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Impact of fishmeal replacement with *Arthrospira platensis* on growth performance, body composition and digestive enzyme activities of the freshwater prawn, *Macrobrachium rosenbergii*

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ABSTRACT

In this study, we assessed the suitable level of replacement of fishmeal with a blue green microalga, *Arthrospira platensis* in feed for the post larvae (PL) of the freshwater prawn, *Macrobrachium rosenbergii* by evaluating the growth performance, prawn proximate composition, feed utilization parameters and the activity of digestive enzymes. The prawns were fed 5 different diets: a control diet and 4 different diets containing *A. platensis* at various levels such as 25%, 50%, 75%, 100%. These diets were fed to the PL for 90 days in triplicates. The growth performance in terms of weight gain, specific growth rate and feed efficiency ratio were found significantly ($P < 0.05$) higher in the prawns fed with the 50% of *A. platensis* feed fed group. At this level of 50% replacement, the prawn proximate composition, such as total protein, amino acids, carbohydrates and lipid contents were significantly ($P < 0.05$) higher than the control. Similarly, feed utilization parameters, such as feeding rate, absorption rate and conversion rate were significantly ($P < 0.05$) higher than the control. Added to that, the activity of the digestive enzymes such as protease, amylase and lipase showed the same trend at the level of 50% replacement. The hierarchy of the growth performance in prawns corresponds to 50 > 25 > 75 > 100% replacement of fishmeal with *A. platensis*. These results concluded that a partial replacement of the fishmeal with *A. platensis* at the level of 50% is beneficial for the growth of prawn *M. rosenbergii*.

1. Introduction

The giant freshwater prawn, *M acrobrachium rosenbergii* known as 'scampi' in commercial parlance, is a highly valued delicious food and commands a very good demand in both domestic and export market. *M. rosenbergii* culture is gradually gaining momentum in the present era owing to its price, taste, fast growth rate, less susceptibility to diseases and its compatibility to grow with carps (New, 2005; Radheyshyam, 2009). It has a wide distribution throughout the Indo-Pacific region and most favoured for farming in tropical and subtropical areas of the world (New, 2002, 2005). This freshwater prawn is farmed chiefly in small to medium-sized earthen ponds in West Bengal, Andhra Pradesh, Tamil Nadu and Kerala in India (Nair and Salin, 2012). Chand et al., 2015 reported and suggested that salinity plays a significant role in the culture of *M. rosenbergii* and the species showed satisfactory growth and survival at wide salinity range (0–15 ppt) and *M. rosenbergii* can be considered as an ideal species to promote. Global production of this prawn has increased from 130,689 t in 2000 to 203,211 t in 2011 (FAO, 2013). The total scampi production from India in 2010–2011 was about 8778 metric tonnes and West Bengal was the leading producer. In 2011–12, India exported 2723 metric tonnes *M. rosenbergii* with an increase of 31.61% than the previous years (MPEDA, 2011).

Paralleling the growth of the aquaculture industry, there has been an expansion in feed production (Tacon and Savas, 2000). Fishmeal (FM) as well as other marine meals are often included in the aquatic feeds as they are considered to be the excellent sources of high quality proteins, highly unsaturated fatty acids (HUFA), vitamins, minerals and attractants (Tacon and Akiyama, 1997; Gatlin et al., 2007; Tacon and Metian, 2008; Naylor et al., 2009). Due to these properties, FM has become one of the primary components of commercial feed formulations. Although, worldwide FM produc-

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tion has attained a relatively stable level, it still could not match the rapid worldwide development of aquaculture (Goytortua-Bores et al., 2006). Due to the limited supply of fishmeal and oil from wild catches, the efficient use and sharing of these products is a major issue for the aquaculture industry (Kaushik and Troell, 2010; Tacon and Metian, 2009). The fish in-fish out ratio (FIFO) is the efficiency at which the aquaculture converts a weight-equivalent unit of wild fish into a unit of cultured fish. Aquaculture converts 65% of the wild fish into fishmeal at a FIFO ratio of between 0.66 (Jackson, 2009; Kaushik and Troell, 2010) and 0.7 (Tacon and Metian, 2008). However, as a result of decreasing the supply of fishery byproducts and various concerns over its quality, the aquaculture industry is now actively investigating the alternative protein sources to be added to the feed.

The need for alternative protein sources to fishmeal has brought attention to many products that are local or regional in nature (Nyina-wamwiza et al., 2012), including single cell proteins, such as yeasts, bacteria, and microalgae (Mukhopadhyay and Ray, 1999; Ng et al., 2002; Skrede et al., 2002; Bairagi et al., 2004; Refstie et al., 2005; Hemaiswarya et al., 2011). In order to be used in aquaculture, a micro algal strain has to meet various criteria, such as ease of culturing, lack of toxicity, high nutritional value with correct cell size and shape and a digestible cell wall to make nutrients available (Raja et al., 2004; Patil et al., 2007). Protein and vitamin content is a major factor determining the nutritional value of microalgae. In addition to it, Polyunsaturated fatty acids (PUFA) e.g. eicosapentaenoic acid (EPA), arachidonic acid (AA) and docosahexaenoic acid (DHA) content is of major importance for an aquaculture organism. Different strategies were being practiced to improve the polyunsaturated fatty acids content in microalgae (Guedes et al., 2010). Also, the crude protein extracts of microalgae rich in amino acids, including the essential amino acids (EAA) isoleucine, leucine, lysine, methionine, phenylalanine, and valine (Becker, 2007; Uslu et al., 2011; Mahboob et al., 2012). It is often advised to use mixed microalgae cultures in order to have a good protein profile, adequate vitamin content and high polyunsaturated fattyacids, mainly EPA, AA and DHA, as they are considered to be essential for the survival and growth during the early stages of life of many marine animals (Volkman et al., 1989). One of the beneficial effects attributed to adding algae is an increase in ingestion rates of food by marine fish larvae which enhance the growth and survival as well as the quality of the larvae of *Hippoglossus hippoglossus* (Naas et al., 1992). In addition, the presence of algae in rearing tanks of European sea bass larvae has been shown to increase the digestive enzyme secretion (Cahu et al., 1998). The green algae *Haematococcus pluvialis,* (Yuang and Chen, 2000), *Chlorella zofingiensis* (Bar et al., 1995) and *C. vulgaris* (Gouveia et al., 2003; Gouveia and Rema, 2005) are used as dietary carotenoid sources for fish and shrimp species. The marine algae *Nanofrustulum* (MAP) and *Tetraselmis* (MAP8) used as alternative feed source and noted the better improvement in the body carcass composition on Atlantic Salmon (*Salmo salar*), common carp (*Cyprinus carpi*) and white leg shrimp (*Litopenaeus vannamei*) (Kiron et al., 2012). Also, the fishmeal replacement with *A. platensis*, *C. vulgaris* and *Azolla pinnata* diet improved the antioxidant status in *M. rosenbergii* post larvae (Radhakrishnan et al., 2014).

