in treatment T0 (1.323 ± 0.052). Overall, result indicates like in growth T3 treatment shows better feed utilization efficiency and least was recorded in control (T0) group.

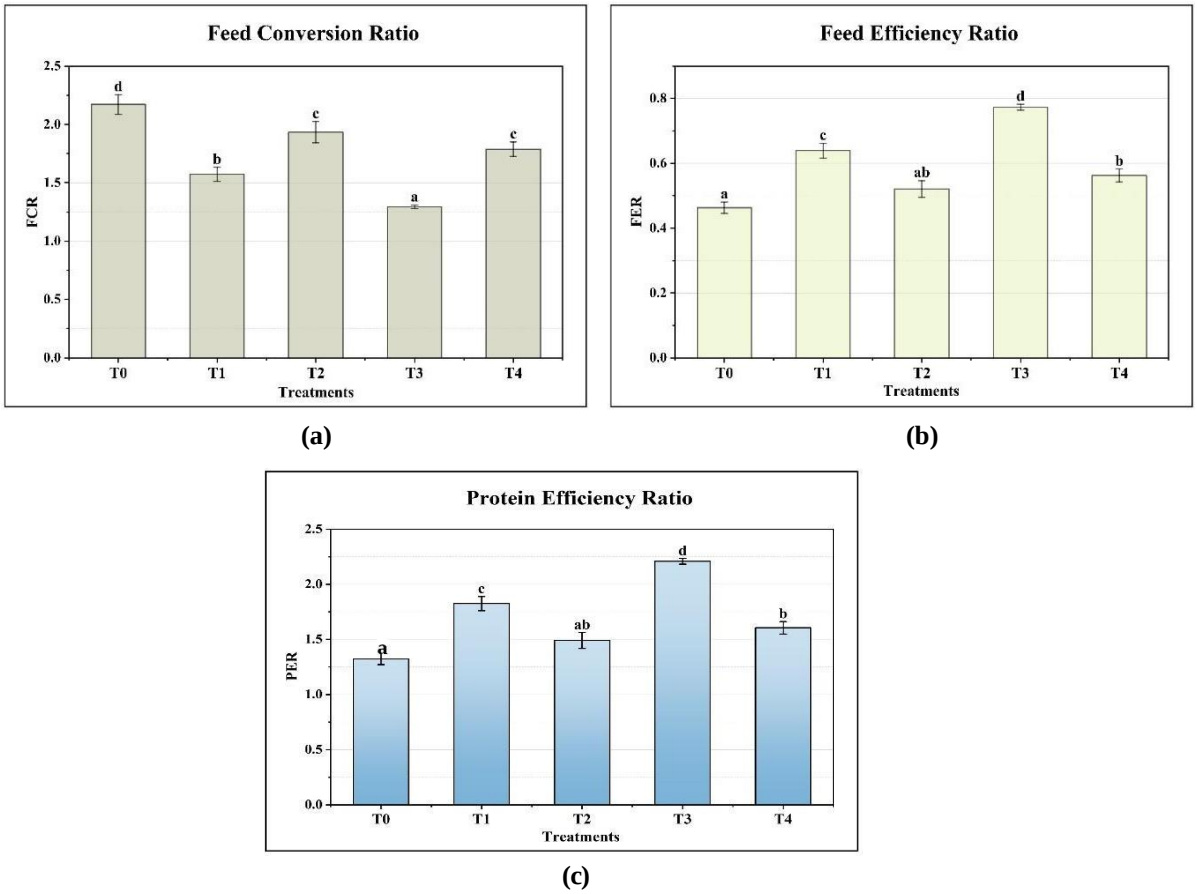


FIGURE 2: Feed utilization of *Penaeus vannamei* (a) Feed conversion ratio (FCR); (b) Feed efficiency ratio (FER); (c) Protein efficiency ratio (PER)

3.3 Survival rate

Survival rate of *Penaeus vannamei* at the end of the 75-day feeding trial is presented in Figure 3. The survival rate was highest in treatment T1 (95.00 ± 1.67%), which was statistically comparable to T3 (90.00 ± 1.92%), but was significantly higher than other treatments ($p < 0.05$). Treatment T4 had a moderate survival rate of 85.00 ± 3.19 % and was statistically similar with T2 (83.33 ± 3.33%), indicating an intermediate survival rate. The lowest survival (73.33 ± 2.72%) was observed in the control treatment (T0), which was significantly lower than all treatments.

