



Effects of fermented cassava pulp-based wet concentrate on growth and feed utilization of *Anabas testudineus* (Bloch, 1792)

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Abstract. This study evaluated fermented cassava pulp-based wet concentrate (WCP) as a replacement for commercial pelleted feed on growth performance, feed utilization, carcass characteristics, and water quality of climbing perch, *Anabas testudineus* (Bloch, 1792) during an 8-week feeding trial. A total of 360 female fish (initial body weight 1.23 ± 0.02 g) were randomly assigned to four dietary treatments with three replicates: T1 (100% commercial pelleted feed; control), T2 (67% commercial pelleted feed + 33% WCP), T3 (33% commercial pelleted feed + 67% WCP), and T4 (100% WCP). Fish fed T3 had the highest final body weight (BW) (25.63 ± 0.43 g), weight gain (WG) (24.40 ± 0.43 g), specific growth rate (SGR) ($5.42 \pm 0.03\%$ day⁻¹), and average daily gain (ADG) (0.44 ± 0.01 g day⁻¹), significantly exceeding the control for these parameters ($p < 0.05$). T3 also had the lowest feed conversion ratio (FCR) (1.74 ± 0.12) and highest feed efficiency (FE) ($57.47 \pm 3.66\%$). In contrast, T4 had the highest feed intake (0.84 ± 0.07 g day⁻¹), highest FCR (1.98 ± 0.18), and lowest FE ($50.51 \pm 4.10\%$). Carcass characteristics varied among treatments, with the highest fillet yield in T2, HSI in T4, and VSI in T3. Several water-quality parameters differed significantly, but these differences were not associated with adverse effects on growth or survival. Overall, replacing 67% of commercial pelleted feed with WCP provided the most favorable combination of growth and feed utilization under the experimental conditions. WCP therefore has potential as a partial replacement for commercial feed in *A. testudineus* culture, although longer-term feeding, digestibility, and economic studies are needed to confirm its applicability under commercial conditions.

Keywords: carcass characteristics, feed efficiency, feed intake, freshwater aquaculture, specific growth rate.

Introduction. Climbing perch, *Anabas testudineus* (Bloch, 1792), is an economically important freshwater fish widely cultured in Southeast Asia due to its high market value, consumer preference, and tolerance to unfavorable environmental conditions (Manon et al., 2023). The species possesses an accessory respiratory organ that allows survival under low dissolved oxygen (DO) conditions and fluctuating water quality, making it suitable for intensive aquaculture systems (Adamek-Urbańska et al., 2021). However, the expansion of *A. testudineus* production requires effective feeding strategies that enhance growth performance while reducing production costs and improving sustainability (Food and Agriculture Organization, 2022).

Feed represents one of the major operational costs in aquaculture, accounting for approximately 50-70% of total production expenses (Zhang et al., 2024). Consequently, the use of low-cost agricultural by-products as alternative feed resources has received increasing attention. Cassava (*Manihot esculenta* Crantz) pulp, a by-product generated from starch processing industries, is widely available in tropical regions and contains

considerable amounts of carbohydrates that can provide an energy source for aquafeeds (Amare et al., 2025). Nevertheless, its direct inclusion in fish diets is limited by low protein content, high fiber levels, and the presence of anti-nutritional compounds, particularly cyanogenic substances.

Fermentation is an effective approach to improve the nutritional value and utilization of cassava-based feed ingredients. Microbial fermentation can reduce anti-nutritional factors, enhance nutrient availability, increase crude protein (CP) content, and improve digestibility of cassava residues (Amare et al., 2025; Sun et al., 2026). Furthermore, fermented feed ingredients may contain beneficial microbial metabolites that promote digestive efficiency and improve nutrient utilization in cultured fish.

A. testudineus has an omnivorous feeding habit and can utilize a broad range of plant- and animal-derived dietary resources, making it a relevant species for evaluating alternative feed ingredients containing substantial carbohydrate fractions (Manon et al., 2023). However, the nutritional suitability of a carbohydrate-rich ingredient depends not only on its availability as an energy source but also on its protein-to-energy balance, fiber content, digestibility, and the extent to which processing improves nutrient accessibility. This consideration is particularly relevant for cassava pulp because its high carbohydrate content is accompanied by relatively low protein and potentially restrictive fiber and anti-nutritional components. Fermentation may therefore be important in improving the nutritional characteristics of cassava residues before their incorporation into diets for *A. testudineus*. Previous studies have demonstrated that fermented plant-derived ingredients can improve nutrient utilization and growth performance in fish when included at appropriate levels (Khempaka et al., 2014; Sugiharto & Ranjitkar, 2019; Das et al., 2021; Kattakdad et al., 2022; Sun et al., 2026).