Spirulina, (*A. platensis*) is a blue-green alga (Cyanobacterium, family Oscillatoriaceae), which is gaining more attention from medical scientists as a nutraceutical and source of potential pharmaceuticals (Kapoor and Mehta, 1994). In 1974, the United Nations World food conference declared *Arthrospira* as "the best for tomorrow" and it has gained popularity in recent years as a food supplement (UNWFC, 1975). Basically, *Arthrospira* consists of 55–70% protein and 5–6% lipid (w/w dried cell). Polyunsaturated fatty acids (PUFAs) constitute 1.5–2% of the total lipid content of this alga. In fact, it is rich in γ-linolenic acid (36% of the total PUFAs), vitamins, essential minerals, antioxidants, carotenoid pigments and enzymes (e.g. lipase) (Belay, 2002; Desai and Sivakami 2007; Demir and Tukel, 2010). Also, *Arthrospira* induces the activity of immune system. It builds up both the cellular and humoral arms of the immune systems and thus improving their ability to function inspite of stresses from environmental toxins and infectious agents (Hayashi et al., 1994; Qureshi et al., 1995), natural biochelated vitamins, especially β-carotene (Seshadri et al., 1991) and an antioxidant enzyme superoxide dismutase (SOD), *S. fusiformis* possess potent antiviral activity (Hayashi et al., 1996), anti cancer effects (Mittal et al., 1999), strengthens immune system (Qureshi et al., 1995, 1996) and significantly reduce the nephrotoxicity (Sharma et al., 2007). In aquaculture, The administration of hot-water extract of *A. platensis* via injection, and immersion routes was reported to enhance the immunity of white shrimp *L. vannamei* and its resistance to *V. alginolyticus* and environmental stress (Tayag et al., 2010; Lin et al., 2010), regulate the antioxidant status in *M. rosenbergii* and *L. vannamei* (Lin et al., 2010; Radhakrishnan et al., 2014), increase the carotenoids and heamato-immunological parameters decrease the stress in *Oncorhynchus mykiss* (Teimouri et al., 2013; Yeganeh et al., 2015) and improve the health status of *Oreochromis niloticus* (Ibrahem and Ibrahim, 2014). Therefore, the biomass of this rich source of nutrients play a vital role in feed and food additives in the agriculture, food, pharmaceuticals, and perfumery industries (Hoseini et al., 2013). The present study was conducted to evaluate the suitable level of replacement of fishmeal with *A. platensis* in diets fed to *M. rosenbergii* postlarvae (PL), and to assess the growth promotion, nutritional and energy utilization parameters, biochemical constituents and digestive capability of *M. rosenbergii* PL.

2. Materials and methods

2.1. Prawns

The postlarvae of *M. rosenbergii* (PL 15) were purchased from the Government prawn hatchery (Thrissur, Kerala) and were safely brought to the laboratory in well-oxygenated plastic bags. They were stocked in a large cement tank ($1.83 \times 1.22 \times 0.91$ m) and were allowed to acclimatize for 2 weeks to the laboratory conditions. During this period, the prawns were fed with boiled egg albumin (egg custard), *Artemia nauplii* and crumble feed (purchased from Rosen fisheries, Thrissur, Kerala) alternatively twice a day. The animals were maintained at natural photoperiods and the room temperature was maintained at $25\,^{\circ}C \pm 2\,^{\circ}C$.The maintenance procedures such as removal of excreta and unused feed, renewal of three fourth of the water were conducted daily. The acclimatization tank was adequately aerated.

2.2. Arthrospira platensis *culture*

The *A. platensis* pure culture was collected from *Spirulina* production research and training centre Kadachanendal, Madurai, Tamil Nadu, India.

2.2.1. *Inoculation of* A. platensis

Inoculation of the microalgae *A. platensis* was done by adding 100 ml of the microalgae mother culture to 900 ml of *Spirulina* medium (Schlosser, 1994) and the cultures were incubated for 15 days at $24 \pm 1\,^{\circ}C$ in a thermo-statically controlled room. The room was illuminated with cool inflorescence lamps (Phillips 40 W, cool daylight 6500 K) at an intensity of 2000 lux in a 12:12 h light dark regime.

Table 1
Proximate composition of experimental feed ingredients.

Ingredients	Proximate composition (%)					
	Protein	Lipid	NFE	Ash	Moisture	Fiber
Fishmeal[a]	64.5	5.1	3.73	21.3	8.3	0.6
Groundnut oil cake[b]	46.9	6.5	35	5.5	8.5	5.4
Soybean meal[b]	43.6	1	30	6.1	8.4	5.5
Wheat bran[b]	10.8	2.5	40.8	3	10.6	9.7
Tapioca flour[b]	2.1	0.2	77.9	2.2	13.5	3.8
Sunflower oil[b]	–	99%	–	–	–	–
Egg albumin[b]	10.6	0.1	0.28	1	87	–
*Vitamin mix[c]	–	–	–	–	–	–
Arthrospira platensis[d]	62.1	4.8	17.3	8.9	6.4	0.5

NFE: nitrogen free extract. *Becosules capsules (each capsule contains): thiamine mononitrate (IP):10 mg; riboflavin (IP): 10 mg; pyridoxine hydrochloride(IP): 3 mg; vitamin B_{12} (as tablets 1:100) (IP): 15 mcg; niacinamide (IP):100 mg; calcium pantothenate (IP): 50 mg; folic acid (IP): 1.5 mg; biotin USP (IP): 100 mcg; ascorbic acid (IP): (150 mg). Source: FAO (2009).

[a] Ingredients purchased from Rosen fisheries, Marathakkara, Thrissur, Kerala, India.
[b] Ingredients purchased from Kannan Departmental Store, Coimbatore, Tamil Nadu, India.
[c] Ingredients purchased from Aruna Medicals, Maruthamalai, Coimbatore, Tamil Nadu, India.
[d] Ingredients purchased from Cultured in Crustacean Biology laboratory, Department of Zoology, Bharathiar University, Coimbatore, Tamil Nadu, India.