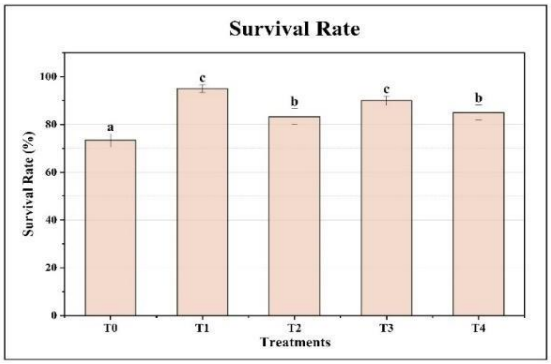


FIGURE 3: Survival rate of *Penaeus vannamei* in different treatments

3.4 Antioxidant enzyme activity

Superoxide dismutase (SOD) activity in *P. vannamei* was measured at the end of the experiment under various dietary treatments. The highest SOD activity was recorded in T1 treatment (11.285 ± 0.186 U/mg protein) among all treatments, followed by T3 (9.582 ± 0.131 U/mg protein) and T4 (8.854 ± 0.290 U/mg protein), which were statistically similar to each other. SOD activity was moderately observed in treatment T2 (7.261 ± 0.335 U/mg

protein), and the lowest activity was noted in T0 (6.110 ± 0.173 U/mg protein), indicating significant differences from the other treatments Figure 4(a).

The antioxidant activities in the hepatopancreas of *P. vannamei* are presented in Figure 4(b). Results showed that the fruit peel treatments showed notably higher antioxidant activities than the control, showing a significant difference. The maximum catalase (CAT) activity (26.994 ± 0.245 U/mg protein), was recorded for T1, which indicates a strong antioxidant response. This was followed by T3 (24.090 ± 0.064 U/mg protein). Treatment T4 obtained an intermediate catalase activity (23.301 ± 0.241 U/mg protein) and was statistically comparable with T3 and T2 treatments. The catalase activity of T2 was 22.579 ± 0.320 U/mg protein. The lowest catalase activity was found in the control group with 19.137 ± 0.365 U/mg protein, statistically significant compared to all treatments.

3.4.1 Antioxidant status (MDA level)

The finding of malondialdehyde (MDA) level was graphically presented in Figure 4(c), with a notably lower level observed in the fruit peel dietary treatment compared to the control and a significant difference. The lowest MDA level was recorded in treatment T1 (0.520 ± 0.029 nmol/mg protein) and the highest level in control T0 (1.377 ± 0.039 nmol/mg protein), showing better antioxidant protection and a statistically significant difference between them. Treatment T3 (0.851 ± 0.050 nmol/mg protein) and T4 (0.877 ± 0.009 nmol/mg protein) had lower and statistically similar MDA levels, while T2 recorded an intermediate level of MDA (1.036 ± 0.048 nmol/mg protein).

