Journal of Academia Vol. 14, Issue 1 (2026) 23 – 33

ASSESSMENT OF ALTERNATIVE PROTEIN SOURCES AS FISHMEAL REPLACEMENTS ON THE GROWTH PERFORMANCE OF JUVENILE *Tor tambroides (*BLEEKER 1854)

Mohd Yusof Hanan[1*], Nur Siti Fatimah Ramli[2], Md. Ali Amatul-Samahah[1], Muhamad Zudaidy Jaapar[1], Mohd Faizzi Mohd Yusof[1]

[1]*Fisheries Research Institute (FRI) Glami Lemi, Department of Fisheries, Titi, 71650 Jelebu, Negeri Sembilan, Malaysia*
[2]*Pusat Asasi Sains Universiti Malaya (PASUM), University of Malaya, 50603 Kuala Lumpur, Malaysia*

[*]*Corresponding author: hanan@dof.gov.my*

Abstract

Fishmeal has traditionally been a primary protein source in aquafeeds due to its high nutritional quality. However, declining availability and rising costs have driven interest in alternative protein sources. This study aimed to evaluate the effects of replacing fishmeal with different protein sources on the growth performance of juvenile Malaysian mahseer, *Tor tambroides*. Four experimental diets were used and formulated using Diet 1 as control, soybean meal (SBM) as Diet 2, Black Soldier Fly Larvae meal (BSFL) as Diet 3, and poultry by-product meal (PBM) as Diet 4. Juvenile *T. tambroides* were reared in outdoor tanks (triplicate groups per diet) over a five-month feeding trial. Growth performance metrics, including body weight gain, specific growth rate (SGR), feed conversion ratio (FCR), protein efficiency ratio (PER), and hepatosomatic index (HSI) were assessed. Results indicated that fish fed the Diet 2 achieved the highest weight gain which is 32.66 ±0.18 g compared to fish fed with Diet 3 (30.29 ±1.41g) and Diet 4 (25.54 ±1.51g). Meanwhile, for SGR, the fish fed with Diet 3 showed highest result (0.42±0.02%/day) compared to those fed Diet 2 (0.41±0.02%/day) and Diet 4 (0.38±0.02%/day). However, the differences were not statistically significant ($P>0.05$). Notably, the Diet 2 (1.06±0.001) resulted in significantly higher PER values ($P<0.05$) compared to the Diet 1 (control) (0.97±0.001). No significant differences ($P>0.05$) were observed in FCR across the different diet treatments. These results highlight the potential of alternative proteins using soybean meal (SBM) and Black Soldier Fly meal (BSFM) as viable substitutes for fishmeal in juvenile *T. tambroides* diets, while emphasizing the need for additional studies on protein utilization and long-term effects.

Keywords: Black soldier fly, diet formulation, Malaysian Mahseer, poultry by-product meal, soybean meal

Article History:- Received: 28 August 2025; Revised: 31 October 2025; Accepted: 2 November 2025; Published: 30 April 2026

DOI: https://doi.org/10.24191/joa.v14i1.8705

Introduction

Tor tambroides or locally known as Malaysian mahseer (Kelah) is from the family of Cyprinidae. Malaysian mahseer is a highly valued freshwater fish prized for its ecological significance, cultural importance, and premium market value (Tan et al., 2019). It commands high prices in the live fish and ornamental markets, sometimes exceeding RM800 per kilogram, making it one of the most economically important indigenous species in Southeast Asia (Bernama, 2021). In addition to its economic value, mahseer is regarded as a flagship species for river conservation and holds cultural significance among local communities, particularly in traditional fisheries and ceremonial uses (Raghavan et al., 2011). Despite its importance, Malaysian mahseer populations are reported to decline rapidly due to several interrelated factors (Asaduzzaman et al., 2016). Studies document that habitat degradation resulting from hydropower dam construction, river engineering, deforestation, and pollution from agriculture and industry, reduces river connectivity and alters critical spawning grounds (Lau et al., 2021). In addition, overfishing, including illegal and unsustainable capture driven by high

market demand, has led to drastic reductions in wild landings across regions such as Peninsular Malaysia and Kenyir Lake (Wong et al., 2019). Successfully induced spawning of captive *Tor tambroides* broodstock has been established to sustain the availability of the juveniles for commercial aquaculture production (Lee et al., 2014). Subsequently, a proper diet rich in proteins is of the utmost importance for the growth and development of *T. tambroides*, as it is known to be a slow-growing fish (Tan et al., 2019). This species allocates a substantial portion of its energy towards maintenance and metabolic processes, thereby requiring high-quality protein sources to support muscle development and optimize growth rates during its extended juvenile phase (Misieng et al., 2011).

Fishmeal has long been a cornerstone ingredient in aquafeed due to its high-quality protein content, ranging between 72-80%, balanced amino acid profile, and excellent digestibility, all of which are essential for optimal growth, feed efficiency, and health of cultured fish species.

However, the rising global demand for fishmeal, coupled with dwindling marine resources, has led to limited availability and a significant increase in feed costs, posing economic and sustainability challenges to the aquaculture industry (Tran et al., 2023). In response, the industry has shifted its focus towards the development of alternative, sustainable protein sources that can partially or fully replace fishmeal without compromising fish performance. These alternative protein sources not only reduce reliance on marine-derived ingredients but also support the goal of producing nutritionally balanced, cost-effective, and environmentally sustainable fish feeds.

Soybean is the second most imported raw ingredient after maize used as animal feed in Malaysia, with 1.35 million MT imports in 2015 (DoA, 2017). The huge demand for SBM is mainly for animal feed requirements. SBM is considered among the cheapest and most available protein sources in Asia. It has long been recognized as the best fit for fishmeal replacement due to its abundant supply, high protein content, and balanced amino acid profile (Ajani et al., 2016; Muin et al., 2015; Silva-Carrillo et al., 2012). Soybean meal, when included at levels of 50% to 60% in formulated diets, has been shown to sufficiently meet the protein requirements of many freshwater fish species due to its high crude protein content (Gatlin et al., 2007). It serves as an effective substitute for more expensive protein sources such as fishmeal, while still supporting optimal fish growth and performance. Consequently, the use of SBM in aquafeeds has contributed to a substantial reduction in overall production costs (Gatlin et al., 2007; Rawles et al., 2011).