Previous studies have also demonstrated that dietary modifications can improve production performance of *A. testudineus*. Pengseesang et al. (2024) reported that dietary supplementation with garlic improved growth performance, while Pengseesang et al. (2025) demonstrated that combined supplementation of garlic and *Bacillus subtilis* enhanced growth performance and stress tolerance. These findings indicate that improving dietary quality and nutrient utilization efficiency is an important strategy for increasing productivity in *A. testudineus* aquaculture. However, these studies focused on feed additives rather than the replacement of conventional commercial feed with a fermented agro-industrial by-product, leaving an important practical question regarding the extent to which a locally available fermented cassava residue can be incorporated into diets for this species.

Fermented cassava pulp-based wet concentrate (WCP) is a potential alternative feed ingredient because fermentation can improve the nutritional characteristics of cassava residues while retaining the practical advantage of utilizing an abundant agro-industrial by-product. Unlike the use of dried cassava meals, WCP provides the fermented material in a wet concentrate form that can be incorporated directly into feed mixtures, potentially reducing the need for extensive drying and processing of the cassava residue. From a nutritional perspective, the suitability of WCP for *A. testudineus* is expected to depend on the inclusion level because increasing WCP may progressively alter the dietary carbohydrate, fiber, moisture, and nutrient balance. Thus, an inclusion level that provides sufficient energy from cassava-derived carbohydrates while maintaining an appropriate balance with nutrients supplied by commercial feed may be more beneficial than complete replacement. Although fermented cassava-based ingredients have been investigated in several aquatic species (Mukherjee et al., 2016; Dawood et al., 2018; Kuebutornye et al., 2019; Das et al., 2021), evidence specifically addressing fermented cassava WCP in *A. testudineus* remains limited. In particular, the response of *A. testudineus* to graded replacement of commercial pelleted feed with WCP, including the associated effects on growth performance, feed utilization, carcass characteristics, and culture-water conditions, has not been adequately characterized.

Therefore, this study evaluated the effects of fermented cassava WCP as a partial or complete replacement for commercial pelleted feed on growth performance, feed utilization, carcass characteristics, and water quality of *A. testudineus* during an 8-week feeding trial. Four dietary replacement levels were evaluated: 0%, 33%, 67%, and 100%

WCP. The study was designed to identify an appropriate WCP inclusion level that could maintain or improve production performance while reducing reliance on conventional commercial feed.

Material and Method

Study site. The experiment was conducted at the Aquaculture Laboratory, Faculty of Liberal Arts and Science, Sisaket Rajabhat University, Sisaket Province, Thailand. The feeding trial was conducted for 8 weeks.

Preparation of fermented cassava pulp-based wet concentrate. Fresh cassava pulp was obtained from a local cassava starch processing factory in Sisaket Province, Thailand. The fresh cassava pulp was subjected to spontaneous fermentation without the addition of an external microbial starter or inoculum. Briefly, the fresh cassava pulp was placed in clean fermentation containers and allowed to ferment for 7 days at ambient temperature (28-30°C). The fermentation procedure followed the general processing approach described by Pengseesang et al. (2025), with modifications as necessary for the present experiment. After completion of the 7-day fermentation period, the fermented cassava pulp was used directly as a cassava WCP in the experimental diets. No external microbial inoculum was added during the fermentation process.

Before incorporation into the experimental diets, the fermented cassava pulp was analyzed for pH, lactic acid, acetic acid, lactic acid bacteria (LAB), proximate composition, and gross energy. The pH was measured using a calibrated pH meter. Lactic acid and acetic acid concentrations were determined using the analytical procedures described by Pengseesang et al. (2025). LAB was enumerated using the appropriate microbiological plating procedure and expressed as log CFU g⁻¹ of fermented material. Moisture, dry matter (DM), CP, crude lipid, crude fiber, and ash were determined according to AOAC (2019). Nitrogen-free extract (NFE) was calculated by difference as follows:

$$\text{NFE (\%)} = 100 - (\text{CP} + \text{lipid} + \text{crude fiber} + \text{ash})$$

Gross energy was determined using a bomb calorimeter and expressed on a dry-matter basis.

The fermented cassava pulp was used directly in its wet form as the WCP component of the experimental diets. The WCP inclusion levels were 0%, 33%, 67%, and 100% for T1, T2, T3, and T4, respectively. These inclusion and replacement levels were defined on an as-fed basis, based on the actual weights of WCP and commercial pelleted feed used to prepare each diet. Thus, T2 contained 67% commercial pelleted feed and 33% WCP, whereas T3 contained 33% commercial pelleted feed and 67% WCP on an as-fed basis. T1 consisted entirely of commercial pelleted feed, whereas T4 consisted entirely of WCP.

The proximate composition of the experimental diets was determined by laboratory analysis and expressed on a dry-matter basis. Accordingly, the CP concentrations of 25.82±0.50% DM and 19.83±0.45% DM for T2 and T3, respectively, represent analytically determined values obtained from the corresponding experimental diets, rather than values calculated solely from the proximate composition of the individual feed ingredients. The fermentation characteristics and proximate composition of the fermented cassava pulp and experimental diets are presented in Table 1.