2.2.2. *Culture of* A. platensis

The plastic culture troughs were cleaned well, first with bleach and rinsed with distilled water. Then the troughs were allowed to dry under the sun for 8 h. Later, the troughs were used to mix the pure nutrient media (N-8) in 25 L of tap water (Vonshak, 1986). Then the nutrient media was inoculated with 1 L mother culture of *A. platensis*. The troughs were vigourously aerated to provide required quantity of oxygen and to maintain cells and media under suspension. The required concentration of algae had developed after 30 days of inoculation. Then the troughs were kept open completely to the outdoor light exposure. A constant temperature of 25–30 °C was maintained throughout the growth period. Later, the culture was filtered using a nylon fabric cloth which had the mesh size between 40 and 60 μm. The filtered algae were dried in a thermostatic hot air oven to remove the water content. The dried algal sample was powdered in an electric pulvalizer and sieved in a 60 μm mesh.

2.3. Feed ingredients and preparation

The feed grade fishmeal powder was purchased from Rosen fisheries, Thrissur, Kerala, India. The other basal feed ingredients such as soybean meal, groundnut oil cake, wheat bran, binding properties (egg and tapioca flour) and sunflower oil were purchased from a local store, Coimbatore, Tamil Nadu, India. The vitamin mix capsules were purchased from a medical shop, Coimbatore, Tamil Nadu, India (Table 1).

As a first step, the basal ingredients namely fishmeal, sun-dried soy meal, groundnut oil cake and wheat bran were ground separately using a micro pulvelizer and sieved through a 60 μm mesh. The powdered and sieved feed ingredients were weighed out and mixed thoroughly in 5 different ratios for preparing five different diets (1 control fishmeal diet and 4 different diets containing microalgae *A. platensis* at various levels such as 25%, 50%, 75%, and 100%). The blends were cooked in a pressure cooker for 15 min at 95–100 °C and were allowed to cool at room temperature. The cooked blends were mixed with the replacement material (*A. platensis* at various concentrations), vitamin tablets (1%), sunflower oil (2%), egg albumin and tapioca flour (12%) for binding and then 10% of boiled water was added and blending well (5 min) until the mixture achieves a dough consistency. The dough was pelletized in a manual pelletizer fixed with 3 mm diameter and the pellets were collected in aluminium trays. Then the feeds were dried in a thermostatic hot air oven (Microsil INDIA, Universal Lab Product Co., Chennai, India) until the moisture content was reduced below 10%. The dried feed pellets were visually examined for their physical appearance, such as uniformity, colour and fragrance. The chemical composition of the ingredients has been provided in Table 1 and the weight of the ingredients and prepared feed proximate composition were provided in Table 2.

Analysis of crude protein, moisture, lipid and ash content of the formulated feed was performed according to the standard AOAC (1995) procedures. Dry matter was determined by drying the feed at 105 °C until a concordance was obtained. Ash content was determined by burning the feed in a muffle furnace at 525 °C for 12 h. Crude protein level (N × 6.25) was analyzed by the Kjeldahl method after acid digestion (KELPLUS-KES 04 LR with automatic digestion system, Pelican Equipments, Chennai, India). Crude lipid content was analyzed by the Soxhlet method. The formulated feed gross energy was determined by using the Oxygen Bomb Calorimeter (230 VAC; Sl. No. 26036; Advance Research Instrument Company, New Delhi, India).

2.4. Experimental setup

Macrobrachium rosenbergii (PL-30; 15.60 ± 2.90 mm and 2.20 ± 0.39 g kg^{-1}) were used for this experiment. Thirty numbers of PL were introduced in a 30 l plastic tank at the rate of 1 PL/l. In each experimental group maintained as triplicate, each group contains 30 PL × 3 = 90 PL, Totally 90 PL × 5 group = 450 PL were maintained in separate experimental aquarium for feeding experiment. A mild aeration was given continuously throughout the period of experiment. The prawns were fed with fishmeal feed replaced with 25, 50, 75 and 100% of *A. platensis* to groups 1–4, respectively. The prawns in group 5 received the control feed. The experimental groups were fed with their respective diets, twice daily at 6:00 am and 6:00 pm. The daily feed ratio was adjusted to 10% of the body weight of PL throughout the feeding period. The feeding period was scheduled for 90 days. In the experimental setup, animals were maintained at natural photoperiods in the laboratory condition at the room temperature at 25 °C ± 2 °C. On the final day of the experimental period, the prawns from each group were sacrificed and various analysis were performed as follows:10 PL were used for analysis of growth and energy utilization parameters (10 PL × 3 = 30 PL/group × 5= 150 PL); 5 PL for proximate composition analyses (5 PL × 3 = 15 PL/group × 5 = 75 PL); 5 PL for digestive enzyme analyses (5 PL × 3 = 15 PL/group × 5 = 75 PL) and 5 PL for amino acid analyses (5 PL × 3 = 15 PL/group × 5 = 75 PL) Therefore, totally 375 PLs (150 + 75 + 75 + 75 = 375 PL) were sacrificed and utilized for analyses.

Table 2
Dietary formulation and proximate composition of experimental diets (g kg^{-1} DWB).

Ingredients (g kg^{-1})	Control (BI + FM)	Diet-1 (BI + FM75 + R25)	Diet-2 (BI + FM50 + R50)	Diet-3 (BI + FM25 + R75)	Diet- 4 (BI + R100)
Fishmeal	250	187.5	125	62.5	0
Groundnut oil cake	250	250	250	250	250
Soybean meal	250	250	250	250	250
Wheat bran	100	100	100	100	100
Egg albumin	70	70	70	70	70
Tapioca flour	50	50	50	50	50
Sunflower oil	20	20	20	20	20
Vitamin mix	10	10	10	10	10
A. platensis	0	62.5	125	187.5	250
Proximate composition (g kg^{-1})					
Protein	420.20	412.40	413.80	415.10	414.50
Carbohydrate	204.80	211.40	213.00	224.50	231.10
Lipid	137.00	136.30	134.60	132.00	131.10
Ash	118.60	128.00	133.00	141.30	143.60
Moisture	99.30	92.00	86.30	79.30	74.00
Gross energy (kJ g kg^{-1})	11.35	11.64	11.93	12.36	12.50
Leaching and stability of fishmeal replaced formulated diet					
Formulated feed	(%)	4 h	6 h	8 h	12 h
Fishmeal (control)	Leaching	8	10	13	15
	Stability	85	82	80	78
25% *A. platensis*	Leaching	8	11	14	16
	Stability	85	81	78	75
50% *A. platensis*	Leaching	9	12	15	17
	Stability	83	79	76	75
75% *A. platensis*	Leaching	11	13	15	18
	Stability	80	77	75	73
100% *A. platensis*	Leaching	11	14	16	20
	Stability	79	77	70	68

BI: basal ingredients; FM: fishmeal; R: replacement. DWB: dry weight basis; h: hours.