Insects are part of the natural diets for wild fish. Therefore, the utilisation of insects as aquaculture feeds mainly in freshwater fish has been studied for their nutritional values (Henry et al., 2015; Taufek et al., 2016). Black soldier fly or *Hermetia illucens* is a species from the order Diptera that also housed the common housefly (Henry et al., 2015). Many studies have focused on insects as an alternative protein source because of their protein enrichment, easy rearing, and availability. Black soldier fly meal (BSFM) is a newly emerging alternative protein industry worldwide including Malaysia (Hasan et al. 2009; Macusi 2023). This sustainable protein source that is reproducible and technically easy and cheap to produce is expected to be among the most suitable alternative protein source in animal feed to substitute fishmeal. The implementation of BSF larvae as feed ingredients for fish and poultry animals has been intensified recently (Belghit et al., 2019; Onsongo et al., 2018).

Poultry by-product meal (PBM) is one of the most readily available and cost-effective protein sources produced locally in Malaysia. In 2018, the country recorded the production of approximately 771 million birds, with projections reaching up to 844 million birds, successfully achieving self-sufficiency in poultry supply for local consumption (DVS, 2019; FLFAM, 2019). PBM, as a rendered animal protein source, has been widely recognized as a viable alternative to fishmeal due to its lower cost and relatively high protein content (Hernandez et al., 2010; Ismail et al., 2013). Several studies have demonstrated that animal-based protein sources such as PBM can effectively support fish growth. For instance, research on Nile tilapia (*Oreochromis niloticus*) fed diets containing PBM reported significant improvements in growth performance (Hernandez et al., 2010; Amm & Aa, 2015).

Research into fishmeal alternatives continues to gain momentum as a strategic effort to reduce the economic burden of aquafeed production, promote resource sustainability, and ensure long-term food security. Identifying effective substitutes not only addresses the limitations of fishmeal supply but also supports environmentally responsible aquaculture practices by utilizing underused or renewable protein sources. In selecting suitable alternatives to fishmeal, several important criteria must be considered, including affordability, consistent availability, nutritional adequacy to support optimal growth, and compatibility with the physiological needs of the target fish species. Although the potential of BSFL and other alternative proteins has been widely reported, studies evaluating their combined use with locally available plant ingredients such as rice bran remain limited. This study investigates the effects of integrating BSFL and rice bran in fish diets on growth performance, feed utilization, and cost efficiency, providing novel data relevant to sustainable aquafeed development in tropical aquaculture systems. Therefore, this study aimed to determine the most suitable alternative protein source among SBM, BSFL, and PBM as a sustainable substitute for fishmeal in the diet of juvenile *T. tambroides*.

Methods

Ethical Statement

This study was approved by the Committee on the Ethics of Animal Experiments, Department of Fisheries Malaysia (DOF) (Protocol Number: DOF/KAKH/03/2020). This study was carried out in strict accordance with the recommendations in the Guide for the Care and Use of Laboratory Animals of the Department of Fisheries Malaysia.

Experimental Diet

Danish fishmeal (DFM), soybean meal (SBM), Black Soldier Fly (BSF) larvae and poultry by-product meal (PBM), were purchased from a local company, Nutrivet Sdn Bhd. and were refrigerated at -20 °C upon arrival at the laboratory. The BSF larvae were dried overnight in the oven at 70 °C and grounded to powder form. The four experimental diets were formulated using the main ingredients DFM, SBM, BSF, and PBM. Diet 1 as control diet. All the main ingredients of raw materials were mixed accordingly with the required materials as described in Table 1. Water was added to the mixture before pellet processing using a mini pelleting machine (KCM, Y132M-4 China) to produce a sinking pellet with a size of 2.0 mm diameter. The produced pellets of each diet were then dried in the oven at 70 °C for 24 hours, properly packed, and later stored in 4 °C.

Table 1. Formulation ingredients in percentage (%) of different protein origin diets used in the diet formulation.

Ingredients (%)	DIET 1 (Control)	DIET 2	DIET 3	DIET 4
Danish fishmeal (DFM)	25.4	0.0	0.0	0.0
Soybean meal (SBM)	20.0	66.5	27.8	20.0
Black soldier fly meal (BSFM)	0.0	0.0	50.2	0.0
Poultry by-product meal (PBM)	0.0	0.0	0.0	20.7
Rice bran	26.0	12.0	2.7	33.8
Maize	20.0	20.0	20.0	20.0
Palm oil	2.0	2.0	2.0	2.0
DCP	1.0	1.0	1.0	1.0
Premix vitamin[a]	0.4	0.4	0.4	0.4
Premix mineral[b]	0.5	0.5	0.5	0.5
Vitamin C	0.1	0.1	0.1	0.1

DFM: Danish fishmeal; SBM: soybean meal; BSFM: Black soldier fly meal; PBM: poultry by-product meal; DCP: dicalcium phosphate; Premix vitamin[a]: vitamin A, 500 IU; vitamin D3, 100 IU; vitamin E, 0.75 mg; vitamin K, 0.02 mg; vitamin B1, 1.0 mg; vitamin B2, 0.5 mg; vitamin B3, 0.3 mg; vitamin B6, 0.2 mg; vitamin B12, 0.001 mg; vitamin C, 0.1 mg; niacin, 0.2 mg; folic acid, 0.1 mg; biotin, 0.235 mg; pantothenic acid, 1.0 mg, inositol, 2.5 mg; Premix mineral[b]: selenium, 0.2 mg; iron, 8.0 mg; manganese 1.0 mg; zinc, 8.0 mg; copper, 0.15 mg; potassium chloride, 0.4 mg; magnesium oxide, 0.6 mg; sodium bicarbonate, 1.5 mg; iodine, 1.0 mg; cobalt, 0.25 mg.