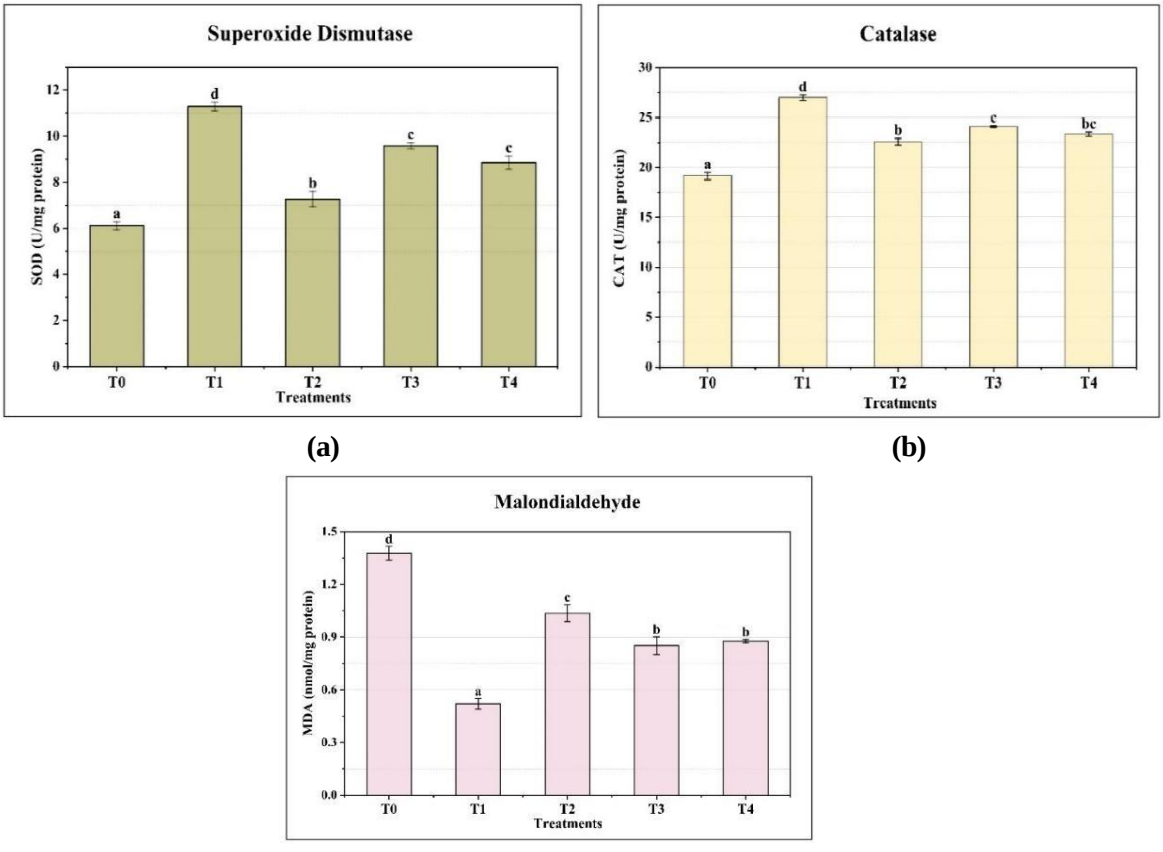


FIGURE 4: Antioxidant activity of *P. vannamei* recorded in different dietary fruit peel powder treatments. (a) Superoxide dismutase activity, (b) Catalase activity and (c) Malondialdehyde level.

4. DISCUSSION

4.1 Growth performance

The present study showed that the growth performance of *P. vannamei* was significantly improved by a diet supplemented with fruit peel powder (orange, apple, papaya, mango peel). The growth (in terms of weight gain and specific growth rate) of shrimp was significantly higher than that obtained in the control. Similar results have been reported in *P. vannamei* fed with a diet incorporated with papain and bromelain enzymes. Likewise, Aarumugam et al. (2013) reported higher weight gain and SGR in *Macrobrachium rosenbergii* fed diets incorporated with papaya peel. Similarly, dietary supplementation of papaya peel or papain resulted in increased growth and SGR in several aquatic species such as *Labeo rohita* (Sundararajan et al., 2024), *Macrobrachium rosenbergii* (Manush et al., 2013) and silver barb (Mahattanee et al., 2021) *Pangasius hypophthalmus*