2.5. Determination of growth parameters

At the end of the feeding trial, the growth parameters such as survival rate, weight gain, specific growth rate, condition factor, feed conversion rate and feed conversion efficiency were individually determined by following equations:-

$$\text{Survival Rate (SR, \%)} = \frac{\text{No. of live prawns}}{\text{No of prawns introduced}} \times 100$$

$$\text{Weight Gain } \left(\text{WG, g kg}^{-1}\right) = \text{Finalweight (g)} - \text{Initialweight (g)}$$

$$\text{Specific Growth Rate (SGR, \%)} = \frac{\text{log of Final weight (g)} - \text{log of Initial weight (g)}}{\text{No. of days}} \times 100$$

$$\text{Condition Factor (CF, \%)} = \frac{\text{Final weight (g)}}{\text{Final length}^3\ \text{(cm)}} \times 100$$

$$\text{Feed Conversion Rate (FCR, g)} = \frac{\text{Feed intake (g)}}{\text{Weightgain (g)}}$$

$$\text{Feed conversion efficiency (FCE, \%)} = \frac{\text{Biomass (g)}}{\text{Total feed intake (g)}}$$

2.6. Energy utilization analysis

The energy content of whole prawns, feeds, moult and faeces were measured using an Oxygen Bomb Calorimeter (230 VAC; Sl. No. 26036; Advance Research Instrument Company, New Delhi, India). The energy utilization was calculated using the equation ($C = (P + E) + R + F + U$) derived by Petrusewicz and Macfadyen (1970); where, C is the energy consumed; P is the growth; R is the material lost as heat due to metabolism; F is the energy lost through faeces; U is the energy lost through NH_3 excretion and; E is the energy lost through exuvia. The daily excretion of ammonia by the prawns after feeding was estimated as per the phenol hypochlorite method of Solorzano (1969) and Elliot, (1976).

On the 90th day, the morphometric data, such as the final length and weight of all prawns in triplicate were measured. The prawns were then sacrificed and dried in an hot air oven as described above to estimate the energy content. The food energy consumed was measured as a difference between the energy content of food offered and that of the unconsumed food. The quantity of absorbed food energy was calculated by subtracting F and U from C. Conversion of growth is the sum of energy channeled to somatic growth (P) and exuvia (E). The efficiency of absorption was calculated by relating the food absorbed to the food consumed. Feeding rate (FR), absorption rate (AR), conversion rate (CR) and metabolic rate (MR) were all calculated by dividing the respective amounts of energy by initial live weight of the prawn per unit time in days. Following the estimations of C, F, U, and P, the metabolism (R = respiration, material lost as heat) was calculated by the following equations. The benzoic acid was used as a standard for the energy budget calculations.

$$\text{Feeding Rate } \left(\text{kJ day}^{-1}\right) = \frac{\text{Mean Food Consumption } \left(\text{kJ day}^{-1}\right)}{\text{Initial live weight of the prawn (g)}}$$

$$\text{Mean Absorption } \left(\text{kJ day}^{-1}\right) = \frac{\text{Mean Food Consumption } \left(\text{kJ day}^{-1}\right)}{\text{Mean Food Excreted as faeces } \left(\text{kJ day}^{-1}\right)}$$

$$\text{Absorption Rate } \left(\text{kJ day}^{-1}\right) = \frac{\text{Mean Absorption } \left(\text{kJ day}^{-1}\right)}{\text{Initial live weight of the prawn (g)}}$$

$$\text{Mean Conversion } \left(\text{kJ day}^{-1}\right) = \text{Mean weight gain } \left(\text{kJ day}^{-1}\right) + \text{Mean exuvial weight } \left(\text{kJ day}^{-1}\right)$$

$$\text{Conversion rate } \left(\text{kJ day}^{-1}\right) = \frac{\text{Mean Conversion } \left(\text{kJ day}^{-1}\right)}{\text{Initial live weight of the prawn (g)}}$$

$$\text{NH}_3 \text{ Excretion Rate } \left(\text{kJ day}^{-1}\right) = \frac{\text{Mean NH}_3 \text{ Excretion } \left(\text{kJ day}^{-1}\right)}{\text{Initial live weight of the prawn (g)}}$$

$$\text{Metabolic Rate} = \text{AR } \left(\text{kJ day}^{-1}\right) - \text{CR } \left(\text{kJ day}^{-1}\right) + \text{NH}_3\text{ER } \left(\text{kJ day}^{-1}\right)$$

where, AR = absorption rate; CR = conversion rate: ER = excretion rate.

2.7. Body carcass composition analyses

Concentration of total protein was estimated by the method of Lowry et al. (1951) using bovine serum albumin as a standard. The content of amino acid was estimated by the method of Moore and Stein (1948), which is based on the deamination reaction undergone by the extracted amino acids when they were heated with ninhydrine. The resulting purple colour was measured at 540 nm against a blank, with Leusine as a standard. The carbohydrate was estimated by the method of Roe (1955) using TCA (trichloroacetic acid) extracted sample. Carbohydrates were hydrolysed into simple sugars by diluted HCl in a hot acidic medium. Glucose is dehydrated into hydroxyl-methyl furfural. This compound reacts with anthrone and produces green colored product, which was measured at 630 nm against a blank. Glucose was used as a standard. Total lipid was extracted with chloroform–methanol mixture following the method of Barnes and Blackstock (1973) and estimated by the method of Folch et al. (1957). Lipid reacts with vanillin in a medium of H_2SO_4 and phosphoric acid to form a pink coloured chromogen, which is proportional to the lipid content of the sample and was measured at 540 nm against a blank. Olive oil was used as a standard.

2.8. Estimation of ash and moisture

Known amount of wet tissue sample was taken individually on previously weighed concave glass and they were kept in a desiccator, maintaining 0.5% relative humidity. The tissues were dried in the desiccator till they reached a constant weight. Then the dried materials were transferred individually in silica crucible and kept in a muffle furnace (Universal Lab Product Co., Chennai, India) and heated at 550–600 °C for 4 h. Finally the ash formed was weighed and calculated by the following equation (APHA, 2005).

$$\text{Moisture } (\%) = \frac{\text{Wet weight (g)} - \text{Dry weight (g)}}{\text{Wet weight (g)}} \times 100$$

$$(\%) = \frac{\text{Weight of ash (g)}}{\text{Weight of sample taken (g)}} \times 100$$

2.9. Analyses of digestive enzymes

Activities of digestive enzymes, such as protease, amylase and lipase were assayed on initial and final day of feeding period. For these tests, in each experiment, prawns were sacrificed (5 PLs in each triplicate) and the whole flesh except eye stalk, appendages and exoskeleton was homogenized in ice cold distilled water and centrifuged at 10,000 rpm under 4 °C for 20 min. The supernatant was used as crude enzyme source.