Proximate Analysis
The four experimental diets were analysed for their proximate compositions according to the standard method of Association of Official Analytical Chemist (Chemists & Analysis 2019). Crude lipid was analysed using petroleum benzene extraction in FOSS Soxtec 2055, while crude protein was determined by titration with hydrochloric acid after the sample was heated up with potassium sulphate and sulphuric acid at 420 °C. Moisture and dry matter were measured by drying the sample in the oven at 105 °C for 8 hours. Meanwhile, ash was measured by placing the sample in the furnace at 600 °C for 4 hours.

Experimental Design
The juveniles of *Tor tambroides* were produced from our own hatchery in Fisheries Research Institute (FRI) Glami Lemi, Jelebu, Negeri Sembilan, Malaysia. Three hundred and sixty juveniles of *Tor tambroides* with mean initial body weight of 34.68 ± 0.26 g and total length of 15.06 ± 0.04 cm were randomly selected and reared into outdoor rectangular-shaped cement tanks. Each tank (volume: 5 x 3 x 0.3 m) contained 3000 L water and equipped with closed-aerated recirculating system (flow rate 10 L/min). Water was changed twice a week and the temperature, pH and dissolved oxygen (dO2) concentrations were maintained at 25 ± 0.05 °C, 72.48 ± 0.01, 9 ± 0.04, and 8.02 ± 0.01 mg/L respectively. Other water parameters monitored include total ammonia, which was kept below 0.18 ± 0.04 mg/L and nitrite at 0.006 ± 0.001 mg/L.

Feeding Trial
At the start of the experiment, the fish underwent initial acclimation for two weeks and were fed with commercial diet twice daily. After a two-day fasting period, 30 fish per tank were randomly assigned to one of four dietary treatments: Diet 1 (control, Danish fishmeal; DFM), Diet 2 (soybean meal; SBM), Diet 3 (black soldier fly meal; BSFM), and Diet 4 (poultry by-product meal; PBM), each serving as a replacement for fishmeal. Each diet groups were assigned in triplicates. Fish were fed on their specific diets twice daily (at 0900 and 1600 H) at the rate of 5 % body weight per day. Thus, the required feeding was calculated depending on the body weight gained after each sampling.

Growth Performance Analysis
Sampling of physical measurement (body weight (g) and total length (cm)) of the fish was done at the end of every month. The fish were captured and anaesthetised with clove oil. Meanwhile, three random fish per tank were sacrificed at the end of the experiment (after 5 months), and their liver were excised and weighed for the determination of hepatosomatic index (HSI). Growth performance and feed utilisation were analysed in terms of body weight gain (BWG), total length gain (TLG), condition factor (K), specific growth rate (SGR), feed conversion ratio (FCR), and protein efficiency ratio (PER). The parameters were calculated as follow:

HSI	= (Liver weight (g) / W1) x 100
BWG	= W1 – W0
TLG	= TL1 – TL0
K	= W100/L3
SGR	= [(lnW1 – lnW0) / t] x 100
FCR	= Df / (W1 – W0)
PER	= BWG / Protein consumed (%)

Where W0 = initial weight (g), W1 = final weight (g), TL0 = initial total length (cm), TL1 = final total length (cm), t = experiment time (day), and Df = dry feed (g).

The condition factor was calculated using Fulton`s equation where K = condition factor, W = weight per month (g), and L = length per month (cm).

Statistical analysis

All data were calculated using Statistical Package for Social Sciences (SPSS) software version 21.0 (IBM, New York, USA) and were expressed as mean ± standard error of the mean (SEM), unless otherwise stated. One-way analysis of variance (ANOVA) was used to compare the mean differences of the dataset between the diet groups. One-way ANOVA was then followed by Tukey's HSD post hoc test was used to determine significant differences among dietary treatments ($P < 0.05$). The differences were considered significant when $P<0.05$. Pearson's correlation coefficient test was used to compare the association between body weight and total length in each diet and the linear regression relationships were presented in the scatterplot.

Result

All four diets have similar isonitrogenous (32% crude protein) and isolipidic (10%) contents. Diet 1 contains the lowest dry matter and the highest moisture compositions of 89.54 % and 10.46 % respectively, while Diet 3 has the highest ash composition (21.31%) among the other diets (Table 2).

Table 2. Proximate nutrient compositions in percentage (%) of different protein origin diets used in the diet formulation.

Ingredients (%)	DIET 1	DIET 2	DIET 3	DIET 4
Crude protein	35.44 ± 1.77	32.13 ± 1.61	31.75 ± 1.59	31.93 ± 1.60
Crude lipid	10.30 ± 0.52	9.80 ± 0.49	11.10 ± 0.56	10.10 ± 0.51
Dry matter	89.54 ± 4.48	91.16 ± 4.56	91.46 ± 4.57	92.49 ± 4.62
Moisture	10.46 ± 0.52	8.84 ± 0.44	8.54 ± 0.43	7.51 ± 0.38
Ash	15.79 ± 0.79	7.54 ± 0.38	21.31 ± 1.07	2.62 ± 0.13

.

Growth performance, feed utilisation, and hepatosomatic index

The results of fish growth, survival rate, condition factor, and hepatosomatic index of *T. tambroides* in response to 5 months feeding trial are presented in Table 3. The survival rate was generally high (95 - 100 %) and no significant differences among diet groups were observed ($p>0.05$). The mean final body weight of fish fed with Diet 2, Diet 3, and Diet 4 was significantly lower than the mean final body weight of fish fed the control diet DFM ($p<0.05$). However, the data of fish fed with Diet 2 (32.66 ± 1.82 g) showed a higher body weight gain than Diet 1 (control) (32.41 ± 1.56 g), although it was not significant. Meanwhile, the fish fed with Diet 4 showed a significant decrease ($p<0.05$) in weight gain when compared to Diet 1 (control). The total length gain of all fish was also not significantly affected ($p>0.05$) among the experimental diets. There was no significant difference observed in the condition factor ($p>0.05$).