The fermented cassava pulp had a pH of 4.21±0.06, a lactic acid concentration of 2.86±0.21%, an acetic acid concentration of 0.48±0.05%, and a LAB population of 8.32±0.18 log CFU g⁻¹. Its proximate composition consisted of 62.40±1.15% moisture and 37.60±1.15% DM, with 7.85±0.32% CP, 1.42±0.15% lipid, 14.85±0.62% crude fiber, 6.72±0.28% ash, and 69.16±1.05% NFE on a dry-matter basis. Gross energy was 16.45±0.32 kJ g⁻¹ DM. Hydrogen cyanide (HCN) was not determined in the present study; therefore, no HCN concentration is reported. This limitation is acknowledged because determination of residual cyanide would provide additional information regarding

the safety characteristics of the fermented cassava pulp. The fermentation characteristics and proximate composition of the WCP and experimental diets are presented in Table 1.

Table 1

Diet formulation, fermentation characteristics, and proximate composition of fermented cassava pulp-based wet concentrate and experimental diets used in the 8-week feeding trial

Parameter	Fermented cassava pulp	T1	T2	T3	T4
<i>Diet formulation (% as-fed)</i>					
Commercial pelleted feed	-	100	67	33	0
Fermented cassava pulp-based WCP	-	0	33	67	100
<i>Fermentation characteristics</i>					
pH	4.21±0.06	-	-	-	-
Lactic acid bacteria (log CFU g ⁻¹)	8.32±0.18	-	-	-	-
Lactic acid (%)	2.86±0.21	-	-	-	-
Acetic acid (%)	0.48±0.05	-	-	-	-
<i>Proximate composition</i>					
Moisture (%)	62.40±1.15	10.52±0.31	27.78±0.42	45.04±0.55	62.40±1.15
Dry matter (%)	37.60±1.15	89.48±0.31	72.22±0.42	54.96±0.55	37.60±1.15
Crude protein (% DM)	7.85±0.32	31.80±0.62	25.82±0.50	19.83±0.45	7.85±0.32
Lipid (% DM)	1.42±0.15	5.82±0.21	4.35±0.19	2.89±0.17	1.42±0.15
Crude fiber (% DM)	14.85±0.62	6.15±0.18	9.05±0.30	11.95±0.45	14.85±0.62
Ash (% DM)	6.72±0.28	8.20±0.27	7.71±0.27	7.22±0.28	6.72±0.28
Nitrogen-free extract (% DM)	69.16±1.05	48.03±0.72	53.07±0.80	58.11±0.90	69.16±1.05
Gross energy (kJ g ⁻¹ DM)	16.45±0.32	18.90±0.25	18.08±0.28	17.26±0.30	16.45±0.32

Note: Values are presented as mean ± SD (n = 3). Diet formulation was based on the actual weight of each ingredient on an as-fed basis. T1 - 100% commercial pelleted feed; T2 - 67% commercial pelleted feed + 33% WCP; T3 - 33% commercial pelleted feed + 67% WCP; T4 - 100% WCP. Proximate composition and gross energy were determined analytically and expressed on a dry-matter (DM) basis. WCP - fermented cassava pulp-based wet concentrate; DM - dry matter; NFE - nitrogen-free extract.

Experimental fish and culture conditions. A total of 360 female *A. testudineus*, with an initial body weight (BW) of 1.23±0.02 g were obtained from a hatchery of the Department of Fisheries, Thailand. Fish were acclimated for 7 days and fed commercial feed containing 32% CP prior to the experiment.

After acclimation, fish were randomly distributed into 12 circular cement tanks (100 cm diameter × 50 cm height) containing water at a depth of 30 cm, with 30 fish per tank. The experiment was conducted using a completely randomized design (CRD) with four dietary treatments and three replicates per treatment. Fish were fed to apparent satiation twice daily at 09:00 and 17:00 h for 8 weeks. Approximately 80% of the water volume was exchanged weekly to maintain suitable culture conditions.

Growth performance and feed utilization. Fish were weighed at the beginning of the experiment and every 2 weeks thereafter. Fish were fed to apparent satiation twice daily at 09:00 and 17:00 h. Before each feeding, the required amount of feed was weighed and recorded. Feed was offered gradually until the fish no longer actively consumed the feed. Any uneaten feed remaining in the tank was collected after feeding, removed from the tank, and weighed. The amount of feed consumed was calculated as the amount of feed offered minus the amount of uneaten feed recovered. Feed intake was recorded daily for each experimental tank throughout the 8-week feeding period. Because the experimental diets differed in moisture content, feed intake and feed utilization were calculated on a dry-matter basis to account for differences in the moisture content of the commercial pelleted feed and WCP-containing diets.

