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ORIGINAL RESEARCH

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Effect of replacing fishmeal with spirulina (*Arthrospira platensis*) powder on growth, survival, body coloration, and meat quality in the freshwater prawn *Macrobrachium caementarius*

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Abstract

Microalgae are a valuable source of nutrients that are widely used in aquaculture to support the growth of aquatic species. This study aimed to assess the effect of replacing fishmeal with Spirulina powder (*Arthrospira platensis*) on the growth, survival, body coloration, and meat quality of *Macrobrachium caementarius*. We investigated the effects of diets containing different levels of fishmeal replaced by Spirulina powder: 0% (control), 25% (S25), 50% (S50), and 75% (S75), using 48 prawns during a 90-day experiment. Our findings indicated that the experimental diets containing Spirulina did not significantly contribute to the growth of the prawns, although the survival rate remained high. The highest ($p < 0.05$) dry hepatosomatic indices were observed in the prawns fed the S25 (2.85%) and S75 (2.56%) diets. We noted distinct color differences based on the diet: prawns fed the S25 exhibited a yellow-orange color, while those on the S50 and S75 diets developed a red-orange hue. Prawns of the S75 diet had significantly higher ($p < 0.05$) numbers (239.00/mm^2), indices (4.76), and coefficients of chromatophores (95.20). Prawns fed the S50 and S75 diets showed higher protein content but lower lipid levels compared to the control diet. Our findings highlight the potential of Spirulina powder to replace fishmeal in diet formulations without compromising prawn *M. caementarius* performance. Diets containing 50% and 75% Spirulina powders are recommended as a fishmeal substitute. These results could contribute positively to the developing aquaculture sector for this valued species.

KEYWORDS

animal nutrition, carotenoids, chromatophores, microalgae meal

1 | INTRODUCTION

Fishmeal (FM) is a key ingredient in aquaculture diets, known for its high levels of proteins, lipids, minerals, and vitamins (Cho & Kim, 2011), representing the main input in aquaculture diets. However, its elevated market value significantly raises production costs, accounting for 40%–60% of the total operating costs in shrimp farming (Sarman et al., 2018). In this context, there is an urgent need for sustainable and cost-effective protein alternatives (Mugwanya et al., 2023).

Microalgae are an excellent source of protein and lipids that can effectively replace FM and fish oil in the expanding aquaculture industry, where the need for sustainable feed ingredients is imperative (Ma & Hu, 2023). Two genera of cyanobacteria, *Arthrospira* and *Spirulina*, commonly referred to as "Spirulina" (Alagawany et al., 2021), are successfully incorporated into diets for fish, shrimp, crabs, and other aquatic organisms. This success is largely due to their high-quality nutrients and bioactive compounds (Ahmad, 2023; Alagawany et al., 2021).

The replacement of FM with *A. platensis* at different concentrations has been shown to effectively enhance the growth performance and survival rates of different crustaceans, including *Macrobrachium rosenbergii* (Radhakrishnan et al., 2016), *Penaeus* (*Penaeus*) *monodon* (Sivakumar et al., 2018), and *P.* (*Litopenaeus*) *vannamei* (Ashour et al., 2024).

Microalgae meal is a rich source of various pigments such as carotenoids, chlorophylls, and phycobiliproteins, which are essential for maintaining the body color of cultivated crustaceans (Chen et al., 2022; Maoka, 2020; Shapawi et al., 2017). In crustaceans, astaxanthin is typically found in complexes with proteins known as carotenoproteins (da Costa & Miranda-Filho, 2020). When these proteins undergo heat treatments, they are denatured, leading to a characteristic orange and deep orange body color (Santos-Sánchez et al., 2020), which is preferred by consumers (Parisenti et al., 2011). In the red swamp crayfish, *Procambarus clarkii*, the intensity of body color improved with the consumption of phycocyanin extracted from *A. platensis* (Yodngam & Hiransuchalert, 2024). Similarly, in the crab *Eriocheir sinensis*, an increase in carotenoid and astaxanthin levels in the carapace was observed when *Haematococcus pluvialis* microalgae powder was added to the diet, resulting in a more intense body color (Long et al., 2017).

Including microalgae in diets also enhances the nutritional quality of the crustaceans (Ashour et al., 2024; Radhakrishnan et al., 2016) by stimulating digestive enzyme activity, which leads to better nutrient absorption (Ashour et al., 2024). For example, *M. rosenbergii* that is fed a diet comprising 50% *Chlorella vulgaris* accumulates higher levels of protein, amino acids, carbohydrates, and lipids in its muscle (Radhakrishnan et al., 2015). In *M. rosenbergii* fed diets containing 50% *A. platensis*, increased enzyme activity is exhibited, resulting in improved nutrient assimilation and decreased feed conversion rates (Radhakrishnan et al., 2016).

The freshwater prawn, *M. caementarius*, is the most significant species in Peru, valued for its high commercial and culinary importance. This is largely due to its excellent meat quality and flavor, its large size, attractive coloration, and the thickness of its chelipeds (Fuentes et al., 2021). Additionally, it has a high nutritional value (Moreno-Reyes et al., 2015). *M. caementarius* is found in Peru and Chile, with the highest population densities located in Peru (Pinazo et al., 2020). The Peruvian populations of this species support an important artisanal fishery, with an estimated harvest of about 1000 t per year (PRODUCE, 2020).