Table 3. Growth performance, feed utilisation, and hepatosomatic index (HSI) of *T. tambroides* fed with different diets[1].

Parameters	DIET 1	DIET 2	DIET 3	DIET 4
Initial body weight (g)	37.56 ± 1.26^{a}	36.64 ± 1.82^{a}	32.71 ± 1.36^{a}	31.78 ± 1.47^{a}
Final body weight (g)	69.98 ± 1.59^{a}	69.30 ± 1.66^{a}	63.00 ± 1.35^{b}	57.32 ± 1.38^{b}
Body weight gain (g)	32.41 ± 1.56^{b}	32.66 ± 1.82^{b}	30.29 ± 1.41^{ab}	25.54 ± 1.51^{a}
Total length gain (cm)	3.24 ± 0.14^{a}	3.46 ± 0.18^{a}	3.40 ± 0.17^{a}	2.95 ± 0.19^{a}
Specific growth rate (%)	0.39 ± 0.02^{a}	0.41 ± 0.02^{a}	0.42 ± 0.02^{a}	0.38 ± 0.02^{a}
Feed conversion ratio	2.21 ± 0.04^{a}	2.49 ± 0.11^{a}	2.44 ± 0.17^{a}	2.82 ± 0.26^{a}
Protein efficiency ratio	0.97 ± 0.001^{a}	1.06 ± 0.001^{b}	1.00 ± 0.003^{c}	0.84 ± 0.003^{d}
Hepatosomatic index	1.03 ± 0.11^{a}	1.14 ± 0.13^{a}	0.95 ± 0.09^{a}	1.25 ± 0.29^{a}

[1]Values are expressed as mean ± SEM. Mean values in the same given row with different superscripts are significantly different ($P < 0.05$).

The specific growth rate (SGR) of fish fed with Diet 2 and Diet 3 was higher than the control Diet 1 but no significant differences were observed ($p>0.05$). The feed conversion ratios (FCR) of the

experimental diets were lower than Diet 1 without any significant difference ($p>0.05$). Interestingly, the Diet 2 group showed the best protein efficiency ratio (PER) reading of 1.06 ± 0.001, followed by Diet 3 with 1.00 ± 0.003, when compared to Diet 4 (0.97 ± 0.001), and they were statistically significant ($p< 0.05$). Diet 4 on the other hand gave a significantly low PER than the other formulated diets ($p<0.05$). Hepatosomatic index (HSI) was calculated based on the ratio of fish liver weight to total body weight, as an indicator of energy reserved in fish. Although the results obtained no significant differences for all experimental diets, fish fed with Diet 2 and Diet 4 did show higher HSI values compared to fish fed with control diet Diet 1 (Table 3).

Correlation between body weight and total length of fish

Pearson's correlation between body weight and total length of all the diet groups were presented in scatter plots with the addition of linear regression analysis to ascertain the strength between these two variables (Figure 1). The body weight and total length in all diet groups showed proportional increments indicating significant positive linear relationships ($p<0.01$). Meanwhile, the correlation coefficient of fish fed with Diet 2 demonstrated the strongest relationship ($r = 0.957$) between body weight and total length, followed by Diet 3 ($r = 0.938$), DFM ($r = 0.923$), and finally the lowest was Diet 4 ($r = 0.867$) (Figure 1).