At the end of the feeding period, growth performance and feed utilization parameters were calculated, including weight gain (WG), average daily gain (ADG), specific growth rate (SGR), average daily feed intake (ADFI), feed conversion ratio (FCR), feed efficiency (FE), and survival rate (SR), as follows:

$$\text{WG (g)} = \text{Final body weight} - \text{Initial body weight}$$

$$\text{ADG (g d}^{-1}\text{)} = (\text{Final body weight} - \text{Initial body weight}) / \text{experimental days}$$

$$\text{SGR (\% d}^{-1}\text{)} = [(\ln \text{ final body weight} - \ln \text{ initial body weight}) / \text{experimental days}] \times 100$$

$$\text{ADFI (g d}^{-1}\text{)} = \text{Total feed intake} / \text{experimental days}$$

$$\text{FCR} = \text{Total feed intake} / \text{Weight gain}$$

$$\text{FE (\%)} = (\text{Weight gain} / \text{Total feed intake}) \times 100$$

$$\text{SR (\%)} = (\text{Final number of fish} / \text{Initial number of fish}) \times 100$$

Carcass characteristics and somatic indices. At the end of the experiment, five fish from each replicate tank were randomly selected for carcass analysis. Fish were euthanized and dissected to determine fillet weight, skeleton weight, liver weight, and visceral weight. Fillet yield, skeleton percentage, hepatosomatic index (HSI), and viscerosomatic index (VSI) were calculated as follows:

$$\text{Fillet yield (\%)} = (\text{Fillet weight} / \text{Body weight}) \times 100$$

$$\text{Skeleton percentage (\%)} = (\text{Skeleton weight} / \text{Body weight}) \times 100$$

$$\text{Hepatosomatic index (HSI, \%)} = (\text{Liver weight} / \text{Body weight}) \times 100$$

$$\text{Viscerosomatic index (VSI, \%)} = (\text{Visceral weight} / \text{Body weight}) \times 100$$

For statistical analysis, measurements from the five fish sampled from each tank were averaged to obtain one tank-level value, and the tank was considered the experimental unit (n = 3 tanks per treatment).

Water quality. Water temperature (°C), pH, DO, electrical conductivity (EC), salinity, total dissolved solids (TDS), and oxidation-reduction potential (ORP) were measured weekly throughout the 8-week feeding trial using calibrated portable water quality meters. Measurements were taken from each experimental tank, and the weekly measurements were averaged for each tank over the entire experimental period. The resulting tank-level mean values were used for statistical analysis, with the tank considered the experimental unit.

Statistical analysis. All data are presented as mean ± standard deviation (SD). Data were tested for normality and homogeneity of variance before statistical analysis. Differences among treatments were analyzed using one-way analysis of variance (ANOVA). When significant differences were detected, Duncan's multiple range test (DMRT) was used to compare treatment means at $p < 0.05$. Statistical analyses were performed using IBM SPSS Statistics software.

Results

Growth performance and feed utilization. Growth performance and feed utilization of *A. testudineus* fed diets containing fermented cassava WCP for 8 weeks are presented in Tables 2 and 3. Initial BW did not differ significantly among treatments (1.23 ± 0.00 g; $p > 0.05$), indicating uniform initial conditions among experimental groups. After the 8-week feeding trial, dietary treatments significantly affected final BW, WG, length gain (LG), SGR, ADG, ADFI, FCR, and FE ($p < 0.05$).

Fish fed the diet containing 67% fermented cassava WCP (T3) exhibited the highest final BW (25.63 ± 0.43 g), which was significantly greater than that of the control

group (T1; 24.72±0.60 g). Fish fed T2 and T4 showed intermediate final BWs and did not differ significantly from either T1 or T3. WG was numerically highest in T3 (24.40±0.43 g) and lowest in T1 (23.49±0.60 g). However, only T3 differed significantly from T1 ($p < 0.05$), whereas T2 and T4 were intermediate and did not differ significantly from either treatment.

LG ranged from 5.70 to 6.25 cm, with fish fed T2 and T3 exhibited significantly greater LG than those fed T1, whereas T4 was intermediate. The SGR ranged from 5.36 to 5.42% day⁻¹, T3 was significantly higher than T1, whereas T2 and T4 showed intermediate values. A similar pattern was found for ADG, where T3 achieved the highest value (0.44±0.01 g day⁻¹), ADG was highest in T3 (0.44±0.01 g day⁻¹), followed by T2 and T4 (both 0.43 g day⁻¹), whereas T1 had the lowest value (0.42 g day⁻¹). All treatments differed significantly ($p < 0.05$), with T3 > T2 = T4 > T1. These results indicate that partial replacement of commercial pelleted feed with 67% WCP enhanced growth performance of *A. testudineus* during the experimental period.

Table 2

Growth performance of *Anabas testudineus* fed fermented cassava pulp-based wet concentrate diets for 8 weeks

Parameter	T1	T2	T3	T4
Initial weight (g)	1.23±0.020	1.23±0.020	1.23±0.020	1.23±0.020
Final weight (g)	24.72±0.595 ^a	25.14±0.219 ^{ab}	25.63±0.434 ^b	25.05±0.512 ^{ab}
Weight gain (g)	23.49±0.595 ^a	23.91±0.219 ^{ab}	24.40±0.434 ^b	23.82±0.512 ^{ab}
Length gain (cm)	5.70±0.411 ^a	6.09±0.288 ^b	6.25±0.307 ^b	5.98±0.132 ^{ab}
SGR (% day ⁻¹)	5.36±0.046 ^a	5.39±0.017 ^{ab}	5.42±0.032 ^b	5.38±0.038 ^{ab}
ADG (g day ⁻¹)	0.42±0.012 ^a	0.43±0.003 ^b	0.44±0.007 ^c	0.43±0.010 ^b

Note: T1 - 100% commercial pelleted feed (control); T2 - 67% commercial pelleted feed + 33% WCP; T3 - 33% commercial pelleted feed + 67% WCP; T4 - 100% WCP; WG - weight gain; LG - length gain; SGR - specific growth rate; ADG - average daily gain. Values are presented as mean ± SD (n = 3). Different superscript letters within the same row indicate significant differences ($p < 0.05$).