Research on the nutrition of this prawn species is recent and primarily focused on growth (Acosta et al., 2018; Cornejo et al., 2015; Díaz et al., 2020; Gallardo-Carril & Reyes-Avalos, 2020; Terrones & Reyes, 2018) and the maintenance of body color (Fuentes et al., 2021), particularly in male prawns from experimental culture (Reyes, 2020). These studies used a basal diet with 30% crude protein, making it necessary to reduce the dependence on FM with inputs with high protein content. Microalgae are good candidates because they contain a high protein content (Apandi et al., 2019), fast growth rate, and high yield, although their high production costs remain a major challenge (Ma & Hu, 2023). However, the effects of dietary microalgae on the growth, coloration, and body chemical composition of *M. caementarius* have not yet been studied, despite the promising results obtained in related species such as *M. rosenbergii* (Radhakrishnan et al., 2016) and *P. (Litopenaeus) vannamei* (Macias-Sancho et al., 2014). Our hypothesis is that Spirulina replacement will have limited effects on growth but may improve coloration and meat quality of *M. caementarius*. Therefore, the aim of this research was to evaluate the effect of replacing FM with Spirulina powder (*A. platensis*) on the growth, survival, body coloration, and meat quality of *M. caementarius*.

2 | MATERIALS AND METHODS

2.1 | Experimental diets

Spirulina powder (*A. platensis*) was acquired from *K y B Natural E.I.R.L.*, Lima, Peru. It is dark green in color and contains 66.50% protein, 13.54% phycocyanin, 1.30% chlorophyll, 0.31% carotenoids, 6.10% moisture, 6.90% ash, 2 CFU/g total bacteria, and < 100 CFU/g yeast. It is certified to be free of total coliforms, *Staphylococcus*, *Salmonella*, and *Shigella*. The FM of the basal diet (control diet) formulated by Reyes-Avalos et al. (2023) was replaced by Spirulina powder in the proportions of 25% (S25), 50% (S50), and 75% (S75), conducted in triplicate. The procedure for preparing the diets was according to the protocol of Reyes-Avalos et al. (2024). The four diets were isoproteic and isolipidic. (Table 1). The organoleptic characteristics of aroma and color of the diets were determined according to Gallardo-Carril and Reyes-Avalos (2020). The proximate composition of the diets is shown in Table 1.

2.2 | Prawn capture, acclimatization, and selection

The prawns ($n = 60$) were collected from the Samanco River (9°15′18″ S, 78°29′42″ W) (Ancash, Peru) and transported in individual vessels according to the technique of Reyes-Avalos (2016). Land transport time lasted 40 min and there was no mortality. The acclimatization stage was conducted in the laboratory during 1 week. Each prawn was put in individual vessels installed inside aquariums (15 prawns/aquarium), where only the control diet was provided. Water changes (30%) were carried out every 2 days and sediment particles were removed from the aquariums. The prawns were identified at the species level with a taxonomic key (Méndez, 1981) and the recently accepted genus name *Macrobrachium caementarius* (DecaNet eds., 2025) was considered. Male prawns were screened based on the presence of a gonopore in each coxopodite of the fifth pair of pereopods. We used only male specimens because their body color is more intense than that of females (Terrones & Reyes-Avalos, 2022).

TABLE 1 Ingredients, physical and organoleptic characteristics, proximal composition (dry basis), and proportion of nutrients of diets with different proportions of Spirulina (S) powder used as fishmeal replacement.

Ingredients (%)	Control	S25	S50	S75
Fishmeal	30.00	22.50	15.00	7.50
Spirulina powder	0.00	7.50	15.00	22.50
Soybean meal	21.00	21.00	21.00	21.00
Corn flour	15.45	15.45	15.45	15.45
Paprika flour	0.25	0.25	0.25	0.25
Fish oil	2.00	2.00	2.00	2.00
Soybean oil	0.50	0.50	0.50	0.50
Corn oil	0.50	0.50	0.50	0.50
Soy lecithin[a]	1.00	1.00	1.00	1.00
Rice powder	20.00	20.00	20.00	20.00
Molasses	2.70	2.70	2.70	2.70
Zeolite	4.00	4.00	4.00	4.00
Common salt	2.00	2.00	2.00	2.00
Vitamins and minerals[b]	0.30	0.30	0.30	0.30
Butylated hydroxytoluene	0.10	0.10	0.10	0.10
Sorbic acid	0.20	0.20	0.20	0.20
Total	100.00	100.00	100.00	100.00
Physical characteristics				
Aroma	Fish	Seaweed	Seaweed	Seaweed
Color	Brown	Green	Dark green	Dark green
Flavor	Salty/Fish	Salty/Fishy	Salty/Fishy	Salty/Bitter
Proximate composition (%)				
Crude protein	29.67	29.85	32.19	33.08
Crude lipid	7.83	8.11	7.92	8.53
Ash	8.39	9.14	8.23	8.54
Crude fiber	2.36	2.19	1.86	1.72
Carbohydrates	51.75	50.71	49.80	48.13
Carbohydrates/Lipid	6.99	6.25	6.29	5.64

Note: S25, S50, and S75: Diets with 25%, 50%, and 75% of Spirulina (S) powder used as fishmeal replacement.
[a]Purified soy lecithin (1200 mg soft capsules with phosphatides ≥250 mg).
[b]Each 100 g contains: Vitamin B1 (Thiamine) 500 mg, vitamin B2 (Riboflavin) 1200 mg, vitamin B6 (Pyridoxine) 900 mg, vitamin B12 (Cyanocobalamin) 1000 μg, biotin 2 mg, nicotinamide 2000 mg, calcium pantothenate 1000 mg, sodium chloride 2000 mg, potassium chloride 8000 mg, magnesium sulfate 1200 mg.

2.3 | Culture and feeding

The prawn culture system consisted of 12 fiberglass tanks (60 × 42 × 45 cm and 100 L), each implemented with a biological trickling filter (2 L) driven by an air-water-lift (1.5 L/min). Four individual prawn culture vessels (19 cm diameter, and 8 cm high, 284 cm^2) were conditioned in each tank, according to the arrangement previously described by Reyes-Avalos et al. (2023). The prawns were randomly assigned individually in each culture vessel (4 prawns/tank = 16 prawns/m^2). All the prawns selected for stocking ($n = 48$) were of similar total weight (6.95 ± 1.49 g), and all possessed complete cephalothoracic appendages and showed no signs of body or cheliped lacerations. The prawns were fed twice a day (40% at 08:00 h and 60% at 18:00 h) and the feeding level was 5% of wet weight, which was adjusted every 30 days after sampling. The feeding was not adaptive, because molts are not synchronous. The experiment lasted 90 days and during this period all diets were accepted by the prawns.