(Rachmawati and Prihanto, 2019), and Juvenile *Acipenser ruthensus* (Wiszniewski et al., 2022). The improved growth recorded in papaya peel-fed groups could be due to the digestive enzyme papain present in papaya peel. Papain enzyme is active in hydrolysing plant proteins, so it can hydrolyse feed protein into simple molecules to improve digestion and absorption of feed by animals, thereby increasing growth (Choi et al., 2015; Sundararajan et al., 2024). Orange peel also has a growth-promoting effect on *P. vannamei*, although its effect is lower than papaya peel. A Similar growth-enhancing effect of orange peel is documented in previous studies. Ashry et al., (2023) found that *P. vannamei* fed with diets containing fermented orange peel exhibited significantly improved weight gain and SGR. Several studies highlighted that positive influence of orange peel derived feed additive on growth in various species. These species includes common carp (Hosseini et al., 2020), *Labeo catla* (Shabana et al., 2019), Mozmbique tilapia (Acar et al., 2015), Nile tilapia (*Oreochromis niloticus*) (Van Doan et al., 2018). The enhanced growth performance is due to the presence of bioactive compounds such as flavonoids, phenolics, pectin, essential oil and antioxidant in orange peel, which improve nutrient digestion, gut health and feed utilization efficiency in aquatic organisms.

Mango peel and apple peel also have a considerable effect on the growth performance of *P. vannamei.* Macusi et al. (2025) reported that moderate inclusion of mango peel meal improved growth performance in nile tilapia. These studies suggest that mango peel can improve fish growth but at moderate levels; but excessive inclusion reduces growth due to its fiber content and lower protein availability, which limits nutrient digestibility and bioavailability (Omojowo et al., 2012; Macusi et al., 2025). Similarly, Saravana Bhavan et al. (2013) demonstrated apple waste enhanced the growth performance of freshwater prawn *Macrobrachium rosenbergii*. Likewise, Qiang et al. (2019) reveals dietary inclusion of apple peel powder (APP) significantly influenced the growth of genetically improved farmed tilapia (*Oreochromis niloticus*). Several studies reported contrasting results like Pourmozaffar et al. (2017) and Jha et al. (2013) reported no significant improvement in growth performance in *P. vannamei* and snow trout (*Schizothorarax richardsonii*) fed apple- derived supplements. These studies suggest that apple peel do improve growth but it is more effectively as antioxidant and pigmentation rather than direct growth promoters. Overall, present study indicates papaya peel and orange peel poses a growth promoting effect on *P. vannamei* shrimp. Mango and apple also have growth-promoting effect but comparatively higher than control and lower than papaya peel and orange peel.

4.2 Feed utilization

The present study demonstrated that papaya peel supplementation poses highest feed utilization efficiency, in terms of feed conversion ratio (FCR), feed efficiency ratio (FER), protein efficiency ratio (PER) in *P. vannamei* followed by orange peel, mango and apple peel. The superior performance of papaya peel may be attributed to papain, a proteolytic enzyme that improves protein digestion and nutrient utilization. A similar finding is reported in *Macrobrachium rosenbergii* following papain and vitamin C supplementation by (Manush et al., 2013) and in *Pangasius hypophthalamus* (Rachmawati and Prihanto, 2019). Pimpimol et al. (2020) also observed improved PER and FER in channel catfish *(Ictalurus punctatus*) fed diets supplemented with dried papaya peel.

The present research shows that shrimp supplemented with orange peel significantly increase feed utilization efficiency. Similar observations were made by Ashry et al. (2023), who observed better FCR in *P. vannamei* shrimp fed fermented orange peel. Tonsy et al. (2019) also found that red tilapia (*Oreochromis mossabicus* × *O. niloticus*) exhibited better feed utilization efficiency when fed with orange peel waste. This improvement may be due to the phytochemicals (eg., Flavonoids, phenolics) present in orange peel, which increase digestive enzyme activity and nutrient absorption of feed. Many researchers have reported a decreased FCR in nile tilapia (Van Doan et al., 2018), *Oreochromis niloticus* (Doan et al., 2018) and in *Cyprinus Carpio* (Hosseini et al., 2020). Similarly, Shabana et al. (2019) also recorded better PER in Catla supplemented with *Citrus sinensis* extract-containing diet. Omojowo et al. (2012) reported that inclusion of mango peel meal in feed also improved feed utilization efficiency in *Oreochromis niloticus*. Likewise, Qiang et al. (2019) reported improved FCR in genetically improved farmed tilapia (*Oreochromis niloticus*) fed diets supplemented with apple peel powder. Similar result was also found by Saravana Bhavan et al. (2013) in *Macrobrachium rosenbergii*.