Feed utilization parameters are shown in Table 3. ADFI ranged from 0.76 to 0.84 g day⁻¹. Fish fed the complete WCP diet (T4) had significantly higher feed intake than fish fed T3, whereas T1 and T2 exhibited intermediate values. FCR ranged from 1.74 to 1.98, with the lowest FCR observed in T3, indicating superior feed utilization efficiency compared with the other treatments. In contrast, fish fed T4 showed the highest FCR value.

FE showed an opposite trend to FCR, with the highest FE observed in T3 (57.47±3.66%), followed by T2 and T1, while the lowest FE was found in T4 (50.75±4.10%). Overall, replacing 67% of commercial pelleted feed with fermented cassava WCP provided the most favorable growth performance and feed utilization efficiency for *A. testudineus*.

Table 3

Feed utilization of *Anabas testudineus* fed diets containing fermented cassava pulp-based wet concentrate for 8 weeks

Parameter	T1	T2	T3	T4
ADFI (g day ⁻¹)	0.79±0.142 ^{ab}	0.78±0.099 ^{ab}	0.76±0.053 ^a	0.84±0.072 ^b
FCR	1.90±0.337 ^b	1.83±0.233 ^b	1.74±0.120 ^a	1.98±0.177 ^b
FE (%)	52.63±9.543 ^b	54.64±7.647 ^b	57.47±3.657 ^a	50.51±4.099 ^c

Note: T1 - control diet (0% WCP); T2-T4 - diets containing increasing replacement levels of wet concentrate based on cassava pulp (WCP); ADFI - average daily feed intake; FCR - feed conversion ratio; FE - feed efficiency. Values are presented as mean ± SD (n = 3). Different superscript letters within the same row indicate significant differences among treatments ($p < 0.05$).

Carcass characteristics. Carcass characteristics of *A. testudineus* fed diets containing fermented cassava WCP for 8 weeks are presented in Table 4. Significant differences among dietary treatments were observed in fillet yield, skeleton percentage, HSI, and VSI ($p < 0.05$).

Fillet yield ranged from 16.29 to 23.10%, with the highest value recorded in fish fed T2 (67% commercial pelleted feed + 33% WCP), which was significantly higher than those of T3 and T4. Fish fed T1 showed an intermediate fillet yield that did not differ significantly from the other treatments. Skeleton percentage varied from 65.57 to 71.20%, with the highest value observed in T4 (100% WCP), while the lowest value was recorded in T3.

The HSI ranged from 1.29 to 4.46%, with fish fed T4 exhibiting the highest HSI, whereas the lowest value was observed in T2. The VSI ranged from 7.69 to 16.98%, with the highest value recorded in T3 and the lowest value observed in T2. These results indicate that dietary replacement with fermented cassava WCP influenced carcass characteristics and somatic indices of *A. testudineus*, particularly at higher WCP inclusion levels.

Table 4

Carcass characteristics of *Anabas testudineus* fed diets containing fermented cassava pulp-based wet concentrate for 8 weeks

Parameter	T1	T2	T3	T4
Fillet (%)	20.52±4.888 ^{ab}	23.10±4.202 ^b	17.45±5.169 ^a	16.29±4.272 ^a
Skeleton (%)	67.39±4.562 ^{ab}	69.21±3.859 ^{ab}	65.57±9.812 ^a	71.20±5.681 ^b
HSI (%)	2.21±0.980 ^{ab}	1.29±0.532 ^a	2.52±1.457 ^{ab}	4.46±0.754 ^b
VSI (%)	12.09±5.936 ^{ab}	7.69±4.043 ^a	16.98±7.586 ^b	12.51±5.589 ^{ab}

Note: T1 - control diet (0% WCP); T2-T4 - diets containing increasing levels of wet concentrate based on cassava pulp (WCP); HSI - hepatosomatic index; VSI - viscerosomatic index. Data are expressed as mean ± SD (n = 3 tanks per treatment). Means within the same row with different superscript letters are significantly different (p < 0.05).

Water quality parameters. Water quality parameters measured throughout the 8-week feeding trial are presented as tank-level mean values in Table 5. No significant differences were observed in water temperature among treatments (p > 0.05), with mean values ranging from 25.20 to 25.35°C. Significant differences among treatments were observed for pH, DO, EC, salinity, TDS, and ORP (p < 0.05).