2.4 | Growth and survival parameters

The total weight (g) of the prawns ($n = 4$ per aquarium) was obtained on the ADAM AQT 600 digital balance (± 0.01 g), and the parameters were calculated with the following formulas:

$$\text{Weight gained (WG, \%)} = [(\text{final weight} - \text{initial weight})/\text{initial weight}] \times 100$$

$$\text{Absolute growth rate (AGR, g/day)} = (\text{final weight} - \text{initial weight})/\text{day cultivation}$$

$$\text{Specific growth rate (SGR, \%/day)} = [(\ln \text{final weight} - \ln \text{initial weight})/\text{day cultivation}] \times 100$$

$$\text{Survival rate (SR, \%)} = (\text{final number prawn}/\text{initial number prawn}) \times 100$$

2.5 | Feed utilization indices

$$\text{Feed conversion rate (FCR)} = \text{feed intake}/\text{biomass gained}$$

$$\text{Feed conversion efficiency (FCE, \%)} = (1/\text{FCR}) \times 100$$

$$\text{Protein efficiency ratio (PER)} = \text{weight gain}/\text{protein intake}$$

2.6 | Organosomatic indices

Organosomatic indices and moisture content of prawn organs ($n = 4$ per treatment) were calculated according to Fotedar (1998). The hepatopancreas and tail muscle were oven dried (100°C for 24 h). Samples were weighed on a JA303P digital balance (± 0.001 g).

$$\text{Tail muscle moisture (TM, \%)} = [(\text{Tw} - \text{Td}) \times 100]/\text{Tw}$$

$$\text{Wet tail muscle indices (TMiw)} = (\text{Tw} \times 100)/\text{BW}$$

$$\text{Dry tail muscle indices (TMid)} = (\text{Td} \times 100)/\text{BW}$$

$$\text{Hepatopancreas moisture (HM, \%)} = [(\text{Hw} - \text{Hd}) \times 100]/\text{Hw}$$

$$\text{Wet hepatosomatic index (HIw, \%)} = (\text{Hw} \times 100)/\text{BW}$$

$$\text{Dry hepatosomatic index (HId, \%)} = (\text{Hd} \times 100)/\text{BW}$$

$$\text{Fulton's K condition indices (K, \%)} = \left(\text{BW}/\text{TL}^3\right) \times 100$$

where: Tw: Tail muscle wet weight; Td: Tail muscle dry weight; Hw: Hepatopancreas wet weight; Hd: Hepatopancreas dry weight; BW: Body weight; TL: Total length.

2.7 | Epidermal chromatophores

The dorsal area of the second abdominal segment of the live prawns ($n = 4$ per treatment) was chosen to photograph the epidermal chromatophores. Photographs were taken with a digital camera (Sony APS-C a6100, 24.2 megapixels) mounted on the eyepiece of a Nikon stereoscope (4X). The digital photographs were analyzed on a computer to count the number of chromatophores/mm^2. The chromatophore index (CI) was determined taking into account the five chromatophore dispersion stages according to Hogben and Slome (1931) (Figure 1) and the following formula was used:

$$\text{CI} = [\Sigma(\text{chromatophore dispersion stage} \times \text{number of chromatophores in a specific stage of dispersion})] / \text{total number of chromatophores observed}$$

The chromatophore coefficient (ChC) was determined with the formula of Ford and Feuerhelm (2020):

$$\text{ChC} = ([\text{n1}/\text{Tc}] \times 20) + ([\text{n2}/\text{Tc}] \times 20) + ([\text{n3}/\text{Tc}] \times 20) + ([\text{n4}/\text{Tc}] \times 20) + ([\text{n5}/\text{Tc}] \times 20)$$

where n1 to n5 is the number of chromatophores in stages 1 to 5 of an individual. This value is divided by the total number of chromatophores (Tc) and multiplied by 20. After calculation of n1 to n5, these values are summed to generate the chromatophore score, which should be between 20 (very light color) and 100 (very dark color).

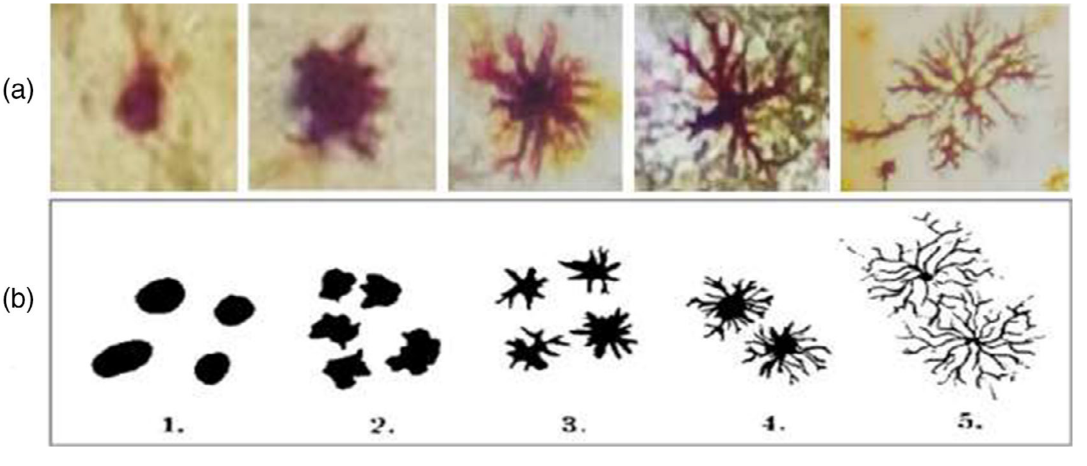

FIGURE 1 (a) Pigment dispersion stage of chromatophores of *M. caementarius*. (b) Melanophore index of the frog *Xenopus laevis*, according to Hogben and Slome (1931), being state 1 small and with pigment concentrated in the chromatophore, up to state 5, which is large and with pigment completely dispersed in the stellate chromatophore.