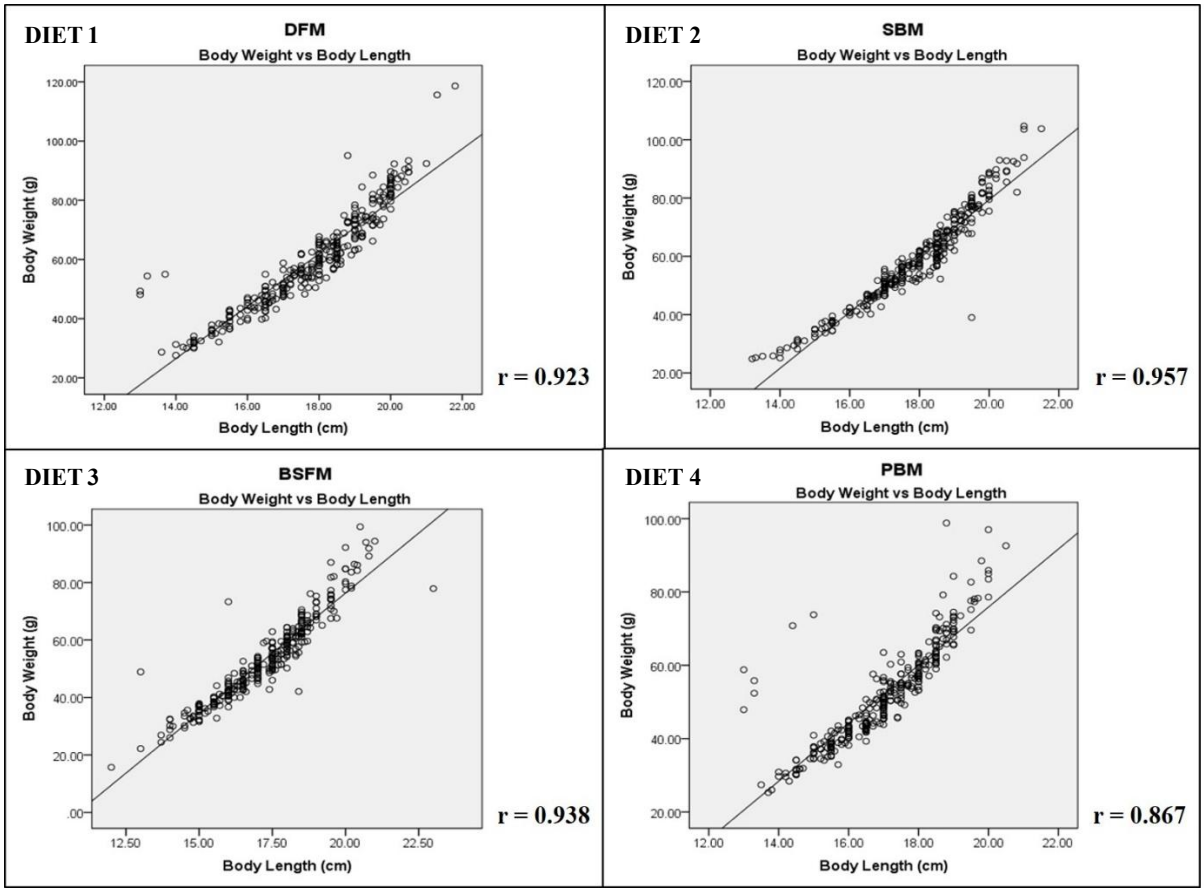

Figure 1. Relationship between body weight (g) and total length (cm) of fish fed with four diets. Pearson's correlation coefficient test were expressed as 'r' for the association between the two variables stated.

Discussion

The crude protein (CP) of the formulated diets was exerted at 31 % to 35 % while crude lipid (CL) was around 9 % to 11 % (Table 1). Initially, there was a large difference in CP and CL values among the experimental diets which caused difficulties in formulating the diet ingredients. The formulation needs to be balanced to achieve isonitrogenous and isolipidic. Thus, the ingredients need to be supported with other protein sources to achieve the target CP and CL for all diets. Therefore, in this study, soybean meal was used to help to contribute sufficient protein amount for each diet. In order to avoid bias, each

diet formulation was standardised to contain 20 % of this same protein source as referred to Ismail et al., 2013. Soybean is an ideal protein source to complement the diet formulation because it contained high CP (47.51 %), and low CL (1.5 %). Meanwhile, BSF contained a low CP value of only 31.37 % but has the highest CL value of 23.78 % compared to the other protein sources. Thus, high inclusion of soybean and BSF for up to 66.5 % and 50.2 % respectively were included on top of 20 % inclusion of the same protein materials in the formulation of Diet 2 and Diet 3. On the other hand, proximate analysis of raw Danish fishmeal and poultry by-product showed a very high CP value of 72.69 % and 82.85 % respectively, while low in CL value of 5.7 % and 7.7 % respectively. Therefore, less inclusion of Danish fishmeal and poultry by-product were added into their respective formulations in order to achieve the targeted nutrient values. However, the present study did not include measurements of amino acid composition, nutrient digestibility, or digestive enzyme activity, which may limit the mechanistic interpretation of growth responses among the diet treatments. Future work incorporating these analyses would help clarify the biochemical basis for observed performance differences and strengthen conclusions on nutrient utilization efficiency.

Previous studies stated that partial replacement of fishmeal with SBM gave a higher body weight gain than the zero replacement diet in juvenile Nile tilapia (*O. niloticus*). The same trend was also shown in the final total body length of the fish (Ajani et al., 2016). In addition, (Silva-Carrillo et al., 2012) also reported that the body weight gained by juvenile spotted rose snapper (*Lutjanus guttatus*) fed with the diet with an inclusion level of 20 % SBM gave the best result compared with the higher inclusions of 40 % and 60 %, but obtained no significant difference with a control diet. This proved that the diets with the incorporation of SBM did influence the growth of fish. A study focussing on dietary phospholipid levels and sources towards growth performance of juvenile *T. tambroides* showed that diets treated with soybean lecithin (2 - 6 %) gave higher body weight gain and SGR values compared to the control diet (without soybean lecithin) (Mian et al., 2017).

Our study showed that *T. tambroides* juveniles fed with Diet 3 had increased body weight and total length over 5 months although not as high as fish fed with Diet 1 (Control) and Diet 2. On the contrary, (Kamarudin et al*.,* 2021) has reported that the growth of lemon fin barb hybrid fish was significantly decreased ($P < 0.05$) with the increase of BSF prepupae meal (25 – 100 % inclusion). Meanwhile, there were no differences in total body weight gain of fish fed at 25 % BSF observed in rainbow trout (*Oncorhynchus mykiss*) (St-Hilaire *et al.,* 2007). Other studies have reported that some inclusion levels of BSF prepupae provided a similar weight gain to that of fish fed fishmeal; 18 % to 36 % (Sealey et al., 2011) and 15 % (St-Hilaire *et al.,* 2007) inclusion in rainbow trout (*O. mykiss*), and 33 % inclusion in juvenile turbot (*Scophthalmus maximus*) (Kroeckel et al., 2012). The findings suggest that the utilisation of insects mainly BSF as alternative protein in the fish diet could be possible without negatively affecting fish growth performance. To date, no studies involving BSF have been reported for *T. tambroides*.

Based on the present results, fish fed with Diet 4 did not show much improvement in growth compared to the other meals. It has the lowest weight gain with 25.54 ± 1.51 g which was significant with Diet 1 (Control) and Diet 2 treatments. This data was in agreement with another study done in the same cyprinid family as *T. tambroides* which is juveniles *Cyprinus carpio,* who obtained the same data as ours, and concluded that the fish growth was negatively affected by Diet 4 levels (Emre et al., 2003). Fish fed with Diet 4 showed the least performed growth in SGR, PER, and FCR compared to the other diet groups. (Webster et al., 2000) also revealed that sunshine bass fed with Diet 4 had no differences in SGR and FCR than the control diet. On the contrary, (Ismail et al., 2013) reported that a diet with 75 % inclusion of poultry offal meal obtained significantly high SGR and low FCR ($p<0.05$) than the fish fed diet with 100 % fishmeal in juvenile *T. tambroides*. The fish in their study was reared in indoor aquaria (65 L) while ours was done in outdoor tanks. This factor may contribute to the differences in the data obtained, as reported by (Islam, 2002) who studied the comparison between the indoor and outdoor phase for the growth performance of *Tor putitora* fed with supplementary diets. The fish in the outdoor ponds showed better growth performance than the ones put in indoor cement tanks.