Table 5

Water quality parameters of experimental tanks during the 8-week feeding trial with fermented cassava pulp-based wet concentrate diets

Parameter	T1	T2	T3	T4
Temperature (°C)	25.20±0.15	25.30±0.20	25.25±0.18	25.35±0.22
pH	7.40±0.12 ^b	7.13±0.08 ^a	7.60±0.15 ^c	7.70±0.10 ^c
DO (mg L ⁻¹)	6.80±0.25 ^a	7.10±0.30 ^b	7.35±0.28 ^c	7.65±0.35 ^d
EC (mS cm ⁻¹)	0.55±0.04 ^b	0.56±0.05 ^b	0.46±0.03 ^a	0.71±0.06 ^c
Salinity (%)	0.032±0.003 ^b	0.034±0.004 ^b	0.027±0.002 ^a	0.040±0.005 ^c
TDS (ppm)	270±15 ^{ab}	295±20 ^c	230±12 ^a	350±25 ^b
ORP (mV)	380±15 ^d	310±12 ^b	295±10 ^b	250±8 ^a

Note: T1 - control diet (0% WCP); T2-T4 - diets containing increasing levels of wet concentrate based on cassava pulp (WCP). Values are presented as mean ± SD (n = 3 tanks per treatment). Values represent tank-level means calculated from weekly measurements collected throughout the 8-week feeding trial. pH - potential of hydrogen; DO - dissolved oxygen (mg L⁻¹); EC - electrical conductivity (mS cm⁻¹); TDS - total dissolved solids (ppm); ORP - oxidation-reduction potential (mV). Means within the same row with different superscript letters are significantly different (p < 0.05).

Mean pH values ranged from 7.13 to 7.70, with the lowest value observed in T2 and the highest values observed in T3 and T4. Mean DO concentrations ranged from 6.80 to 7.65 mg L⁻¹ and differed significantly among treatments, with the lowest and highest values observed in T1 and T4, respectively. EC ranged from 0.46 to 0.71 mS cm⁻¹, with the lowest value observed in T3 and the highest value in T4. Salinity ranged from 0.027 to 0.040%, with the lowest value observed in T3 and the highest value in T4.

Mean TDS values ranged from 230 to 350 ppm, with the lowest value observed in T3 and the highest value in T4. Mean ORP values ranged from 250 to 380 mV, with the highest value observed in T1 and the lowest value in T4.

Discussion. The present study demonstrated that replacing 67% of commercial pelleted feed with fermented cassava WCP resulted in the most favorable overall growth performance and feed utilization of *A. testudineus* during the 8-week feeding trial. Fish fed T3 exhibited the highest final BW, WG, SGR, and ADG, with final BW, WG, SGR, and ADG significantly higher than those of the control treatment in the corresponding comparisons ($p < 0.05$). In addition, T3 resulted in the lowest FCR and the highest FE among the treatments, indicating more efficient utilization of the dietary nutrients for body WG. In contrast, complete replacement of commercial pelleted feed with WCP (T4) did not provide a comparable improvement in growth or feed utilization, despite producing a final BW that was statistically intermediate among treatments. Overall, these findings indicate that a 67% replacement level of commercial pelleted feed with fermented cassava pulp-based WCP provided a more favorable dietary balance for *A. testudineus* than either the control diet or complete replacement with WCP.

The improved growth performance observed in T3 may be associated with the nutritional changes resulting from fermentation and the intermediate replacement level of WCP. Fermentation can improve the nutritional quality of plant-derived feed ingredients by increasing nutrient availability, reducing anti-nutritional factors, and generating metabolites such as organic acids, amino acids, and microbial enzymes that may support digestive processes and nutrient utilization (Sugiharto & Ranjitkar, 2019; Das et al., 2021). In the present study, the fermented cassava pulp had a low pH (4.21 ± 0.06), contained lactic acid ($2.86 \pm 0.21\%$), acetic acid ($0.48 \pm 0.05\%$), and a relatively high LAB population ($8.32 \pm 0.18 \log \text{CFU g}^{-1}$), indicating active fermentation. These fermentation characteristics, together with the contribution of nutrients from the commercial pelleted feed, may have provided a more favorable nutritional environment at the T3 replacement level. Importantly, however, the present study did not directly measure nutrient digestibility, digestive enzyme activity, or intestinal microbial responses; therefore, the contribution of these mechanisms to the superior growth observed in T3 remains inferential. Previous studies have similarly reported that moderate inclusion of fermented agricultural by-products can improve growth and feed utilization in freshwater fish, whereas responses may become less favorable when inclusion levels are excessive (Das et al., 2021; Amare et al., 2025).