2.8 | Body coloration

Following the methodology of Fuentes et al. (2021), the prawns ($n = 4$ per treatment) were boiled (100°C for 2 min) and the body color was determined by capturing digital images with a Sony APS-C a6100 digital camera (24.2 megapixels), according to the methodology of Terrones and Reyes-Avalos (2022). The image was analyzed with the RGB (red, green, and blue) color mode of the Adobe Photoshop 2020 program. In addition, the color index (IColor) of the body was determined according to Díaz-Jiménez et al. (2018):

$$\text{IColor} = (\text{R} + \text{G} + \text{B})/765$$

where R, G, and B were assigned values from 0 to 255, obtained with Adobe Photoshop, and 765 is a constant resulting from the sum of the maximum RGB values.

2.9 | Proximal analysis of diets and prawn muscle

Proximal chemical analysis of the diets and prawn muscle (1 per treatment) was carried out at the Laboratorio Físico Químico Ambiental Perú S.A.C (FQA PERÚ), according to the Peruvian Technical Standard (NTP, https://www.inacal.gob.pe/cid/categoria/normas-tecnicas-peruanas), which included crude protein (NTP 205.005/79), crude lipid (NTP 205.006/80), crude fiber (NTP 205.002/79), ash (NTP 205.004/79), and moisture (NTP 205.002/79). For carbohydrates, the formula by difference was used: 100 – (% protein + % fat + % fiber + % ash).

2.10 | Water quality

The water quality parameters were similar ($p > 0.05$) among dietary treatments. Water temperature averaged (24.21 ± 0.13°C) was recorded using a digital thermometer. Dissolved oxygen (8.03 ± 0.19 mg/L) was determined with a Hanna HI 9146 Oximeter. The pH (7.98 ± 0.24 units) was measured with a PH-222 digital pH meter. Total ammonia nitrogen (0.12 ± 0.09 mg/L), ammonia nitrogen (0.06 ± 0.05 mg/L), ammonium nitrogen (0.06 ± 0.05 mg/L), nitrite nitrogen (0.18 ± 0.43 mg/L), and nitrate nitrogen (4.31 ± 1.94 mg/L) were measured once a week with a commercial kit (Hanna®), whose reading was carried out with the Hanna® multiparameter photometer HI 83399.

2.11 | Statistical analysis

The data were processed using a completely randomized statistical design and the Shapiro–Wilk normality test and Levene's test were performed. Differences between treatment means were determined by one-way analysis of variance (ANOVA) and Duncan's test, with a significance level of $p < 0.05$. The number, index, and coefficient of epidermal chromatophores were evaluated by Pearson correlation with significance of $p < 0.01$. The analyses were determined with the SPSS software version 26 for Windows and the results were expressed as mean ± standard deviation.

3 | RESULTS

3.1 | Growth and survival

The obtained growth parameters, feed utilization, and organosomatic indices are shown in Table 2. Weight growth prawn (Figure 2), feed utilization indices, the condition index K, and survival were similar ($p > 0.05$) among the

TABLE 2 Growth, survival, feed utilization, and organosomatic indices of *M. caementarius* fed diets containing different proportions of Spirulina (S) powder used as fishmeal replacement.

Parameters	Control	S25	S50	S75
Growth and survival				
Initial weight (g)	7.25 ± 1,94	6.07 ± 0,43	6.90 ± 1,35	6.76 ± 0,83
Final weight (g)	9.22 ± 1,53	8.20 ± 1.10	9.71 ± 2.44	8.34 ± 1, 12
Weight gained (WG, %)	29.92 ± 15.61	34.79 ± 11.63	39.54 ± 9.74	33.69 ± 9.19
Absolute growth rate (AGR, g/day)	0.022 ± 0.006	0.024 ± 0.009	0.031 ± 0.012	0.018 ± 0.007
Specific growth rate (SGR, %/day)	0.286 ± 0.133	0.329 ± 0.098	0.368 ± 0.079	0.234 ± 0.084
Survival rate (SR, %)	58.33 ± 14.43	66.67 ± 14.43	66.67 ± 14.43	66.67 ± 14.43
Feed utilization				
Feed conversion rate (FCR)	2.70 ± 1.16	3.62 ± 3.03	1.90 ± 0.96	7.26 ± 5.94
Feed conversion efficiency (FCE, %)	42.57 ± 19.93	40.70 ± 23.89	67.76 ± 45.84	25.34 ± 24.02
Protein efficiency ratio (PER)	0.18 ± 0.35	0.08 ± 0.68	0.37 ± 0.95	0.16 ± 0.67
Organosomatic indices				
Tail muscle moisture (TM, %)	76.46 ± 3.52	76.62 ± 4.69	76.39 ± 2.34	76.79 ± 5.08
Wet tail muscle indices (TMiw)	19.38 ± 1.17	17.60 ± 4.42	18.00 ± 4.54	21.56 ± 4.32
Dry tail muscle indices (TMid)	4.52 ± 0.40	4.16 ± 1.55	4.22 ± 0.97	4.91 ± 1.04
Hepatopancreas moisture (HM, %)	51.29 ± 18.78	46.66 ± 5.35	62.14 ± 7.69	44.58 ± 4.28
Wet hepatosomatic index (HIw, %)	4.72 ± 1.24^{ab}	5.27 ± 0.84^{b}	3.04 ± 1.02^{a}	4.66 ± 1.09^{ab}
Dry hepatosomatic index (HId, %)	2.29 ± 1.15^{ab}	2.85 ± 0.29^{b}	1.17 ± 0.48^{a}	2.56 ± 0.44^{b}
Fulton's K condition indices (K, %)	3.97 ± 0.28	3.88 ± 0.64	4.34 ± 0.11	4.52 ± 1.13

Note: S25, S50, and S75: Diets with 25%, 50%, and 75% of Spirulina (S) powder used as fishmeal replacement. Data are expressed as the mean ± standard deviation ($n = 3$). Data with different superscript letters in the same row indicate a significant difference ($p < 0.05$).

dietary treatments, compared to the control diet. Mortality was due to unknown causes and difficulty in completing the ecdysis. The organosomatic indices, HIw and Hid, were significantly lower ($p < 0.05$) only with the S50 diet, relative to the other diets.