Total protease activity was determined by the casein-hydrolysis method described by Furne et al., 2005. The assay buffer consisted of 0.1 M glycine–NaOH (pH 10.0). The reaction mixture consisted of casein at 1% (w/v) (0.25 ml), buffer (0.25 ml) and supernatant from the homogenates (0.1 ml). The mixture was incubated for1 h at 37 °C. The reaction was stopped by adding 0.6 ml 8% (w/v) trichloroacetic acid solution and kept for 1 h at 2 °C, then centrifuged at 1800 g for 10 min. The supernatant obtained was used to measure the absorbance at 280 nm against blank. For the blank preparation, the supernatant from the homogenates was added at the end of the incubation period, just before adding trichloroacetic acid. Tyrosine solution was used as a standard. One unit of enzyme activity was defined as the amount of enzyme needed to catalyze the formation of 1.0 μmol of tyrosine per min.

Amylase activity was determined by the starch-hydrolysis method of Bernfeld (1955). The reaction mixture consisted of 2% (w/v) starch solution (0.125 ml), 0.1 M citrate–phosphate buffer at pH 7.5 (0.125 ml) and supernatant from the homogenates (0.5 ml). The mixture was incubated for 1 h at 37 °C. The absorbance was measured at 600 nm against a blank. For the blank, the supernatant from the homogenate was added just after the incubation period. Maltose solution was used as standard. One unit of amylase activity was defined as the amount of enzyme that produced 1.0 μmol of maltose per min.

Lipase activity was determined following the method of Furne et al. (2005) by degrading triacylglycerol to free fatty acids. A solution of 1% polyvinyl alcohol (PVA) and 5 ml of 0.1 N HCl in 1L of distilled water was heated to 75–85 °C, cooled, filtered and adjusted to pH 8.0 with 0.1 N NaOH. Virgin olive oil was added to an aliquot of this solution obtaining a substrate concentration of 0.1 M. This mixture was emulsified for 5 min. Parallely, McIlvaine buffer was prepared from 0.1 M citric acid and 0.2 M disodium phosphate. A reaction mixture containing PVA solution-emulsified substrate (1 ml), McIlvaine buffer at pH 8.0 (0.5 ml), and supernatant from the homogenate (0.5 ml) was incubated for 4 h at 37 °C. Afterwards, 3 ml of 1:1 ethanol–acetone solution was added to stop the reaction and break the emulsion. Phenolphthalein in ethanol 1% (w/v) was added to the reaction mixture and titrated with 0.01 M NaOH. For the blanks, the same procedure was followed but boiled enzyme was used. One unit of lipase activity was defined as the hydrolysis of 1.0 microequivalent of fatty acids from triacylglycerols in 1 h at pH 8.00 and 37 °C.

2.10. Amino acid profile analysis

The profiles of amino acids of the experimental prawn PL were performed following the high performance thin layer chromatographic (HPTLC) method of Hess and Sherma (2004). The exoskeleton removed prawns were dried at 60 °C for 48 h in a thermostatic hot air oven, the dried samples were digested with 6 M aqueous hydrochloric acid and dried under vacuum. The powdered sample was dissolved in distilled water and 5 μl of sample was loaded on 8 mm thick pre-coated silica gel 60F254 TLC plate (20 cm × 15 cm) and processed in CAMAG-LINOMAT 5 instrument. The plate was developed in butane–ammonia–pyridine–water (3.9:1:3.4:2.6) mobile phase. The plate was sprayed with ninhydrin reagent prepared in propane-2-ol and dried. The developed plate was documented using photo-documentation chamber (CAMAG-REPROSTAR 3) at UV 254 nm and UV366 nm lights. Finally, the plate was scanned at 500 nm using CAMAG-TLC SCANNER 3. The peak area of the sample was compared with standard amino acids and quantified. Four groups of standard amino acids were also run in parallel. Group-I: proline, serine, asparagine, glutamine and methionine; Group-II: aspartic acid, glutamic acid, alanine, valine and

phenyl alanine; Group-III: lysine, glycine, threonine, isoleucine and tyrosine; Group-IV: arginine, cystine, histidine, leucine and tryptophan.

2.11. Statistical analyses

The results were expressed as Mean ± SD by using Microsoft Excel sheet of Windows 2007. Statistical analyses were carried out by Analysis of Variance (one way ANOVA and subsequently post hoc multiple comparison with DMRT) considered as indicative of significance at $P < 0.05$, as compared to the control group. All calculations were performed using SPSS, version 16.0for Windows (SPSS, Michigan Avenue, Chicago, IL, USA).

3. Results

3.1. Growth parameters and energy utilization

The growth performance of the prawns, *M. rosenbergii* PL fed by the fishmeal replaced with *A. platensis* was provided in Table 3. At end of the feeding period, the final length and weight gain, specific growth rate, survival rate and feed conversion efficiency were found to be significantly higher ($P < 0.05$) in the prawns fed with the fishmeal replaced with 50% *A. platensis*, followed by the 25% and 75% when compared to the control. The feed conversion ratio and condition factor showed a reverse trend, which reveals that the given feed is well utilized by the prawn PL. There is no significant difference in the growth performance of the prawns feds with the fishmeal completely replaced with (100%) *A. platensis*.

The feeding rate, absorption rate and conversion rate were found significantly higher ($P < 0.05$) in prawns fed with the fishmeal replaced with 50% of *A. platensis* followed by the 25% and 75% when compared to the control diet. The metabolic rate and ammonia excretion rate showed no significant difference when compared to the control group (Table 3).

3.2. Body carcass composition and amino acid profile

The biochemical constituents, such as total protein and amino acid profile were significantly ($P < 0.05$) improved in the prawns fed with the fishmeal replaced with 50% of *A. platensis*, followed by 25% and 75% diets when compared to the control group. The carbohydrate content showed a significance ($P < 0.05$) improvement in 50% and 75% *A. platensis* inclusion diets. But, the lipid content shows no significant difference up to 50% inclusion level of *A. platensis* when compared with control (Table 4). Also, the amino acids lysine, arginine, glycine, asparagine, alanine, glutamic acid, cysteine, threonine, isoleucine, tyrosine and phenyl alanine were detected in experimental and control feed fed PL groups (Table 5). In these amino acids, lysine, arginine, alanine, glutamic acid, cysteine, isoleucine and tyrosine were detected in all groups. Glycine was detected in the control group; asparagine and phenyl alanine were detected in 100% fishmeal replacement diet; threonine was detected in 75% fishmeal replacement diet. Overall, the detected essential amino acids were significantly ($P < 0.05$) higher in the prawns fed with fishmeal replaced with 50% of *A. platensis.*

3.3. Digestive enzymes

The activity of the digestive enzymes, protease and amylase were significantly higher in the prawns fed with the diet containing 50% of *A. platensis* followed by the 25% and 75% diets when compared to the control. The lipase activity showed a decreasing trend when the composition of microalgae was increased in the diet. The present digestive enzymes activity were significantly lower in 100% *A. platensis* incorporated feed fed group when compared with control. The statistical analysis (DMRT) revealed that the activities of the enzymes protease and amylase between control and the experimental diets fed prawns were statistically significant ($P < 0.05$) (Table 4).