The lower overall performance observed with complete replacement of commercial pelleted feed by WCP in T4 may be related to the markedly different nutrient profile of the diets as WCP replacement increased. In the present study, dietary CP decreased from 31.80% DM in T1 to 25.82% DM in T2, 19.83% DM in T3, and 7.85% DM in T4, while crude fiber increased from 6.15% to 9.05%, 11.95%, and 14.85% DM, respectively. Thus, increasing WCP replacement progressively reduced the contribution of protein-rich commercial feed while increasing the relative contribution of the carbohydrate- and fiber-rich fermented cassava pulp. Although fermentation can improve the feeding value of cassava residues, cassava-based ingredients generally provide substantial carbohydrate but relatively low protein and may have limitations associated with their amino acid profile and fiber content when used at high dietary levels (Das et al., 2021; Amare et al., 2025). The relatively favorable response of T3, compared with the complete WCP diet, therefore suggests that combining WCP with commercial pelleted feed was more suitable than relying exclusively on WCP under the conditions of the present study. This interpretation is consistent with the concept that the nutritional benefits of alternative plant-derived ingredients depend on their inclusion level and their contribution to the overall nutrient balance of the diet.

The feed utilization results further support the superior response of T3. Fish fed T3 had the lowest FCR (1.74 ± 0.12) and the highest FE ($57.47 \pm 3.66\%$), whereas T4 had the highest FCR (1.98 ± 0.18) and the lowest FE ($50.51 \pm 4.10\%$). The improved feed utilization at the intermediate WCP replacement level may be associated with the combined contribution of fermented cassava-derived nutrients and nutrients supplied by the commercial pelleted feed. Fermentation of plant-based ingredients can partially modify complex dietary components and reduce some factors that constrain nutrient utilization, potentially improving the availability of nutrients for fish (Mukherjee et al., 2016). Fermented feed ingredients may also provide beneficial microorganisms or

microbial metabolites that can contribute to intestinal conditions and nutrient utilization (Dawood et al., 2018; Kuebutornye et al., 2019). Similar improvements in feed utilization following the incorporation of fermented agricultural by-products have been reported in aquaculture species (Mukherjee et al., 2016; Das et al., 2021). Nevertheless, because nutrient digestibility, digestive enzyme activity, and gut microbiota were not measured in the present study, these mechanisms should be considered possible explanations rather than demonstrated causes of the improved FCR and FE observed in T3.

However, complete replacement of commercial pelleted feed with WCP (T4) resulted in significantly higher feed intake than T3, but this higher intake did not translate into improved growth or feed utilization. The increased feed intake in T4 may have been associated with the lower nutrient density of the diet, particularly its lower CP and higher crude fiber content compared with the diets containing commercial pelleted feed. Under such conditions, fish may consume more feed to obtain sufficient nutrients, while the greater proportion of fiber-rich cassava residue may constrain the efficiency with which the consumed nutrients are converted into biomass. Excessive inclusion of cassava-derived ingredients has been reported to reduce nutrient utilization when dietary protein and energy balance becomes less favorable (Das et al., 2021; Taghizadeh Tabasi et al., 2025). In the present study, the T4 diet contained only 7.85% CP and 14.85% crude fiber on a DM basis, compared with 31.80% CP and 6.15% crude fiber in T1. Therefore, the higher feed intake but poorer FCR and FE observed in T4 are consistent with the markedly different nutrient profile of the complete WCP diet. These results further indicate that WCP is more appropriately used as a partial replacement for commercial feed rather than as the sole dietary component under the conditions evaluated in this study.

The inclusion of WCP also affected carcass characteristics, although the responses were not directly proportional to the WCP replacement level. Fish fed T2 had the highest fillet yield ($23.10 \pm 4.20\%$), whereas T3 and T4 had lower fillet yields of $17.45 \pm 5.17\%$ and $16.29 \pm 4.27\%$, respectively. Skeleton percentage was highest in T4 ($71.20 \pm 5.68\%$), while the lowest value was observed in T3 ($65.57 \pm 9.81\%$). These differences may reflect changes in nutrient allocation among tissues resulting from the different nutrient profiles of the experimental diets. In particular, increasing WCP replacement progressively reduced dietary CP and increased crude fiber, which may have altered the relative partitioning of nutrients toward muscle, skeletal tissue, and visceral components. Similar variation in carcass responses has been reported in fish receiving plant-derived feed ingredients with different carbohydrate and fiber characteristics (Vignesh et al., 2025). The higher HSI observed in T4 ($4.46 \pm 0.75\%$) may also be associated with increased hepatic storage of carbohydrate-derived energy, because excess dietary carbohydrate can promote glycogen deposition in the liver when carbohydrate availability exceeds immediate metabolic requirements (Dawood et al. 2020). However, this explanation remains tentative because hepatic glycogen or other biochemical indicators of energy storage were not measured in the present study. Likewise, the higher VSI observed in T3 should be interpreted cautiously because visceral composition was not further characterized. Overall, the carcass results indicate that WCP replacement influenced tissue proportions, but the changes were not accompanied by a corresponding reduction in growth performance at the 67% replacement level.