3.2 | Epidermal chromatophores

Significantly higher ($p < 0.05$) values of chromatophore number, CI, and ChC were obtained only in the prawns fed the S75 diet, but the correlation was highly significant (Table 3).

3.3 | Body coloration

The color of chromatophores was mainly yellow-orange in prawns fed the control diet and the S25 diet, whereas red-orange colored prawns were observed with the S50 (RGB = 213–137–70) and S75 (RGB = 204–135–82) diets. However, some blue chromatophores were also observed in smaller proportions, showing a direct relation with increasing Spirulina powder concentration in the diets (Figure 3a). The color of boiled prawns was directly related to the appearance of their chromatophores (Figure 3b). The RGB values indicated shades ranging from light yellow-orange (240–176–81) to red-orange (204–135–82) (Figure 3c).

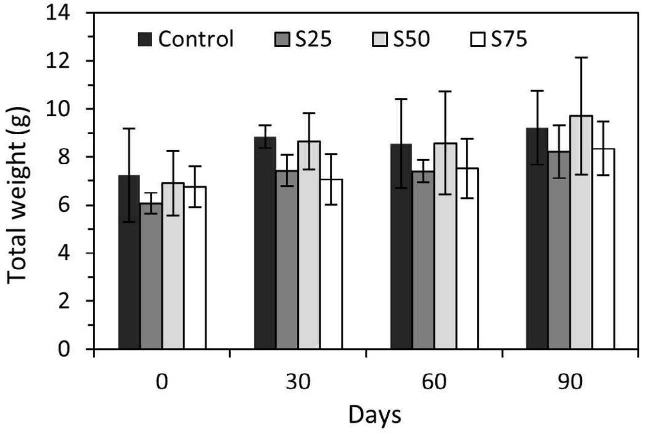

FIGURE 2 Weight growth of *M. caementarius* prawn over culture time. Data are expressed as the mean ± standard deviation ($n = 3$).

TABLE 3 Number, index, and coefficient of epidermal chromatophores of *M. caementarius* fed different proportions of Spirulina (S) powder used as fishmeal replacement.

Chromatophore	Control	S25	S50	S75	*r*
Number chromatophore/mm^2	88.50 ± 14.85^a	92.00 ± 8.49^a	122.50 ± 33.23^a	239.00 ± 26.87^b	0.851**
Chromatophore index (CI)	2.85 ± 0.92^a	3.46 ± 0.20^a	4.12 ± 0.30^a	4.76 ± 0.10^b	0.898**
Chromatophore coefficient (ChC)	56.90 ± 18.24^a	69.20 ± 3.96^a	82.40 ± 5.94^a	95.20 ± 1.98^b	0.899**

Note: S25, S50, and S75: Diets with 25%, 50%, and 75% of Spirulina (S) powder used as fishmeal replacement.
**The correlation is highly significant ($p < 0.01$). Data are expressed as the mean ± standard deviation ($n = 3$). Data with different superscript letters in the same row indicate a significant difference ($p < 0.05$).

The RGB values of the cephalothorax and abdomen decreased ($p < 0.05$) with increasing amounts of Spirulina powder in the diets, with red being the highest, and green and blue being the lowest (Table 4). IColor was significantly lower ($p < 0.05$) with the diets containing Spirulina powder compared to the control diet (Table 4).

3.4 | Meat quality

The protein and fiber from the prawn muscle increased as the concentration of Spirulina powder in the diet increased; in contrast, the opposite was true for lipids, ash, and carbohydrates (Table 5).

4 | DISCUSSION

4.1 | Growth and survival

This study found that the diets containing different proportions of Spirulina powder did not affect the growth, feed utilization, or survival rates of *M. caementarius*, even though the water quality in each dietary treatment was similar to that of the natural environment (Zacarías & Yépez, 2015). Genetic diversity varies among prawn individuals