The current study showed that the SGR and PER were higher in the Diet 2 and Diet 3 groups compared to the control Diet 1 (Control). This finding was in agreement with (Ajani et al., 2016) who have reported the same results in their 46 % soybean inclusion in the diet of juvenile Nile tilapia (*O. niloticus*), but there were no significant differences reported in SGR and PER of fish fed with BSF compared to control diet (without BSF) in juvenile mirror carp (Xu et al., 2020). High PER suggested that the fish were able to digest the formulated diets and absorb their nutrients efficiently (Misieng et al., 2011). In addition, the FCR in the present study was not significantly different among all groups, suggesting that the experimental diets were palatable for juvenile *T. tambroides.*

There is a linear relationship between body weight and length of the fish for all experimented groups as indicated by the 'r' values (Figure 1). Fish fed with Diet 2 demonstrated the strongest relationship (r = 0.957) as the 'r' value was the closest to 1. This indicated the healthy well-being of the juveniles as they were growing in both weight and total length over the feeding period. The condition factor (K) measures the suitability of the environmental factor for fish growth and is strongly correlated with the length-weight relationship (Abd Hamid et al., 2015). The mean of the K value in the current study was between 1.02 ± 0.01 and 1.05 ± 0.02, thus it was considered to be a good condition for the fish. Our data obtained was similar to other studies reported in *Tor* species with the condition factor range of 1.07 to 1.14 in *T. tambroides* (Wan, 2015) and 0.98 to 1.09 in *Tor tor* (Saini *et al.,* 2014).

Hepatosomatic index (HSI) was calculated based on the ratio of fish liver weight to total body weight, as an indicator of energy reserved in the liver. HSI was also referred to as the development of gonad and has been vastly used alongside gonadosomatic index (GSI) in breeding and vitellogenesis studies. The HSI obtained in the present study was within the normal range of 0.95 to 1.25 and no significant difference was observed.

Crude economic analysis showed that Diet 2 is the most cost-effective meal compared to other diets. The cost of feeding *T. tambroides* using Diet 2 can be reduced by up to 47 % compared to the Diet 1 (Control) diet and 55 % compared to the Diet 3 diet for each production per kg fish. This is due to the raw material cost factor for the protein source from Diet 3 which is currently expensive at around RM 8000 (USD 1900) per MT, while imported fishmeal or Diet 1 (Control) is priced at RM 12,000 (USD 2900) per MT. However, if the price of Diet 3 can be significantly lowered by the manufacturer, then, it has the potential to be one of the best alternative sources for *T. tambroides* diet and replacement of fish meal. In addition, for the purpose of commercialization, the production of alternative protein sources must be consistent to meet market demand.

Several factors could influence the present results including fish species, types of protein sources used, and method of diet processing. In this study, we performed the feeding treatment in juveniles *T. tambroides*. The fish is omnivorous and known to have reduced growth rate as it matures (Ingram et al., 2007). This species' natural food comprises fruits, plant parts, insects, prawns, and aquatic invertebrates (Ingram et al., 2007). Thus, incorporation of different protein origins in the fish diet of this study were palatable for *T. tambroides*.

Conclusion

In conclusion, the current study results showed that SBM and BSFM could be taken as options for substituting fishmeal in diet formulations. Both of the diets have shown positive improvements in the growth of *T. tambroides* juveniles. SBM seems to be a relevant alternative protein source due to its abundant supply, affordable price, and is highly nutritious. While all of these benefits also apply to BSFM, the inclusion of SBM in the diet formulation is still needed. Furthermore, a supplementary study on the digestibility and amino acid profiles in the suggested diet substitution of fishmeal should be considered.

Acknowledgements/Funding

This study was conducted within the framework of research project No. BKP001; University of Malaya and co-funded by research project under 11th Malaysian Plan Development Fund Project (Project code: 22501 037)

Department of Fisheries, Malaysia. The authors would like to thank all of the staff members of Fisheries Research Institute (FRI) Glami Lemi, Negeri Sembilan, Malaysia for their assistance in completing this project.

Author Contribution

MY Hanan - Conceptualization, Investigation, Data Analysis, Project Administration, Writing -original draft, review and editing; NSF Ramli - Supervision, Investigation, Project Administration, Writing -review and editing; MA Amatul-Samahah - Investigation, Project Administration, Writing -review and editing; MZ Jaapar- Investigation, Project Administration, Writing -review and editing; MF Mohd-Yusof – Supervision, Investigation, Project Administration, Writing -review and editing.

Conflict of Interest

The authors declare that they have no known competing financial interests or personal relatioships that could have appeared to influence the work reported in this paper.

Declaration of the Use of Generative AI

Authors declare that AI tools such as ChatGPT and Grammarly were used only to improve the language and grammar of this manuscript. They were not used to generate content, analyze data, or make scientific decisions. All ideas, analysis, and conclusions are the authors' own work.

References

Abd Hamid M., Mansor, M. & Siti-Azizah M. (2015). Length-weight relationship and condition factor of fish populations in Temengor Reservoir: Indication of Environmental Health. *Sains Malaysiana*, 44, 61-66. http://doi.10.17576/jsm-2015-4401-09

Ajani, E.K., Orisasona, O., Omitoyin, B.O. & Osho, E.F. (2016). Total replacement of fishmeal by soybean meal with or without methionine fortification in the diets of Nile Tilapia, *Oreochromis niloticus*. *Journal of Fisheries and Aquatic Science*, 11,238-243. http://doi.10.3923/jfas.2016.238.243

Amm, Y. & Aa, M. (2015). Effects of fish meal substitution with poultry by-product meal on growth performance, nutrients utilization and blood contents of juvenile Nile Tilapia (*Oreochromis niloticus*). *Journal of Aquaculture Research & Development*, 7,1. http://doi.10.4172/2155-9546.1000389

AOAC International. (2019). *Official Methods of Analysis of AOAC International* (21st ed.). AOAC International: pp. 920–950.