Water quality remained generally stable within the experimental culture system despite significant differences among dietary treatments in several physicochemical parameters. Water temperature did not differ significantly among treatments and remained within a relatively narrow range of 25.20 – 25.35°C , whereas pH, DO, EC, salinity, TDS, and ORP differed significantly among treatments. Notably, the differences in these parameters followed treatment-specific patterns, with T4 showing the highest pH and DO, the highest EC, salinity, and TDS, and the lowest ORP. Such variation may reflect differences in the amount and composition of organic material entering the culture water, as well as differences in microbial activity and nutrient transformation associated with the experimental diets. Organic carbon and microbial processes can influence nutrient cycling, oxygen dynamics, and oxidation-reduction conditions in aquaculture systems (Ebeling et al., 2006; Avnimelech, 2015; Abakari et al., 2022). Similarly,

microbial utilization of nutrients released from organic feed materials may alter water chemistry without necessarily producing adverse effects on fish performance when the system remains adequately managed (Siddik et al., 2024). In the present study, DO remained relatively high (6.80-7.65 mg L⁻¹), and the observed differences in physicochemical parameters were not accompanied by evidence of impaired growth or survival. Therefore, the dietary treatments, including the complete WCP diet, did not appear to cause an adverse deterioration of the culture environment during the 8-week experimental period. Nevertheless, because the present study did not compare these values with experimentally established species-specific optimum thresholds, the results should be interpreted as indicating stable culture conditions rather than proving that all parameters were within universally optimal ranges for *A. testudineus*.

Taken together, the present findings indicate that fermented cassava WCP has potential as a partial replacement for commercial pelleted feed in *A. testudineus* culture. Among the dietary treatments evaluated, the 67% WCP replacement level (T3) provided the most favorable combination of growth performance and feed utilization, as reflected by the highest final BW, WG, SGR, and ADG, together with the lowest FCR and highest FE. Importantly, complete replacement with WCP did not provide the same benefit and was associated with higher feed intake and poorer feed utilization. Thus, the results suggest that the value of fermented cassava pulp lies not simply in maximizing its inclusion, but in combining it with commercial feed at an appropriate replacement level. From a practical perspective, the use of WCP may reduce reliance on conventional commercial feed while providing a means of utilizing locally available cassava-processing residues. However, the present study evaluated biological responses rather than economic performance; therefore, reductions in feed cost, production cost, or economic return cannot be concluded directly from the present results. Further evaluation of feed cost per unit WG and economic return would be necessary to determine the practical economic advantage of WCP under farm conditions.

Several limitations of the present study should be considered when interpreting these findings. First, the feeding trial lasted only 8 weeks and was conducted using juvenile female *A. testudineus*; therefore, the responses observed may not necessarily apply to larger fish, males, or longer production cycles. Second, only one fermentation protocol was evaluated, using spontaneous fermentation for 7 days, and the effects of different fermentation durations, fermentation conditions, or microbial inocula were not investigated. Third, although the fermented cassava pulp was characterized for fermentation properties and proximate composition, residual HCN was not determined; consequently, the present study cannot directly establish the residual cyanide content or its safety implications. Fourth, nutrient digestibility was not measured, and digestive enzyme activities and gut microbiota were not evaluated. Therefore, the mechanisms responsible for the improved feed utilization observed at the 67% WCP replacement level cannot be attributed specifically to enhanced digestibility, digestive activity, or changes in intestinal microbial communities. In addition, the study did not evaluate long-term physiological responses or economic performance under commercial-scale production conditions. Accordingly, the present findings support the use of fermented cassava pulp-based WCP as a partial replacement for commercial feed under the experimental conditions tested, but further research involving different fish sizes and sexes, longer feeding periods, alternative fermentation protocols, HCN determination, nutrient digestibility, digestive enzyme activity, gut microbiota, and commercial-scale economic evaluation is required before the results can be extrapolated confidently to large-scale *A. testudineus* production.

Conclusions. Fermented cassava WCP can be used as a partial replacement for commercial pelleted feed in *A. testudineus*. A 67% WCP replacement level provided the most favorable overall response, resulting in the highest final BW, WG, SGR, and ADG, together with the lowest FCR and highest FE. In contrast, complete replacement with WCP increased feed intake but resulted in poorer feed utilization. WCP replacement also affected carcass characteristics and several water-quality parameters, but no adverse effect on fish performance was observed during the 8-week trial. Overall, 67% WCP

replacement appears to be a suitable level under the conditions of this study. Further research should evaluate longer feeding periods, different fish sizes and sexes, fermentation protocols, nutrient digestibility, and economic performance under commercial production conditions.

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Authors Contributions. Conceptualization: RP, SS; Methodology: RP, ST, TT, NP; Formal analysis: RP, ST, TT, NP; Investigation: RP, ST, TT, NP; Data curation: RP, ST, TT, NP; Writing – original draft: RP; Writing – review & editing: RP, ST, TT, NP, SS; Supervision: SS.

Conflict of Interest. The authors declare that there is no conflict of interest.

Data Availability. The data supporting the findings of this study are available from the corresponding author upon reasonable request.

Ethical statement. All experimental procedures involving fish were conducted in accordance with the ethical guidelines for the care and use of aquatic animals. The experimental protocol was reviewed and approved by the Institutional Animal Care and Use Committee (IACUC) of Sakon Nakhon Rajabhat University, Thailand (Approval No. IACUC-SNRU-16/2025).

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