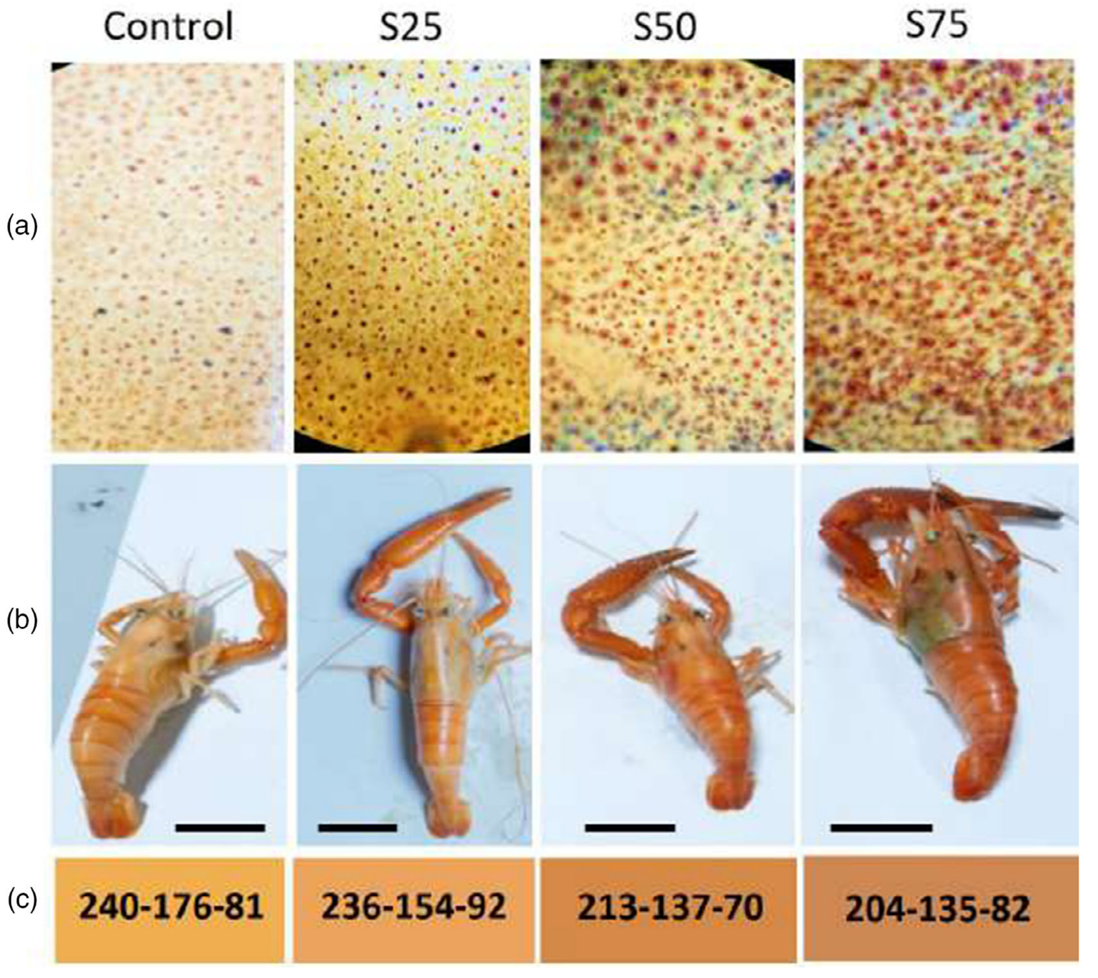

FIGURE 3 *M. caementarius* prawn fed control, S25, S50, and S75 diets. (a) Epidermal chromatophores of the dorsal area of the second abdominal segment, observed in live prawn with stereoscope at 4X. (b) Body color of boiled prawns from each treatment. (c) Average RGB of the cephalothorax and abdomen; the color of each rectangle represents the color tone. Bar = 2 cm.

(Zhang et al., 2024), which can influence our experimental outcomes. However, prawn mortality due to difficulty completing ecdysis has been reported in this species (Reyes, 2018) and attributed to lipid reduction (Acosta et al., 2018; Reyes-Avalos et al., 2023). Notably, with the S50 diet, the GP and SGR were interesting biologically, but not statistically supported. These findings were similar to those reported for the same prawn species, although they depend on the type of stimulus used (Acosta et al., 2018; Reyes-Avalos et al., 2024). However, they diverge somewhat from related studies involving other crustacean species. For instance, in *M. rosenbergii*, a diet containing 50% *A. platensis* resulted in significant improvements in growth parameters and feed utilization while maintaining a high survival rate (Radhakrishnan et al., 2016). Similarly, in *P. (Litopenaeus) vannamei*, even a low inclusion level (0.80%) of *A. platensis* powder in the diet significantly increased body weight and TCE (Ahmed et al., 2025). Furthermore, incorporating 1% lipid-free *A. platensis* biomass in the diet resulted in superior feed efficiency for *P. (Litopenaeus) vannamei* (Ashour et al., 2024).

FCR, PER, and SGR are sensitive indicators of protein quality of a diet (Goytortúa-Bores et al., 2006; Tacon, 1989). In the present research, these indicators showed improvement in *M. caementarius* fed the S50 diet,

TABLE 4 Body color (cephalothorax and abdomen) of *M. caementarius* fed diets containing different proportions of Spirulina (S) powder used as fishmeal replacement, according to RGB, and color index (IColor) values.

Body color	Control	S25	S50	S75
Cephalothorax				
Red (R)	246.00 ± 2.83^{b}	216.00 ± 5.66^{a}	227.00 ± 5.66^{ab}	208.50 ± 14.85^{a}
Green (G)	187.00 ± 26.87^{b}	152.00 ± 4.24^{ab}	141.00 ± 7.07^{ab}	108.00 ± 16.97^{a}
Blue (B)	64.50 ± 23.33	62.50 ± 13.14	42.00 ± 15.56	35.50 ± 4.95
IColor	0.65 ± 0.06^{b}	0.56 ± 0.03^{ab}	0.54 ± 0.04^{ab}	0.46 ± 0.05^{a}
Abdomen				
Red (R)	246.50 ± 7.78^{b}	214.00 ± 2.83^{a}	222.50 ± 3.54^{a}	215.50 ± 6.36^{a}
Green (G)	155.00 ± 26.87	131.50 ± 3.54	123.00 ± 5.66	117.00 ± 11.31
Blue (B)	72.00 ± 18.38^{b}	53.50 ± 4.95^{ab}	38.50 ± 0.71^{a}	36.00 ± 1.41^{a}
IColor	0.62 ± 0.05^{b}	0.52 ± 0.01^{a}	0.50 ± 0.01^{a}	0.48 ± 0.02^{a}

Note: S25, S50, and S75: Diets with 25%, 50%, and 75% of Spirulina (S) powder used as fishmeal replacement. Data are expressed as the mean ± standard deviation ($n = 3$). Data with different superscript letters in the same row indicate a significant difference ($p < 0.05$).

TABLE 5 Proximal chemical composition (dry basis) determined in the muscle of *M. caementarius* fed diets containing different proportions of Spirulina (S) powder used as fishmeal replacement.