4. Discussion

The use of alternative protein sources in the feed for the crustacean species is commercially important and it reduces the feed production costs. But, applying the strategies in optimizing the content of the alternative protein sources in the basal feed, favouring the growth of the crustaceans is also very essential to give the best output. In this study, the prawns fed with the feed containing 50% of the microalgae, *A. platensis* have significantly improved in length, weight gain, survival and nutritional utilization. Similarly, Hanel et al., 2007 reported in partial replacement of fishmeal with *A. platensis* meal feed fed Pacific white shrimp, *L. vannamei* had significantly improved in survival and weight gain. Also, they suggesting that partial replacement of fishmeal by *A. platensis* is beneficial for shrimp culture, because it improves the body pigmentation and carcass composition without any synthetic addition. (Jaime et al. (2006) reported, *Chaetoceros muelleri* replaced with 25% *A. platensis* feed fed *Litopenaeus. Schmitti* larvae had significant improvement in survival, length and weight gain in. Consequently, Ghaeni et al. (2011) suggested that *Arthrospira* is a best supplement for shrimp larval culture; it does improve the significant growth and survival of *Peneaus semisulcatus*. Also, Chien and Shiau (2005), reported that *A. pacifica* with various astaxanthin (algae and synthetic) incorporated diets fed kuruma prawns, *Marsupenaeus japonicas* gained significant improvement in growth, survival and stress resistance. Addition of *Arthrospira* in the diet of giant freshwater prawn (*M. rosenbergii*) significantly improved growth, survival and feed utilization regardless of supplementation level in range of 5–20% (Nakagawa and Gomez-Diaz, 1995). Ibrahim (2013) and Dawah et al. (2002) found that the addition of microalgae in diets for fish improved the growth performance of the fish Nile tilapia (*O. niloticus*). EL-Shake (2012) observed, inclusion of algae in diets for fish significantly improved the live body weight and SGR. Similarly, The optimum level of partial replacement of fishmeal with *Arthrospira* accompanied by various levels of vitamin C incorporated diet enhanced the feed consumption, weight gain and specific growth rate in *Carassius auratus* (Vasudhevan and James, 2011) and, in Mekong Giant Catfish *Pangasius paucidens* (Tongsiri et al., 2010). Also, in the present study, the energy utilizations parameters like feeding rate, absorption rate and conversion rate were significantly higher in the group of prawns fed with partial replacement of fishmeal with 50% of *A. platensis* feed when compared with control group. The similar results were reported in total replacement of fishmeal by *Chlorella vulgaris* feed fed *M. rosenbergii* PL had significantly improved the energy utilization (Radhakrishnan et al., 2015). In this study, the 100% *A. platensis* incorporated feed showed very high leaching and poor stability in water, when compared with other diets (Table 2). Also, it is easily soluble and produces turbidity in experimental aquarium system. May be due to the reason of that feed high leaching and low stability decreases the nutritional utilization and survival in 100% *A. platensis* diet fed group. Also, the same trends of results were observed in *M. rosenbergii* PL fed with *C. vulgaris* incorporated feed (Radhakrishnan et al., 2015).

In this study the experimental prawns, which were fed with the fishmeal replaced with 50% of *A. platensis,* had shown significantly higher body chemical composition such as, total protein, amino acid, carbohydrate and lipid than the control and other groups. This was in coordination with the results of Jaime et al. (2006) who reported that *L. Schmitti* larvae, when fed with *A. platensis*

Table 3
Growth, nutritional indices and energy utilization parameters of *M. rosenbergii* PL fed formulated diets for 90 days.

Tests	Parameters		Control	% of Replacement			
				(BI + FM75 + R25)	(BI + FM50 + R50)	(BI + FM25 + R75)	(BI + R100)
Morphometric data	Length (mm)	Initial	15.60 ± 2.90	15.60 ± 2.90	15.60 ± 2.90	15.60 ± 2.90	15.60 ± 2.90
		Final	43.00 ± 3.10^{d}	49.90 ± 4.00^{b}	55.00 ± 0.20^{a}	51.00 ± 1.00^{b}	45.80 ± 1.50^{c}
	Weight (g kg^{-1})	Initial	2.20 ± 0.39	2.20 ± 0.39	2.20 ± 0.39	2.20 ± 0.39	2.20 ± 0.39
		Final	18.30 ± 1.00^{c}	24.90 ± 3.00^{b}	27.80 ± 1.60^{a}	23.80 ± 2.50^{b}	16.40 ± 2.30^{c}
Nutritional indices	Survival rate (%)		86.66 ± 3.50^{b}	90.00 ± 2.00^{ab}	93.00 ± 2.00^{a}	87.00 ± 3.00^{b}	76.00 ± 2.00^{c}
	Weight gain (g kg^{-1})		16.10 ± 0.70^{c}	22.70 ± 1.20^{b}	25.60 ± 1.30^{a}	21.60 ± 1.40^{b}	14.20 ± 1.80^{c}
	Specific growth rate (%)		0.96 ± 0.15^{c}	1.17 ± 0.13^{b}	1.22 ± 0.18^{a}	1.14 ± 0.15^{b}	0.96 ± 0.10^{c}
	Feed intake (%)		66.00 ± 0.12^{d}	83.33 ± 0.35^{b}	86.66 ± 0.45^{a}	80.00 ± 0.39^{c}	62.00 ± 0.42^{e}
	Condition factor (%)		1.90 ± 0.14^{a}	2.00 ± 0.19^{a}	1.67 ± 0.16^{c}	1.79 ± 0.1^{b}	2.06 ± 0.12^{a}
	Feed conversion rate (g)		1.62 ± 0.09^{b}	1.50 ± 0.1^{bc}	1.47 ± 0.17^{b}	1.66 ± 0.11^{bc}	1.70 ± 0.15^{a}
	Feed conversion efficiency (%)		2.05 ± 0.17^{b}	2.21 ± 0.11^{a}	2.37 ± 0.14^{a}	2.20 ± 0.12^{ab}	1.95 ± 0.08^{c}
Energy Utilization (kJ day^{-1})	Feeding rate		1.65 ± 0.25^{d}	1.85 ± 0.15^{b}	2.01 ± 0.13^{a}	1.67 ± 0.05^{c}	1.50 ± 0.16^{e}
	Absorption rate		1.36 ± 0.17^{d}	1.74 ± 0.16^{b}	1.89 ± 0.11^{a}	1.58 ± 0.09^{c}	1.33 ± 0.18^{e}
	Conversion rate		0.88 ± 0.09^{d}	1.21 ± 0.05^{b}	1.33 ± 0.04^{a}	1.11 ± 0.07^{c}	0.88 ± 0.06^{d}
	NH_3 excretion		0.07 ± 0.01^{e}	0.08 ± 0.01^{c}	0.09 ± 0.01^{a}	0.08 ± 0.01^{b}	0.07 ± 0.01^{d}
	Metabolic rate		0.52 ± 0.06^{c}	0.61 ± 0.07^{ab}	0.65 ± 0.04^{a}	0.54 ± 0.08^{c}	0.52 ± 0.01^{c}