Asaduzzaman, M., Kader, M.A., Bulbul, M., Abol-Munafi, A.B., Ghaffer, M.A. & Verdegem, M. (2016). Biochemical composition and growth performances of Malaysian Mahseer *Tor tambroides* larvae fed with live and formulated feeds in indoor nursery rearing system. *Aquaculture Reports*, 4,156-163. https://doi.org/10.1016/j.aqrep.2016.09.003

Belghit, I., Liland, N.S., Gjesdal, P., Biancarosa, I., Menchetti, E., Li, Y., Waagbø, R., Krogdahl, Å. & Lock, E-J. (2019). Black soldier fly larvae meal can replace fish meal in diets of sea-water phase Atlantic salmon (*Salmo salar*). *Aquaculture*, 503, 609-619. https://doi.org/10.1016/j.aquaculture.2018.12.032

Bernama (2021). Siakap vs Empurau: Which is more expensive? https://www.astroawani.com/berita-malaysia/siakap-vs-empurau-which-more-expensive-330842.
.[Access online 4 March 2024]

Bilen, S. & Bilen, A. (2013). Effects of different protein sources on growth performance and food consumption of goldfish, Carassius auratus. *Iranian Journal of Fisheries Sciences,* 12,717-722.

DoA, Department of Agricultural Malaysia. (2017). Corn & Animal Feed & Corn Industry in Malaysia. http://www.doa.gov.my/index/resources/aktiviti_sumber/sumber_awam/penerbitan/kertas_pembentangan/seminar_jagung_bijian_2017/kertas_pembentangan13.pdf. [Access online 23 July 2023].

DVS, Department of Veterinary Services Malaysia. (2019). Livestock Statistics 2019/2020. http://www.dvs.gov.my/dvs/resources/user_1/2019/BP/Perangkaan%20Ternakan%2020182019/1)_Malaysia_Perangkaan_Ternakan.pdf. [Access online 23 August 2022].

Emre, Y., Sevgili, H., Diler, İ.J.T & Sciences A. (2003) Replacing fish meal with poultry by-product meal in practical diets for mirror carp (*Cyprinus carpio*) fingerlings. *Turkish Journal of Fisheries and Aquatic Sciences*, 3,81-85.

FLFAM, Federation Of Livestock Farmers' Associations Of Malaysia. (2019). Industry statistic. http://www.flfam.org.my/index.php/industry-statistics. [Access online 23 September 2021].

Gatlin, D.M., Barrows, F.T., Brown, P., Dabrowski, K., Gaylord, T.G., Hardy, R.W., Herman, E., Hu, G., Krogdahl, Å., Nelson, R., Overturf, K., Rust, M., Sealey, W., Skonberg, D.J., Souza, E., Stone, D., Wilson, R. & Wurtele E. (2007) Expanding the utilization of sustainable plant products in aquafeeds: a review. *Aquaculture Research*, 38, 551-579. https://doi.org/10.1111/j.1365-2109.2007.01704.x

Hasan, M. R., Rana, K. J., & Siriwardena, S. (2009) Impact of rising feed ingredient prices on aquafeeds and aquaculture production (FAO Fisheries and Aquaculture Technical Paper No. 541). Food and Agriculture Organization of the United Nations. https://www.fao.org/4/i1143e/i1143e00.htm

Henry, M., Gasco, L., Piccolo, G. & Fountoulaki, E. (2015) Review on the use of insects in the diet of farmed fish: Past and future. *Animal Feed Science and Technology*, 203, 1-22. https://doi.org/10.1016/j.anifeedsci.2015.03.001

Hernandez, C., Olvera-Novoa, M.A., Hardy, R.W., Hermosillo, A., Reyes, C., & Gonzalez B. (2010) Complete replacement of fish meal by porcine and poultry by-product meals in practical diets for fingerling Nile tilapia, Oreochromis niloticus: digestibility and growth performance. *Aquaculture Nutrition*, 16, 44-53. https://doi.org/10.1111/j.1365-2095.2008.00639.x

Ingram, B., Sungan, S., Tinggi, D., Sim, S.Y. & De Silva, S.S. (2007). Breeding performance of Malaysian mahseer, *Tor tambroides* and *T. douronensis* broodfish in captivity. *Aquaculture Research*, 38, 809-818. https://doi.org/10.1111/j.1365-2109.2007.01716.x

Islam, M.S. (2002). Evaluation of supplementary feeds for semi-intensive pond culture of mahseer, *Tor putitora* (Hamilton). *Aquaculture*, 212(1-4), 263-276.

Ismail, S., Kamarudin, M.S. & Ramezani-Fard, E. (2013). Performance of commercial poultry offal meal as fishmeal replacement in the diet of juvenile Malaysian Mahseer, *Tor tambroides*. *Asian Journal of Animal and Veterinary Advances*, 8, 284-292. https://doi.org/10.3923/ajava.2013.284.292

Kamarudin, M.S., Rosle, S. & Md. Yasin, I.S. (2021). Performance of defatted black soldier fly pre-pupae meal as fishmeal replacement in the diet of lemon fin barb hybrid fingerlings. *Aquaculture Reports*, 21, 100775.

Kroeckel, S., Harjes, A.G.E., Roth, I., Katz, H., Wuertz, S., Susenbeth, A. & Schulz, C. (2012). When a turbot catches a fly: Evaluation of a pre-pupae meal of the Black Soldier Fly (Hermetia illucens) as fish meal substitute — Growth performance and chitin degradation in juvenile turbot (Psetta maxima). *Aquaculture* ,364-365, 345-352. https://doi.org/10.1016/j.aquaculture.2012.08.041

Lau, M. M. L., Lim, L. W. K., Ishak, S. D., Abol Munafi, A. B., & Chung, H. H. (2021). A review on the emerging Asian aquaculture fish, the Malaysian mahseer (Tor tambroides): Current status and the way forward. *Proceedings of the Zoological Society,* 74(2), 227–237. https://doi.org/10.1007/s12595-021-00368-4

Lee, K., Lihan, S., Dasthagir, F.F.G., Mikal, K.M., Collick, F. & Ng, K.H. (2014). Microbiological and physicochemical analysis of water from Empurau fish (*Tor tambroides*) farm in Kuching, Sarawak, Malaysian Borneo. *International Journal of Scientific & Technology Research*, 3, 285-292.