Proximate composition	Control	S25	S50	S75
Moisture (%)	2.41	3.23	3.11	3.06
Crude protein (%)	21.06	23.18	24.00	24.37
Crude lipid (%)	6.89	6.64	6.57	6.48
Ash (%)	7.86	7.74	7.70	7.65
Crude fiber (%)	2.19	2.71	2.54	2.61
Carbohydrates (%)	62.00	59.73	59.19	58.89

Note: S25, S50, and S75: Diets with 25%, 50%, and 75% of Spirulina (S) powder used as fishmeal replacement.

although this finding lacked statistical significance. This behavior may be related to the pronounced organoleptic characteristics of the diet, which likely promoted better consumption, digestion, and assimilation. Diets containing low proportions of Spirulina meal have proven to be effective dietary attractants for other crustacean species, including *P. (Litopenaeus) vannamei* (Ahmed et al., 2025; Silva-Neto et al., 2012). Additionally, the increase in PER observed with the S50 diet, although not statistically significant compared to the other diets, suggests that prawns from this dietary group could have improved protein utilization, leading to enhanced growth. This suggests that carbohydrates served as the primary energy source for the normal metabolism of the prawns fed the S50 diet. This species is renowned for its ability to effectively utilize a high carbohydrate content (47%) in its diet for energy (Gallardo-Carril & Reyes-Avalos, 2020), demonstrating high digestibility (93%) (Baltodano et al., 2022). For *M. amazonicum*, maintaining a balanced intake of protein and energy is vital to maximize growth, as dietary energy levels can influence overall performance (Ballester et al., 2023). Therefore, it is essential to determine the optimal protein requirements to maximize growth in *M. caementarius*.

The hepatopancreas serves as the main storage organ for enzymes and nutrients, supplying the energy needs for the metabolism of various organisms, including crustaceans (Vogt, 2019). In our study, the *M. caementarius* individuals fed the S50 diet exhibited an increase in HM, although this was not statistically supported. This suggests a depletion of energy reserves in the hepatopancreas. This observation is consistent with findings from research on the shrimp *P. (Oleopenaeus) latisulcatus* (Sang & Fotedar, 2004), which reported lower HIw and HId values with a S50 diet, whereas different diets resulted in higher HIw and Hid values. Although the improvements in growth and feed utilization in *M. caementarius* fed the S50 diet were not statistically significant, they can be attributed to enhanced nutrient availability and improved digestibility of this diet.

On the other hand, *M. caementarius* fed the S75 diet exhibited the lowest growth parameters and poor feed utilization. However, there were no significant differences compared to the other diets. Interestingly, the prawns in this dietary group showed a small but notable increase in TMiw, TMid, and K-index values, which was an unexpected finding. It is known that high TMiw and TMid values in *Cherax tenuimanus* are linked to efficient nutrient absorption and subsequent energy storage in their abdominal muscles (Ambas et al., 2013; Sang et al., 2011). Therefore, the effects of the S75 diet observed in our results suggest that the excessive amount of Spirulina powder in this diet may have negatively affected protein quality and, as a result, the growth of the prawns. Nonetheless, the high FCR obtained with these diets affects the practical viability of Spirulina as a replacement for FM in diets for *M. caementarius*. A similar effect was also observed in *P. (Litopenaeus) vannamei* when fed a diet containing more than 20% green water meal (Shapawi et al., 2017), which led to poor growth attributed to deficiencies in lysine and tryptophan, coupled with low digestibility of such diet (Shapawi et al., 2017).

In our study, we found that the high levels of pigments in the S75 diet, particularly phycocyanin, may have negatively affected the prawn's nutrient metabolism. Similar effects have been observed in various crustaceans that fed diets supplemented with high levels of Spirulina. For example, a diet containing excessive *A. platensis* powder negatively affected the growth performance of *M. rosenbergii* (Radhakrishnan et al., 2016). Additionally, in *P. clarkii*, it has been suggested that phycocyanin from *A. platensis* could alter metabolic pathways, thereby influencing growth (Yodngam & Hiransuchalert, 2024). Conversely, it has been reported that up to 75% of FM can be replaced by *A. platensis* without adversely affecting survival or causing growth depression in *P. (Litopenaeus) vannamei* (Ahmed et al., 2025).

4.2 | Body coloration

The Spirulina powder used in this study had a high concentration of phycocyanin, while it contained low levels of chlorophyll and carotenoids. Phycocyanin is a bright blue phycobiliprotein found in cyanobacteria (Dagnino-Leone et al., 2022). Typically, male *M. caementarius* specimens exhibit a standard coloration of light and dark brown, with the dactyl and fixed finger of their chelipeds showing a light blue hue (Terrones & Reyes-Avalos, 2022). However, in the present study, we observed low levels of blue chromatophores in the prawn bodies. These levels increased with higher concentrations of Spirulina powder in the diets, which may explain the accumulation of phycocyanin in certain chromatophores within the prawn's body.

In the body of *M. caementarius*, the main color observed in the chromatophores was yellow-orange and red-orange. This indicates a higher conversion and accumulation of carotenoids and other pigments. Spirulina is rich in carotenoids and zeaxanthin (Yu et al., 2012), and in fish zeaxanthin can be converted to astaxanthin by the idoxanthin and adonixanthin pathways (Maoka, 2020). The latter pathway is also present in many crustaceans (Maoka, 2011). *M. caementarius* prawns rapidly metabolize dietary carotenoids (Fuentes et al., 2021), and they likely also metabolize zeaxanthin from Spirulina powder, given that astaxanthin is often bound to a multimeric protein complex called crustacyanin (Wade et al., 2012). This carotenoprotein complex masks purple, yellow, and blue colors (Hossain & Shahidi, 2024), contributing to the diverse range of colors seen in the crustacean exoskeleton (Hamdi et al., 2020). The role of dietary phycocyanin in the coloration of *M. caementarius* requires further investigation. The astaxanthin found in the exoskeleton and hypodermal tissue of crustaceans accounts for over 90% of the total carotenoids in their bodies (Wade et al., 2012).