Each value is a mean ± SD of three replicates, means with different superscripts letters within each row are statistically significant. (BI: basal ingredients; FM: fishmeal, R: replacement).

Table 4
Concentration of total biochemical constituents and activities of digestive enzymes of *M. rosenbergii* PL fed formulated diets for 90 days.

Assays	Parameters		Control	% of Replacement			
				(BI + FM75 + R25)	(BI + FM50 + R50)	(BI + FM25 + R75)	(BI + R100)
Biochemical constituents (g kg^{-1} WB)	Protein		575.00 ± 13.60^{c}	610.00 ± 11.30^{b}	678.00 ± 13.10^{a}	632.00 ± 15.80^{b}	552.00 ± 18.60^{c}
	Amino acid		311.10 ± 12.40^{d}	390.90 ± 16.30^{b}	420.70 ± 17.00^{a}	353.40 ± 11.40^{c}	272.30 ± 11.10^{e}
	Carbohydrate		213.50 ± 12.30^{c}	238.60 ± 08.90^{b}	260.50 ± 02.70^{a}	255.60 ± 05.90^{a}	226.50 ± 06.90^{bc}
	Lipid		171.60 ± 03.10^{a}	169.00 ± 02.10^{a}	168.50 ± 02.90^{a}	156.20 ± 04.50^{b}	140.90 ± 06.10^{c}
	Ash (%)		12.03 ± 0.15^{d}	12.59 ± 0.22^{c}	13.66 ± 0.24^{a}	13.11 ± 0.19^{b}	12.06 ± 0.31^{d}
	Moisture (%)		75.20 ± 1.20^{a}	73.35 ± 1.11^{ab}	72.39 ± 1.17^{b}	74.86 ± 1.23^{b}	74.58 ± 1.16^{ab}
Digestive enzyme activity (U mg^{-1} protein)	Protease	Initial	0.39 ± 0.09	0.39 ± 0.09	0.39 ± 0.09	0.39 ± 0.09	0.39 ± 0.09
		Final	1.14 ± 0.09^{bc}	1.19 ± 0.08^{ab}	1.27 ± 0.09^{a}	1.22 ± 0.07^{abc}	1.10 ± 0.08^{c}
	Amylase	Initial	0.27 ± 0.12	0.27 ± 0.12	0.27 ± 0.12	0.27 ± 0.12	0.27 ± 0.12
		Final	0.78 ± 0.08^{c}	1.01 ± 0.05^{b}	1.18 ± 0.09^{a}	0.92 ± 0.08^{bc}	0.85 ± 0.07^{c}
	Lipase	Initial	0.28 ± 0.07	0.28 ± 0.07	0.28 ± 0.07	0.28 ± 0.07	0.28 ± 0.07
		Final	0.83 ± 0.08^{a}	0.76 ± 0.07^{ab}	0.70 ± 0.11^{ab}	0.67 ± 0.06^{b}	0.61 ± 0.03^{c}

Each value is a Mean ± SD of three replicates, means with different superscripts letters within each row are statistically significant. WB: wet basis; DWB: dry weight basis. BI: basal ingredients; FM: fishmeal; R: replacement.

Table 5
Concentration of essential amino acids (g kg^{-1}) in formulated diet fed *M. rosenbergii* PL.

Assigned substance	Control	% of Replacement			
		(BI + FM75 + R25)	(BI + FM50 + R50)	(BI + FM25 + R75)	(BI + R100)
Lysine[a]	9.30 ± 0.21^{a}	8.40 ± 0.15^{b}	9.20 ± 0.10^{a}	8.20 ± 0.14^{b}	7.60 ± 0.17^{c}
Arginine[a]	17.20 ± 0.11^{b}	16.40 ± 0.09^{c}	18.40 ± 0.05^{a}	14.60 ± 0.20^{d}	13.60 ± 0.22^{e}
Glycine[a]	2.60 ± 0.27	–	–	–	–
Asparagine[a]	–	–	–	–	4.60 ± 0.19
Alanine[b]	7.00 ± 0.05^{d}	8.60 ± 0.10^{b}	8.60 ± 0.08^{a}	8.20 ± 0.07^{c}	5.60 ± 0.20^{e}
Glutamic acid[b]	7.00 ± 0.07^{c}	8.60 ± 0.04^{a}	8.60 ± 0.06^{a}	8.20 ± 0.10^{b}	5.60 ± 0.15^{d}
Cysteine[b]	14.40 ± 0.18^{c}	17.80 ± 0.13^{a}	17.80 ± 0.15^{a}	16.80 ± 0.11^{b}	11.60 ± 0.12^{d}
Threonine[a]	–	–	–	3.60 ± 0.22	–
Isoleucine[a]	9.60 ± 0.11^{b}	9.60 ± 0.13^{b}	12.60 ± 0.12^{a}	4.20 ± 0.19^{c}	3.40 ± 0.15^{d}
Tyrosine[a]	13.80 ± 0.25^{d}	20.40 ± 0.20^{c}	26.40 ± 0.18^{a}	23.60 ± 0.15^{b}	–
Phenyl alanine[a]	–	–	–	–	5.30 ± 0.23

Each value is a mean ± SD of three replicates, means with different superscripts letters within each row are statistically significant. BI: basal ingredients; FM: fishmeal; R: replacement.