Macusi, E. D. (2023). Protein fishmeal replacement in aquaculture: A review. *Sustainability*, 15(16), 12500. https://doi.org/10.3390/su151612500

Mian, S., Kader, M. & Abol-Munafi A. (2017). Effects of dietary phospholipid levels and sources on growth performance, fatty acid composition and oxidative responsiveness of juvenile Malaysian mahseer, *Tor tambroides. AACL Bioflux*, 10, 1127-1139.

Misieng, J., Kamarudin, M.S. & Musa, M. (2011). Optimum dietary protein requirement of Malaysian Mahseer (*Tor tambroides*) Fingerling. *Pakistan Journal of Biological Sciences*, 14, 232-235. https://doi.org/10.3923/pjbs.2011.232.235

Muin, H., Mohd. Taufek, N., Abiodun, R.A., Mohd. Yusof, H. & Abdul Razak S. (2015). Effect of partial and complete replacement of fishmeal with mushroom stalk meal and soy bean meal on growth performance of Nile Tilapia, *Oreochromis niloticus* fingerlings. *Sains Malaysiana*, 44, 511-516. https://doi.org/10.17576/jsm-2015-4404-05

Onsongo, V.O., Osuga, I.M., Gachuiri, C.K., Wachira, A.M., Miano, D.M., Tanga, C.M., Ekesi, S., Nakimbugwe, D. & Fiaboe, K.K.M. (2018) Insects for income generation through animal feed: effect of dietary replacement of soybean and fish meal with black soldier fly meal on broiler growth and economic performance. *Journal of Economic Entomology,* 111, 1966-1973. https://doi.org/10.1093/jee/toy118

Raghavan, R., Ali, A., Dahanukar, N., & Rosser, A. (2011) Is the fishery for the Deccan Mahseer, *Tor khudree* (Sykes, 1839) in the Western Ghats Hotspot sustainable? A participatory approach to stock assessment. *Fisheries Research*, 110, 29–38.

Rawles, S.D., Thompson, K.R., Brady, Y.J., Metts, L.S., Aksoy, M.Y., Gannam, A.L., Twibell, R.G., Ostrand, S.& Webster, C.D. (2011) Effects of replacing fish meal with poultry by-product meal and soybean meal and reduced protein level on the performance and immune status of pond-grown sunshine bass (*Morone chrysops* × *M. saxatilis*). *Aquaculture Nutrition*, 17, e708-e721. https://doi.org/10.1111/j.1365-2095.2010.00831.x

Saini, V.P., Ojha, M. & Gupta M. (2014) Selected morpho physiographic features of Tor Maheer (*Tor tor*) from Rana Pratap Sagar reservoir, Rajasthan (India). *International Journal of Fisheries and Aquatic Studies*, 1, 168-172.

Sealey, W.M., Gaylord, T.G., Barrows, F.T., Tomberlin, J.K., McGuire, M.A., Ross, C., & St-Hilaire, S. (2011) Sensory analysis of rainbow trout, *Oncorhynchus mykiss*, fed enriched black soldier fly prepupae, *Hermetia illucens*. *Journal of the World Aquaculture Society*, 42(1), 34-45.

Silva-Carrillo, Y., Hernández, C., Hardy, R.W., González-Rodríguez, B., & Castillo-Vargasmachuca S. (2012) The effect of substituting fish meal with soybean meal on growth, feed efficiency, body composition and blood chemistry in juvenile spotted rose snapper *Lutjanus guttatus* (Steindachner, 1869). *Aquaculture*, 364-365, 180-185. https://doi.org/10.1016/j.aquaculture.2012.08.007

St-Hilaire, S., Sheppard, C., Tomberlin, J.K., Irving, S., Newton, L, McGuire MA, ... & Sealey, W. (2007) Fly prepupae as a feedstuff for rainbow trout, *Oncorhynchus mykiss*. *Journal of the World Aquaculture Society*, 38(1), 59-67.

Taufek, N.M., Muin, H., Raji, A.A., Razak, S.A., Yusof, H.M. & Alias, Z. (2016). Apparent digestibility coefficients and amino acid availability of cricket meal, *Gryllus bimaculatus,* and fishmeal in African catfish, *Clarias gariepinus*, diet. *Journal of the World Aquaculture Society*, 47, 798-805. https://doi.org/10.1111/jwas.12302

Tran, T. N., Phan, L. T., Nguyen, H. V., & Le, T. N. (2023). Sustainable alternatives to fishmeal and fish oil for aquafeed production: A global perspective. *Sustainability,* 15(16), 12500. https://doi.org/10.3390/su151612500

Wan, M. S. (2015). Investigation of alternative ingredients for the replacement of fish meal in formulation of feed for Malaysian mahseer fingerlings, Tor tambroides. Graduate Master's Thesis, Universiti Tunku Abdul Rahman. Retrieved from http://eprints.utar.edu.my/1719/

Webster, C.D., Thompson, K.R., Morgan, A.M., Grisby, E.J. & Gannam, A. L. (2000). Use of hempseed meal, poultry by-product meal, and canola meal in practical diets without fish meal for sunshine bass (*Morone chrysops*× *M. saxatilis*). *Aquaculture,* 188(3-4), 299-309.

Xu, X., Ji, H., Yu, H. & Zhou, J. (2020). Influence of dietary black soldier fly (*Hermetia illucens* Linnaeus) pulp on growth performance, antioxidant capacity and intestinal health of juvenile mirror carp (*Cyprinus carpio* var. specularis). *Aquaculture Nutrition*, 26, 432-443.