Carotenoproteins are denatured by heat, which allows the visualization of the characteristic red color of astaxanthin (Fuentes et al., 2021; Terrones & Reyes-Avalos, 2022). In boiled male *M. caementarius* prawns, the most intense reddish hue in the body was observed with the S75 diet, where the pigments from Spirulina powder are concentrated in the pigmented layer of the cuticle and in the chromatophores of the male specimens (Fuentes et al., 2021). Similar results were observed in male specimens of *M. amazonicum* (Costa et al., 2022) and *M. caementarius* (Terrones & Reyes-Avalos, 2022), where a higher concentration of astaxanthin pigments was detected in the abdominal muscles and carapace due to the lower demand for pigments for reproduction. In *P. (Litopenaeus) vannamei*, the intense red-orange color of the cooked body results from a diet that includes green water meal (Shapawi et al., 2017).

The RGB values of boiled *M. caementarius* bodies decreased proportionally with increasing levels of Spirulina powder in the diet. This resulted in a more intense yellow-orange color when the prawns were fed the S50 and S75 diets, as evidenced by the lower IColor values. According to Díaz-Jiménez et al. (2018), this color represents the most intense hue observed in the body of *Lysmata wurdemanni*.

Recent research has shown that in male specimens of *M. caementarius*, body color varies with the stage of the molting cycle (Terrones & Reyes-Avalos, 2022). In the present study, we used male prawns in the intermolt stage and observed that the number of chromatophores, the CI, and the chromatophore coefficient increased with the addition of Spirulina powder to their diet. Although these parameters were significantly higher only with the S75 diet, a strong positive correlation was found among the different dietary treatments. These findings suggest that the pigments in Spirulina powder stimulated the proliferation and dispersion of epidermal chromatophores, resulting in the enhanced red-orange coloration of *M. caementarius*, as a redder coloration increases marketability. Similarly, *N. davidi* fed with the microalgae *H. pluvialis* and *A. platensis*, also exhibited increased chromatophore dispersion and chromatophore coloration intensity (Luna-Vivaldo et al., 2024). However, our results contrast with those reported by Fuentes et al. (2021), who found that an increase in carotenoids in the diet did not affect the condition of the epidermal chromatophores in *M. caementarius*. This discrepancy can be attributed to the feeding period, which in their research was 30 days, while in the present study, it was extended to 90 days.

4.3 | Meat quality

In the present study, the inclusion of Spirulina powder in the diets resulted in increased protein accumulation in the muscle of male *M. caementarius* prawns. These results suggest that the high protein concentration of Spirulina powder was effectively metabolized and stored in the muscle tissues of the prawns. Similar results have been observed in other species, such as *M. rosenbergii* (Radhakrishnan et al., 2016), and *P. (Litopenaeus) vannamei* (Ahmed et al., 2025; Ashour et al., 2024), which were fed varying proportions of *A. platensis* meal in their diet. The unique structure of this microalga, characterized by a thin and fragile cell wall, allows for the release of up to 75% of its proteins (Safi et al., 2014). In contrast, a diet containing phycocyanin extracted from *A. platensis* did not affect the protein levels in *P. clarkii* (Yodngam & Hiransuchalert, 2024).

The results of this research indicate that including Spirulina powder in the diets of *M. caementarius* reduced the lipid and carbohydrate content in their muscle. This suggests that dietary proteins were not utilized as an energy source; instead, carbohydrates (CHO) and lipids (LIP) from these diets supported the prawn's metabolism. This conclusion is reinforced by the fact that *M. caementarius* uses a high carbohydrate content of its diet serving as an energy source (Gallardo-Carril & Reyes-Avalos, 2020). However, the specific level of lipids used in the metabolism of this species remains unknown. In the experimental diets, there was a lower accumulation of lipids in the muscle and a low CHO/LIP ratio. In contrast, the control diet, which had a higher CHO/LIP ratio, resulted in an increased concentration of lipids in the prawn muscle, suggesting that lipogenesis was favored with this diet. In *P. (Litopenaeus) vannamei*, increasing dietary carbohydrate levels leads to a greater accumulation of lipid reserves in muscle (Ruvalcaba-Márquez et al., 2021; Zainuddin & Aslamyah, 2014). In *M. rosenbergii*, the body lipid level decreased in individuals fed diets containing 75% and 100% *A. platensis*. This reduction is primarily attributed to the lower lipid content in *A. platensis* compared to FM (Radhakrishnan et al., 2016). In contrast, *P. clarkii* showed an increase in lipid content when fed a diet containing phycocyanin extracted from *A. platensis*, due to the lipid fraction present in the phycocyanin extract itself (Yodngam & Hiransuchalert, 2024). Consequently, the results of the higher protein content and lower lipid content of the experimental diets indicate improved product quality for the benefit of consumer health.

5 | CONCLUSION

The main result of this study is the possibility of replacing FM with spirulina in diet formulations without compromising prawn (*M. caementarius*) performance. However, it is important to highlight that feeding diets containing 50% and 75% spirulina powders as a FM substitute led to an increase in protein content associated with body coloration. In contrast, lipid and carbohydrate content in prawn muscle decreased. Further research on amino acid and fatty acid profiles is needed to confirm the nutritional quality of prawns reared on these dietary treatments.

CONFLICT OF INTEREST STATEMENT

The authors declare that they have no conflict of interest.

DATA AVAILABILITY STATEMENT

All data generated or analyzed during this study are included in this published article.

ANIMAL WELFARE

All experiments performed in this study were conducted in accordance with the Peruvian Animal Protection and Welfare Law (Law 30,407, Ley de Protección y Bienestar Animal), and every effort was made to minimize suffering during the exposure of individuals to nutritional stress.

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