[a] Essential amino acid.
[b] Non essential amino acid.

incorporated diet, had significantly gained the growth and body proximate composition. Similarly, Bhavan et al. (2010) noted in *M. rosenbergii* PL had significantly improved the body biochemical composition when fed with a meal containing *Artemia* nauplii enriched with *Arthrospira*. Shuli and Baoqing (1992) used *C. muelleri* and *Arthrospira sp.* to feed the larvae *of P. orientalis* and suggest that fishmeal and *Arthrospira* are the most suitable proteins for inclusion in their practical diets. In addition, it has been reported that, the replacement of artificial diet for postlarvae of abalone, *Haliotis discus discus* with *Arthrospira sp.*, gives a good growth performance (Stott et al., 2004). Abdulrahman and Hamad Ameen (2014), found that fishmeal replacement with *Arthrospira,* up to 20%, significantly

improves the growth, nutritional utilization and body carcass composition in *Cyprinus carpio*. It has significantly affected the protein and lipid contents in whole-fish body. Also, Hernandez et al. (2012); reported the possibility of using a mixture of 75% *Arthrospira* powder and 25% soybean meal to replace 100% fishmeal in diets for juvenile rainbow trout (*O. mykiss*). The palatability of *Arthrospira* meal in fish diets can improve the feed consumption which directly increases the growth rate and body carcass composition. It may be due to the reason of, *Arthrospira* contains a high level of protein (53%) and all the essential amino acids, vitamins and fatty acids, etc., (James and Sampath, 2004; James et al., 2006). On the other hand, changes in the protein and lipid contents in the fish body could be linked to changes in their synthesis, deposition rate in muscle and different growth rates (Kyewalyanga, 2003; Abdel-Tawwab et al., 2006; Karakatsouli, 2012). In the present study, the formulated feed with 50% fishmeal replaced by 50% of *A. platensis,* has significantly affected the prawn PL, which showed a significant improvement in body carcass composition. The lipid level was found decreased in the prawns, fed with the diet containing 75% and 100% of *A. platensis*. This may be due to the lipid content which was lower in *A. platensis* instead with the fishmeal.

Moreover, we found that the activities of digestive enzymes such as, protease, amylase and lipase, were found significantly higher in the prawns fed with 50% of *A. platensis* when compared to the control group. Similarly, Anand et al., 2013 reported the similar increment of digestive enzymes, that was noted in *Penaeus monodan* fed with peripyton supplemented feed. They also suggested that the diet containing microalgae, stimulated the growth of beneficial microbes in the digestive tract and the production of endogenous digestive enzymes by the shrimp. The presence of microalgae and its components, even at low concentration in the gut, can trigger the production of digestive enzymes (Reitan et al., 1993; Sheele, 1993). It has been demonstrated that the consumption of the biota present in the farming systems improves the activity of digestive enzymes and physiological status of farmed aquatic animals (Brito et al., 2004). Le Vay et al. (1993), noticed that microalgae enhances trypsin activity in *Pariibacus japonicas*, as it contain large amounts of free amino acids. However, it is important to make distinction between enzymes produced endogenously by the shrimp and exogenous enzymes synthesized by resident gut flora (Harris, 1993). Nandeesha et al. (1994, 1998) reported that the diets with 25%, 50%, 75% and 100% level of fishmeal replaced with *A. platensis*, significantly improved the digestive activity in catla, rohu and common carp mixed culture. Mustafa and Nagakawa (1995) suggested that the algae contribute to an increase in protein assimilation and feed utilization. Also it has been proved that, the diet included with 50% of *A. platensis*, significantly improved the protein digestibility in the common carp *C. carpio* (Umesh et al., 1994).

In the present study, the significant level of amino acids was found in experimental feed fed *M. rosenbergii* PL compared with the control group. The similar results were found in *Arthrospira* enriched with *Artemia* fed *M. rosenbergii* PL (Bhavan et al., 2010). Carcea et al., 2015; reported that *Arthrospira* (dihe) is a good raw material for nutritional purpose and it is rich in more than 15 amino acids like thrionine, serine, glutamine, proline, glycine, alanine, cystine, valine, methionine, isolucine, leucine, tyrosine, phenylealanine, histidine, lysine and arginine. The incorporation of *Arthrospira* powder improve the growth of the juvenile trout, it's may be that *Arthrospira* contains indispensable amino acids for most fish species, as reported (Olvera-Novoa et al., 1998; NRC, 2011). This agrees with studies of this microalgae as a substitute for fishmeal in diets for silver sea bream (El-Sayed, 1994), tilapia (Olvera-Novoa et al., 1998), and sturgeon (Palmegiano et al., 2005). Also, in the present study, the *Arthrospira* supplemented feed fed prawns gained the significant level of amino acid composition in body tissue.

As a feed additive, dried algae improve the growth, feed efficiency, carcass quality, and physiological resistance towards stress and disease in several species of fish (Mustafa and Nakagawa, 1995). In addition, it has been proved that it is an effective immune-modulator (Takeuchi et al., 2002). Several studies have been conducted using dried *Arthrospira* as a feed supplement (Chow and Woo, 1990; Watanabe et al., 1990) and *A. platensis* has been considered for the partial substitution of microalgae in the feed for white shrimp *L. schmitti* protozoans (Jaime et al., 2004). Similarly, El-Sayed (1994) used the fishmeal replaced with *Arthrospira* to feed silver sea bream (*Rhabdosargus sarba*). Hernandez et al. (2012) suggested that the possibility of using a mixture of 75% *Arthrospira* powder and 25% soybean meal to replace 100% fishmeal in aquaculture had shown beneficial towards growth performance, the dissolved phosphorus and phosphorus in faeces, and lysozyme activity.

5. Conclusion

Our results indicate that, *A. platensis* can be used for the partial replacement of fishmeal protein in aqua feeds. It was revealed that the level of 50% *A. platensis* replacement in the diet can be significantly utilized by *M. rosenbergii* PL for their growth. At 75% inclusion level, growth and nutritional utilization was reduced significantly, and it has been sharply reduced at 100% inclusion level. Additionally, such a high inclusion level, showed a decreasing trend in nutritional indices, survival, body carcass composition and digestive enzyme activity. The prawns fed with the diet containing an equal composition of fishmeal and *A. platensis* (50%) showed a significant improvement in feed utilization, survival and their carcass composition. However, the prawns fed with the diet containing microalgae more than 50% showed an increasing level of leaching and decreasing trend of stability when compared with other diets. It may affect the feed utilization, consumption and growth of experimental group prawn PL, when they were fed with the diet in which the fishmeal has been replaced above 50%. Therefore, our study suggests that, 50% replacement of fishmeal by *A. platensis* in the diet will be an ideal solution to get a better feed utilization, growth output and a sustainable culture of *M. rosenbergii* PL.

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