4.3 Survival rate

In the present study, dietary supplementation with orange peels enhanced the survival rates in *P. vannamei.* This outcome aligns with results of Ashry et al. (2023). Arisa et al. (2021) reported that the combination of papain and bromelain enzymes (derived from papaya and pineapple) positively affected survival in *P. vannamei* PL. Similar positive effects were observed in *Labeo rohita* fingerlings (Sundararajan et al., 2024). Macusi et al. (2025) observed improved survival rate in red tilapia fed with mango peel powder. Similar finding reported in nile tilapia (Outama et al., 2022). Saravana Bhavan et al. (2013) demonstrated that incorporating apple waste in the diet of *Macrobrachium rosenbergii* significantly improves survival rate. However, Qiang et al. (2019) observed that dietary supplementation with apple peel powder did not significantly influence the survival of genetically improved farmed tilapia (*Oreochromis niloticus*).

4.4 Antioxidant enzyme activity

Antioxidant are providing the protective effect against free radicals and reactive oxygen species (ROS) that ability of the fight against this compound due the available certain bioactive compounds in the present study fruit

peel that is a natural antioxidant (Vives-Bauza et al., 2007). In the present study, superoxide dismutase (SOD), catalase (CAT), and Malondialdehyde (MDA) level were evaluated, and the strongest protective effect was observed in the citrus fruit orange peel, followed by the papaya, mango peel, and the lowest effect was observed in the apple peel. The higher SOD and CAT enzyme activities, both with lower MDA levels, indicate preventing effect against cellular damage from free radicals. Similar findings were reported by Eslami et al. (2025), who demonstrated that dietary lemon peel extract significantly increased SOD and CAT activities in *P. vannamei*, particularly at 0.5% and 1% supplementation, indicating enhanced antioxidant defence. Vicente et al. (2019) also reported increased SOD activity in Nile tilapia fed orange peel, while Salem et al. (2019) observed improved antioxidant responses in gilthead sea bream supplemented with orange peel. Furthermore, Cai et al. (2025) reported that mango leaf supplementation increased SOD and CAT activities and reduced MDA levels in *P. vannamei*, suggesting reduced oxidative damage. Similarly, Yang et al. (2020) found that apple polyphenols enhanced SOD and CAT activities and reduced MDA concentration in grass carp. The present findings therefore suggest that fruit peel supplementation, particularly orange peel, can strengthen the antioxidant defence system of shrimp, probably through the synergistic action of its phenolic and flavonoid compounds, thereby improving the ability of *P. vannamei* to neutralize ROS and protect cellular membranes from oxidative damage.

5. CONCLUSION

The present study demonstrated that dietary supplementation of fruit peel powders enhanced the growth performance, survival and feed utilization efficiency of *P. vannamei*. Papaya peel is most effective supplement for growth and feed utilization efficiency, whereas orange peel was superior for antioxidant activity and survival. Mango and apple peel also improved shrimp performance but at a moderate level and comparatively higher than the control treatment. The finding of this study indicates that fruit peel can serve as sustainable phytogenic feed additives, and their utilization may provide alternative to synthetic growth promoters in shrimp aquaculture. Further studies should evaluate different inclusion levels of fruit peel powders to clarify dose-dependent effects and both higher and lower concentration that establish the optimum level of fruit peel powders for sustainable shrimp production.

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ETHICAL APPROVAL

All applicable international, national and/or institutional guidelines for the care and use of animals were followed in this study.

CONFLICT OF INTEREST

The author declares no conflict of interest.

AUTHORS' CONTRIBUTION

Conceptualisation: DCT, KHV; Methodology & Experimental Design: DCT, KHV, RVC; Data analysis: DCT, RVC, KDP; Manuscript preparation: DCT, JMT; Review & Editing: KHV, RMD, RVC, KDP.

DATA AVAILABILITY STATEMENT

The data supporting the findings of this study are available within the article.

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